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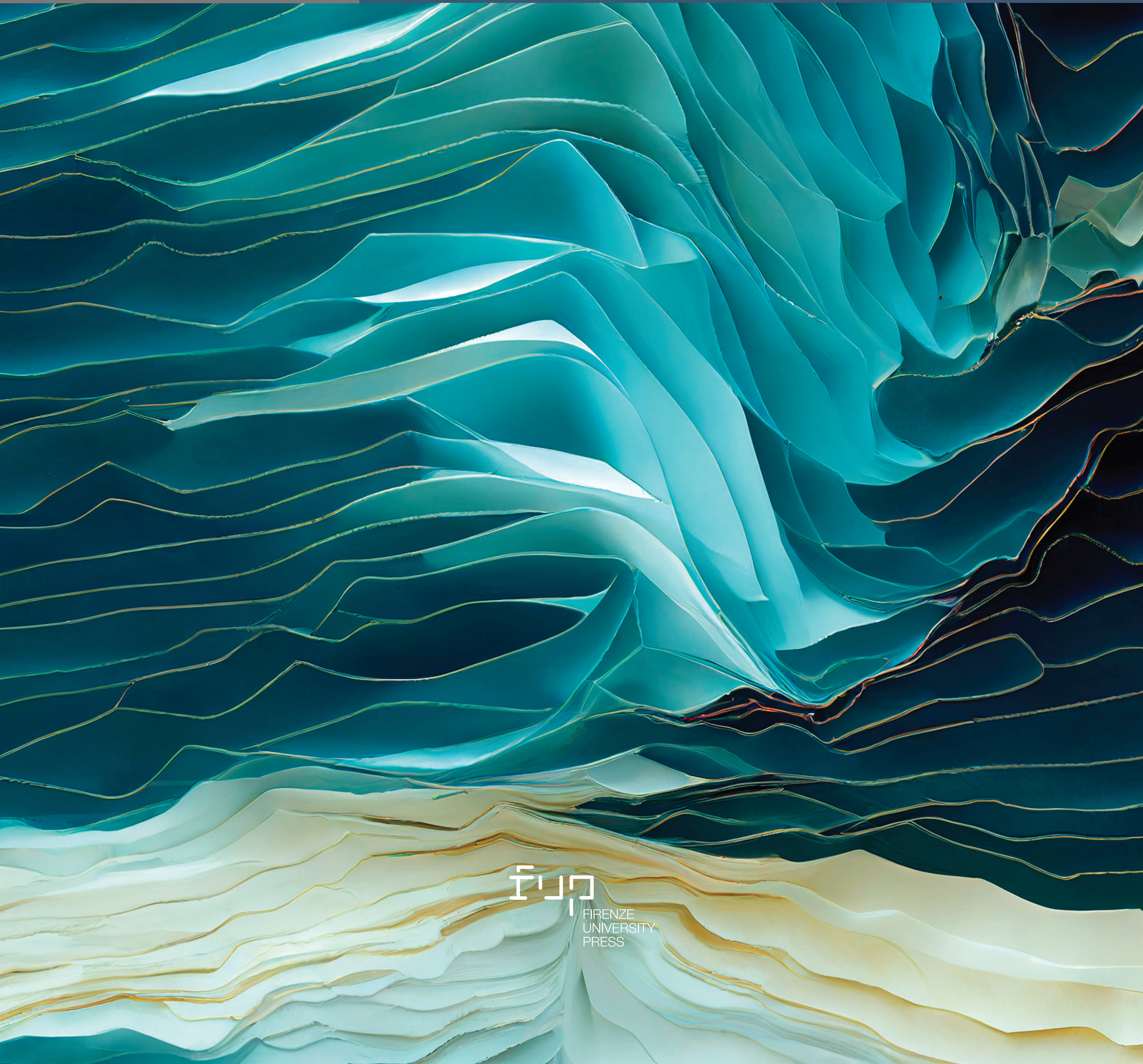

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Editorial

Mirror, mirror on the wall...

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While seeking for newer and wider horizons in science and technology, some scientists appear to be challenging common life in a way that - in time - may eventually destroy life on Earth and therefore mankind. For the moment this risk is still far away, but it may become one of the worse scenarios on the planet.

To introduce this discussion, we need a mirror.

"Mirror, mirror on the wall, who's the fairest of them all?" was the resounding question that the Evil Queen uttered when consulting her magic mirror to be reassured that she was still the most beautiful lady in the land. (At least until Snow White entered the scene).

But "our" mirror will be used in another way.

We know that a chiral molecule can exist in two different forms, which are perfectly specular (here is the mirror) but they cannot be superposed. Hands and feet are the most common example we teach chirality to our students. In fact in Greek χειρ means hand.

Most of biomolecules such as amino acids and sugars possess at least a chiral center and produce R and S enantiomers. And what is particularly intriguing is that their biological activity is totally different. So, most of natural proteins are composed of L-amino, although some contain D-amino acids or after some time this homochirality is lost in aged organisms.[1]

These weird D-proteins pave the way to unique applications in molecular biology, structural biology, bioactive compound discovery, and pharmacology. In fact according to Lander "D-enantiomers of protein drug targets can be used in mirror-image phage display allowing discovery of non-proteolytic D-peptide ligands".[2] Similarly, the biologically active form of ascorbic acid is the levo isomer (vitamin C), while its epimer D-isoascorbic acid retains only 5% of the biological activity, perhaps because of different hydration properties.[3]

On the other hand, Nature builds up polysaccharides such as amylose, glycogen and cellulose from D-sugars. While it is still possible to synthesize L-glucose, it does not have any function in living organisms as a source of energy because it cannot be phosphorilated by hexokinase, however we know since 1979 that the plant pathogenic bacterium *Burkholderia caryophill* can oxidize L-glucose.[4] Thus, natural exceptions exist but they are extremely confined to tiny environments and they are not healthy.

DNA and RNA homochirality, which is essential for life, is based on the presence of D-nucleotides in these macromolecules.[5]

Uppalapati and coworkers found that a D-protein is much more stable in different animals (including humans). In particular they synthesized a variant of a D-protein antagonist of natural vascular endothelial growth factor A (VEGF-A) with an extra receptor blocking capability.[6] Now, while the L-protein triggers a massive immune response, the D-protein was found to be nonimmunogenic in mice. Hence, its potential use as an alternative to therapeutic antibodies.

With these premises, mirror-molecules do have some potential interesting applications, for example in pharmacology due to their specific pharmacokinetic features.

However, a very recent paper, published in December 2024 and authored by 38 experts is not only an informative review on the business but also a superb alert of what may occur if this kind of biotechnology research is not guided, controlled and monitored by appropriate authorities.[7] An issue that lately has become particularly pressing. The authors belong to quite different fields, particularly synthetic biology, human, animal, and plant physiology, immunology, microbial ecology,

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evolutionary biology, planetary life detection, biosecurity global health, and policy-making groups.

In that paper the authors state:

> *Driven by curiosity and plausible applications, some researchers had begun work toward creating lifeforms composed entirely of mirror-image biological molecules.*

Our immune defenses are strongly based on chiral recognition, but in the presence of a mirror bacterium our "normal" immune recognition would be significantly weakened if not completely useless. And

> *even partial impairment of either innate or adaptive immunity can leave patients vulnerable to bacterial infection. Similar evidence is seen in a wide variety of animal and plant immune systems.*[8]

The number of potential harms that may derive from such laboratory activities are scary indeed. Accidental or deliberate misuse, leakage of contaminated material, spillover, uncontrolled infections and particularly the lack of effective and timely countermeasures are part of the nightmare we may have to confront soon or later. Particularly because we have almost no idea of what would happen, the relevant literature is extremely poor and once a catastrophic accident occurs, we have no history or memory to refer to.

Apparently at this moment we do not even have a clear idea of what we are really looking for and for sure the real serious risks that we may be facing in the future are quite uncertain.

The perspective is definitely challenging. It seems that the collaboration of different players at different level is absolutely necessary. But this is another story and, given the current circumstances, more unrealistic that Snow White and the Seven Dwarfs.

1 N. Fujii, T. Takata, N. Fujii, K. Aki, H. Sakaue, D-Amino acids in protein: The mirror of life as a molecular index of aging. *Biochim. Biophys. Acta* **2018**, *1866*, 840-847. https://doi.org/10.1016/j.bbapap.2018.03.001

2 A.J. Lander, Y. Jin, L.Y.P. Luk, D-Peptide and D-Protein Technology: Recent Advances, Challenges and Opportunities. *ChemBioChem* **2023**, *24*, e202200537. https://doi.org/10.1002/cbic.202200537

3 C. Dolle, P. Magrone, S. Riva, M. Ambrosi, E. Fratini, N. Peruzzi, Symmetric and Asymmetric Bolaamphiphiles from Ascorbic Acid, *J. Phys. Chem. B* **2011**, *115*, 11638-11649. https://doi.org/10.1021/jp204920y.

4 K.I. Sasajima, A.J. Sinskey, Oxydation of L-glucose by a Pseudomonad, *Biochim. Biophys. Acta* **1979**, *571*, 120-126. https://doi.org/10.1016/0005-2744(79)90232-8

5 Y. Chen. W. Ma, The origin of biological homochirality along with the origin of life. *PLoS Comput. Biol.* **2020**, *16*, e1007592. https://doi.org/10.1371/journal.pcbi.1007592

6 M. Uppalapati, D.J. Lee, et al. A Potent d-Protein Antagonist of VEGF-A is Nonimmunogenic, Metabolically Stable, and Longer-Circulating in Vivo, *ACS Chem. Biol.* **2016**, *11*, 1058-1065.https://doi.org/10.1021/ acschembio.5b01006

7 K.P. Adamala, D. Agashe, et al. Confronting risks of mirror life. *Science* **2024**, *386*, 1351-1353. https://doi.org/ 10.1126/science.ads9158

8 K. P. Adamala et al., "Technical report on mirror bacteria: Feasibility and risks" (Stanford Digital Repository, 2024); https://doi.org/10.25740/cv716pj4036 (last accessed, Feb 11, 2025).

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Research Article

PFAS Toxicity and Female Reproductive Health: A Review of the Evidence and Current State of Knowledge

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Abstract. Per- and polyfluoroalkyl substances (PFAS) constitute a class of synthetic chemicals extensively utilized in various consumer products and industrial applications. Characterized by their remarkable persistence, PFAS chemicals resist degradation, perpetuating their presence in the environment for an indefinite period. Human exposure to PFAS occurs through multiple pathways, including contaminated food, water, air, and products, resulting in widespread detection in biological matrices such as blood and urine. Exposure to PFAS has also been linked to adverse reproductive outcomes, yet the impact on female reproductive health remains poorly understood. This review synthesizes recent findings on the PFAS-female reproductive health connection, highlighting the effects on ovarian function, hormone regulation, and pregnancy outcomes. The evidence suggests that PFAS exposure is associated with reduced fertility, increased risk of miscarriage, and altered menstrual cycle dynamics. The review also explores the underlying mechanisms, including endocrine disruption and oxidative stress. The implications of these findings on female reproductive health are discussed, emphasizing the need for further research and policy changes to mitigate PFAS exposure and protect female reproductive physiology.

Keywords: PFAS; Female fertility; Pregnancy; Endocrine disruption; Menstrual cycle

INTRODUCTION

Per- and polyfluoroalkyl substances (PFAS), also known as forever chemicals are a large family of synthetic organofluorine chemicals with diverse applications across various industries. First discovered in the 1930s, they entered commercial production on a large scale in the 1940s (OECD, 2021; ITRC, 2020). Comprising over 4,730 identified chemicals (Zhang et al., 2022), PFAS are characterized by the presence of at least one fluorine atom bonded to carbon atoms, forming a robust carbon-fluorine bond. This unique property renders PFAS versatile for various applications, including electronics, aerospace, construction, and consumer products (Gaines, 2022). However, it also confers exceptional resistance to degradation in the environment, requiring temperatures above 1,100 °C for destruction (OECD, 2021). PFAS can exist in various ionic states, including cations, anions, and zwitterions, which determine their electrical charge, physical, and chemical properties. These properties control their fate and transport in the environment. The carbon-fluorine groups in PFAS can be linked to various chemical groups, making them useful for a wide range of applications. However, the widespread use of PFAS has led to various environmental release pathways, including wear and tear of consumer products, fugitive emissions, manufacturing waste streams, disposal of PFAS-containing materials, and spills. This has resulted in the aggregation of PFAS in wastewater treatment plant effluent and sludges, posing a threat to wildlife and human health. Exposure to PFAS can occur through direct involvement in the manufacturing process, professional use of materials containing PFAS, use of consumer products containing PFAS, and exposure to environmental releases (Brunn et al., 2023; De Silva et al., 2021). The persistence and mobility of PFAS in the environment, bioaccumulation in humans and wildlife, and potential toxicity raise significant concerns, highlighting the need for policy changes and mitigation strategies to protect human and environmental health.

The persistence of PFAS is a notable property, characterized by their resistance to degradation under natural conditions, posing a significant threat to the environment as long-lasting pollutants (Cousins et al., 2020). The half-life of a chemical, defined as the time required for its concentration to decrease by half in a specific medium, is a criterion for persistence. According to EU chemical regulation, chemicals with a half-life exceeding 60 days in water and 180 days in sediments or soil are considered very persistent (Cousins et al., 2020). Although the half-life of most PFAS is unknown, estimates suggest that some PFAS polymers can persist for over 1,000 years in soil, while non-polymeric PFAS can persist for over 40 years in water (Xu et al., 2020). Even when PFAS degrade, their degradation products often include other persistent PFAS (Bridger, 2023), earning them the name forever chemicals. PFAS are also bioaccumulative, building up in the human body and wildlife by binding to proteins in blood, such as albumin and fatty acid binding protein, rather than accumulating in fatty tissues like most bioaccumulative chemicals (Forsthuber et al., 2020; Lai et al., 2020; Khazaee, 2021). This bioaccumulation occurs through absorption, without excretion, leading to increased concentrations higher up the food chain.

The processing of PFAS varies across organisms, with differences observed between sexes, species, and structures. In humans, long-chain PFAS are eliminated slowly, taking years (e.g., PFHxS has a half-life in blood of up to 8.5 years), and tend to accumulate in protein-rich compartments like blood, liver, kidney, and bones (Cousins, 2022). In contrast, short-chain PFAS are eliminated more quickly (e.g., PFBS has a half-life in blood of 26 days), accumulating in different organs and tissues like lungs, kidneys, and the brain. While the behavior of PFAS in wildlife is not well understood, reports indicate bioaccumulation in water birds, wild boars, polar bears, and dolphins (Sudarshan et al., 2022; Fenton, 2021). Due to their high water solubility, PFAS are highly mobile in the environment, rapidly migrating through soil and leaching into groundwater (McMahon et al., 2022). Additionally, they can easily pass through conventional drinking water treatment facilities, contaminating drinking water. Furthermore, short-chain PFAS have been shown to migrate from soil to plants, accumulating in edible parts of fruits and vegetables (Xu et al., 2022). This highlights the potential for PFAS to enter the food chain and have far-reaching environmental and health impacts.

Exposure to PFAS has been linked to various adverse health effects, including immune system dysfunction, thyroid impairment, neurodevelopmental issues, cardiovascular disease, and reproductive problems (Mueller et al., 2020; Zheng et al., 2024). The human endocrine system, which regulates fertility hormones such as estrogen, Izumo1, Juno, and sperm receptors, is crucial for conception and ovulation. However, interference by PFAS and other endocrine-disrupting chemicals (EDCs) could lead to reproductive health challenges like hormone imbalance and infertility (Kim et al., 2019). While various natural and environmental factors influence reproductive success, evidence suggests that environmental chemicals like EDCs decrease reproductive success in many species, including vertebrates and invertebrates (Kim et al., 2020; Xie et al., 2021; Rickard et al., 2022). EDCs disrupt reproductive development by altering the function of endocrine system components. The risks posed by EDCs to wildlife and humans are a widespread concern, and global efforts are underway to develop advanced assessment methodologies (Barton-Maclaren et al., 2022).

METHODS

Literature Search

A comprehensive literature search was conducted using major databases, including PubMed, Scopus, Web of Science, and Google Scholar. The search strategy employed a combination of keywords and Medical Subject Headings (MeSH) terms related to per- and polyfluoroalkyl substances (PFAS), reproductive health, endocrine disruption, and female reproductive disorders. The search terms used included "PFAS," "reproductive health," "menstrual cycle irregularities," "polycystic ovary syndrome (PCOS)," "endometriosis," "pregnancy outcomes," and "female reproductive tract disorders." The search was limited to English-language articles published between 2007 and 2024.

Paper Selection

Studies were selected based on their relevance to the research question, study design, and publication date. Two independent reviewers screened the titles and abstracts of retrieved articles, and potentially relevant studies were selected for full-text review.

Inclusion and Exclusion Criteria

Studies were included if they:

1. Investigated the association between PFAS exposure and female reproductive health outcomes.
2. Examined the effects of PFAS on hormone regulation and endocrine function.
3. Were published in English.
4. Were peer-reviewed articles.
5. Were published between the year 2000 and 2024.

Studies were excluded if they:

1. Were conference proceedings, or editorials.
2. Did not focus on human subjects.
3. Did not examine PFAS exposure.
4. Were not relevant to female reproductive health.
5. Were published before the year 2000.

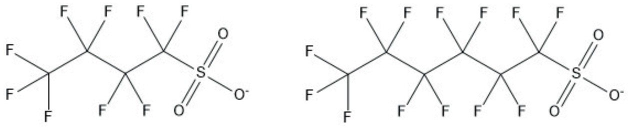
Study Selection

A total of 461 studies were identified through the literature search. After removing duplicates and applying the inclusion and exclusion criteria, 227 studies remained. Two independent reviewers screened the titles and abstracts, and 148 studies were selected for full-text review. Following full-text review, 106 studies met the final inclusion criteria.

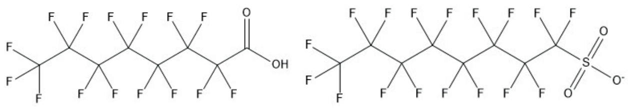
SIZE-TOXICITY RELATIONSHIP OF PFAS MOLECULES

Per- and polyfluoroalkyl substances (PFAS) are distinct from other fluorinated compounds, such as Freon (chlorofluorocarbons) and Teflon (polytetrafluoroethylene, PTFE), due to their unique chemical structure. PFAS are characterized by a hydrophobic fluorinated chain linked to diverse functional groups, exhibiting variability in chain length and chemical functionality. In contrast, Freon features chlorine and fluorine bonded to carbon, while Teflon comprises polymerized tetrafluoroethylene monomers (-CF2-CF2-), with both possessing fixed molecular structures. The sizes and structures of PFAS molecules vary widely, ranging from short-chain compounds with seven or less carbon atoms to long-chain compounds with eight or more carbon atoms (Peritore et al., 2023). This variability in molecular size and structure contributes to their differing properties and potential environmental and health impacts.

Short-chain PFAS (Figure 1), such as PFBS (perfluorobutane sulfonate) and PFHxS (perfluorohexane sulfonate), have smaller molecular sizes and are more water-soluble than their long-chain counterparts (Grgas et al., 2023). While considered less toxic, short-chain PFAS have still been linked to environmental contamination and human health concerns. However, it is the long-chain PFAS that have created the most significant problems due to their persistence, bioaccumulation, and toxicity. Long-chain PFAS, such as PFOA (perfluorooctanoic acid) and PFOS (perfluorooctane sulfonate), have larger molecular sizes and are more persistent in the environment (Grgas et al., 2023). These compounds have been widely used in industrial applications and have raised concerns due to their potential health impacts, including cancer, reproductive issues, and immunological problems.



Perfluorobutane sulfonate **Perfluorohexane sulfonate**



Perfluorooctanoic acid **Perfluorooctane sulfonate**

Figure 1: Some short-chain and long-chain PFAS

SOURCES AND PATHWAYS OF PFAS EMISSION

PFAS are ubiquitous in everyday products, having been used in a wide range of applications since the 1940s due to their unique carbon-fluorine bond properties (Glüge et al., 2021). This bond structure makes PFAS the most persistent chemicals known today (Kurwadkar et al., 2022). As a result, people and wildlife are exposed to hundreds of PFAS simultaneously through various environmental routes. The major routes of exposure to PFAS include:

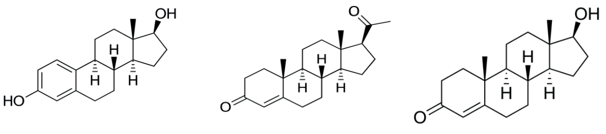
- Oil and gas refineries, where PFAS are employed as viscosity reducers in crude oil processing (ITRC, 2020; Brunn et al., 2023; NRDC, 2021; Neuwald et al., 2020; USEPA, 2022).
- Household products, including food coverings, cables, coated woods, solar panels, textiles, leather, and glasses (Korzeniowski, 2022; Fiedler et al., 2020).
- Firefighting facilities and training areas where fluorine-containing firefighting materials are stored, used, or released, such as those utilizing firefighting foams to extinguish liquid fires (Mazumder et al., 2023).
- Waste management facilities and areas of bio-solids production and application, with significant impacts associated with industrial wastewater discharges (Saliu and Sauvé, 2024).
- Certain pesticides contain PFAS as co-formulants, specifically as wetting agents, which can lead to PFAS contamination in agricultural and environmental settings (PAN Europe, 2023).
- Personal care products, such as skin and hair care cosmetics that contain PFAS for water-repellent properties (Whitehead et al., 2021).
- PFAS used in surface treatment for paper and printed products for food packaging (Straková et al., 2021).

These various exposure routes highlight the widespread presence and persistence of PFAS in our environment.

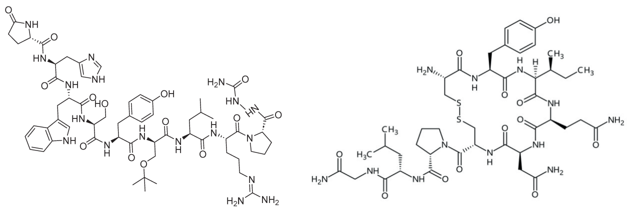
PFAS have been detected in rivers unrelated to manufacturing sites, indicating emissions from widespread sources (Pan et al., 2018). These diffuse sources include consumer products like: cleaning products, food packaging, hydraulic fluids, textiles, cabling and wiring, metal finishing and plating. These products release PFAS through washing and wear and tear (Commission for Environmental Cooperation, 2017). Additional sources include coatings like anti-graffiti paints (Zhang et al., 2021). From 1970, DuPont utilized the telomerization process to produce eight-carbon chain perfluoro compounds, specifically perfluorooctanoic acid (PFOA) and perfluorooctane sulfonate (PFOS), by combining two key reactants, an organoiodine compound, known as the telogen, which served as the initiator molecule, and tetrafluoroethylene (TFE), a highly reactive gas referred to as the taxogen. The telomerization reaction involved the sequential addition of TFE units to the telogen, resulting in a growing polymer chain that ultimately yielded the desired eight-carbon chain perfluoro compounds, possessing exceptional properties such as non-stick surfaces, water and oil repellency, thermal stability, and chemical resistance (Travis, 2024). Fluorotelomer alcohols, such as 6:2 FTOH and 8:2 FTOH, are volatile and have been detected in high concentrations in indoor air at stores selling sports, textiles, and carpets materials (Schlummer, 2013). This is a significant source of human exposure to PFAS (Huang et al., 2019). The stability of the C-F bond poses a problem for waste disposal and incineration at very high temperatures is the only way to ensure PFAS destruction (Berg et al., 2022).

ENDOCRINE DISRUPTING CHEMICALS (EDC)

The endocrine system is a complex network of tissues and glands that produce and secrete hormones, which play a crucial role in regulating various bodily functions, including growth and development, sexual function, metabolism, reproduction, mood, and sleep (Endocrine Society, 2023). This intricate system comprises specialized endocrine glands composed of tissues and cells that have a secondary endocrine function, secreting a diverse range of hormones. Hormones have specific target cells, characterized by the presence of receptors that bind to the hormone, either on the cell surface or intracellularly. The hormone-receptor interaction initiates a cascade of biochemical reactions within the target cell, ultimately modifying its function or activity. The reproductive hormones (Figure 2) produced primarily in the ovaries (females) and testes (males), play a crucial role in regulating reproductive cycles (Atiya et al., 2021).



Estrogen **Progesterone** **Testosterone**



Gonadotropin releasing hormone **Oxytosin**

Figure 2: Human reproductive hormones

Hormones exert their effects on target cells through binding to specific receptors, which are cell proteins. The binding of a hormone to its corresponding receptor forms a hormone-receptor complex, triggering various responses within the target cells. Certain compounds can bind to hormone receptors, blocking them and leading to adverse effects in non-target organisms through an endocrine mode of action. These substances, known as endocrine disrupting chemicals (EDCs), are exogenous compounds that can disrupt the normal functioning of the endocrine system, potentially inducing adverse health effects (Ercan et al., 2022). EDCs mimic endogenous hormones, such as thyroid hormones, estrogens, and androgens by exhibiting similar activities or structures, influencing their metabolism and synthesis. These chemicals can impact various bodily systems by altering hormone regulation or normal function. Out of the estimated 85,000 synthetic chemicals in existence, a significant subset of over 1,000 compounds have been recognized as potential endocrine disruptors (Endocrine Society, 2023). The most well-studied EDCs include bisphenol A (BPA), phthalates, atrazine, polybrominated diphenyl ethers (PBDE), polychlorinated biphenyls (PCBs), perchlorate, and per- and polyfluoroalkyl substances (PFAS) (Nian et al., 2020; Lee et al., 2021). PFAS are recognized endocrine disrupting chemicals that have been linked to pervasive human health and environmental concerns. Research has demonstrated that PFAS could interfere with hormone regulation, exhibiting both agonist and antagonist properties that could disrupt endogenous hormone activity, thereby impairing reproductive, thyroid, and immune system function. Notably, their persistence, bioaccumulation, and widespread contamination of water, food, and consumer products necessitate rigorous scientific investigation and scrutiny, underscoring the importance of continued research into the implications of PFAS exposure.

PFAS AND THEIR ASSOCIATION WITH DISORDERS OF THE FEMALE REPRODUCTIVE SYSTEM

The female reproductive system is governed by a complex interplay of endocrine mechanisms, susceptible to interference by per- and polyfluoroalkyl substances (Figure 3). PFAS like PFOA, PFOS, and PFHxS have been associated with imbalances at the hormonal, metabolic, and reproductive systems levels, indicating their potential to disrupt normal hormone regulation and endocrine function (Fenton et al., 2021). Some of these PFAS are thought to disrupt crucial reproductive hormones, including progesterone, estradiol (E2), testosterone, follicle-stimulating hormone (FSH), and luteinizing hormone (LH) (Lee et al., 2021). Moreover, there are suggestions that PFOS can alter important targets of the reproductive system, including thyroid hormone, human chorionic gonadotropin (hCG) levels, and prolactin, directly and indirectly by affecting reproductive tissues like breast and placental tissue (Pierozan and Karlsson, 2018; Bangma et al., 2020).

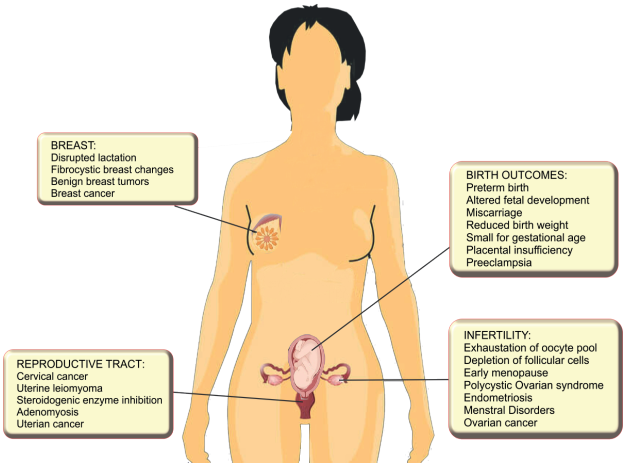


Figure 3: Disorders caused by PFAS to the female reproductive system

The ovary, a vital endocrine organ and female gonad, is a significant target for PFAS toxicity in females. It is responsible for folliculogenesis, oocyte maturation, and the production of female sex steroid and peptide hormones that regulate reproductive and non-reproductive functions (Athar et al., 2024). Environmental exposures to PFNA, PFOA, and PFOS have been linked to exhaustion of the oocyte pool, depletion of follicular cells, and earlier age at menopause, premature ovarian failure, and infertility (Ning et al., 2020). Furthermore, women exposed to PFNA, PFOS, PFDeA, and PFBS during IVF treatment have been found to have lower numbers of retrieved eggs, fertilized eggs, mature eggs, and high-quality embryos, suggesting that such exposure may negatively impact IVF success rates (Shen et al., 2024).

Research has established a link between PFAS exposure and fertility issues in pregnant women from Greenland, Poland, and Ukraine (Jørgensen et al., 2014). Higher levels of PFNA in the blood were associated with longer times to conceive and increased odds of infertility, both in the combined international group and in Greenlandic women specifically (Chen et al., 2021). Consistently, other studies have found that women with higher levels of PFOA and PFNA in their blood had a lower chance of getting pregnant compared to those with lower levels (Lum et al., 2017). While the evidence linking PFAS exposure to miscarriage is conflicting, a clear association has been found between increased umbilical cord concentrations of PFOS and preterm birth (Chen et al., 2021; Green et al., 2021). Furthermore, PFAS have been shown to promote endometriosis, a major cause of female infertility (Hammarstrand et al., 2021; Zhang et al., 2018). Women residing in heavily contaminated regions are particularly vulnerable to multiple reproductive dysfunctions (Rumph et al., 2022). Additionally, PFAS can affect human reproductive capacity by altering fetal development (Erinc et al., 2021). Elevated levels of PFOA in blood have been linked to an increased risk of miscarriage in early pregnancy (first trimester), a common complication in pregnancy (Wikstrom, 2021).

Short-chain PFAS, such as PFBS, PFHxS, PFTrDA, and GenX, have been found to impact hormone activities and physiology in non-occupationally exposed humans and other living organisms (Nian et al., 2020). Long-term exposure to PFOA, PFOS, and PFHxS leads to accumulation of these chemicals in women of child-bearing age, increasing exposure to the fetus and breastfed babies (Enyoh et al., 2023). The Minnesota Department of Health (MDH) recommends that women who are breastfeeding, pregnant, or planning to become pregnant take steps to reduce their exposure to PFAS. This is based on research showing that long-term exposure to PFAS could impact human health, particularly vulnerable populations like fetuses and infants. The Minnesota Pollution Control Agency (MPCA) and MDH have been investigating PFAS since 2002, and ongoing research continues to inform their guidance (https://www.pca.state.mn.us/sites/default/files/p-gen1-22g.pdf). Infant formula mixed with PFAS-contaminated water can result in higher exposure to PFAS, as babies consume more water per body weight than adults (Zhang et al., 2018). Research has shown an inverse association between perinatal exposure to certain PFAS (PFOS, PFOA, PFNA, PFHxS, or PFDA) and placental function, fetal growth, and infant birth weight (Kashino et al., 2020; Yao et al., 2021; Pearce et al., 2021). A study analyzing follicular fluid samples from Australian women detected 32 different types of PFAS in all samples. The study found correlations between specific PFAS levels and infertility issues, such as genital tract infections, polycystic ovary syndrome (PCOS), and endometriosis, although no significant link was found with fertilization success rates (Kim et al., 2020). Higher levels of PFNA, PFOA, and PFOS were linked to infertility issues, including PCOS, endometriosis, and genital tract infections, but were not significantly associated with fertilization rates (Kang et al., 2020; Campbell et al., 2016).

PFAS and Menstrual Cycle Irregularities

Research has consistently shown that exposure to PFAS can disrupt normal menstrual cycles, posing a potential threat to reproductive health and fertility (Lum et al., 2017). Animal studies have demonstrated that PFAS exposure can alter estrous cycles, leading to reduced frequency and disrupted reproductive function (Feng et al., 2015; Kato et al., 2015). Laboratory animals receive controlled, high-dose exposures, which may not accurately reflect real-world human exposure scenarios. Humans on the other hand, are exposed to lower doses over prolonged periods, often spanning decades. This chronic exposure leads to bioaccumulation, with PFAS detected in human blood, breast milk, and umbilical cord blood. In human populations, exposure to PFAS has been linked to irregular menstrual cycles (Lum et al., 2017), with a higher risk of cycles lasting seven days or more. Specifically, PFOS, PFNA, PFOA, and PFHxS have been associated with longer menstrual cycles, while higher levels of PFOA, PFOS, PFNA, and PFHxS have been linked to decreased reports of heavy or prolonged menstrual periods (menorrhagia) and increased reports of short or light menstrual periods (hypomenorrhea) (Zhou et al., 2017). The correlation between PFAS exposure and menstrual cycle irregularities has been further supported by studies examining menstrual cycle characteristics in women from the Norwegian Mother and Child Cohort (Singer et al., 2018) and in girls (Di Nisio et al., 2020). The exact mechanisms underlying these associations are not fully understood, but reduced menstrual bleeding and altered hormone regulation may contribute to the observed effects (Zhou et al., 2017). These findings suggest that PFAS exposure may have significant impacts on reproductive health and fertility.

The relationship between PFAS exposure and menstrual cycle length in couples attempting to conceive has been investigated. Findings showed that certain PFAS are associated with longer menstrual cycles, while others are associated with shorter cycles. Specifically, high serum PFOA levels were linked to longer menstrual cycles (25-31 days) compared to shorter cycles (≥32 days). Additionally, increased cycle length was observed with higher PFDA exposure, while decreased cycle length was seen with higher PFOA exposure (Lum et al., 2017). Similarly, another study found that women with higher PFOA exposure had longer menstrual cycles (≥32 days) and increased cycle irregularity (Lyngsø et al., 2014). Exposure to PFOA has also been linked to earlier age at menarche and a higher risk of irregular menstruation (Di Nisio et al., 2020). Overall, these findings suggest that PFAS exposure may impact menstrual cycle regulation and reproductive health.

PFAS and Female Reproductive Tract Disorders

Polycystic ovary syndrome (PCOS) is a prevalent hormonal disorder affecting women of reproductive age, significantly contributing to female infertility and reproductive health issues (Zeng et al., 2022). Research has established a link between PCOS and infertility (Meng et al., 2022; Kim et al., 2020; Zhan et al., 2023). The exact causes and mechanisms of PCOS are not yet fully understood, but it is believed to result from interplay between genetic and environmental factors (Escobar-Morreale, 2018). Women with PCOS often experience ovulation dysfunction, high androgen levels, and multiple small ovarian cysts, leading to symptoms such as irregular menstrual periods, excess hair growth, acne, infertility, and weight gain. Studies have shown that patients with PCOS have higher serum concentrations of PFOS compared to controls (Zhan et al., 2023). Additionally, high PFOS levels were associated with menstrual irregularities in both PCOS patients and controls, suggesting that PFAS may increase the risk of PCOS and related complications in women of reproductive age due to their endocrine-disrupting capabilities (Heffernan et al., 2018). A positive association was identified between mixture of 6:2 Cl-PFESA, HFPO-DA, PFOS, and PFDoA with an elevated odd of polycystic ovarian syndrome and PCOS-related infertility risk (Zhan et al., 2023). However, an inverse association was found between PFUdA levels and PCOS-related infertility, while no associations were observed for other PFAS compounds. These findings suggest that exposure to these compounds may play a role in the development and progression of PCOS and related infertility issues.

Exposure to certain PFAS has also been linked to an increased risk of ovarian cancer and endometriosis (Wang et al. 2017). The mean levels of PFOA, PFNA and PFOS were found to be higher in women who reported having endometriosis compared to women who did not (Campbell et al., 2016). The 2003-2006 NHANES study found a potential link between PFAS exposure and endometriosis in US women. Analyzing data from 753 women aged 20-50, researchers discovered that those with endometriosis had higher average levels of PFNA, PFOA, and PFOS compared to women without the condition (Campbell et al., 2016). However, the study's cross-sectional design, self-reported endometriosis diagnosis, and single-spot serum samples may limit its findings. Additionally, confounding variables such as age, BMI, parity, smoking status, and family history of endometriosis were identified. Despite these limitations, the study suggests a potential association between PFAS exposure and endometriosis, warranting further investigation. In a study of Chinese women with confirmed endometriosis, higher levels of PFBS in their blood were linked to a greater risk of infertility related to endometriosis, even in women who had never been pregnant or had no other gynecological conditions (Wang et al., 2017). Evidences has shown positive associations between exposure to PFAS in contaminated communities and the risk of developing ovarian cancer (Chang et al., 2023). Association has been found to exist between high exposure to PFOS and PFHxS (mostly used in firefighting foam) and increased risk for PCOS and uterine leiomyoma (Hammarstrand et al., 2021).

PFAS Exposure in Pregnancy and Impact on Birth Outcomes

Prenatal exposure to certain PFAS has been linked to reduced birth weight, with studies indicating a more pronounced effect in female newborns. Research by Wikström et al. (2020) revealed a significant correlation between maternal PFAS levels and lower birth weight, particularly in girls. Specifically, increased exposure to PFOS, PFOA, and PFHxS has been associated with reduced birth weight, with some studies suggesting a 50-100 gram reduction in birth weight per unit increase in maternal PFAS levels. This is concerning, as low birth weight (less than 2,500 grams) is linked to increased risks of infant mortality, respiratory distress, and long-term health consequences, including cardiovascular disease and metabolic disorders (Lin et al., 2007). Furthermore, PFAS exposure has also been implicated in preterm birth, which can exacerbate birth weight issues (Qin et al., 2023). While additional research is necessary to fully understand the relationship between PFAS and birth weight, existing evidence underscores the importance of minimizing PFAS exposure during pregnancy to protect fetal development and promote healthy birth outcomes (Qin et al., 2023). PFAS exposure has also been associated with neurodevelopmental toxicity, although the evidence from epidemiological studies on specific neurodevelopmental effects is inconsistent and requires further research. A study investigating the relationships between prenatal PFAS exposure and symptoms of attention-deficit/hyperactivity disorder (ADHD), cognitive functioning (language skills, estimated IQ, and working memory) in preschool children, and effect modification by child sex found some associations between PFAS and working memory, specifically negative relationships with nonverbal working memory and positive relationships with verbal working memory. However, these relationships were weak for both positive and negative, suggesting no clear association and the need for further studies to fully understand the effects of PFAS exposure on fetal development and neurodevelopmental outcomes (Skogheim et al., 2020).

Research has identified a level of association between prenatal exposure to PFOS and PFDA and an increased risk of preterm birth and miscarriage, with a direct linear relationship observed between the two (Gao et al., 2021; Chen et al., 2021; Green et al., 2021). Additionally, the presence of PFAS compounds has been detected in over 50 % of follicular fluid samples from Chinese women, highlighting their widespread presence (Kang et al., 2020). Exposure to PFAS has also been shown to alter placental cell function in mice and affect gene expression in liver and fat tissue in a sex-specific manner in offspring (Bangma et al., 2020; Szilagyi et al., 2020). A meta-analysis of 30 studies found associations between prenatal PFAS exposure and increased childhood body mass index (BMI) and waist circumference (WC), while childhood exposure was linked to reduced BMI (Frigerio et al., 2023). However, the evidence is not yet conclusive, and further research is necessary to validate these findings. The persistent presence of PFAS in the environment and human bodies, combined with their transfer from mother to child during pregnancy and breastfeeding, poses a potential threat to fetal growth and development (Gao et al., 2021; Padula et al., 2023).

Disruption of Ovarian Hormonal Regulation and Feedback Mechanisms by PFAS

The reproductive system is governed by an intricate hormonal process involving the pituitary gland, adrenal cortex, and gonads. At puberty, the hypothalamus initiates the release of gonadotropin-releasing hormone (GnRH), which stimulates the pituitary gland to produce and secrete follicle-stimulating hormone (FSH) and luteinizing hormone (LH). These gonadotropins play a vital role in regulating the function of the gonads (testes in males and ovaries in females), driving their development, maturation, and reproductive capacity (Marques et al., 2022). In both males and females, FSH and LH play crucial roles in regulating reproductive function. FSH promotes gametogenesis (sperm and egg production), while LH stimulates the gonads to produce sex hormones (testosterone and estrogen). As sex hormone levels rise, they exert a negative feedback effect on the hypothalamus, suppressing GnRH production and thereby regulating the reproductive axis. This feedback loop maintains a delicate balance in the reproductive system, preventing overstimulation.

Studies have demonstrated that PFAS, specifically PFOS accumulation in the hypothalamus can interfere with reproductive hormone regulation, leading to changes in sex hormone production and steroidogenesis (Starnes et al., 2022; Yang et al., 2022). Notably, studies have identified profound long-term effects, including reduced follicular estradiol levels in childless women (Barrett et al., 2015), impaired fetal reproductive development due to altered gonadotropin levels during pregnancy (Nian et al., 2020), increased risk of reproductive disorders, and altered gene expression in reproductive tissues. These changes have been shown to be persistent, even after exposure cessation, and are associated with adverse health outcomes, such as infertility, reduced fertility, pregnancy complications, endocrine disruption, and developmental issues. Specifically, the identified profound long-term effects include disrupted menstrual cycles, reduced fertility, increased risk of endometriosis and PCOS, birth defects, low birth weight, preterm birth, and increased cancer risk (Rickard et al., 2022).

Research has yielded inconsistent findings on the relationship between PFAS exposure and hormone levels, posing a challenge for understanding their impact on hormone regulation. In vitro studies have shown that PFOA and PFOS can exhibit both estrogenic and antiestrogenic properties, disrupting steroid hormone production and potentially affecting reproductive and endocrine systems. While some studies have found that PFOA and PFOS exhibit estrogenic properties, others have observed an inverse association between PFOS exposure and estradiol levels (Knox et al., 2011), suggesting that PFOS may both mimic and disrupt estrogenic activity. Cell-based assays have revealed that high concentrations of PFOA and PFOS can increase estrone and progesterone production (Behr et al., 2018), potentially leading to hormone imbalances. However, some studies have found no association between PFOA exposure and sex hormone levels, indicating a complex relationship that requires further investigation. Notably, PFOA has been shown to bind directly to progesterone, counteracting its effects on genes involved in embryo implantation and endometrial growth, suggesting a potential mechanism for reduced fertility and reproductive issues. Overall, the evidence suggests that PFAS exposure may have significant impacts on hormone regulation, but further research is needed to fully understand these effects.

The female reproductive tract is a complex system that relies heavily on hormonal feedback loops to regulate various physiological processes, including ovulation, menstruation, and pregnancy. These feedback loops are driven by changes in circulating hormone levels, which are precisely regulated by the hypothalamic-pituitary-gonadal axis. However, exposure to select PFAS has been shown to disrupt these delicate feedback loops, potentially leading to reproductive dysfunction. These PFAS can interfere with hormone regulation by altering hormone production, metabolism, or clearance, even if they do not directly activate receptors. For example, PFAS have been shown to:

- Inhibit the production of steroidogenic enzymes, leading to decreased hormone synthesis.
- Alter the expression of hormone receptors, affecting hormone signaling.
- Disrupt the balance of hormone-metabolizing enzymes, leading to changes in hormone clearance.
- Interact with hormone-binding proteins, affecting hormone transport and delivery.

These disruptions could have far-reaching consequences, including altered menstrual cyclicity and fertility, changes in ovulation and implantation, increased risk of pregnancy complications, and disrupted lactation and breast development. Moreover, PFAS exposure has been linked to changes in circulating hormone levels, including decreased estradiol and progesterone levels, increased androgen levels, and altered thyroid hormone levels.

Despite suggestions of a potential link between PFAS exposure and reproductive health concerns, scientific evidence remains inconclusive, with some studies indicating no significant correlation between these compounds and reproductive health. For instance, it was reported that there is inconclusive evidence for an association between PFAS and fetal growth (Säve-Söderbergh et al., 2024). Furthermore, many existing studies are limited by factors such as small sample sizes, incomplete exposure assessments, and methodological inconsistencies, which underscore the need for rigorous, well-designed investigations to definitively ascertain the relationship between PFAS exposure and reproductive health outcomes.

Table 1: Overview of epidemiological studies on PFAS and female reproductive health

Study year	Population	PFAS measured	Key findings
Barrett et al. (2015)	178 women between 25-35 years	PFOS	Positive association between PFOS and decreased production of estradiol in reproductive age women.
Nian et al. (2020)	752 mother-infant pairs	PFBS, PFHpA	Inverse association between PFBS and follicle stimulating hormone, and between PFHpA and Free Androgen Index. PFBS and PFHpA were associated with the disturbance of fetal gonadotropins and free androgen level.
Knox et al. (2019)	25 957 women between 18-65 years	PFOA, PFOS	Significant inverse association between PFOS and estradiol in perimenopausal and menopausal age groups but not between PFOA and estradiol.
Chen et al. (2021)	255 pregnant women	Five PFAS	No association between the PFAS and birth weight or head circumference was observed.
Lum et al. (2017)	501 couples	Seven PFAS	Serum levels of PFOA and PFDeA were associated with changes in menstrual cycle length.
Zhou et al. (2017)	950 women	Ten PFAS	PFOA and PFOS are associated with abnormal menstruation in humans.
Zhan et al. (2023)	366 women	Twenty three PFAS	Mixture of 6:2 Cl-PFESA, HFPO-DA, PFOS, and PFDoA was associated with an elevated odd of polycystic ovarian syndrome
Wikstrom et al. (2021)	78 women	Eight PFAS	PFOA was associated with first trimester miscarriage.
Säve-Söderbergh et al. (2024)	248 804	Four PFAS	PFOS was associated with increased risk of Large-for-gestational-age.

FUTURE DIRECTIONS IN PFAS-FEMALE REPRODUCTIVE HEALTH RESEARCH

Despite growing evidence linking PFAS to adverse female reproductive health outcomes, significant knowledge gaps remain. To better understand the complex relationships between PFAS exposure and female reproductive health, further research is necessary. Elucidating the molecular mechanisms underlying PFAS-induced reproductive toxicity is crucial. Investigating PFAS interactions with hormone receptors, gene expression, and epigenetic modifications will provide valuable insights. Additionally, developing more accurate methods to quantify PFAS exposure in humans, particularly during critical windows of vulnerability, is essential.

Large-scale, longitudinal studies examining PFAS exposure and reproductive health outcomes in diverse populations are needed to confirm associations and establish dose-response relationships. Research focusing on specific reproductive stages, such as follicular development, implantation, and pregnancy, will help identify critical periods of susceptibility. Also, investigating PFAS interactions with other endocrine-disrupting chemicals will provide a more comprehensive understanding of cumulative risks. Examining PFAS exposure during fetal development and early life stages will shed light on potential long-term reproductive health consequences.

To advance PFAS research and policy, collaboration between scientists, policymakers, and stakeholders is essential. Prioritizing research areas, translating findings into policy, and promoting public awareness would mitigate any potential harmful effects of PFAS on female reproductive health.

CONCLUSION

The persistence and bioaccumulation of PFAS, combined with their extensive use and release, may pose a significant threat to human and environmental health. There is growing concern that these "forever chemicals" could be polluting our environment and accumulating in humans, potentially impacting reproductive health and overall well-being. The ability of these substances to accumulate in the food chain raises concerns about long-term risks to human health and the environment, highlighting the need for continued monitoring. PFAS exposure raises concerns about female reproductive health, as they may disrupt hormone balance and fertility. This issue affects not only humans but also the broader ecosystem, with the potential to disrupt reproductive success in various species. To address this challenge, collaborative efforts from governments, industries, and individuals are necessary. A multidisciplinary approach can help manage PFAS usage, develop effective remediation technologies, and promote sustainable practices prioritizing human and environmental well-being.

LIST OF ABBREVIATIONS

6:2 FTOH	6:2 Fluorotelomer Alcohol Glucuronide
8:2 FTOH,	8:2 Fluorotelomer Alcohol Glucuronide
ADHD	Attention-Deficit/Hyperactivity Disorder
EDCs	Endocrine-Disrupting Chemicals
FSH	Follicle-Stimulating Hormone
GnRH	Gonadotropin-Releasing Hormone
hCG	Human Chorionic Gonadotropin Hormone
HPGA	Hypothalamic-Pituitary-Gonadal Axis
BMI	Body Mass Index
IPEN	International Pollutants Elimination Network
ITRC	Interstate Technology & Regulatory Council
IVF	In Vitro Fertilization
LH	Luteinizing Hormone
NHANES	National Health and Nutrition Examination Survey
NRDC	Natural Resources Defense Council
OECD	Organization for Economic Cooperation and Development
PCOS	Polycystic Ovary Syndrome
PFAS	Per- and Polyfluoroalkyl Substances
PFBS	Perfluorobutane Sulfonate
PFDA	Perfluorodecanoic Acid
PFHxS	Perfluorohexane Sulfonate
PFNA	Perfluorononanoic Acid
PFOA	Perfluorooctanoic Acid
PFOS	Perfluorooctane Sulfonate
PFDeA	Perfluorodecanoic Acid
PFTrDA	Perfluorotridecanoic Acid
PFUdA	Perfluoro-undecanoic Acid
Cl-PFESA	Chlorinated Polyfluoroalkyl Sulfonates
HFPO-DA	Hexafluoropropylene Oxide Dimer Acid
THDCs	Thyroid Disrupting Compounds
USEPA	United States Environmental Protection Agency
EU	European Union

REFERENCES

Athar, F.; Karmani, M.; Templeman, N. Metabolic Hormones Are Integral Regulators of Female Reproductive Health and Function. *Biosci. Rep.* **2024**, 31 (1), BSR20231916. https://doi.org/10.1042/BSR20231916

Atiya, A.; Somia, G. Reproductive Hormones and Implication of Synthetic Hormonal Preparations: Current Status. Biomed. *J. Sci. Tech. Res.* **2021**, 38 (2), 30156–30159.

Bangma, J.; Eaves, L. A.; Oldenburg, K.; Reiner, J. L.;

Manuck, T.; Fry, R. C. Identifying Risk Factors for Levels of Per- and Polyfluoroalkyl Substances (PFAS) in the Placenta in a High-Risk Pregnancy Cohort in North Carolina. *Environ. Sci. Technol.* **2020**, 54 (13), 8158–8166.

Bangma, J. J.; Szilagyi, B. E.; Blake, C.; Plazas, S.; Kepper, S. E.; Fenton, R. C. An Assessment of Serum-Dependent Impacts on Intracellular Accumulation and Genomic Response of Per- and Polyfluoroalkyl Substances in a Placental Trophoblast Model. *Environ. Toxicol.* **2020**, 35 (12), 1395–1405.

Barrett, E. S.; Chen, C.; Thurston, S. W.; Haug, L. S.; Sabaredzovic, A.; Fjeldheim, F. N.; Frydenberg, H.; Lipson, S. F.; Ellison, P. T.; Thune, I. Perfluoroalkyl Substances and Ovarian Hormone Concentrations in Naturally Cycling Women. *Fertil. Steril.* **2015**, 103 (5), 1261–1270.

Barton-Maclaren, T. S.; Wade, M.; Basu, N.; Bayen, S.; Grundy, J.; Marlatt, V.; Moore, R.; Parent, L.; Parrott, J.; Grigorova, P.; Pinsonnault-Cooper, J.; Langlois, V. S. Innovation in Regulatory Approaches for Endocrine Disrupting Chemicals: The Journey to Risk Assessment Modernization in Canada. *Environ. Res.* **2022**, 204, 112225.

Behr, A. C.; Lichtenstein, D.; Braeuning, A.; Lampen, A.; Buhrke, T. Perfluoroalkylated Substances (PFAS) Affect Neither Estrogen and Androgen Receptor Activity nor Steroidogenesis in Human Cells In Vitro. *Toxicol. Lett.* **2018**, 291, 251–260.

Berg, C.; Crone, B.; Gullett, B.; Higuchi, M.; Krause, M. J.; Lemieux, P. M.; Martin, T.; Shields, E. P.; Struble, E.; Thoma, E.; Whitehill, A. Developing Innovative Treatment Technologies for PFAS-Containing Wastes. *J. Air Waste Manag. Assoc.* **2022**, 72 (6), 540–555.

Bridger, J. R.; Thackray, C. M.; Butt, C. M.; LeBlanc, D. R.; Tokranov, A. K.; Vecitis, C. D.; Sunderland, E. M. Centurial Persistence of Forever Chemicals at Military Fire Training Sites. *Environ. Sci. Technol.* **2023**, 57 (11), 8096–8106.

Brunn, H.; Arnold, G.; Körner, W.; Rippen, G.; Steinhäuser, K. G.; Valentin, I. PFAS: Forever Chemicals—Persistent, Bioaccumulative, and Mobile. Reviewing the Status and the Need for Their Phase-Out and Remediation of Contaminated Sites. *Environ. Sci. Eur.* **2023**, 35, 20. https://doi.org/10.1186/s12302-023-00721-8

Campbell, S.; Raza, M.; Pollack, A. Z. Perfluoroalkyl Substances and Endometriosis in US Women in NHANES 2003–2006. *Reprod. Toxicol.* **2016**, 65, 230–235.

Chang, V. C.; Rhee, J.; Berndt, S. I.; Moore, S. C.; Freedman, N. D.; Jones, R. R.; Silverman, D. T.; Gierach, G. L.; Hofmann, J. N.; Purdue, M. P. Serum Perfluorooctane Sulfonate and Perfluorooctanoate and Risk of Postmenopausal Breast Cancer According to Hormone Receptor Status: An Analysis in the Prostate, Lung, Colorectal and Ovarian Cancer Screening Trial. *Int. J. Cancer* **2023**, 153 (4), 775–782.

Chen, L.; Tong, C.; Huo, X.; Zhang, J.; Tian, Y. Prenatal Exposure to Perfluoroalkyl and Polyfluoroalkyl Substances and Birth Outcomes: A Longitudinal Cohort with Repeated Measurements. *Chemosphere* **2021**, 267, 128899.

Commission for Environmental Cooperation. Furthering the Understanding of the Migration of Chemicals from Consumer Products – A Study of Per- and Polyfluoroalkyl Substances (PFASs) in Clothing, Apparel, and Children's Items; Project Report; Montreal, Canada, **2017**. http://www.cec.org/files/documents/publications/11777-furthering-understanding-migration-chemicals-from-consumer-products-en.pdf

Cousins, I. T.; Johansson, J. H.; Salter, M. E.; Sha, B.; Scheringer, M. Outside the Safe Operating Space of a New Planetary Boundary for Per- and Polyfluoroalkyl Substances (PFAS). *Environ. Sci. Technol.* 2022, 56 (16), 11172–11179.

Cousins, J. T.; DeWitt, J. C.; Glüge, J.; Goldenman, G.; Herzke, D.; Lohmann, R.; Ng, C. A.; Scheringer, M.; Wang, Z. The High Persistence of PFAS Is Sufficient for Their Management as a Chemical Class. *Environ. Sci. Process Impacts* **2020**, 22, 2307–2312.

De Silva, A. O.; Armitage, J. M.; Bruton, T. A.; Dassuncao, C.; Heiger-Bernays, W.; Hu, X. C.; Kärrman, A.; Kelly, B.; Ng, C.; Robuck, A.; Sun, M.; Webster, T. F.; Sunderland, E. M. PFAS Exposure Pathways for Humans and Wildlife: A Synthesis of Current Knowledge and Key Gaps in Understanding. *Environ. Toxicol. Chem.* **2021**, 40 (3), 631–657.

Di Nisio, A.; Rocca, M. S.; Sabovic, I.; De Rocco-Ponce, M.; Corsini, C.; Guidolin, D. Perfluorooctanoic Acid Alters Progesterone Activity in Human Endometrial Cells and Induces Reproductive Alterations in Young Women. *Chemosphere* **2020**, 242, 125208. https://pubmed.ncbi.nlm.nih.gov/31896193/

Endocrine Society. Common EDCs and Where They Are Found. https://www.endocrine.org/topics/edc/what-edcs-are/common-edcs (accessed 2023).

Enyoh, C. E.; Ovuoraye, P. E.; Qingyue, W.; Weiqian, W. Examining the Impact of Nanoplastics and PFAS Exposure on Immune Functions through Inhibition of Secretory Immunoglobulin A in Human Breast Milk. *J. Hazard. Mater.* **2023**, 459, 132103. https://pubmed.ncbi.nlm.nih.gov/37527590/

Ercan, O.; Tarcin, G. Overview on Endocrine Disruptors in Food and Their Effects on Infant's Health. Global

Pediatr. **2022**, 2, 100019. https://www.sciencedirect.com/science/article/pii/S2667009722000136

Erinc, A.; Davis, M. B.; Padmanabhan, V.; Langen, E.; Goodrich, J. M. Considering Environmental Exposures to Per- and Polyfluoroalkyl Substances (PFAS) as Risk Factors for Hypertensive Disorders of Pregnancy. *Environ. Res.* **2021**, 197, 111113.

Escobar-Morreale, H. F. Polycystic Ovary Syndrome: Definition, Aetiology, Diagnosis and Treatment. *Nat. Rev. Endocrinol.* **2018**, 14 (5), 270–284.

Feng, X.; Wang, X.; Cao, X.; Xia, Y.; Zhou, R.; Chen, L. Chronic Exposure of Female Mice to an Environmental Level of Perfluorooctane Sulfonate Suppresses Estrogen Synthesis Through Reduced Histone H3K14 Acetylation of the StAR Promoter Leading to Deficits in Follicular Development and Ovulation. *Toxicol. Sci.* **2015**, 148 (2), 368–379.

Fenton, S. E.; Ducatman, A.; Boobis, A.; DeWitt, J. C.; Lau, C.; Ng, C.; Smith, J. S.; Roberts, S. M. Per- and Polyfluoroalkyl Substance Toxicity and Human Health Review: Current State of Knowledge and Strategies for Informing Future Research. *Environ. Toxicol. Chem.* **2021**, 40 (3), 606–630.

Fiedler, H.; Kennedy, T.; Henry, B. J. A Critical Review of a Recommended Analytical and Classification Approach for Organic Fluorinated Compounds with an Emphasis on Per- and Polyfluoroalkyl Substances. *Integr Environ. Assess. Manag.* **2021**, 17 (2), 331–351.

Forsthuber, M.; Kaiser, A. M.; Granitzer, S. H.; Hengstschläger, M.; Stangl, H.; Gundacker, C. Albumin Is the Major Carrier Protein for PFOS, PFOA, PFHxS, PFNA, and PFDA in Human Plasma. *Environ. Int.* **2020**, 137, 105324.

Frigerio, G.; Ferrari, C. M.; Fustinoni, S. Prenatal and Childhood Exposure to Per-/Polyfluoroalkyl Substances (PFASs) and Its Associations with Childhood Overweight and/or Obesity: A Systematic Review with Meta-Analyses. *Environ. Health* **2023**, 22 (1), 56.

Gaines, L. G. T. Historical and Current Usage of Per- and Polyfluoroalkyl Substances (PFAS): A Literature Review. *Am. J. Ind. Med.* **2022**, 66 (5), 353–378.

Gao, X.; Ni, W.; Zhu, S.; Wu, Y.; Cui, Y.; Ma, J.; Liu, Y.; Qiao, J.; Ye, Y.; Yang, P.; Liu, C.; Zeng, F. Per- and Polyfluoroalkyl Substances Exposure during Pregnancy and Adverse Pregnancy and Birth Outcomes: A Systematic Review and Meta-Analysis. *Environ. Res.* **2021**, 201, 111632. https://doi.org/10.1016/j.envres.2021.111632

Glüge, J.; Scheringer, M.; Cousins, I. T.; DeWitt, J. C.; Goldenman, G.; Herzke, D.; Lohmann, R.; Ng, C. A.; Trier, X.; Wang, Z. An Overview of the Uses of Per- and Polyfluoroalkyl Substances (PFAS). *Environ. Sci. Process Impacts* **2021**, 22 (12), 2345.

González-Comadran, M.; Jacquemin, B.; Cirach, M.; Lafuente, R.; Cole-Hunter, T.; Nieuwenhuijsen, M.; Brassesco, M.; Coroleu, B.; Checa, M. A. The Effect of Short-Term Exposure to Outdoor Air Pollution on Fertility. *Reprod. Biol. Endocrinol.* **2021**, 19 (1), 151. https://doi.org/10.1186/s12958-021-00838-6

Green, M. P.; Harvey, A. J.; Finger, B. J.; Tarulli, G. A. Endocrine Disrupting Chemicals: Impacts on Human Fertility and Fecundity during the Peri-Conception Period. *Environ. Res.* **2021**, 194, 110694. https://pubmed.ncbi.nlm.nih.gov/33385395/

Grgas, D.; Petrina, A.; Štefanac, T.; Bešlo, D.; Landeka Dragičević, T. A Review: Per- and Polyfluoroalkyl Substances—Biological Degradation. *Toxics* **2023**, 11 (5), 446.

Hammarstrand, S.; Jakobsson, K.; Andersson, E.; Xu, Y.; Li, Y.; Olovsson, M.; Andersson, E. M. Perfluoroalkyl Substances (PFAS) in Drinking Water and Risk for Polycystic Ovarian Syndrome, Uterine Leiomyoma, and Endometriosis: A Swedish Cohort Study. *Environ. Int.* **2021**, 157, 106819. https://doi.org/10.1016/j.envint.2021.106819

Heffernan, A. L.; Cunningham, T. K.; Drage, D. S.; Aylward, L. L.; Thompson, K.; Vijayasarathy, S.; Mueller, J. F.; Atkin, S. L.; Sathyapalan, T. Perfluorinated Alkyl Acids in the Serum and Follicular Fluid of UK Women with and without Polycystic Ovarian Syndrome Undergoing Fertility Treatment and Associations with Hormonal and Metabolic Parameters. *Int. J. Hyg. Environ. Health* **2018**, 221 (7), 1068–1075.

Huang, M. C.; Robinson, V. G.; Waidyanatha, S.; Dzierlenga, A. L.; DeVito, M. J.; Eifrid, M. A.; Gibbs, S. T.; Blystone, C. R. Toxicokinetics of 8:2 Fluorotelomer Alcohol (8:2-FTOH) in Male and Female Hsd:Sprague Dawley SD Rats after Intravenous and Gavage Administration. *Toxicol. Rep.* **2019**, 6, 924–932.

Innocent, T. M. Bioaccumulation of Emerging Micro Pollutants in Aquatic Biota Per- and Polyfluoroalkyl Substances; Donghua University: Shanghai, China, **2022**. https://www.researchgate.net/publication/362668596

Interstate Technology and Regulatory Council (ITRC). Naming Conventions and Physical and Chemical Properties of Per- and Polyfluoroalkyl Substances (PFAS); **2020**. https://pfas-1.itrcweb.org/fact-sheets

IPEN. Stockholm Convention COP-9 White Paper: The Global PFAS Problem: Fluorine-Free Alternatives As Solutions; **2019**. https://ipen.org/sites/default/files/documents/the_global_pfas_problem-v1_5_final_18_april.pdf

Jørgensen, K. T.; Specht, I. O.; Lenters, V.; Bach, C. C.; Rylander, L.; Jönsson, B. A.; Lindh, C. H.; Giwercman, A.; Heederik, D.; Toft, G.; Bonde, J. P. Perfluoroalkyl Substances and Time to Pregnancy in Couples from Greenland, Poland and Ukraine. *Environ. Health* **2014**, 13, 116. https://doi.org/10.1186/1476-069X-13-116

Kang, J. S.; Choi, J. S.; Park, J. W. Transcriptional Changes in Steroidogenesis by Perfluoroalkyl Acids (PFOA and PFOS) Regulate the Synthesis of Sex Hormones in H295R Cells. *Chemosphere* **2016**, 155, 436–443.

Kang, Q.; Gao, F.; Zhang, X.; Wang, L.; Liu, J.; Fu, M.; Zhang, S.; Wan, Y.; Shen, H.; Hu, J. Nontargeted Identification of Per- and Polyfluoroalkyl Substances in Human Follicular Fluid and Their Blood-Follicle Transfer. Environ. Int. **2020**, 139, 105686. https://www.sciencedirect.com/science/article/pii/S0160412019347439

Kashino, I.; Sasaki, S.; Okada, E.; Matsuura, H.; Goudarzi, H.; Miyashita, C. Prenatal Exposure to Perfluoroalkyl Substances and Fetal Growth: A Large-Scale Prospective Birth Cohort Study. *Environ. Int.* **2020**, 136, 105355. https://pubmed.ncbi.nlm.nih.gov/32029284/

Kato, H.; Fujii, S.; Takahashi, M.; Matsumoto, M.; Hirata-Koizumi, M.; Ono, A.; Hirose, A. Repeated Dose and Reproductive/Developmental Toxicity of Perfluorododecanoic Acid in Rats. *Environ. Toxicol.* **2015**, 30, 1244–1263.

Khazaee, M.; Christie, E.; Cheng, W.; Michalsen, M.; Field, J.; Ng, C. Perfluoroalkyl Acid Binding with Peroxisome Proliferator-Activated Receptors alpha, gamma, and delta, and Fatty Acid Binding Proteins by Equilibrium Dialysis with a Comparison of Methods. *Toxics* **2021**, 9 (1), 1–15.

Kim, D. H.; Kim, U. J.; Kim, H. Y.; Choi, S. D.; Oh, J. E. Perfluoroalkyl Substances in Serum from South Korean Infants with Congenital Hypothyroidism and Healthy Infants—Its Relationship with Thyroid Hormones. *Environ. Res.* **2016**, 147, 399–404.

Kim, Y.; Pacella, R.; Harden, F.; White, N.; Toms, L. A Systematic Review: Impact of Endocrine Disrupting Chemicals Exposure on Fecundity as Measured by Time to Pregnancy. *Environ. Res.* **2019**, 171, 119–133.

Kim, Y. R.; White, N.; Bräunig, J.; Vijayasarathy, S.; Mueller, J. F.; Knox, C. L.; Harden, F. A.; Pacella, R.; Toms, L. L. Per- and Poly-Fluoroalkyl Substances (PFASs) in Follicular Fluid from Women Experiencing Infertility in Australia. *Environ. Res.* 2020, 190, 109963.

Knox, S. S.; Jackson, T.; Javins, B.; Frisbee, S. J.; Shankar, A.; Ducatman, A. M. Implications of Early Menopause in Women Exposed to Perfluorocarbons. *J. Clin. Endocrinol. Metab.* **2011**, 96, 1747–1753.

Korzeniowski, S. H.; Buck, R. C.; Newkold, R. M.; El Kassmi, A.; van Laganis, E.; Matsuoka, Y.; Dinelli, B.; Beauchet, S.; Adamsky, F.; Weilandt, K.; Soni, V. K.; Kapoor, D.; Gunasekar, P.; Malvasi, M.; Brinati, G.; Musio, S. A Critical Review of the Application of Polymer of Low Concern Regulatory Criteria to Fluoropolymers II: Fluoroplastics and Fluoroelastomers. *Integr Environ. Assess. Manag.* **2022**, 18 (4), 4646.

Kurwadkar, S.; Dane, J.; Kanel, S. R.; Nadagouda, M. N.; Cawdrey, R. W.; Ambade, B.; Struckhoff, G. C.; Wilkin, R. Per- and Polyfluoroalkyl Substances in Water and Wastewater: A Critical Review of Their Global Occurrence and Distribution. *Sci. Total Environ.* **2022**, 809, 151003. https://doi.org/10.1016/j.scitotenv.2021.151003

Lai, T. T.; Eken, Y.; Wilson, A. K. Binding of Per- and Polyfluoroalkyl Substances to the Human Pregnane X Receptor. *Environ. Sci. Technol.* **2020**, 54 (24), 15986–15995.

Lee, Y. J.; Jung, H. W.; Kim, H. Y.; Choi, Y. J.; Lee, Y. A. Early-Life Exposure to Per- and PolyFluorinated Alkyl Substances and Growth, Adiposity, and Puberty in Children: A Systematic Review. *Front. Endocrinol.* **2021**, 9 (12), 683297.

Lin, C. M.; Chen, C. W.; Chen, P. T.; Lu, T. H.; Li, C. Y. Risks and Causes of Mortality among Low-Birthweight Infants in Childhood and Adolescence. *Paediatr. Perinat. Epidemiol.* **2007**, 21 (5), 465–472.

Lum, K. J.; Sundaram, R.; Barr, D. B.; Louis, T. A.; Buck Louis, G. M. Perfluoroalkyl Chemicals, Menstrual Cycle Length, and Fecundity: Findings from a Prospective Pregnancy Study. *Epidemiology* **2017**, 28 (1), 90–98.

Lyngsø, J. H.; R-HC; Høyer, B. B.; Støvring, H.; Bonde, J. P.; Jönsson, B.; Lindh, C. H.; Pedersen, H. S.; Ludwicki, J. K.; Zviezdai, V.; Toft, G. Menstrual Cycle Characteristics in Fertile Women from Greenland, Poland, and Ukraine Exposed to Perfluorinated Chemicals: A Cross-Sectional Study. *Hum. Reprod.* **2014**, 29 (22), 359–367.

Marques, P.; Skorupskaite, K.; Rozario, K. S. Physiology of GnRH and Gonadotropin Secretion; **2022**. https://www.ncbi.nlm.nih.gov/books/NBK279070/

Mazumder, N. U.; Hossain, M. T.; Jahura, F. T.; Girase, A.; Hall, A. S.; Lu, J.; Ormond, R. B. Firefighters' Exposure to Per- and Polyfluoroalkyl Substances (PFAS) as an Occupational Hazard: A Review. *Front. Mater.* **2023**, 10, 1143411.

McMahon, P. B.; Tokranov, A. K.; Bexfield, L. M.; Lindsey, B. D.; Johnson, T. D.; Lombard, M. A.; Watson, E. Perfluoroalkyl and Polyfluoroalkyl Substances in Groundwater Used as a Source of Drinking Water in the Eastern United States. *Environ. Sci. Technol.* **2022**, 56 (4), 2279–2288.

Meng, L.; Xiangyan, R.; Alfred, O.; Mueck. Management Strategy of Infertility in Polycystic Ovary Syndrome. *Glob. Health J.* **2022**, 6 (2), 70–74.

Minnesota's PFAS Blueprint. Protecting families and communities from PFAS pollution. https://www.pca.state.mn.us/sites/default/files/p-gen1-22g.pdf

Mueller, R.; Kate, E. S. Human and Ecological Health Effects and Risk Assessment of Per- and Polyfluoroalkyl Substances (PFAS); **2020**. https://pfas-1.itrcweb.org/wp-content/uploads/2020/10/human_and_eco_health_508_20200918.pdf

Neuwald, I. J.; Zahn, D.; Knepper, T. P. Are (Fluorinated) Ionic Liquids Relevant Environmental Contaminants? High-Resolution Mass Spectrometric Screening for Per- and Polyfluoroalkyl Substances in Environmental Water Samples Led to the Detection of a Fluorinated Ionic Liquid. *Anal. Bioanal. Chem.* **2020**, 412, 4881–4892.

Nian, M.; Luo, K.; Luo, F.; Aimuzi, R.; Huo, X.; Chen, Q.; Tian, Y.; Zhang, J. Association between Prenatal Exposure to PFAS and Fetal Sex Hormones: Are the Short-Chain PFAS Safer? *Environ. Sci. Technol.* **2020**, 54, 8291–8299.

Ning, D.; Siobán, D.; Harlow, J. F.; Randolph, J.; Rita, L.; Sung, K. P. Perfluoroalkyl and Polyfluoroalkyl Substances (PFAS) and Their Effects on the Ovary. *Hum. Reprod. Update* **2020**, 26 (5), 724–752.

NRDC (Natural Resources Defense Council). Engaging the Textile Industry as a Key Sector in SAICM—A Review of PFAS as a Chemical Class in the Textile Sector; **2021**. https://saicmknowledge.org/sites/default/files/resources/SAICM%20report_PFAS%20in%20Textile_final_May%202021.pdf

Organisation for Economic Co-operation and Development (OECD). Reconciling Terminology of the Universe of Per- and Polyfluoroalkyl Substances: Recommendations and Practical Guidance, Health and Safety Publications Series on Risk Management; **2021**. www.oecd.org/chemicalsafety

Padula, A. M.; Ning, X.; Bakre, S.; Barrett, E. S.; Bastain, T.; Bennett, D. H.; Bloom, M. S.; Breton, C. V.; Dunlop, A. L.; Eick, S. M.; Ferrara, A.; Fleisch, A.; Geiger, S.; Goin, D. E.; Kannan, K.; Karagas, M. R.; Korrick, S.; Meeker, J. D.; Morello-Frosch, R.; O'Connor, T. G. Birth Outcomes in Relation to Prenatal Exposure to Per- and Polyfluoroalkyl Substances and Stress in the Environmental Influences on Child Health Outcomes (ECHO) Program. *Environ. Health Perspect.* **2023**, 131 (3), 37006. https://doi.org/10.1289/EHP10723

PAN Europe. Europe's Toxic Harvest - Unmasking PFAS Pesticides Authorized in Europe; **2023**. https://www.pan-europe.info/sites/pan-europe.info/files/public/resources/reports/PFAS%20Pesticides%20report%20November%202023.pdf

Pan, Y. Worldwide Distribution of Novel Perfluoroether Carboxylic and Sulfonic Acids in Surface Water. *Environ. Sci. Technol.* **2018**, 52 (14), 7621–7629.

Pearce, J. L.; Neelon, B.; Bloom, M. S.; Buckley, J. P.; Ananth, C. V.; Perera, F. Exploring Associations between Prenatal Exposure to Multiple Endocrine Disruptors and Birthweight with Exposure Mapping. *Environ. Res.* **2021**, 200, 111386.

Peritore, A. F.; Gugliandolo, E.; Cuzzocrea, S.; Crupi, R.; Britti, D. Current Review of Increasing Animal Health Threat of Per- and Polyfluoroalkyl Substances (PFAS): Harms, Limitations, and Alternatives to Manage Their Toxicity. *Int. J. Mol. Sci.* **2023**, 24 (14), 11707.

Pierozan, P.; Karlsson, O. PFOS Induces Proliferation, Cell-Cycle Progression, and Malignant Phenotype in Human Breast Epithelial Cells. *Arch. Toxicol.* **2018**, 92, 705–716.

Qin, X. D.; Zhou, Y.; Bloom, M. S.; Qian, Z. M.; Geiger, S. D.; Vaughn, M. G.; Chu, C.; Li, Q.; Yang, B. Y.; Hu, L. W.; Yu, Y.; Zeng, X. W.; Dong, G. H. Prenatal Exposure to PFAS, Associations with Preterm Birth and Modification by Maternal Estrogen Levels: The Maoming Birth Study. Environ. *Health Perspect.* **2023**, 131 (11), 117006.

Rickard, B. P.; Rizvi, I.; Fenton, S. E. Per- and Poly-Fluoroalkyl Substances (PFAS) and Female Reproductive Outcomes: PFAS Elimination, Endocrine-Mediated Effects, and Disease. *Toxicology* **2022**, 465, 153031. https://pubmed.ncbi.nlm.nih.gov/34774661/

Rumph, J. T.; Stephens, V. R.; Martin, J. L.; Brown, L. K.; Thomas, P. L.; Cooley, A.; Osteen, K. G.; Bruner-Tran, K. L. Uncovering Evidence: Associations between Environmental Contaminants and Disparities in Women's Health. *Int. J. Environ. Res. Public Health* **2022**, 19 (3), 1257. https://doi.org/10.3390/ijerph19031257

Saliu, T. D.; Sauvé, S. A Review of Per- and Polyfluoroalkyl Substances in Biosolids: Geographical Distribution and Regulations. *Front. Environ. Chem.* **2024**, 5, 1383185.

Säve-Söderbergh, M.; Gyllenhammar, I.; Schillemans, T.; Lindfeldt, E.; Vogs, C.; Donat-Vargas, C.; Halldin Ankarberg, E.; Glynn, A.; Ahrens, L.; Helte, E.; Åkesson, A. Per- and Polyfluoroalkyl Substances (PFAS) and Fetal Growth: A Nation-Wide Register-Based Study on PFAS in Drinking Water. *Environ. Int.* **2024**, 187, 108727. https://doi.org/10.1016/j.envint.2024.108727

Schlummer, M.; Gruber, L.; Fiedler, D.; Kizlauskas, M.; Müller, J. Detection of Fluorotelomer Alcohols in Indoor Environments and Their Relevance for Human Exposure. *Environ. Int.* **2013**, 57–58, 42–49.

Shen, J.; Mao, Y.; Zhang, H.; Lou, H.; Zhang, L.; Moreira, J. P.; Jin, F. Exposure of Women Undergoing In-Vitro Fertilization to Per- and Polyfluoroalkyl Substances: Evidence on Negative Effects on Fertilization and High-Quality Embryos. *Environ. Pollut.* **2024**, 359, 124474. https://doi.org/10.1016/j.envpol.2024.124474

Singer, A. B.; Whitworth, K. W.; Haug, L. S.; Sabaredzovic, A.; Impinen, A.; Papadopoulou, E.; Longnecker, M. P. Menstrual Cycle Characteristics as Determinants of Plasma Concentrations of Perfluoroalkyl Substances (PFASs) in the Norwegian Mother and Child Cohort (MoBa Study). *Environ. Res.* **2018**, 166, 178–185.

Skogheim, T. S.; Villanger, G. D.; Weyde, K. V. F.; Engel, S. M.; Surén, P.; Øie, M. G.; Skogan, A. H.; Biele, G.; Zeiner, P.; Øvergaard, K. R.; Haug, L. S.; Sabaredzovic, A.; Aase, H. Prenatal Exposure to Perfluoroalkyl Substances and Associations with Symptoms of Attention-Deficit/Hyperactivity Disorder and Cognitive Functions in Preschool Children. *Int. J. Hyg. Environ. Health* **2020**, 223 (1), 80–92.

Starnes, H. M.; Rock, K. D.; Jackson, T. W.; Belcher, S. M. A Critical Review and Meta-Analysis of Impacts of Per- and Polyfluorinated Substances on the Brain and Behavior. *Front. Toxicol.* **2022**, 4, 881584. https://doi.org/10.3389/ftox.2022.881584

Straková, J.; Schneider, J.; Cingotti, N. Throwaway Packaging, Forever Chemicals: European Wide Survey of PFAS in Disposable Food Packaging and Tableware; **2021**. https://www.env-health.org/wp-content/uploads/2021/05/FINAL_pfas_fcm_study_web.pdf

Sudarshan, K.; Jason, D.; Sushil, R.; Kanel; Mallikarjuna, N.; Nadagouda; Ryan, W.; Cawdrey, B. A.; Garrett, C.; Struckhoff, R. W. Per- and Polyfluoroalkyl Substances in Water and Wastewater: A Critical Review of Their Global Occurrence and Distribution. *Sci. Total Environ.* **2022**, 809 (25), 151003.

Szilagyi, J. T.; Freedman, A. N.; Kepper, S. L.; Keshava, A. M.; Bangma, J. T.; Fry, R. C. Per- and Polyfluoroalkyl Substances Differentially Inhibit Placental Trophoblast Migration and Invasion In Vitro. *Toxicol. Sci.* **2020**, 175 (2), 210–219.

Travis, A. S. The Discovery and Analysis of PFAS ('Forever Chemicals') in Human Blood and Biological Materials. *Substantia* **2024**, 8 (1), 7–24.

United States Environmental Protection Agency (USEPA). Per- and Polyfluoroalkyl Substances (PFAS) in Pesticide and Other Packaging; **2022**. https://www.epa.gov/pesticides/pfas-packaging#packaging

Wang, Y. K.; Bashashati, A.; Anglesio, M. S.; Cochrane, D. R.; Grewal, D. S.; Ha, G.; McPherson, A.; Horlings, H. M.; Senz, J.; Prentice, L. M.; Karnezis, A. N.; Lai, D.; Aniba, M. R.; Zhang, A. W.; Shumansky, K.; Siu, C.; Wan, A.; McConechy, M. K.; Li-Chang, H.; Tone, A.; Shah, S. P. Genomic Consequences of Aberrant DNA Repair Mechanisms Stratify Ovarian Cancer Histotypes. *Nat. Genet.* **2017**, 49 (6), 856–865.

Whitehead, H. D.; Venier, M.; Wu, Y.; Eastman, E.; Urbanik, S.; Diamond, M. L.; Shalin, A.; Schwartz-Narbonne, H.; Bruton, T. A.; Blum, A.; Wang, Z.; Green, M.; Tighe, M.; Wilkinson, J. T.; McGuinness, S.; Peaslee, G. F. Fluorinated Compounds in North American Cosmetics. *Environ. Sci. Technol. Lett.* **2021**, 8, 538–544.

Wikstrom, S.; Hussein, G.; Lingroth, K. A.; Lindh, C. H.; Bornehag, C. G. Exposure to Perfluoroalkyl Substances in Early Pregnancy and Risk of Sporadic First Trimester Miscarriage. *Sci. Rep.* **2021**, 11, 3568. https://www.nature.com/articles/s41598-021-82748-6

Wikström, S.; Lin, P. I.; Lindh, C. H.; Shu, H.; Bornehag, C. G. Maternal Serum Levels of Perfluoroalkyl Substances in Early Pregnancy and Offspring Birth Weight. *Pediatr. Res.* **2020**, 87 (6), 1093–1099.

Xie, X.; Weng, X.; Liu, S. Perfluoroalkyl and Polyfluoroalkyl Substance Exposure and Association with Sex Hormone Concentrations: Results from the NHANES 2015–2016. *Environ. Sci. Eur.* **2021**, 33, 69. https://doi.org/10.1186/s12302-021-00508-9

Xu, B.; Qiu, W.; Du, J.; Wan, Z.; Zhou, J. L.; Chen, H.; Liu, R.; Magnuson, J. T.; Zheng, C. Translocation, Bioaccumulation, and Distribution of Perfluoroalkyl and Polyfluoroalkyl Substances (PFASs) in Plants. *iScience* **2022**, 25 (4), 104061. https://doi.org/10.1016/j.isci.2022.104061

Xu, Y.; Fletcher, T.; Pineda, D.; Lindh, C. H.; Nilsson, C.; Glynn, A. Serum Half-Lives for Short- and Long-Chain Perfluoroalkyl Acids after Ceasing Exposure from Drinking Water Contaminated by Firefighting Foam. *Environ. Health Perspect.* **2020**, 128, 77004.

Xuping, G.; Wanze, N.; Sui Zhu; Yanxin, W.; Yunfeng, C.; Junrong, M.; Yanhua, L.; Jinlong, Q.; Yanbin, Y.; Pan, Y.; Chaoqun, L.; Fangfang, Z. Per- and Polyfluoroalkyl Substances Exposure during Pregnancy and Adverse Pregnancy and Birth Outcomes: A Systematic Review and Meta-Analysis. *Environ. Res.* **2021**, 201, 111632.

Yang, M.; Lee, Y.; Gao, L.; Chiu, K.; Meling, D. D.; Flaws, J. A.; Warner, G. R. Perfluorooctanoic Acid Disrupts Ovarian Steroidogenesis and Folliculogenesis in Adult Mice. *Toxicol. Sci.* **2022**, 186 (2), 260–268.

Yao, P. L.; Ehresman, D. J.; Rae, J. M. C.; Chang, S. C.;

Frame, S. R.; Butenhoff, J. L.; Kennedy, G. L.; Peters, J. M. Comparative in Vivo and in Vitro Analysis of Possible Estrogenic Effects of Perfluorooctanoic Acid. *Toxicology* **2014**, 326, 62–73.

Zeng, L. H.; Rana, S.; Hussain, L.; Asif, M.; Mehmood, M. H.; Imran, I.; Younas, A.; Mahdy, A.; Al-Joufi, F. A.; Abed, S. N. Polycystic Ovary Syndrome: A Disorder of Reproductive Age, Its Pathogenesis, and a Discussion on the Emerging Role of Herbal Remedies. *Front. Pharmacol.* **2022**, 13, 874914. https://doi.org/10.3389/fphar.2022.874914

Zhan, W.; Qiu, W.; Ao, Y.; Zhou, W.; Sun, Y.; Zhao, H.; Zhang, J. Environmental Exposure to Emerging Alternatives of Per- and Polyfluoroalkyl Substances and Polycystic Ovarian Syndrome in Women Diagnosed with Infertility: A Mixture Analysis. Environ. *Health Perspect.* **2023**, 131 (5), 57001. https://doi.org/10.1289/EHP11814

Zhang, S.; Tan, R.; Pan, R.; Xiong, J.; Tian, Y.; Wu, J. Association of Perfluoroalkyl and Polyfluoroalkyl Substances with Premature Ovarian Insufficiency in Chinese Women. *J. Clin. Endocrinol. Metab.* **2018**, 103, 2543–2551.

Zhang, W.; Cao, H.; Liang, Y. Plant Uptake and Soil Fractionation of Five Ether-PFAS in Plant-Soil Systems. Sci. Total Environ. **2021**, 771, 144805. https://pubmed.ncbi.nlm.nih.gov/33529820/

Zhang, Z.; Sarkar, D.; Biswas, J. K.; Datta, R. Biodegradation of Per- and Polyfluoroalkyl Substances (PFAS): A Review. Bioresource Technol. **2022**, 344, 126223. https://doi.org/10.1016/j.biortech.2021.126223

Zheng, Q.; Yan, W.; Gao, S.; Li, X. The Effect of PFAS Exposure on Glucolipid Metabolism in Children and Adolescents: A Meta-Analysis. *Front. Endocrinol.* **2024**, 15. https://doi.org/10.3389/fendo.2024.1261008

Zhou, W.; Zhang, L.; Tong, C.; Fang, F.; Zhao, S.; Tian, Y.; Tao, Y.; Zhang, J. Plasma Perfluoroalkyl and Polyfluoroalkyl Substances Concentration and Menstrual Cycle Characteristics in Preconception Women. *Environ. Health Perspect.* **2017**, 125 (6), 067012. https://doi.org/10.1289/EHP1203

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Research Article

A Facile Synthesis of Montmorillonite Supported $CdAl_2O_4$ Nanocomposites with Photocatalytic and Hydrophobic Properties

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Abstract. The hydrothermal co-precipitation method was successfully employed to synthesize $CdAl_2O_4$ supported by natural clay, montmorillonite to reduce the toxicity of solid and liquid wastes that cause serious harm to livestock and humans due to the discharge of dye wastes into water bodies. Characterization of the montmorillonite/ $CdAl_2O_4$ (*MMT/*$CdAl_2O_4$) composite was carried out using various techniques, including X-ray powder diffraction (XRD), scanning electron microscopy (SEM), high-resolution transmission electron microscopy (HR-TEM), photoluminescence spectroscopy (PL), diffuse reflectance spectroscopy (DRS), and BET surface area analysis. Experimental results indicate that $CdAl_2O_4$ supported by montmorillonite at a 9% wt concentration exhibited superior photocatalytic activity compared to undoped $CdAl_2O_4$ in the removal of the azo dye rhodamine-B (rh-B) under solar light irradiation. The FE-SEM images clearly reveal the formation of nanoclusters and nanoflakes interconnected via edge-to-flat-surface conjunction. Furthermore, BET analysis demonstrated that the surface area of MMT/$CdAl_2O_4$ surpasses that of undoped $CdAl_2O_4$. Remarkably, montmorillonite-supported $CdAl_2O_4$ exhibited excellent photocatalytic activity for up to four consecutive uses. A notable feature of montmorillonite/ $CdAl_2O_4$ is its high hydrophobicity, as evidenced by a contact angle of 113.8°. This hydrophobic nature is particularly advantageous in the production of self-cleaning materials.

Keywords: Montmorillonite clay; $CdAl_2O_4$ nanoflakes; Rhodamine-B; Photodegradation; Contact angle

1. INTRODUCTION

In recent decades, heterogeneous photocatalysis has gained recognition as an eco-friendly method for energy conversion. Semiconductor photocatalysts, in particular, have shown great promise for environmentally cleaning by facilitating the degradation of organic contaminants [1-3]. Semiconductor-mediated photocatalysis is one of the most successful methods available for cleaning and dye purification procedures. Metal oxide coupling has been proven to be an effective method to improve the photocatalytic activities of nanomaterials, which can reduce the recombination of the photogenerated electron/hole pairs and lengthen their lifetime [4,5]. A number of surface active materials such as activated carbon, zeolites, graphene and clay materials have been used as photocatalyst supports [6-8].

Natural clay materials, such as coal fly ash, bentonite, kaolinite, sepiolite, and montmorillonite, have demonstrated excellent support for semiconductor oxide materials in various photocatalytic studies [9-14]. Due its softness, plasticity, porosity, tangibility, pliability, and climatic adaptability, all at affordable cost, were viable characteristics to recognize its usefulness, and introduced several products. Among the many kinds of clay minerals, montmorillonite has been recognized as an effective adsorbent due to its high surface area, expandability, and swellability, montmorillonite within the smectite group has been found to be an effective adsorbent for the removal of dyes and cations. The structure and cation exchange capacity (CEC) of montmorillonite are linked to its adsorption capability. Because of its structure, montmorillonite clay interacts with cations and accommodates them in the voids between layers, giving it the properties of a molecular sieve. In general, the mineral's net negative charge determines Montmorillonite's adsorption capacity. MMT can also be used as a coagulant in ponds when it is finely powdered. When added to water, it causes the water to become "clouded," draws in tiny particles, and eventually sinks to the bottom. Although MMT effectively absorbs heavy metals, its impact on human health is still unknown [16-20].

Spinel oxides, denoted by the formula AB_2O_4 with trivalent (B) and divalent (A) cations occupy octahedral and tetrahedral positions, while oxygen ions create a cubic unit cell. These materials are known for their chemical and thermal stability, finding applications in various fields like humidity sensors, ceramic catalysis, and magnetic materials [21]. Spinel compounds (general formula AB_2O_4) are part of a large family of inorganic materials and have become the subject of extensive research in numerous industrial processes, as well as in the realm of nanoscience and technology [22]. Their unique properties, such as high electrical resistivity, high coercivity, and moderate saturation magnetization, make them promising candidates for advanced applications in drug delivery systems, power electronics, sensors, catalysts, and spintronics [23-25]. Furthermore, aluminum spinel can be used as a suitable charge carrier and photocatalyst for a range of applications due to its exceptional optical, thermal, electrical, and magnetic properties. Cadmium aluminate ($CdAl_2O_4$) is a significant spinel material and it has crucial applications in microwave devices, sensors, light helmets, light-emitting diodes, luminescence, pigment, gas sensors, and photocatalysts [26-29].

Rafiq et al. in their review assessed that, the ability of several nanomaterials to clean up dye-contaminated aquatic systems by focussing on essential parameters that affect the photocatalytic decolorisation of dyes. Numerous semiconductor nanocatalysts have demonstrated the ability to function as photocatalysts in the treatment of wastewater that contains textile colours. Operational characteristics are significantly impacted by photocatalyst efficiency. The nature of the sample to be degraded must be investigated because the impact of different parameters has occasionally proven contentious. Higher pH causes some reactive dyes to deteriorate, while lower pH causes others to do so. For photocatalytic dye degradation in wastewater, the reaction should therefore be conducted at the proper pH. The photocatalytic decolorisation of any dye is discovered to be influenced by a variety of factors, including reaction temperature, photocatalyst concentration, light intensity and irradiation duration, and dopant impact [30].

The co-precipitation technique has been widely used to prepare nanoparticles. It entails the simultaneous precipitation of multiple substances from a solution. With the help of a base and a solvent, metal hydroxides precipitate from salt precursors in an eco-friendly and economical manner. Co-precipitation is an easy method with controlled size distribution, high yield, and solvents that are safe for the environment for creating inorganic and metal-based nanoparticles. Given these advantages, we utilized co-precipitation techniques to prepare $CdAl_2O_4$. Herein, we successfully synthesized montmorillonite-supported $CdAl_2O_4$ and assessed its photocatalytic ability through the removal of rhodamine-B by solar-light radiation. Additionally, we investigated the hydrophobic nature of montmorillonite/$CdAl_2O_4$.

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

$Al(NO_3)_3.9H_2O$, $Cd(CH_3COO)_2.2H_2O$, NaOH, EtOH and Montmorillonite clay $(Na,Ca)_{0.3}$ $(Al,Mg)_2Si_4O_{10}(OH)_2.nH_2O)$ were supplied by Himedia chemicals, Rhodamine-B (Rh-B; $C_{28}H_{31}ClN_2O_3$; M.W- 479·01-CAS 81-88-9)) dye was supplied by CDH (P) Limited. The purity of all the purchased chemicals were AR grade (99 % purity). Distilled water was used over the experiments.

2.2 Characterization Techniques

X-Ray diffraction (XRD) pattern of catalysts were obtained using a Siemens D5005 diffractometer with Cu Kα (λ = 0.151418 nm) radiation. The diffractograms were recorded in 2θ range between 10 and 80° in steps of 0.02° with count time of 20s at each point. The morphology of catalyst was examined using Model ULTRA-55 field emission scanning electron microscope (FE-SEM). Samples were mounted on a gold platform placed in the scanning electron microscope for taking images at various magnifications. HR-TEM images were taken using the 200kV Ultra High Resolution Transmission Electron Microscope JEOL-2010, having high resolution Optical microscope and Leica microscope. A small quantity of catalyst suspension was dropped onto copper grids with holey carbon film. The grids were dried under natural conditions and examined. The diffuse reflectance spectra of all the catalysts were recorded in Shimadzu UV 2450 model equipped with an integrating sphere and using powdered $BaSO_4$ as a reference. The photoluminescence (PL) spectrum at room temperature was recorded using a Perkin-Elmer LS 55 fluorescence spectrometer. The specific surface areas of the catalysts were determined using a Micromeritics ASAP 2020 sorption analyzer. The samples were degassed at 423K for 12 hours and analysis was performed at 77K with N_2 gas as the adsorbate. The Brunauer-Emmett-Teller (BET) multipoint method least-square fit provided the specific surface area. The hydrophobicity of catalyst coating was obtained from the water contact angle. A drop shape analyser (DSA) (Kruss GmbH Germany) was used to record the contact angle.

2.3 Synthesis of montmorillonite supported $CdAl_2O_4$

The synthesis of both pure $CdAl_2O_4$ and *MMT* supported $CdAl_2O_4$ samples were carried out through a co-precipitation method. Initially, 50mL of 0.5M $Al(NO_3)_3.9H_2O$ and 50mL of 0.3M $Cd(CH_3COO)_2.2H_2O$ were separately dissolved in 50mL of distilled water. After that, the $Cd(CH_3COO)_2.2H_2O$ solution was slowly added to $Al(NO_3)_3.9H_2O$ solution with vigorous stirring. Subsequently, 0.500g (9% wt) of MMT powder, mixed with 10mL of ethanol, was introduced to the previous solutions with constant stirring. Following this, 0.2M of NaOH was dissolved in 30mL of distilled water followed by slowly adding to the combined aqueous solutions. The resulting solution was stirred at 80°C for 5 hours. The resulting product was collected by washing with ethanol and distilled water, and dried at 100°C for 5 hours. Finally, the *MMT s*upported $CdAl_2O_4$ nanoparticles were calcinated for 6 hours at 700°C, denoted as 9% wt *MMT/*$CdAl_2O_4$ for easy understanding. The pure $CdAl_2O_4$ nanoparticles and 5% wt *MMT* supported $CdAl_2O_4$ were synthesized by the similar process for comparison.

2.4 Photocatalytic degradation

Photocatalytic degradation experiments were performed in summer days from 11 am to 2 pm under a direct sunlight illumination(Tiruvannamalai, Tamilnadu, India). A transparent borosilicate glass tube with 40 cm in height and 20mm in diameter was employed as reaction vessel. After 30 minutes of magnetic stirring in the dark, the dye and *MMT/*$CdAl_2O_4$ solutions reached equilibrium of adsorption and desorption. The photodegradation process was carried out outdoors. To ensure thorough mixing of the reaction solution and to introduce oxygen, a pump was used to continuously aerate a 50mL dye solution containing *MMT/*$CdAl_2O_4$. There was no significant solvent evaporation during the lighting period. The first sample was collected during the adsorption phase while it was still dark. A two-milliliter sample was retained, and at predetermined intervals, it was centrifuged and diluted to ten milliliters. The solar light intensity, evaluated with the LT Lutron LX-10/A Digital Lux meter, was I_{solar} = 1250×100Lux ± 10.

2.5 Contact angle measurements

Water contact angles were measured on a Drop Shape Analyzer (DSA) from Kruss GmbH in Germany. The volume of the water droplet was about 4μL based on at least four measurements. The average of these values represented the substrate's water contact angle (WCA). Spin coating was successfully applied to glass substrates to produce *MMT/*$CdAl_2O_4$-modified silane coatings at room temperature. *MMT/*$CdAl_2O_4$ photocatalyst-coated substrates were heated at 125°C for 2 hours in a controlled furnace to confirm the densification of the gel network.

3. RESULTS AND DISCUSSION

3.1 Characterization of nanocomposite

X-ray diffraction (XRD) measurement was conducted to assess the crystallinity of the synthesized photocatalysts. Figure 1a-c present the XRD patterns for undoped $CdAl_2O_4$, 5% wt *MMT/*$CdAl_2O_4$, and 9% wt *MMT/*$CdAl_2O_4$ photocatalysts. In Figure 1a, the undoped $CdAl_2O_4$ photocatalyst displays the diffraction peaks at 13.75°, 19.25°, 26.37°, 31.02°, 33.56°, 37.50°, 38.85°, 55.18°, and 67.60°, which were corresponding to the (012), (300), (131), (042), (241), (431), (223), (520), and (713) planes of monoclinic primitive $CdAl_2O_4$ (JCPDS No. 34-0071). [31, 32]. Fig. 1b represents the diffraction peaks of montmorillonite clay alone. Meanwhile, in Figure 1c and 1d, newly formed peaks indicated by star symbols corresponded well with raw *MMT* for SiO_2, Fe_2O_3, MgO and Al_2O_3, confirming the presence of MMT clay in the *MMT*/$CdAl_2O_4$ photocatalysts [33].

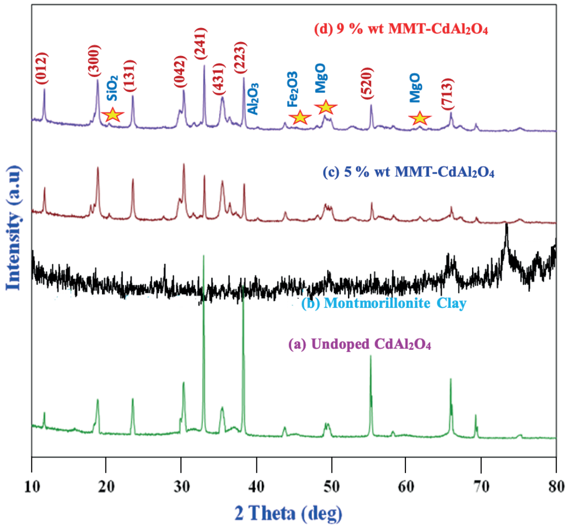

Fig. 1 XRD patterns of the (a) undoped $CdAl_2O_4$ (b) Montmorillonite clay (c) 5% wt *MMT*/$CdAl_2O_4$ and (d) 9% wt *MMT*/$CdAl_2O_4$ photocatalysts

A typical diffraction peak of montmorillonite clay is observed around 7.25° corresponding to basal spacing of 13.46A° (Fig. SI 1). And after intercalation of $CdAl_2O_4$ this peak disappears. Importantly, no crystalline impurities or typical diffraction peaks were observed, indicating the good purity of the synthesized photocatalysts. Furthermore, the Scherrer equation was utilized to calculate the crystallite sizes of $CdAl_2O_4$.

$$\Phi = \frac{K\lambda}{\beta cos\theta} \tag{1}$$

In this equation, Φ, λ, K, β and θ represents the crystalline size, the wavelength of the X-ray used, the shape factor, the full line width at the half-maximum height of the peak, and the Bragg angle, respectively. Using this equation, the average crystallite size of hexagonal $CdAl_2O_4$ was determined to be 36.8 nm.

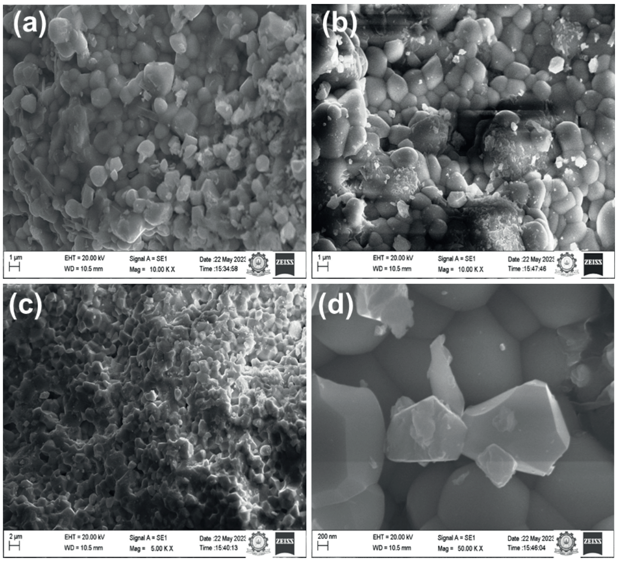

Fig. 2 FE-SEM images of the 9% wt *MMT*/$CdAl_2O_4$ photocatalyst at various magnifications: (a, b) 1 µm (c) 2 µm and (d) 200 nm

The surface structure and particle sizes of the synthesized samples were investigated using scanning electron microscopy (SEM) and high-resolution transmission electron microscopy (HR-TEM) techniques. SEM images of the 9% wt *MMT*/$CdAl_2O_4$ photocatalyst are presented in Figure 2a-d, revealing a flake-like morphology with intermittent nanosponge like particle. The presence of MMT particles is likely responsible for the observed surface morphologies. The elements present in the montmorillonite clay were confirmed by EDAX image (Fig. SI 3). The microstructures exhibit a nanosponge-like structure with high porosity. Micro-size hierarchical shape composed of nano-sized building blocks offer several advantages, including numerous mesopores that enhance the transfer of organic materials.

Moreover, Figure 3a-d displays HR-TEM images of 9% wt *MMT*/$CdAl_2O_4$ photocatalyst, showing spherical and hexagonal particle shapes with uniform element distribution. The dark areas in the images indicate the presence of MMT clay on the surface of $CdAl_2O_4$. From the lattice fringes (Figure 3c), the d spacing value calculated as 0.214 nm. The selected area electron diffraction (SAED) image in (Figure 3d) confirms the occurrence of (131) plane of $CdAl_2O_4$ structure.

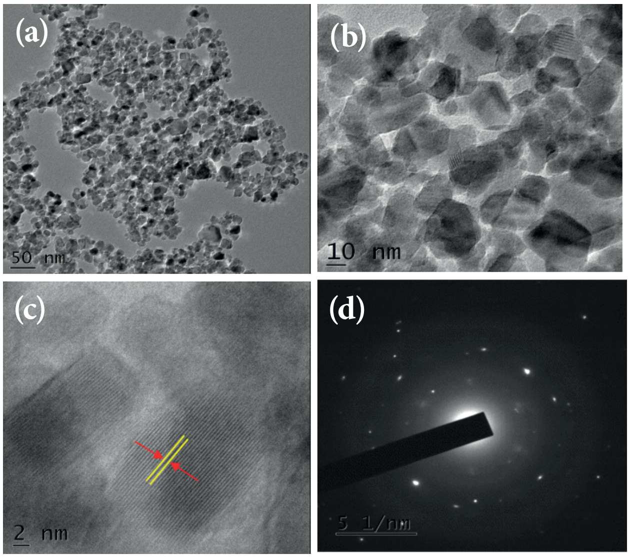

Fig. 3 HR- TEM images of the 9% wt $MMT/CdAl_2O_4$ photocatalyst at (a) 50 nm, (b) 10 nm and (c) Lattice fringes at 2 nm and (d) SAED pattern on 5 1/nm

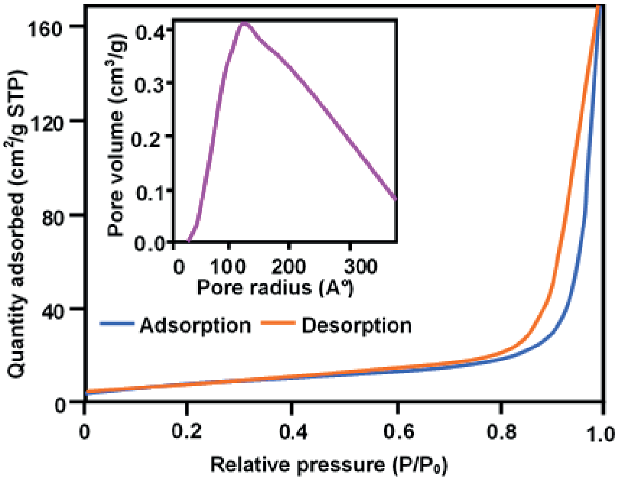

Fig. 4 BET adsorption–desorption isotherms and pore size distribution plots of the 9% wt $MMT/CdAl_2O_4$ photocatalyst

The pore structure and surface area of 9% wt $MMT/CdAl_2O_4$ photocatalystwere examined using nitrogen adsorption–desorption isotherms. The Barrett–Joyner–Halenda (BJH) technique was employed to analyze the pore size distribution. Notably, as shown in Figure 4, a significant enhancement in the adsorption volume of N_2 in the P/P_0 range of 0.65 to 0.90, indicating good homogeneity and macroporous size in the catalyst. In addition, the average pore radius, as indicated by the pore size distribution plot presented in the insert of Figure 4, is 125.7Å for the 9% wt $MMT/CdAl_2O_4$ and bare $CdAl_2O_4$ has pore radius of 90.23Å. The resultant 9% wt $MMT/CdAl_2O_4$ photocatalyst exhibits a large specific surface area of $42.15 m^2g^{-1}$, compared to undoped $CdAl_2O_4$ ($20.4 m^2g^{-1}$) contributing to its improved photocatalytic activity.

To gain further insight into the photocatalytic mechanism, various factors, including optical absorption, emission properties, and specific area, were considered. An efficient material requires an appropriate bandgap to absorb light efficiently, generating charge carriers that catalyze the breakdown of dye molecules. In addition, the diffuse reflectance spectra of undoped $CdAl_2O_4$ and 9% wt $MMT/CdAl_2O_4$ photocatalysts are shown in Figure 5a and 5b, respectively.

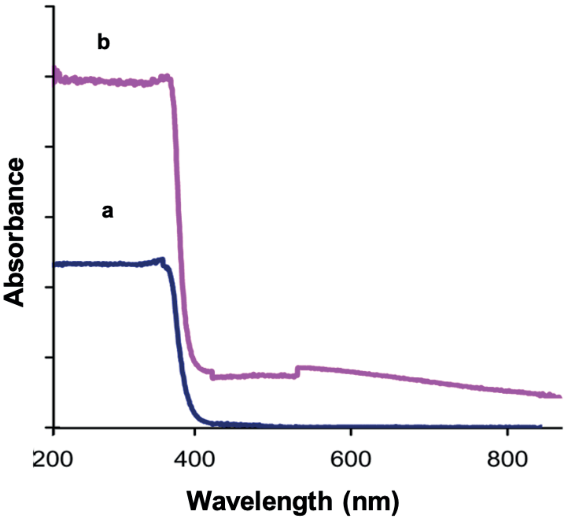

Fig. 5: Diffuse reflectance spectra of the (a) undoped $CdAl_2O_4$ and (b) 9 % wt $MMT/CdAl_2O_4$ photocatalysts

In the UV region, 9% wt $MMT/CdAl_2O_4$ photocatalyst exhibits double the intensity of UV absorption compared to undoped $CdAl_2O_4$. The addition of 9% wt MMT results in a significant rise in the resultant peak intensities and a little decrease in E_g values. The energy gap of $MMT/CdAl_2O_4$ nanoparticles is computed using the Tauc plot, which is afforded in the following equation

$$\alpha = A(h\nu - Eg)^{1/2} / h\nu$$

Where Eg, α, A and hν signify energy gap, photon energy, absorption coefficient, and the proportionality constant, The Eg of all synthesized materials were computed by linear fitting $(\alpha h\nu)^2$ versus photon (hν) energy as 3.35 [30, 32] and 2.81eV for undoped $CdAl_2O_4$ and $MMT/CdAl_2O_4$ nanoparticles respectively as displayed in Fig. SI.2

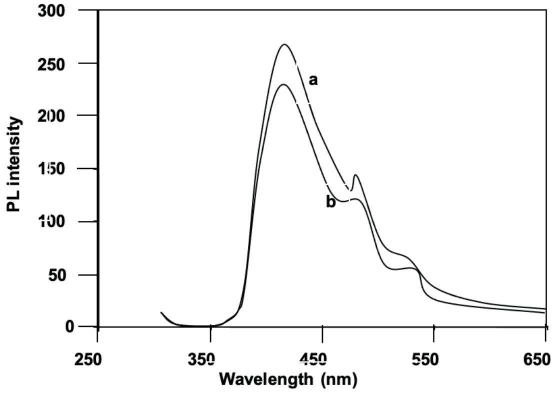

Fig. 6 Photoluminescence spectra of the (a) undoped $CdAl_2O_4$ and (b) 9% wt $MMT/CdAl_2O_4$ photocatalysts

In addition, Figure 6a and 6b display the photoluminescence spectra of undoped $CdAl_2O_4$ and 9% wt $MMT/CdAl_2O_4$ photocatalysts, respectively. The photoluminescence intensity of 9 % wt $MMT/CdAl_2O_4$ photocatalyst is lower than that of undoped $CdAl_2O_4$, indicating reduced electron-hole pair recombination due to the presence of loaded MMT on $CdAl_2O_4$. This reduction in fluorescence intensity contributes to enhanced photocatalytic activity and reduced electron-hole pair recombination.

3.2. Photocatalytic Study

Figure 7 illustrates the photocatalytic activity of the synthesized undoped $CdAl_2O_4$, 5% wt $MMT/CdAl_2O_4$ and 9% wt $MMT/CdAl_2O_4$ towards the removal of rh-B dye by direct solar-light radiation. The dye is resistant to self photolysis and for the same experiment with $MMT/CdAl_2O_4$ in the dark, a decrease (10%) in dye concentration was observed due to the adsorption of dye on the catalyst. The degradation outcomes display there is no significant changes in dye concentration under the absence of light and catalyst. With 9% wt $MMT/CdAl_2O_4$ photocatalyst, rh-B dye almost completely degrades within 45 minutes. However, under the same conditions, undoped $CdAl_2O_4$ and 5% $MMT/CdAl_2O_4$ achieved 68% and 78.5% degradation of rh-B dye, respectively. The montmorillonite clay shows nearly 35% of adsorption. These results indicate that the prepared 5% $MMT/CdAl_2O_4$ photocatalystis more effective than other catalysts in degrading rh-B when exposed to direct solar-light. This underscores higher photocatalytic activity of $MMT/CdAl_2O_4$ photocatalyst, attributed to the loaded MMT clay. Under UV light irradiation $MMT/CdAl_2O_4$ produce maximum dye degradation percentage compare other pure MMT clay and undoped $CdAl_2O_4$ (Fig. SI.4).

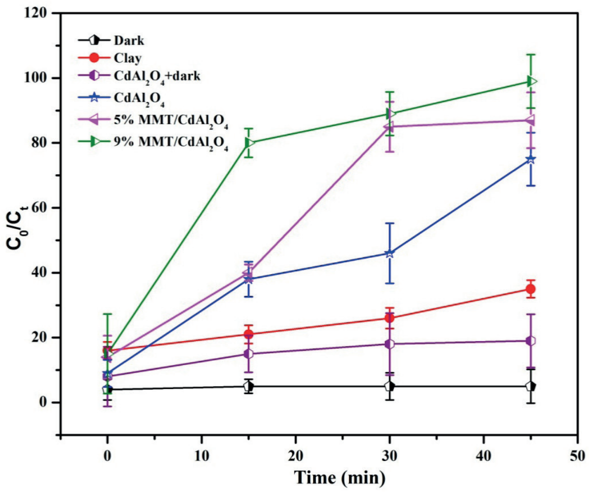

Fig. 7 Photodegradation of rh–B dye with different catalysts: dye concentration = 3×10^{-4} M, catalyst suspended = 3 g L^{-1}, pH = 7, airflow rate = 8.1 mL s^{-1}, I_{solar} = 1250×100 Lux ± 10. Irradiation time = 45 min)

Under visible irradiation, the e^- on MMT doped $CdAl_2O_4$ sample are photoexcited into their particular (CB), leaving h^+ on the valence band (VB), thereby making photoexcited charge carriers. The photoexcited e^- on the CB of *MMT* doped $CdAl_2O_4$ rapidly transformed into that of trapping level of $MMT/CdAl_2O_4$, while the photoexcited h^+ on the VB of $MMT/CdAl_2O_4$ reacts with the water molecules to produce the $OH^\cdot$ radicals and generated e^- near to CB of the trap state can interact with the dissolved O_2 to produce $O_2^{\cdot-}$ (superoxide radicals). Finally, these two radicals $O_2^{\cdot-}$ and $OH^\cdot$ react with the dye molecule and convert the dye into CO_2 and H_2O. Further, the increased degradation efficiency can be attributed to two primary factors such as (i) MMT's ability to absorb UV light, doubling the absorption compared to undoped $CdAl_2O_4$ and (ii) the larger surface area of $MMT/CdAl_2O_4$ photocatalyst compared to undoped $CdAl_2O_4$ photocatalyst. In fact, MMT is an electron acceptor due to the presence of Lewis acids. The electrons from these aluminum sites are then transferred to the O_2 molecules in the $CdAl_2O_4$ solution, which result in delay in the recombination reaction. Additionally, the presence of metal oxides in loaded MMT contributes to this electron transfer and may lead to the formation of sporadic energy levels, reducing the band gap energy and preventing electron-hole recombination [34]. A proposed mechanism in Figure 8 views MMT as an impurity in the attached with $CdAl_2O_4$ photocatalyst.

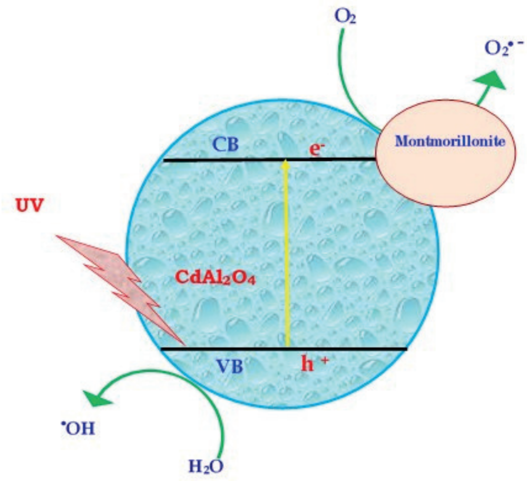

Fig. 8 A possible photodegradation mechanism of the $MMT/CdAl_2O_4$ photocatalyst

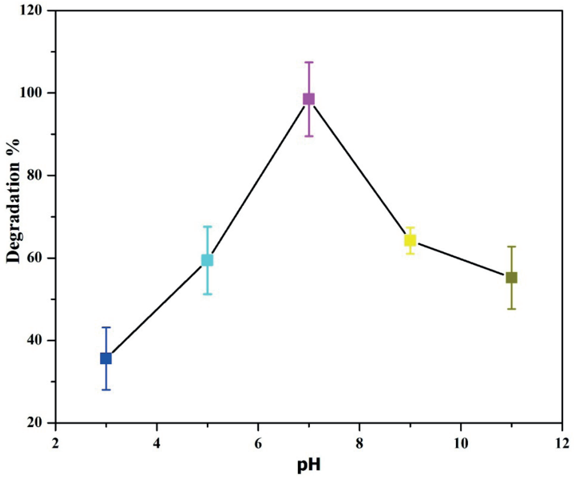

Fig. 9 Effect of solution pH on rh–B degradation using 9% wt $MMT/CdAl_2O_4$ photocatalyst (dye concentration = 3×10^{-4}M, catalystsuspended = $3gL^{-1}$, airflow rate = $8.1mLs^{-1}$, I_{solar} = 1250 ×100 Lux ± 10, irradiation time = 45 min)

The pH of the solution significantly affects photocatalytic degradation. Figure 9 illustrates the impact of pH on the photocatalytic removal of rh-B within the pH range of 3–11. The results indicate that increasing the pH from 3 to 7 enhances rh-B removal ability, with a neutral pH of 7 being the optimal condition for effective rh-B removal using the 9% wt $MMT/CdAl_2O_4$ photocatalyst.

To evaluate the reusability of the 9% wt $MMT/CdAl_2O_4$ photocatalyst for photoreactions, the catalyst was removed and cleaned with methanol following the complete dye degradation. The recovered catalyst, after drying for 90 minutes at 100°C, was utilized in the subsequent cycle. Five consecutive cycles of rh-B degradation under solar light demonstrated the photocatalyst's good stability and reusability (Figure 10). Until the fourth cycle, there is a slight decrease in degradation; after that, no further change is observed. In the fourth run, at 45 minutes of radiation, a degradation efficiency of 95.5% was achieved. These findings demonstrate that 9% wt $MMT/CdAl_2O_4$ photocatalyst exhibits excellent stability and reusability.

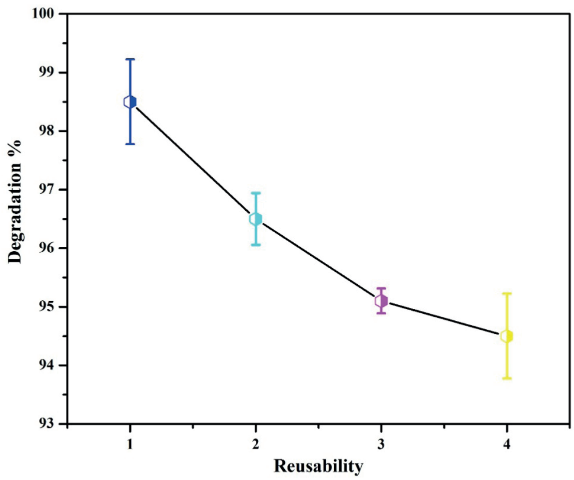

Fig. 10 Reusability of 9 % wt $MMT/CdAl_2O_4$ photocatalyston rh–B degradation (dye concentration = 3×10^{-4} M, pH =7, catalyst suspended= $3gL^{-1}$, airflow rate = $8.1mLs^{-1}$, I_{solar}= 1250 ×100Lux ± 10, irradiation time= 45min)

3.3 Hydrophobicity property

Water-repellent materials find a wide range of applications, and this property was studied using water contact angle assessments (Figure 11). The contact angle of an uncoated glass slide is 37.2° (Figure 11a), which increases progressively with increasing TEOS values (58.4°, Figure 11b), TEOS + $CdAl_2O_4$ (80.5°, Figure 11c), TEOS + 5% wt $MMT/CdAl_2O_4$ (100.2°, Figure 11d), and TEOS + 9% wt $MMT/CdAl_2O_4$ (113.8°, Figure 11e). The extreme hydrophobicity properties of TEOS (Tetraethyl orthosilicate)-containing $MMT/CdAl_2O_4$ is evident, with the O-Si-O groups being altered and the surface becoming rougher due to the presence of 9% wt MMT with $CdAl_2O_4$. In general, the catalyst's hydrophobic nature results in water contact angle of 90° [35]. This hydrophobic nature results in a water contact angle of 113.8°, further demonstrating its extreme hydrophobicity property.

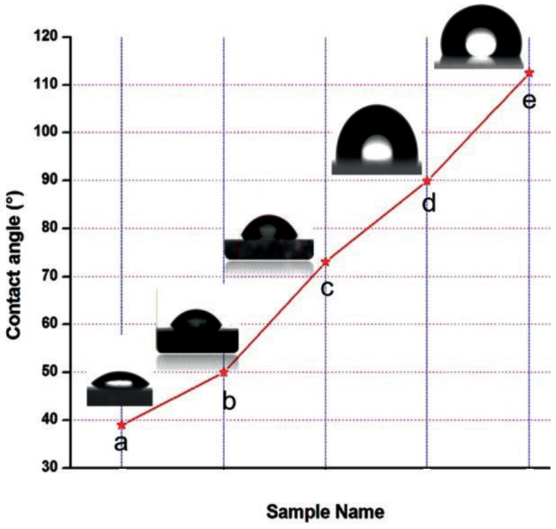

Fig. 11 Water contact angle assessments (a) uncoated glass slide, (b) TEOS coated glass slide, (c) TEOS+$CdAl_2O_4$ and (d) TEOS+5% wt $MMT/CdAl_2O_4$ and (e) TEOS/9% wt $MMT/CdAl_2O_4$

4. CONCLUSIONS

In summary, a facile hydrothermal co-precipitation technique was employed to produce montmorillonite-supported $CdAl_2O_4$ ($MMT/CdAl_2O_4$) without using any surfactants or organic solvents. The FE-SEM results showed that the as synthesized photocatalyst consists of microflakes and nanoclusters structures. The HR-TEM results confirmed the homogeneous spreading of elements, indicating the strongly attached within oxides, where particles take on hexagonal and spherical structures. Photodegradation results reveal that 9% MMT-supported $CdAl_2O_4$ is much effective at degrading rh-B dye than both raw and undoped $CdAl_2O_4$, particularly at neutral pH 7. Reduced fluorescence intensity in $MMT/CdAl_2O_4$ leads to decreased electron-hole pair recombination and improved photocatalytic activity. Moreover, the montmorillonite/$CdAl_2O_4$ showed a water contact angle of 113.8°, showing its extreme hydrophobicity nature. This study introduces a novel application for montmorillonite, naturally occurring clay, in semiconductor oxide materials for energy and environmental applications.

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SUPPLEMENTARY INFORMATION

To learn more about the characterization data of montmorillonite supported $CdAl_2O_4$ nanocomposites, refer to the supplementary information file, which contains XRD pattern of 9% wt Montmorillonite clay supported $CdAl_2O_4$, Tauc energy plot, EDAX image of Montmorillonite clay and Photodegradability of Rh–B dye with different catalysts.

DECLARATIONS

Ethical Approval
Not applicable

Competing interests
There was no indication of potential conflicts of interest by the authors.

Authors' contributions
The data was collected by K. Kalpana
The data analysis was done by Dr. K. Rajathi.

Funding
Not applicable

Availability of data and materials
The writers are unwilling to share the data they used.

REFERENCES

1. S. Boudiaf, N. Nasrallah, M. Mellal, C. Belabed, B. Belhamdi, D. Meziani, M. Trari, *Optik* **2020**, *219*, 165038.
2. G. Wu, W. Wang, L. Cai, Y. Hong, Y. Jiang, C. Wang, *React. Kinet. Mech. Cat*. **2016**, *119*, 523-535.
3. J. Li, S. Wang, G. Sun, H. Gao, X. Yu, S. Tang, Y. Wei, *Mater. Today Chem*. **2021**, *19*, 100390.
4. X. D. Hou, X. Q. Guan, Y. F. Cao, Z. M. Weng, Q. Hu, H. B. Liu, J. Hou, *Int. J. Biol. Macromol*. **2020**, *145*, 620.
5. G. Panthi, O. H. Kwon, Y. S. Kuk, K. R. Gyawali, Y. W. Park, M. Park, *Catalysts* **2020**, *10*, 348.

6. H. Fan, X. Zhao, J. Yang, X. Shan, L. Yang, Y. Zhang, *Catal. Commun*. **2012**, *29*, 29-33.
7. M. Bahrami, A. Nezamzadeh Ejhieh, *Mater. Sci. Semicond. Process* **2015**, *30*, 275-284.
8. K. Thirumalai, S. Balachandran, M. Swaminathan, *Materials Chemistry and Physic* **2016**, *183*, 191-200.
9. N. Nadeem, M. Zahid, Z. A. Rehan, M. A. Hanif, M. Yaseen, *International Journal of Environmental Science and Technology* **2021**, *19*, 3045-3060.
10. K. Thirumalai, E. T. Deva Kumar, R. Aravindhan, J. Raghava Rao, and M. Swaminathan, *Surfaces and Interfaces* **2016**, *5*, 30-38.
11. G. K. Zhang, X. M. Ding, F. S. He, X. Y. Yu, J. Zhou, Y. J. Hu, J. W. Xie, *Langmuir* **2008**, *24* (3), 1026-1030.
12. M. Mylarappa, N. Raghavendra, N. R. Bhumika, C. H. Chaithra, B. N. Nagalaxmi, K. N. Shravana Kumara, *Chem.Phys.Mater.* **2023**, *3*, 83-93.
13. M. Mylarappa, N. Raghavendra, B. S. Surendra, K. N. Shravana Kumar, S. Kantharjau, *Applied Surface Science Advances* **2022**, *10*, 100268.
14. M. Mylarappa, S. Chandruvasan, B. Thippeswamy, K. N. Shravana Kumara, S. Kantharaju, *Sustainable Chemistry for the Environment* **2023**, *2*, 100007.
15. F. Uddin, *j. intech. open* **2018**, 77987.
16. H. Liang, Z. Wang, L. M. Liao, L. Chen, Z. Li, J. Feng, *Optics*. **2017**, *136*, 44-51.
17. I. Fatimah, T Huda, *Applied Clay Science* **2013**, 74, 115-120.
18. N. Yaghmaeiyan, M. Mirzaei, R. Delghavi, *Results in Chemistry* 2022, 4, 100549.
19. K. G. Bhattacharyya, S.S. Gupta, Adv. Colloid Interface Sci. 2008, 140, 114.
20. K. Fuminao, W. Toru, and O. Tatsuya, *ACS Appl Mater Interfaces*. **2020**, *12*(6), 7021-7029.
21. Karthika, V. Srivastava, *E3S Web of Conferences* **2023**, 453, 01058.
22. P.R. Ashok, S. Grigorly, B. Subhash, *Current Pharmaceutical Biotechnology* **2021**, 22(6), 773-792.
23. A. Bouhemadou, F. Zerarga, A. Almuhayya, S. Bin-Omran, *Mater. Res. Bull*. **2011**, *46* (12), 2252-2260.
24. L. K. Kurihara, S. L. Suib, *Chem. Mater* **1993**, *5*(5), 609-613.
25. A. Manzar, G. Murtaza, R. Khenata, M. Yousaf, S. Muhammad, Hayatullah, *Chin. Phys. Lett*. **2014**, *31*(6), 67401-67404.
26. Z. Lv, Q. Chen, Y. Guo, *Solid State Sciences* **2020**, *109*, 106393.
27. R. Kumar, M. A. Barakat, B. A. Al Mur, F. A. Alseroury, J. O. Eniola, *J. Clean. Prod.* **2020**, *246*, 119076.
28. Y. Ma, W. Ran, *J. Alloys Compd*. **2020**, *842*, 155787.
29. G. Rajesh, P. Senthil Kumar, R. Gayathri, S. Akilandeswari, Mandal, A. Aindrila, V. Uma Shankar, M. Ramya, K. Nirmala, K. Thirumalai, *Mol. Catal*. **2023**, *535*, 112835.
30. A. Rafiq, M. Ikram, S. Ali, F. Niaz, M. Khan, Q. Khan, M. Maqbool, *Journal of Industrial and Engineering Chemistry* **2021**, 97, 111-128,
31. W. Ran, L. Wang, Q. Liu, G. Liu, D. Qu, X. Pan, J. Shi, *RSC Adv*. **2017**, 7, 17612.
32. P. Rajesh, P. Senthil Kumar, S. Akilandeswari, G. Rangasamy, S. Lohita, V. Uma Shankar, M. Ramya, K. Thirumalai, *Chemosphere* **2023**, *32*, 138232.
33. T. Watanabe, T. Sato, *Clay Sci*. **1988**, *7*, 129-138.
34. S. Mahalaxmi, G. Rajesha, P. Senthil Kumara, S. Akilandeswari, M. Arul Joshua, V. Uma Shankar, M. Ramya, K. Thirumalai, R. Gayathri, *Chemosphere* **2023**, *322*, 138178.
35. V. Maja, L. Mika, L. Kai, H. Tommi, H. A. R. Ras, *Soft Matter* **2019**, *15*, 7096.

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Research Article

Fluorescence Quenching of Aromatic Amino Acids by Rhodium Nanoparticles

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Abstract. In this paper, the fluorescence quenching of the aromatic amino acids tyrosine and tryptophan by rhodium nanoparticles has been investigated. The choice of rhodium nanoparticles was determined by the fact that the plasmonic maximum of the nanoparticles and the absorption range of the amino acids are in the UV. The quenching constants and types of quenching were estimated using Stern-Volmer dependencies. The fluorescence intensity of amino acids was found to decrease with nanoparticle concentration, with different types of quenching observed: tryptophan-nanoparticle system showed static quenching, while dual quenching (static and dynamic) occurred in tyrosine-nanoparticle system. Calculation of parameters of quenching efficiency were done: diffusion coefficient, diffusion rate parameter and quenching activation energy. Opportunities to exploit quenching mechanisms to realise optical sensing effects in UV have been shown.

Keywords: aromatic amino acids, tyrosine, tryptophan, fluorescence spectroscopy

INTRODUCTION

Fluorescence spectroscopy is actively used to study the structure and dynamics of proteins and other biological macromolecules [1,2]. The intrinsic fluorescence of proteins is due to the presence of aromatic amino acids: tyrosine (Tyr), tryptophan (Trp) and phenylalanine (Phe) [3]. The optical activity of these aromatic amino acids has long been of interest to scientists and has been actively used to study protein aggregation and conformation [4,5]. Trp, Tyr and Phe also play the role of internal fluorescent probes of protein conformation, dynamics and intermolecular interactions [6,7].

The contribution of Phe to protein fluorescence is small due to of its low absorbance and quantum yield [8] Tyr fluorescence in native proteins is often suppressed by energy transfer to Trp, therefore, Tyr is less frequently used for protein studies [9,10]. However, protein unfolding can lead to partial elimination of Tyr quenching, making it to be a useful indicator for protein conformational changes [11,12].

The possibility of Tyr fluorescence use to monitor conformational changes in proteins that are not detected by Trp fluorescence was investigated by a group of authors Zhdanova et.al. [13] where human and bovine serum albumin were chosen as model objects.

Trp is most often used as a probe because it is the dominant absorber at λ ~ 280 nm and the emission source at λ ~ 350 nm [9]. Trp fluorescence has been found to be very sensitive and responsive to changes in its microenvironment. For example, denaturation of bovine serum albumin (BSA) under the action of sodium dodecyl sulfate (SDS) was investigated on the basis of Trp fluorescence quenching [14]. The fluorescence intensity change of free Trp and Trp attached to the membrane of *Escherichia coli* and *Bacillus subtilis* was determined [15]. It is shown that for Trp being in a free state and not attached to the protein, there is no increase in fluorescence intensity. Fluorescence intensity enhancement can be explained by an additional contribution of Trp fluorescence formed when the protein unfolds, breaking the bond attaching Trp to the membrane of the bacterial protein. Studies on the application of metal surfaces and particles to investigate the fluorescence intensity enhancement/quenching are being actively developed and published.

Metal-enhanced fluorescence (MEF) can lead up to 100 times intensity increase due to the plasmon-enhanced local field. Meanwhile, fluorescence emission can be quenched for fluorophores at short distance (<5 nm) from the metal surface or in direct contact with the metal surface, in which the quenching effect overwhelms the enhancement effect. Different mechanisms for the fluorescence enhancement and quenching of metal nanoparticles have been suggested, but the precise mechanism is still unknown due to the complexity of metal–fluorophore interactions [16]. Metal nanostructures can be useful as fluorescence signal amplifiers for DNA detection [17,18]. Traditionally, nanoparticles of noble metals have been synthesized for fluorescence studies of protein compounds [19–24]. However, the development of science indicates the importance of investigating the use of other metals whose application is possible for the UV-range. Suitable metals for UV-range studies are represented by aluminum [25,26], copper [27]. However, there are plenty of works in which it is shown that the presence of oxide film together with high sensitivity to temperature and humidity result in the difficulty of using such metals for work in the UV range [28,29]. The noble metals such as rhodium and platinum are increasingly attracting the attention of scientific groups. Such metals are considered to be perspective for this area due to their characteristics, namely, resistance to environmental conditions, biocompatibility, and absence of oxide film [30]. Platinum nanoparticles (PtNPs) are increasingly used to enhance the capabilities of modern sensor technologies. The use of Pt nanostructures for the implementation of the UV–MEF method has been studied. Akbay et al. studied MEF of nucleic acids using platinum nanostructured substrates [31]. In the presence of Pt nanostructures, guanosine monophosphate exhibited a higher fluorescence intensity compared to control samples on a quartz substrate. An optical sensor was used for determining oxygen concentration based on a Pt(II) complex and silver-coated SiO2 nanoparticles embedded in a sol–gel matrix [32].

Fluorescence quenching is also actively used to investigate the interaction between fluorophore molecules and nanoparticles. For such purposes, already familiar metals, namely gold and silver, are most often used [33]. The hydrophilic ferroferric oxide nanoparticles and hydrophobic nanoparticles were used to study the interaction mechanism of bovine serum albumin. The presence of dynamic quenching with the first type of nanoparticles and static quenching in the second type was determine [34]. AuNP with diameters of 10 nm were used to study the possibility of controlling fluorescence of fluorophores. The authors were able to experimentally quench and enhance Cypate fluorescence by changing the distance between the fluorophore and GNP [35].

The continuous transition from fluorescence enhancement to fluorescence quenching on a single molecule was measured as a function of distance from a laser-irradiated gold nanoparticle [36].

The quenching of chlorophyll fluorescence by silver nanoparticles with different concentrations and diameters was investigated. It was shown that AgNPs strongly reduced the fluorescence intensity of Chl at 678 nm , which depended on the nanoparticle size with an exponential decrease as a function of the nanoparticle diameter. The authors determined that the fluorescence quenching was caused by a dynamic quenching process, the Stern-Folmer constant being linearly dependent on the nanoparticle size [37].

There is a paper describing the interactions of tyrosine, tryptophan and phenylalanine with biologically synthesized silver nanoparticles. The silver nanoparticles have the ability to quench the intrinsic fluorescence of these amino acids through a dynamic quenching process [38]. In such a case, the issues related to the study of fluorescence of analytes using another noble UV-active metal - rhodium are relevant. A literature review revealed the only paper describing the use of rhodium complexes to study the fluorescence quenching of Trp residue in human serum albumin (HSA) [39].

In the present research we have demonstrated the possibility of fluorescence quenching in amino acid-RhNPs systems, which can be considered as a fundamental basis for the creation of new methods based on ultraviolet plasmonics for biophysical matters.

MATERIALS AND METHODS

Reagents

The following reagents were used for the experiment: sodium borohydride $NaBH_4$ (LTD “Lenreaktiv”, Russia, purity 97,5%), rhodium chloride $RhCl_3$ (LTD “Aurat”, Russia, purity 36 %), aromatic amino acids Tyr (Sigma-Aldrich, EU, purity 99%) and Trp (Sigma-Aldrich, EU, purity 99%). Ultrapure water produced by the Direct-Q 3 UV (Merck, Germany) water purification system was used during synthesis.

RhNPs synthesis

The synthesis was carried out at room temperature. As a first step, 1.26 g of polyvinylpyrrolidone (PvP) (Mw ~ 55,000) was dissolved in 50 mL of water. Then 200 μl of 0.1 M rhodium salt solution was added to 10 ml of PvP solution. Then 400 μl of freshly prepared and cooled 0.1 M sodium borohydride was added dropwise. After adding the total volume of reducing agent, the solution was stirred for half an hour and left for 24 hours. The synthesised solution was brown in colour. After 24 hours, 5 ml of the solution was centrifuged once at 13000 rpm for 30 minutes. The supernatant was removed and the precipitate was diluted (dispersed) to 5 ml with distilled water. Finally, the concentration of the nanoparticles was calculated:

The volume of the nanoparticle was calculated to determine the concentration:

$V_{np} = \frac{4}{3} \cdot \pi \cdot r^3$, where r - nanoparticle radius, cm^3

Then, we calculated the mass of one particle using the value of volume and density of nanoparticle:

$m_{np} = V_{np} \cdot \rho_{Rh}$, where ρ_{Rh}- rhodium metal density (12,41 g/cm^3).

The number of nanoparticles is then calculated by the ratio of the total mass of recovered rhodium to the mass of one particle. The amount of rhodium substance is equivalent to the amount of rhodium (III) chloride

$N_{np} = \frac{m_{Rh}}{m_{np}}$, where m_{Rh} = 2,1 mg.

Next, we found the mole number of nanoparticles through Avogadro’s number:

$n_{np} = \frac{N_{Rh}}{N_A}$, N_A where = $6{,}02 \cdot 10^{23}$ M^{-1}

The nanoparticle concentration was calculated using the formula:

$C_{np} = \frac{n_{np}}{V}$, where V -nanoparticle solution volume,

The results of calculations are presented in the Table 1:

Table 1. The results of calculation the RhNPs concentration

Parameter	Value
V_{np}, cm^3	$6{,}24 \cdot 10^{-16}$
m_{np}, g	$7{,}74 \cdot 10^{-15}$
N_{np}	$2{,}66 \cdot 10^{11}$
n_{np}, M	$4{,}42 \cdot 10^{-13}$
c_{np}, M	$4{,}17 \cdot 10^{-11}$

The concentration of synthesized RhNPs was $4{,}17 \cdot 10^{-11}$ M. We obtained lower concentrations of nanoparticles by diluting the initial concentration with distilled water.

Sample preparation

Tyr and Trp solutions with a concentration of 10^{-5} M were prepared. The amino acid was mixed with RhNPs of different concentrations in a 4 mL optically transparent quartz cuvette (Q-1 grade) at room temperature and normal pressure. For this purpose, 3 ml of amino acid and 1 ml of RhNPs were added to the cuvette. After adding the RhNPs to the amino acid, the solution was stirred for 15-30 seconds.

Experiment

The absorption maxima of each aromatic amino acid were determined using a UV-2600 spectrophotometer (Shimadzu). 3 ml of the amino acid was placed in a cuvette and then placed in the holder of the spectrophotometer. The spectrum was recorded in the range 200 - 500 nm. The baseline of the absorption spectrum of amino acids was recorded after subtracting the absorption spectrum of distilled water. The absorption spectrum of the synthesised NPs was obtained in a similar way.

The average hydrodynamic radius and zeta-potential of RhNPs were determined using a Photocor Compact-Z (Photocorr) dynamic light scattering spectrometer.

The influence of the presence of RhNPs on the fluorescence intensity of aromatic amino acids was investigated using a spectrofluorometer RF-5301PC (Shimadzu). The fluorescence intensity of aromatic amino acids with RhNPs was investigated using excitation wavelengths corresponding to the absorption maxima of aromatic amino acids (280 nm and 275 nm for Trp and Tyr, respectively).

The fluorescence lifetime of aromatic amino acids was measured on a Fluorolog-3 FL3-22 (Horiba Jobin Yvon) using Data Station software. The spectrum of the empty cuvette was used as the decay for each AA + RhNPs sample. A NanoLED pulsed laser diode operating at λ = 284 nm (Horiba Jobin Yvon) with a nanosecond pulse duration of 1.2 ns was used as an excitation source.

RESULTS AND DISCUSSION

The absorption spectra of aromatic amino acids were obtained as a result of the study (Figure 1). The absorption spectra of the amino acids showed two peaks. The peaks at 280 and 220 nm correspond to Trp, whereas the peaks at 275 and 225 nm correspond to Tyr. Unfortunately, the ability to study kinetics using short wavelength excitation near 220 nm is not currently available worldwide due to the lack of suitable pulsed radiation sources. Therefore, 280 nm and 275 nm, corresponding to the absorption band of the indole ring for Trp and the phenol ring for Tyr, respectively, were chosen as the excitation wavelengths for obtaining fluorescence spectra of amino acids in the presence of RhNPs to further comply with spectral and time-resolved fluorescence studies.

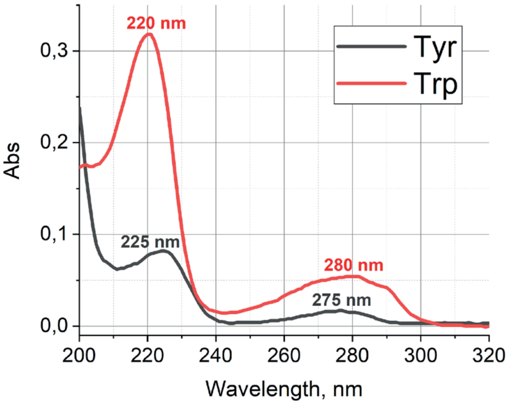
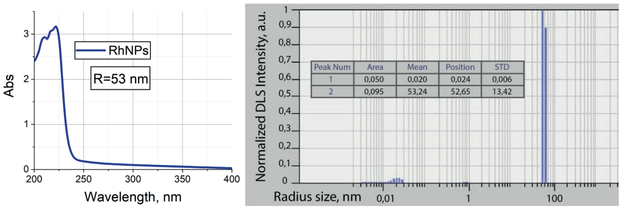


Figure 1. Absorption spectrum of aromatic amino acids: Tyr (gray line), Trp (red line).

Figure 2 shows the absorption spectrum of the synthesized RhNPs. The synthesized RhNPs show a narrow size distribution with an average hydrodynamic radius of 53 nm. The measurement error is 7 per cent (± 4 nm). The plasmonic absorption maximum of the synthesized NPs is in the UV-range at a wavelength of 220 nm. The value of Z-potential = -14.2698019 mV.



Peak Num	Area	Mean	Position	STD
1	0,050	0,020	0,024	0,006
2	0,095	53,24	52,65	13,42

Figure 2. Absorption spectrum (left) and size distribution (right) of RhNPs.

One way to determine the shape of nanoparticles is the absorption spectrum of nanospheres, consistent with literature data [40]. The spectrum of the synthesised nanoparticles by us differs from the spectra of rhodium nanoparticles of other geometries[41–43]. The SEM image of the RhNPs was also taken to demonstrate the spherical shape of the synthesised RhNPs (Figure 3). As can be seen in the Figure 3, the RhNPs have a spherical shape. The value of the hydrodynamic radius coincides with the value obtained with the Photocor Compact-Z (Photocorr).

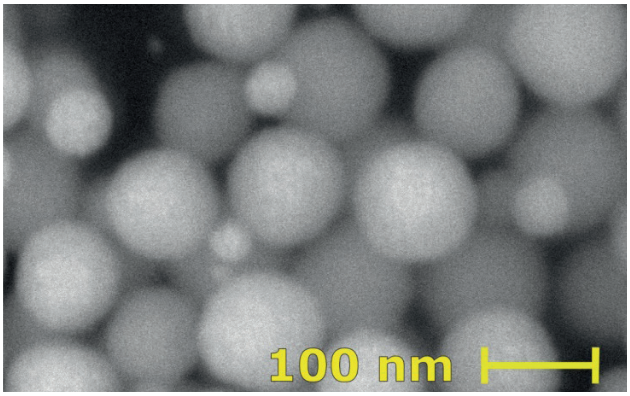


Figure 3. SEM image of synthesized spherical RhNPs.

We obtained the fluorescence and absorption spectra of Trp (Fig.4-5) and Tyr (Fig. 6-7).

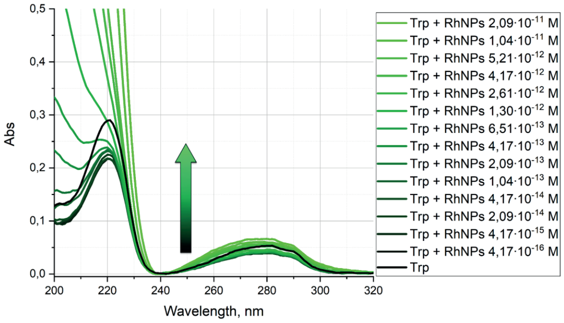


Figure 4. Absorption spectra of Trp with RhNPs of different concentration. Black spectrum - spectrum of Trp without RhNPs, bright green spectrum-spectrum of Trp with RhNPs of highest concentration ($2{,}09 \cdot 10^{-11}$ M).

As can be seen in Figure 4 the absorption spectrum shows two absorption maxima at 220 nm and maxima at 280 nm which do not change their position. The absorption of Trp decreases and then increases with increasing concentration of RhNPs.

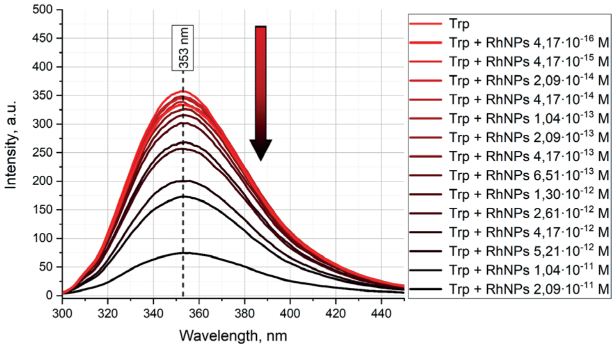


Figure 5. Fluorescence spectra of Trp in the presence of RhNPs with different concentrations. Bright red spectrum-spectrum of Trp without RhNPs, black spectrum - spectrum of Trp with RhNPs of highest concentration (2,09 · 10^{-11} M).

The fluorescence intensity of Trp decreases almost sevenfold with increasing concentration of RhNPs. The fluorescence maximum occurs at a wavelength of 353 nm and does not change its position.

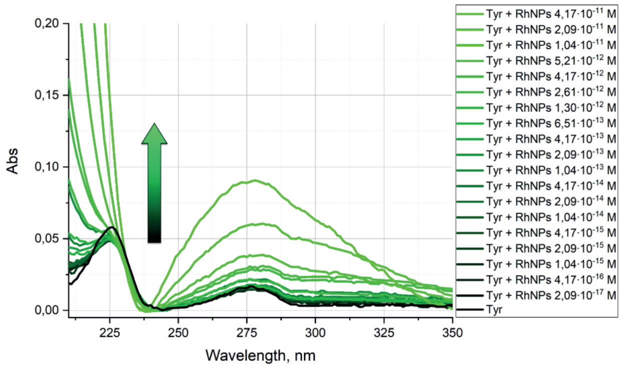


Figure 6. Absorption spectra of Tyr with RhNPs of different concentration. Black spectrum - spectrum of Tyr without RhNPs, bright green spectrum-spectrum of Try with RhNPs of highest concentration (2,09 · 10^{-11} M).

The absorption of Tyr increases with increasing concentration of RhNPs. The absorption maxima occur at a wavelengths of 220 nm and 275 nm. The positions of the maxima do not change throughout the experiment.

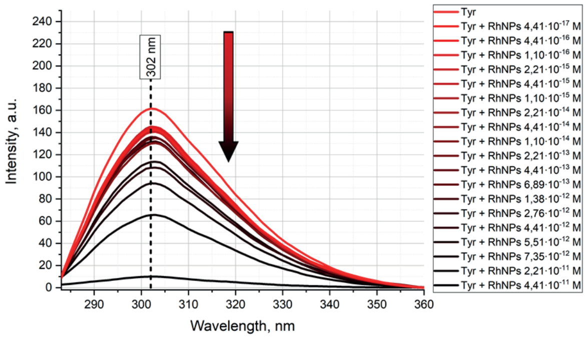


Figure 7. Fluorescence spectra of Tyr in the presence of RhNPs with different concentrations. Bright red spectrum-spectrum of Tyr without RhNPs, black spectrum - spectrum of Tyr with RhNPs of highest concentration (4,41 · 10^{-11} M).

The fluorescence intensity of Tyr decreases almost 16 times with increasing concentration of RhNPs. The fluorescence maximum occurs at a wavelength of 302 nm and does not change its position.

In order to identify the mechanisms of Trp and Tyr fluorescence quenching by RhNPs Stern-Volmer dependencies (Figure 8) were plotted. Each fluorescence spectrum was integrated to calculate the area under the curve to construct a more accurate dependence of fluorescence intensity. The decay kinetics of the excited state of the both amino acids were recorded (Table 1) and also used to determine the mechanisms.

Lifetimes of each aromatic amino acids in the presence of RhNPs were measured. It was determined that Trp fluorescence lifetime was practically unchanged and the average value was equal to 2.6 ns. Tyr fluorescence lifetime decreased linearly with RhNPs concentration increase and was in the range of values 3.14 - 1.99 ns (see Table 2).

Table 2. Time resolved characteristics for Rh NPs-Tyr/Trp complexes.

C_{NPS}, M	Lifetime,ns*	
	Trp	Tyr
0	2,63	3,14
4,4·10^{-15}	2,62	2,93
4,4·10^{-14}	2,65	2,99
2,2·10^{-13}	2,77	2,96
4,4·10^{-13}	2,71	3,02
5,5·10^{-13}	2,58	2,8
1,1·10^{-12}	2,75	2,58
2,2·10^{-12}	2,63	2,47
4,4·10^{-12}	2,64	2,41
5,5·10^{-12}	2,55	2,57
1,1·10^{-11}	2,43	2,3
2,2·10^{-11}	2,34	1,99

* The measurement error for TCSPC measurements was ± 0.1 ns for Tyr and ± 0,06 ns for Trp.

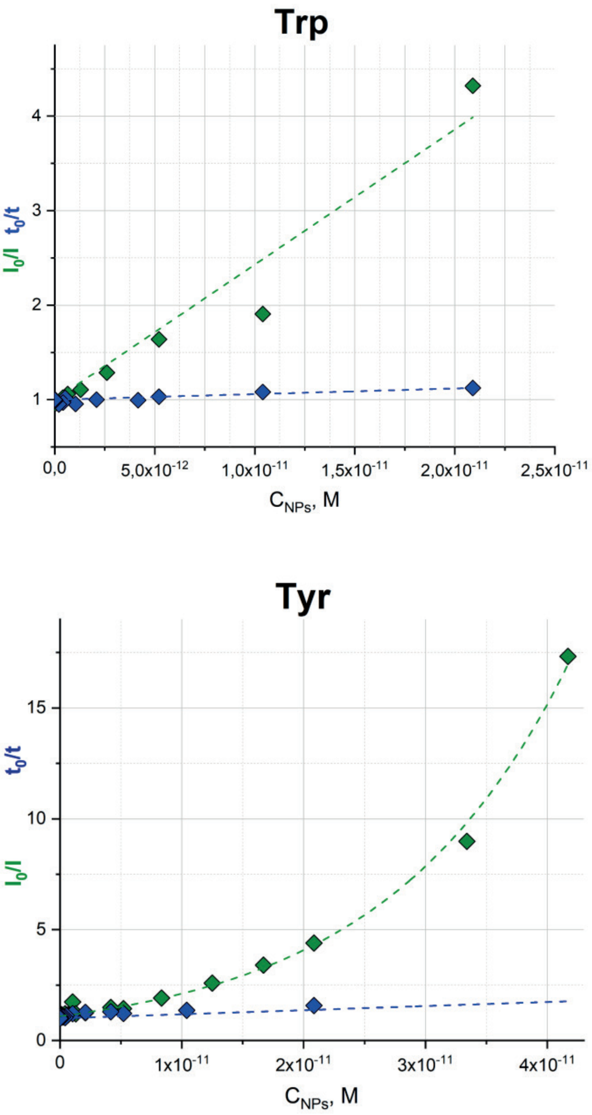


Figure 8. Stern-Volmer plots for the systems: (a) Trp + NPs and (b) Tyr + NPs, where I_0, I (t_0,t) denote integral fluorescence intensities (lifetime) of aromatic amino acids in the absence and presence of quencher

As can be seen from Figure 4(a) Trp lifetime t does not practically change with increasing RhNPs concentration, while its inverse fluorescence relative intensity $\frac{I_0}{I}$ linearly increases, which provided evidence of a static quenching mechanism (ground state complex formation model). In this connection, Coulomb interaction results in non-fluorescent complexes formation between a Trp and negative charged RhNPs. The number of such complexes increases with the quencher concentration raise. The association constant was calculated according to formula (1):

$$\frac{I_0}{I} = 1+K_s[Q] \tag{1}$$

where [Q] denotes concentration of quencher, K_s is an association constant, which can be estimated as a slope of (I_0/I – 1) vs [Q] graph. The association constant was found to be $1.73 \cdot 10^{11}$ M^{-1} for Trp+RhNPs system.

As can be seen from Figure 4(b), the dependence of lifetime ratios $\frac{t_0}{t}$ for the Tyr+RhNPs system is linear and increases with RhNPs concentration increase. Dependence of fluorescence intensity ratio $\frac{I_0}{I}$ is nonlinear. Deviation from linearity for the obtained dependencies showed that Stern-Volmer model is not optimal for describing the fluorescence quenching within static mechanism and can indicate the simultaneous existence of both static and dynamic quenching mechanism in the Tyr-RhNPs system. In this case, fluorescence quenching occurs due to the formation of nonfluorescent complexes of the amino acid with RhNPs and diffuse collision between tyrosine molecules and RhNPs. The modified form of the Stern-Volmer equation for combined quenching is a second-order equation, which accounts for the upward curvature of the line characteristic of the combined quenching mechanism (2):

$$\frac{I_0}{I} = (1+K_D[Q])(1+K_s[Q]) \tag{2}$$

The contribution of dynamic K_D quenching was determined using a linear approximation of the lifetime ratios from equation (3):

$$\frac{t_0}{t} = 1+K_D[Q] \tag{3}$$

The slope coefficient of the straight line determined the value of the dynamic quenching constant and was equal to $K_D = 3{,}05 \cdot 10^{10}$ M^{-1}. The value of the dynamic extinguishing constant was used to estimate the static extinguishing constant in equation (2) and $K_s = 7{,}5 \cdot 10^{10}$ M^{-1}.

In addition, the following parameters of quenching efficiency were calculated: diffusion coefficient, diffusion rate parameter, quenching activation energy. Since the size of the quencher (RhNPs) and the fluorophore (Tyr or Trp) differed by an order of magnitude, the calculation formulas were determined as follows.

The diffusion coefficient was calculated using the formula (4):

$$D = \frac{k_B T}{6\pi\eta R} \tag{4}$$

where k_B denotes the Boltzmann constant ($1, 38 \cdot 10^{-23}$ J/K), T is the temperature, η is the viscosity, R is the RhNPs radius.

The experiment was carried out at T = 20 °C. After centrifugation, removal of the supernatant and dilution of the precipitate, the amount of PVP in the precipitate became negligible, thus eliminating its influence on the experiment therefore η = 1,005 was used to determine the diffusion coefficient. The deffusion coefficient was equal to $4{,}03 \cdot 10^{-12}$ m^2/sec.

Diffusion rate parameter was calculated using the formula (5):

$$K = \frac{4\pi RDN}{1000} \tag{5}$$

where N is Avogadro's number ($6{,}02 \cdot 10^{23}$ M^{-1}). The diffusion rate constant was equal to 1616,84 m^3 /sec·M

The activation energy was calculated using the Arrhenius equation (6):

$$K = A \cdot \exp^{\frac{-E_a}{RT}} \tag{6}$$

where A is the multiplier, E_a is the activation energy, R is the gas constant (8,31 J/M·K). The viscosity of water was indicated in the temperature range of 5-55 °C every 5 degrees and the corresponding parameters were calculated. All information you can find from Table 3.

Table 3. Parameters of quenching efficiency.

T, °C	T, K	η, mPa·sec	D · 10^{-15}, m^2/sec	K, m^3 / sec·M	ln(K)	1/T
5	278,15	1,519	2,53	1,01	6,92	0,003595
10	283,15	1,308	2,99	1,20	7,09	0,003532
15	288,15	1,14	3,49	1,40	7,24	0,00347
20	293,15	1,005	4,03	1,62	7,39	0,003411
25	298,15	0,8937	4,61	1,85	7,52	0,003354
30	303,15	0,8007	5,23	2,10	7,65	0,003299
35	308,15	0,7225	5,90	2,36	7,77	0,003245
40	313,15	0,656	6,60	2,65	7,88	0,003193
45	318,15	0,5988	7,35	2,95	7,99	0,003143
50	323,15	0,5494	8,13	3,26	8,09	0,003095
55	328,15	0,5064	8,96	3,59	8,19	0,003047

As a result, Figure 9 was plotted and the -E_a/R was estimated as a slope of ln(K) vs (1/T). The activation energy was calculated as 19 080,5 J/mol or ≈19 kJ/M.

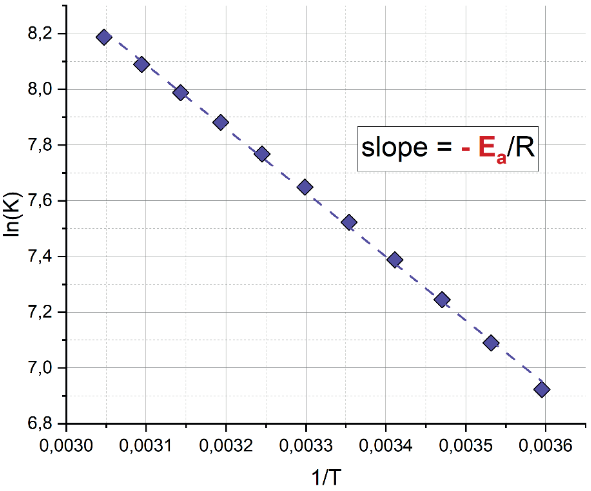


Figure 9. Dependency of ln(K) versus 1/T.

It is worth noting that quenching can also occur as a result of the overlap between the absorption spectrum of the quencher and the fluorescence spectrum of the donor. The size of the NPs and the geometrical arrangement of the donor and acceptor influence the energy transfer mechanism. The close proximity of two molecules (or two parts of a molecule) can lead to an overlap in the electronic state functions of the molecules. In this region, the electrons of the molecules are indistinguishable, which can lead to two-way electron exchange. Such energy transfer occurs in an exchange resonance manner (charge (electron) energy transfer mechanism) [44] (Dexter's mechanism). Realisation of the exchange-resonance mechanism of energy transfer occurs in the near field at small distances between the donor and acceptor, which usually does not exceed 1 nm [45]. Another transport mechanism is inductive resonance energy transfer (dipole-dipole energy transfer (FRET)). The Forster model explains the mechanism of energy transfer between distant molecules at such a distance that no overlapping of electron orbitals occurs. The Ferster mechanism is based on the long-range dipole-dipole Coulomb interaction between electrons or excited donor molecule and acceptor molecule, initially in the ground state, using the coupling of their respective transition dipole moments [45].

Energy transfer can be described by nanometal surface energy transfer (NSET) [46,47]. In the case of NSET, the PM is considered as a two-dimensional (2D) dipole array [48] is a nanosurface having many single dipoles. This transition from a one-dimensional dipole to a two-dimensional dipole array, occurs due to the small size of the NPs, on the order of < 80 nm [46,49,50], where surface and volume are indistinguishable. As is known, the energy transfer rate is related to the donor-acceptor interaction. The key point is that, unlike FRET, NSET does not require a resonant electronic transition. The process of energy transfer via NSET arises from the interaction of the electromagnetic field of the donor dipole with the free conduction electrons of the host metal [51]. In this theoretical model it is assumed that the field reflected from the surface is negligibly small and does not interfere with the dipole field [52], and also does not depend on the size, shape of nanoparticles and the degree of spectral overlap between the donor and acceptor [53]. The inclusion of size dependence in NSET was proposed in the Chance, Prock and Silby-Kuhn (CPS-Kuhn) model, which is discussed below. According to the literature, the process of quenching of fluorophore emission is usually described by FRET, NSET, G-N, and CPS - Kuhn models. To summarise, we assume NSET quenching in the investigated complexes and this model will be tested in our further studies.

Since there are no data on the use of rhodium nanoparticles with proteins, it is possible to assume the possibility of using such particles in transport proteins, such as HSA [54]. In this case, the nanoparticle can be embedded in the binding centres of the protein [55], thus changing its conformation and photophysical properties. The structure of HSA contains several metal binding sites. These binding sites play an important role in the transport of metal ions during certain physiological or pathological processes in vivo, allowing the reversible binding of various metal ions. The existence of these metal binding sites has led to extensive research on HSA as a template for the synthesis of inorganic metal nanomaterials, including silver sulphide (Ag2S), gadolinium oxide (Gd2O3), manganese dioxide (MnO2) and copper sulphide (CuS) [56]. The results obtained in this article demonstrate the possibility of controlling photophysical processes in nanosytems, as well as the prospects for the application of such systems in biophysics.

CONCLUSIONS

RhNPs with a hydrodynamic radius of 53 nm and plasmonic absorption in the UV range have been synthesized. Trp and Tyr fluorescence spectra and decay kinetics in the presence of RhNPs have been recorded. It was shown that static quenching of Trp fluorescence takes place in Trp + RhNPs system while Tyr fluorescence is quenched by RhNPs due to dual mechanisms. Parameters of quenching efficiency: diffusion coefficient, diffusion rate parameter and quenching activation energy were calculated. At present there are known works with quenching of fluorescence of amino acids with other metals, for example, with silver and gold. Researchers are also investigating complexes based on such nanoparticles, for example [38]. However, since the plasmonic maximum of silver and gold is in the visible region, researchers apply FRET-based models to calculate the energy transfer in this case. In our case, the NSET model is assumed in the case of quenching, while the PIRET model is assumed in the case where enhancement would be observed. The addition of metal-containing compounds and nanoparticles can alter the photophysical properties of the complex through the effects of quenching and fluorescence enhancement. The use of rhodium nanoparticles for such applications may be useful for isolating a specific fluorescent protein in urine, selectively binding to it and then quenching it. It is also possible to modify the nanoparticles with specific linkers, for example to the P2Y12 receptors on platelets, and evaluate the conformation of its receptor environment. The main idea for possible future applications of rhodium nanoparticle-based nanosystems is the spectral overlap described above. This provides an opportunity to exploit quenching mechanisms to realise optical sensing effects in UV.

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CONFLICT OF INTEREST

Authors state no conflict of interest.

REFERENCES

1. S. Basak, K. Chattopadhyay, *Phys. Chem. Chem. Phys.*, **2014**, 16, 11139.
2. M.C. Murphy, I. Rasnik, W. Cheng, T.M. Lohman, T. Ha, *Biophysical Journal,* **2004**,86, 2530–2537
3. C.A. Royer, *Chem. Rev*. **2006**, 106, 1769–1784
4. J.T. Vivian, P.R. Callis, *Biophysical Journal*, **2001,** 80, 2093–2109.
5. A. Biswas, R.K. Swarnkar, B. Hussain, S.K. Sahoo, P.I. Pradeepkumar, G.N. Patwari, R. Anand, J. *Phys. Chem. B*, **2014**, 118, 10035–10042.

6. A. Ghisaidoobe, S. Chung, *IJMS* , **2014**, 15, 22518–22538.
7. M. Clerici, G. Colombo, F. Secundo, N. Gagliano, R. Colombo, N. Portinaro, D. Giustarini, A. Milzani, R. Rossi, I. Dalle-Donne, **2014**, 52, 166–174.
8. J.R. Lakowicz, Principles of Fluorescence Spectroscopy, Spinger: Berlin/Heidelberg, Germany, **2006**.
9. Y. Chen, M.D. Barkley, *Biochemistry*, **1998,** 37, 9976–9982.
10. F.W.J. Teale, *Biochemical Journal,* **1960***,* 76, 381–388.
11. J. Steinhardt, J. Krijn, J.G. Leidy, *Biochemistry*, **1971**, 10, 4005–4015.
12. R.W. Cowgill, *Biochimica et Biophysica Acta (BBA)-Protein Structure*, **1968**, 168, 417–430.
13. N.G. Zhdanova, E.G. Maksimov, A.M. Arutyunyan, V.V. Fadeev, E.A. Shirshin, *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **2017**, 174, 223–229.
14. M.A. Rub, J.M. Khan, A.M. Asiri, R.H. Khan, K., *Journal of Luminescence,* **2014**, 155, 39–46.
15. R. Li, D. Dhankhar, J. Chen, T.C. Cesario, P.M. Rentzepis, *Proc. Natl. Acad. Sci. U.S.A.* , **2019**, 116, 18822–18826.
16. E. Demishkevich, A. Zyubin, A. Seteikin, I. Samusev, I. Park, C.K. Hwangbo, E.H. Choi, G.J. Lee, *Materials,* **2023,** 16, 3342.
17. Y. Jeong, Y.M. Kook, K. Lee, W.-G. Koh, *Biosensors and Bioelectronics*, **2018**, 111, 102–116.
18. K. Aslan, I. Gryczynski, J. Malicka, E. Matveeva, J.R. Lakowicz, C.D. Geddes , *Current Opinion in Biotechnology*, **2005**, 16, 55–62.
19. T. Ribeiro, C. Baleizão, J.P.S. Farinha, *Sci Rep*, **2017**, 7, 2440.
20. J.-H. Choi, J.-W. Choi, *Nano Lett*., **2020**, 20, 7100–7107.
21. J. Chen, Y. Jin, N. Fahruddin, J.X. Zhao, *Langmuir*, **2013**, 29, 1584–1591.
22. B. Della Ventura, M. Gelzo, E. Battista, A. Alabastri, A. Schirato, G. Castaldo, G. Corso, F. Gentile, R. Velotta, *ACS Appl. Mater. Interfaces*, **2019**, 11, 3753–3762.
23. K. Aslan, S.N. Malyn, C.D. Geddes, *J Fluoresc*, **2006**, 17, 7–13.
24. K. Aslan, J.R. Lakowicz, C.D. Geddes, *Anal Bioanal Chem*, **2005**, 382, 926–933.
25. M.H. Chowdhury, K. Ray, S.K. Gray, J. Pond, J.R. Lakowicz, *Anal. Chem*., **2009**, 81, 1397–1403.
26. J.M. McMahon, G.C. Schatz, S.K. Gray, *Phys. Chem. Chem. Phys*., **2013**, 15, 5415–5423.
27. Y. Zhang, K. Aslan, M.J.R. Previte, C.D. Geddes, *Applied Physics Letters*, **2007**, 90, 173116.
28. M.W. Knight, N.S. King, L. Liu, H.O. Everitt, P. Nordlander, N.J. Halas, *ACS Nano,* **2014***,* 8, 834–840.
29. Y. Gutierrez, D. Ortiz, J.M. Sanz, J.M. Saiz, F. Gonzalez, H.O. Everitt, F. Moreno, *Opt. Express*, **2016**, 24, 20621.
30. Y. Gutiérrez, R. Alcaraz De La Osa, D. Ortiz, J. Saiz, F. González, F. Moreno, *Applied Sciences*, **2018**, 8, 64.
31. N. Akbay, F. Mahdavi, J.R. Lakowicz, K. Ray, *Chemical Physics Letters*, **2012**, 548, 45–50.
32. C.-S. Chu, T.W. Sung, Y.L. Lo, *Sensors and Actuators B: Chemical*, **2013,**185, 287–292.
33. D. Ghosh, N. Chattopadhyay, *Journal of Luminescence*, **2015**, 160, 223–232.
34. C. Hao, G. Xu, Y. Feng, L. Lu, W. Sun, R. Sun, *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, 2017, 184, 191–197.
35. K.A. Kang, J. Wang, J.B. Jasinski, S. Achilefu, *J Nanobiotechnol*, **2011**, 9, 16.
36. P. Anger, P. Bharadwaj, L. Novotny, *Phys. Rev. Lett*., **2006**, 96,113002.
37. A.M. Queiroz, A.V. Mezacasa, D.E. Graciano, W.F. Falco, J.-C. M'Peko, F.E.G. Guimarães, T. Lawson, I. Colbeck, S.L. Oliveira, A.R.L. Caires, *Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy*, **2016**, 168, 73–77.
38. S. Roy, T.K. Das, *J Appl Spectrosc*, **2015**, 82, 598–606.
39. B.P. Espósito, A. Faljoni-Alário, J.F.S. De Menezes, H.F. De Brito, R. Najjar, *Journal of Inorganic Biochemistry*, **1999**, 75, 55–61.
40. M.Z. Bellus, M. Li, S.D. Lane, F. Ceballos, Q. Cui, X.C. Zeng, H. Zhao, *Nanoscale Horiz.,* **2017**, 2, 31–36.
41. S. Kundu, K. Wang, H. Liang, *J. Phys. Chem*., **2009**, 113, 18570–18577.
42. T. Wakita, H. Yao, *Chemical Physics Letters*, **2021**, 779,138866.
43. G. Kumar, R.K. Soni, *J Raman Spectroscopy*, **2022**, 53, 1890–1903.
44. D.L. Dexter, A Theory of Sensitized Luminescence in Solids, *The Journal of Chemical Physics*, **1953**, 21, 836–850.
45. H.V. Demir, S.V. Gaponenko, Applied Nanophotonics, *Cambridge University Press*, **2018**.
46. C.S. Yun, A. Javier, T. Jennings, M. Fisher, S. Hira, S. Peterson, B. Hopkins, N.O. Reich, G.F. Strouse, *J. Am. Chem. Soc*. **2005**, 127, 3115–3119
47. P.F. Gao, Y.F. Li, C.Z. Huang, *TrAC Trends in Analytical Chemistry*, **2020**, 124, 115805.
48. S. Rakshit, S.P. Moulik, S.C. Bhattacharya, *Journal of Colloid and Interface Science*, **2017**, 491, 349–357.
49. T.L. Jennings, J.C. Schlatterer, M.P. Singh, N.L. Greenbaum, G.F. Strouse, *Nano Lett*., **2006**, 6, 1318–1324.
50. M.P. Singh, G.F. Strouse, *J. Am. Chem. Soc*, **2010**, 132, 9383–9391.

51. T. Sen, S. Sadhu, A. Patra, *Applied Physics Letters*, **2007**, 91, 043104.
52. C. Chen, N. Hildebrandt, Resonance energy transfer to gold nanoparticles: NSET defeats FRET, TrAC Trends in Analytical Chemistry 123 (2020) 115748.
53. C.J. Breshike, R.A. Riskowski, G.F. Strouse, Leaving Förster Resonance Energy Transfer Behind: Nanometal Surface Energy Transfer Predicts the Size-Enhanced Energy Coupling between a Metal Nanoparticle and an Emitting Dipole, J. Phys. Chem. C 117 (2013) 23942–23949.
54. K. Bolaños, M.J. Kogan, E. Araya, Capping gold nanoparticles with albumin to improve their biomedical properties, IJN Volume 14 (2019) 6387–6406.
55. W. Bal, M. Sokołowska, E. Kurowska, P. Faller, Binding of transition metal ions to albumin: Sites, affinities and rates, Biochimica et Biophysica Acta (BBA) - General Subjects 1830 (2013) 5444–5455.
56. H. Iqbal, T. Yang, T. Li, M. Zhang, H. Ke, D. Ding, Y. Deng, H. Chen, Serum protein-based nanoparticles for cancer diagnosis and treatment, Journal of Controlled Release 329 (2021) 997–1022.

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Research Article

Chemical Engineer George L. Standart – between Science and Ideology

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Abstract. George L. Standart was one of the few western scientists who decided to emigrate to communist Czechoslovakia. He was driven to do that by his leftist mindset, his conviction that there lay a more just and peaceful world behind the Iron Curtain, and also the feeling that he was at risk in his homeland, unsettled by McCarthyism. Although he was recruited as an informer first by the Soviet, and subsequently also the Czech intelligence services, they never found his work satisfactory as an agent, a situation exacerbated by his increasing academic workload. Most of all, George L. Standart was an outstanding scientist, one of the best in his discipline and at that time, who made an unexpected and unprecedented contribution to the Czechoslovak chemical sciences and their practical industrial applications. The almost twenty years that George L. Standart and his wife spent in Prague nevertheless provides an opportunity to trace their personal and academic lives, the development of their attitudes shaped by their ideology and idealism and, partially, also the process by which the Czechoslovak security and intelligence services were established from the close of the 1940s to the late 1960s and the view they took of foreign nationals.

Keywords: George L. Standart; science in communist regime; history of chemical engineering; political emigration West-to-East

INTRODUCTION

The so-called Iron Curtain was raised in Europe in 1948, and, as a result of the wartime and post-war development, uncompromisingly divided the world up into two alien camps that entered into a conflicting situation, known as the Cold War. This had an impact mainly on the economy, the arms industry, science and technology, ideology, space exploration and other areas, with propaganda and espionage used as inevitable instruments of warfare. With their intelligence activities both sides of the conflict were on thin ice, as opportunities to provide relevant and reliable information were limited, and seeking persons suitable for such jobs was quite a delicate task. Someone coming from the other side of the Iron Curtain therefore personified both the risk of being an agent of a hostile country and the hope of becoming an agent of the host country. With intelligent, well-educated people with good language skills, understandably including scientists, this was even more relevant.

While cases of academicians fleeing communist Czechoslovakia to the West were by no means exceptional, with dozens to hundreds having being documented, hardly anyone went in the other direction. Still, such situations did happen, usually for reasons of family, work, ideology and, last but not least, idealism; however, they usually involved more or less conscious collaboration with the intelligence services. And it is not unlikely that a certain combination of all of the above came together in the chemical engineer George Lenell Standart, undoubtedly one of the most complicated figures in Czechoslovak science.

This paper strives to present the phenomenon of West-to-East political emigration across the Iron Curtain concerning a specific social, and supremely intellectual, group, i.e. members of the science and research community. There were not many scientists who opted for this way of expanding not only their academic, but especially their personal prospects; as yet, there are only a few documented cases. All of them, however, were becoming a difficult challenge for the intelligence services of all (potentially) involved countries. The case of George L. Standart and his wife is highly specific, partly because they had come to Czechoslovakia in 1948, i.e., only a few months after the communist coup d'état which definitely forged the rule of a single party in the country. It was also the time when the scope and work of the Czechoslovak State Police, which was *de facto* a political force, was just forming and adapting to the Soviet model. The almost twenty years that the Standarts spent in Prague thus provides an opportunity to trace their personal and academic lives, and also the development of their attitudes formed by their ideological and idealistic motivations and, partially, also the process by which the Czechoslovak security and intelligence services were established from the close of the 1940s to the late 1960s. The source base for this comprises archival materials, especially those lodged with the Security Services Archive (originally with the Archives of the Ministry of the Interior), witness recollections and the available literature.

THE EMIGRATION OF THE "WESTERN" INTELLECTUAL ELITE TO POST-WAR CZECHOSLOVAKIA

Even though far more people emigrated from East to West than in the opposite direction, the number of those coming to the countries of the Eastern Bloc cannot be described as negligible. A more apparent imbalance, however, was seen with the true intellectual elites who were able to positively influence the development of society or make an important contribution in their academic field. In this respect, the East is much worse off. In Czechoslovakia alone, the estimated number of those who fled the country in 1948–1989 is relatively wide, assumed to be somewhere between 260 and 550 thousand[1], including hundreds of elite scientists. Communist Czechoslovakia, on the other hand, became an attractive destination for some fifteen thousand western émigrés, including only a few scientists.[2]

There were basically two principal waves of emigration from communist Czechoslovakia to so-called West. The first began at the end of the 1940s and waned in the early 1960s, while the other, responding to the invasion of Czechoslovakia by the Warsaw Pact armies, peaked in 1968–1969 and only lessened in the next two decades.

The reasons that led hundreds of thousands of people to decide to leave their homes for good lay primarily in extensive repressive measures aimed at those who were real or alleged opponents of the power monopoly of the Communist Party of Czechoslovakia and the rule of law it implemented, including the subordinate, vassal relationship to the Soviet Union, and also in the desire to provide themselves and their offspring with the possibility to live in a free country. Hundreds of top Czech scientists thus not only enriched western academic research, but often, having become citizens in their new country, also won some prestigious academic awards, significantly boosting the academic reputation and statistical data of other countries. George Jiří Schulz, for instance, became the very first holder of the Davisson-Germer Prize in Atomic or Surface Physics for the development of techniques for the study of electron collisions on gases, and for the discovery of resonances in electron scattering by helium and atomic hydrogen, in 1965.

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[1] Vojtěch Jeřábek et al., *Českoslovenští uprchlíci ve studené válce* [Czechoslovak Émigrés in the Cold War], Brno: Stilus, 2005; Petr Koura, „Svědomí mi nedovolí šířit takové lži". Český exil mezi únorem 1948 a srpnem 1968 ["Conscience would not Allow Me to Spread Such Lies". Exiled Czechs between February 1948 and August 1968]. In Lucie Wittlichová (ed.), *Vzkazy domů. Příběhy Čechů, kteří odešli do zahraničí (emigrace a exil 1848–1989)* [Messages to Their Homes. Stories of the Czechs who Left Abroad (Emigration and Exile 1848–1989)], Praha: Dny české státnosti, Labyrint, 2012: 203–215; Libuše Paukertová, Několik základních údajů o odchodech z Československa, 1948–1991 [Some Basic Facts about Escapes from Czechoslovakia, 1948–1991]. In Stanislav Brouček, Karel Hrubý (eds.), *Češi za hranicemi na přelomu 20. a 21. století: Sympozium o českém vystěhovalectví, exulantství a vztazích zahraničních Čechů k domovu 29.–30. června 1998*, Praha: Karolinum – Etnologický ústav AV ČR, 2000: 25–31.

[2] Soňa Štrbáňová, Antonín Kostlán (eds.), *Sto českých vědců v exilu* [One Hundred Czech Scientists in Exile], Praha: Academia, 2011.

Jan and Eva Roček were very successful at the Department of Chemistry of the University of Illinois, Chicago, where Jan Roček also played a major role in the development of doctoral programmes and research into the oxidation of organic compounds, and Eva Ročková's successful pedagogical career was twice awarded the Silver Circle Award for Excellence in Teaching. In 2001, Professor of civil engineering Zdeněk P. Bažant received the award of Highly Cited Researcher, which is given by the Institute for Scientific Information to only 250 authors worldwide across all engineering fields.

Most of the people who came to communist Czechoslovakia as political émigrés were from Greece, Italy and Spain, and even though there were many leftist intellectuals among them, true members of the academic community would be very hard to find. During the initial period under the rule of a single party subordinate to the arbitrariness of the Soviet Union, besides George L. Standart and his wife, the cases of the economist G. S. Wheeler, Hispanist A. C. García, mathematician M. Nadler and Anglicist I. F. Milner are also documented. Three of them are therefore representatives of the humanities and social sciences, while another three worked in the natural and technical sciences. George Shaw Wheeler was an economist and a former US Army colonel, who together with his wife requested political asylum in Czechoslovakia in the late 1940s and, after the establishment of the Czechoslovak Academy of Sciences, was employed in its Institute of Economy.[3] He graduated from Reed College, Portland and in the 1930s was deputy head of the Department of Economics at the University of Chicago. In 1936 he became a civil servant, and in 1944 he was transferred to London to head a denazification unit. In May 1945 he became the Chief of the Section of the Office of Military Government for Germany (United States).[4] Shortly after the end of the war, he was found ineligible to work for government agencies, as he had always toed the Communist line from before 1939. So, in 1947, he and his family moved to Czechoslovakia where he began to lecture in economics at Charles University, where he was awarded a doctoral degree in 1961. In 1950 the Wheelers submitted a public application in Prague for political asylum in the form of a statement condemning the American policy.[5] Despite that, the Czech State Security still suspected them of spying for the American intelligence services, amongst other things because of their regular contacts with the American Embassy in Prague the purpose of which was unclear, and other possibly suspicious contacts, but also due to the legal rehabilitation the Wheelers were granted by the American authorities.[6] In the mid-1950s G. Wheeler began to work for the ČSAV Institute of Economics, where he was involved in preparing the economic reform that constituted the specific Czechoslovak approach to the state economy. After the August 1968 invasion of Czechoslovakia by the armies of the Warsaw Pact, the Wheelers decided to return to the USA, where George S. Wheeler became a Professor of Washington State University in 1971. In 1990, Wheeler returned to Prague alone (his wife had died in 1981) and lived with one of his daughters until his death in 1998.

Another political émigré to come to communist Czechoslovakia was Spanish general and former deputy defence minister Antonio Cordón García. He was a leading figure in the Communist Party of Spain and a close collaborator of the Republican Prime Minister Juan Negrín, who was defeated in the Spanish Civil War by General Francisco Franco's troops.[7] Antonio Cordón García first left for the Soviet Union, later spent some time working in Tito's Yugoslavia, and eventually ended in Czechoslovakia where, amongst other things, he lectured on Spanish literature at the Faculty of Arts and Humanities of Charles University.[8] He died in Rome in 1968, but his remains were not transferred to his native country until after Franco's death in 1975 and the subsequent political changes in Spain.

The natural and technical sciences were represented by Morton Nadler, an American, whose position in Czechoslovakia turned out to be a complicated one. He was born into a Jewish family in New York in 1921 as Mandel Nadler. He joined the Communist Party USA when he was just 15, and his strong left-wing focus also led him to study at the Illinois Institute of Technology. This was a traditional centre of leftism after the war, hence also a sphere of interest for the Soviet intelligence

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[3] Doubravka Olšáková, "V krajině za zrcadlem. Političtí emigranti v poúnorovém Československu a případ Aymonin" ["In the Landscape Behind the Mirror. Political Émigrés in Post-February Czechoslovakia and the Aymonin Case"], *Soudobé dějiny* 4 (2007): 719–743, here: 738.

[4] Patrick Wright, *Iron Curtain: From Stage to Cold War*, Oxford: Oxford University Press, 2009: 360–361.

[5] "Prohlášení George Wheelera, bývalého amerického plukovníka" ["Statement by George Wheeler, a Former American Colonel"], *Rudé právo*, 8 April 1950: 2; Kathleen Geaney, "At Home among Strangers: The Extraordinary Year 1950 in the Life of an Ordinary American Family in Communist Czechoslovakia", *Comenius: Journal of Euro-American Civilization* 1 (2015): 25–42.

[6] Jiří Bašta, "Propagandistické využití kausy amerického emigranta G. S. Wheelera: Dezinformace StB v sdělovacích prostředcích" ["Propagandistic Use of the Case of American Émigré G. S. Wheeler: Disinformation by the Secret Police in the Mass Media"], *Securitas Imperii* 7 (2001): 224–251, here: 225.

[7] Matilde Eiroa, *Españoles tras el Telón de Acero: El exilio republicano y comunista en la Europa socialista,* Madrid: Marcial Pons Historia, 2018.

[8] Josef Petráň, *Filozofové dělají revoluci* [Philosophers Doing the Revolution], Praha: Karolinum, 2015: 186.

services.[9] The fact that he was at least contacted by the KGB can be seen in his post-war European career. Having obtained a passport in 1947 to travel to do a doctorate at the Sorbonne in Paris, he indeed left for France and enrolled; however, soon afterwards, in March 1948, just a few days after the February 1948 Communist coup d'état in Czechoslovakia, he moved to Prague. His wife and daughter joined him a few months later. As early as at the end of the 1940s, Nadler was already suspected of being one of the two spies around Julius Rosenberg[10] and also of passing sensitive strategic information regarding U.S. Army radars to the Czechoslovaks. The US Embassy therefore confiscated his passport, stating that he could only get it back to travel directly to the USA. However, given the circumstances, that was not in Nadler's interests, and so he applied for Czechoslovak citizenship. Nadler was allegedly contacted by two real spies while they were staying in Czechoslovakia in 1950–1956 and doing research into electronics for the military.[11] They may have tried to persuade him to accompany them back to the USSR to work on the development of microcomputers controlling military and industrial systems. At that time, however, in consequence of the events in Hungary and probably also due to the abject political and economic situation in Czechoslovakia, he found himself becoming less enamoured with communism, and moving to the Soviet Union was by no means an appealing prospect.[12]

There is evidence that in 1951, he was recruited as an informer of the Czechoslovak intelligence services. In his personal file, he is codenamed "Nový", and his main task was to inform about the Křižík national enterprise where he was employed, and about the people who worked there.[13]

From the mid-1950s, he worked for the Research Institute for Mathematical Machines of the Czechoslovak Academy of Sciences, but in 1957 he informed his bosses of his intention to take his family back to the USA because of their younger daughter, who was hard of hearing and, allegedly at the recommendation of a doctor, would need to be taught in English as that suited her better.[14] Unlike Nadler himself, the rest of his family retained their American citizenship, and so the children went to the USA first, followed by their mother, and their father joined them later, travelling via India[15] where he worked for 15 months at the Indian Statistical Institute in Calcutta on the development of the first digital computers.[16] He died in the USA in 2013, aged 92.

A peculiar character among the academicians who came from the so-called Western countries was Ian Frank Milner, a New Zealander and a graduate in the political sciences, philosophy and economics from New College, Oxford. After the end of World War II he entered the service of the United Nations, during which he was relocated to New York in 1947. In the following year, the Anglo-American code-breaking team managed to crack communications between Moscow and the Soviet Embassy in Canberra, Australia through which the intelligence gathered by Milner from classified British postwar documents relating to the situation in the Australian region was despatched.[17] Milner, who engaged in extensive espionage for the Soviet Union,[18] probably learnt about it. As a matter of fact, he and his wife travelled to Czechoslovakia in 1950, stating that there was a spa where his wife, who suffered from severe osteoarthritis, could be cured.[19] With the help of the State Police, he got a position at Charles University where he lectured in English Studies, became an associate professor in 1964, and defended his doctoral thesis in 1971. Even though he sympathised with the renewal process and Alexander Dubček during the Prague Spring,[20]

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[9] Cf. e.g. Sergei Ivanovich Zhuk, *The KGB, Russian Academic Imperialism, Ukraine, and Western Academia, 1946–2024*, Lexington Books, 2024: 71–90.

[10] Steven T. Usdin, *Engineering Communism: How Two Americans Spied for Stalin and Founded the Soviet Silicon Valley*, Yale University Press, 2005: 19.

[11] According to Nadler's Recollections, they were Filipp Staros and Joe Berg, see Morton Nadler, No regrets. In Petr Golan, René Kolliner et al., *Almanach historie Výzkumného ústavu matematických strojů 1950–1997, Díl V.: Osobnosti VÚMS a vzpomínky aktérů*, Praha, 2021: 258–285, here: 267.

[12] Mark Kuchment, "The American Connection to Soviet Microelectronics", *Physics Today*, 9 (1985): 44–50, here: 47.

[13] Archiv bezpečnostních složek (Security Services Archive, ABS), Svazky tajných spolupracovníků [Informer Files], sign. TS 114876 MV, Morton Nadler.

[14] Masaryk Institute and Archives of the Czech Academy of Sciences, Archiv Akademie věd ČR, Ústav matematických strojů ČSAV, box 11, inv. č. 24, Morton Nadler's Letter to Section I of the ČSAV of 28 March 1957.

[15] Helena Durnová and Doubravka Olšáková, "Academic Asylum Seekers in the Communist Czechoslovakia". In Marco Stella, Soňa Štrbáňová, Antonín Kostlán (eds.), *Scholars in Exile and Dictatorship of the 20th Century*, Praha: Centre for the History of Sciences and Humanities of the Institute for Contemporary History of the Czech Academy of Sciences, 2011: 90–103, here: 95.

[16] Nikhil Menon, MENON, 'Fancy Calculating Machine': Computers and planning in independent India. *Modern Asian Studies* 2 (2018): 421–457, here: 422.

[17] Phillip Deery, Cold War Victim or Rhodes Scholar Spy? Revisiting the Case of Ian Milner. *Overland* 47 (1997): 9–12.

[18] Desmond Ball and David Horner, *Breaking the Codes: Australia's KGB Network, 1944–1950*, St Leonards, NSW: Allen & Unwin, 1998.

[19] Richard C. S. Trahair and Robert L. Miller, *Encyclopaedia of Cold War, Espionage, Spies, and Secret Operations*, New York: Enigma Books, 2009): 248–250.

[20] Aaron Robert Jackson, Socialism Tells its Own Story: Ian Milner and the Dream of a Redeemed Socialism in the Prague Spring, *New Zealand Journal of History* 2 (2021): 3–31.

he did not leave the country after it was invaded by the armies of the Warsaw Pact, unlike many other foreign nationals. The reason was of course obvious: he would have gained nothing from going to an Eastern Bloc country, and he would have faced actual arrest and extradition to the Australian courts in the West. He eventually lived to see the 1989 Velvet Revolution in Czechoslovakia, and died in Prague in 1991. His collaboration with the Soviet and Czechoslovak intelligence services was definitively proven in the late 1990s when Czech and Australian documents were declassified.[21] It is worth noting for this study that Ian Frank Milner's Prague contacts included also the chemical engineer George Lenell Standart.

All of these immigrants arrived in Czechoslovakia within the relatively short period of 1947–1950, and although each of them can be presumed to have possessed a considerable degree of leftist idealism, the reasons for their emigration varied. Although they can all be seen as representatives of the academic community, there were major differences in their research quality and results.

G. S. Wheeler, Morton Nadler and the Standarts can be considered American intellectuals who radically opposed the US policy in the latter half of the 1940s and who, most probably through Soviet agents trying to approach left-wing American figures and institutions, gained the impression that the Eastern Bloc was a peaceful world, full of social empathy and justice. The McCarthyist anti-communist campaign just getting under way in the USA, which saw them risk being accused of treason and espionage, only confirmed their belief. Nadler and the Standarts became so convinced of their views that they accepted collaboration with the eastern intelligence services, probably expecting to profit from it as a result. Being a former US Army Colonel, Wheeler probably raised too much suspicion[22] and never became an informer of the Czechoslovak State Police. Ian Frank Milner, on the other hand, was the only one in this group of foreign nationals who had acted consciously and systematically as a high-ranking spy against the West before coming to Czechoslovakia, which he then continued in coordination with the Czechoslovak intelligence services.[23] General Antonio Cordón García was in the most dangerous situation on his home turf – his life was literally at risk. After the end of the Spanish civil war, he was forced to flee into exile and never returned to his homeland. His stay in Czechoslovakia, however, was not linked to the Secret Police in any way. It should be said that except for Milner, none of these western émigrés eventually remained in Czechoslovakia, even though Wheeler did return after the events of 1989, and that each of them eventually more or less sobered up from their initial left-wing idealism, quite certainly partly due to the financial hardship they faced in a country with a centrally controlled economy. For G. S. Wheeler and probably also I. F. Milner, the impetus for their deep personal transformation came with the occupation of Czechoslovakia by the Warsaw Pact armies in 1968.

While George Shaw Wheeler was seen as an outstanding economist from the 1930s and enjoyed considerable academic renown throughout his life, Antonio Cordón García's engagement at Charles University almost solely consisted of giving classes in Spanish language and literature. He was not involved in any academic work and published no works. Although Ian Frank Milner compiled an anthology of English fiction and poetry for the students at the Faculty of Arts of Charles University[24] and also made a mark as a translator, his work can in no way be described as academic. Morton Nadler was involved in electrical engineering and IT in the industrial enterprises he worked for, as well as in the Institute for Mathematical Machines after joining the Czechoslovak Academy of Sciences,[25] and was considered a well-established scientist. One truly outstanding member of the academic elite was George Standart, and his wife, too, undoubtedly had great academic potential, but her health and later also her pregnancy basically prevented her from taking her research activities any further, and so any assessment of her academic qualities would be nothing more than speculation.

GEORGE LENELL STANDART

George Lenell Standart (Figure 1) was born on 29 January 1921 in Detroit as the only child of a construction engineer and an official, Lillian, nee Clark. His parents divorced in 1935 and George lived with his mother. He studied chemistry at the California Institute of

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[21] Cf. Tom Heenan, "Milner, Ian Frank (1911–1991)". In Melanie Nolan, Malcolm Allbrook (eds.), *Australian Dictionary of Bibliography*, Volume 19: 1991–1995, Canberra: ANU Press, 2021: 587–588; Petr Hrubý, *Nebezpeční snílci: Australská levice a Československo* [Dangerous Dreamers: Australian Left and Czechoslovakia], Brno: Stilus, 2007: 123–228.

[22] ABS, Svazky kontrarozvědného rozpracování [Operative Files], sign. KR 638164 MV, George Shaw Wheeler.

[23] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 621743 MV, John Frank Milner.

[24] Ian Milner, English Prose and Poetry: Reader for Language and Literature Courses, Praha: SPN, 1974.

[25] His publications include, for instance, Morton Nadler, *Elektronkový oscilograf: instruktivní pomůcka technikům a zájemcům* [Valve Oscillograph: Instructive Guide for Technicians and Interested Parties], Praha: SNTL, 1954; Morton Nadler, Vilém Nessel, Vilém. *Elektronkový osciloskop* [Valve Oscilloscope], Praha: SNTL, 1960.

Technology in Pasadena, taking lectures, among others, with the famous quantum chemist and biochemist Linus Pauling, later a Nobel Prize laureate in chemistry in 1954 and Nobel Peace Prize winner in 1962, who strongly advocated the peaceful coexistence of all states and a ban on nuclear experiments.[26]

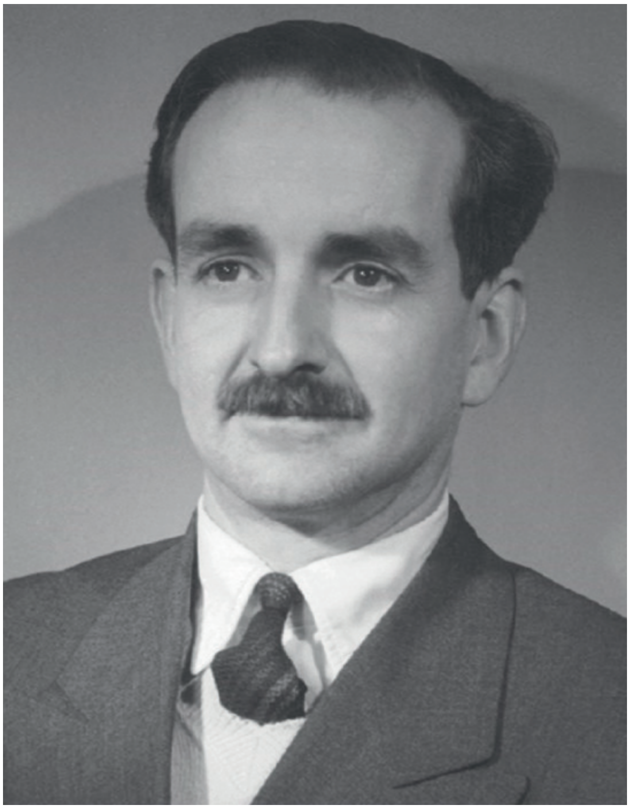

Figure 1. George L. Standart, the First Foreigner Appointed As a University Professor in Czechoslovakia (University of Chemistry and Technology, Prague, Czech Republic)

Back during his university studies Standart had participated in chemical and physical, and chemical and engineering machinery research, and in 1944–1946 was employed by Standard Oil of California in Richmond, namely in laboratories that focused on military research. After the nuclear bombs were dropped on Japan, he began to side with the American peace initiatives, perhaps partly influenced by Linus Pauling.

January 1944, he joined American Youth for Democracy, a communist youth organisation, and in June he became a member of the Communist Party of America. In March 1946, however, he was allegedly instructed to quit his membership. His engagement in a research institution that, besides other things, worked for the US Army was seen by the communist comrades as a potential reason for a communist charge of espionage. The same circumstances led to the departure from the party of his wife Phoebe Anne Standart, the eldest of the two daughters of the astronomer Ferdinand Johannes Neubauer, a Czechoslovak-born German of Mariánské Lázně, West Bohemia, and his wife Margaret. Phoebe A. Standart was born on 27 February 1920 in Palo Alto. She was a graduate in medicine from Stanford University and worked as a bacteriologist and also lectured on biometry, medicinal statistics and epidemiology. Her younger sister Margaret Nancy Neubauer died tragically in 1944 aged 20 while serving in the US Navy.

Figure 2. Phoebe A. Standart (University of Chemistry and Technology, Prague, Czech Republic)

George Standart continued his studies at the California Institute of Technology in 1946–1948 and was allegedly involved in military research topics there.

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[26] Unless otherwise noted, all biographical information for George L. Standard and his wife is quoted from ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart and sign. TS 442023 MV, Phoebe Standart.

After the death of Phoebe's mother Margaret (1947), the Standarts applied for a tourist passport for a trip to France. They did not stay long in Paris, however, and on 5 August arrived in Prague, where they were initially only allowed to stay until 20 June 1949. They applied for political asylum in Czechoslovakia, stating that in the USA research in their academic fields was primarily linked with warfare machinery, and they would prefer to live and work in a people's democracy. Their left-wing convictions, moreover, had discredited them strongly in their native land where the McCarthyist witch hunt targeting communists and their sympathisers was gradually stepping up. Unlike the aforementioned Linus Pauling, who was banned from taking a business trip to Great Britain for a conference in 1950, the reason being that his journey could have threatened US interests,[27] the Standarts were lucky in a way. They had left soon enough before the main witch hunt for American communists and communist sympathisers had got going, and also compared to L. Pauling, an influential figure with a high public profile, they were not, like Morton Nadler, seen as such a great risk.

Their first steps in seeking employment took them to the general directorate of the Czechoslovak Chemical Enterprises, where they arrived on 17 August. George Standart took advantage of a favourable word from one Karel Neubauer, a member of the Commission for National Economy of the Central Committee of the Communist Party of Czechoslovakia. The question that naturally arises is whether this Neubauer could have been a relative of Standart's wife Phoebe; however, this remains unanswered. The Standarts were screened by the plant's specialists, who concluded that they were a highly qualified and skilled pair of experts who could make a major contribution to the development of Czechoslovak chemical sciences and industry.[28]

George Standart initially worked under a so-called expert contract as a consultant and organised expert lectures for employees. His wider involvement, however, was hindered not only by his American citizenship and initial poor language skills, but more importantly by the ongoing reliability check, which certainly was not helped by the fact that his application for a Soviet citizenship in 1949 had been denied, although the reasons for this remained unknown to the Czechoslovak intelligence services.

In 1949, they were both assigned posts at the University of Chemical and Technological Engineering, then part of the Czech Technical University (from 1952 a separate University of Chemistry and Technology, VŠCHT). George Standart took a position at the Institute of Inorganic Chemistry headed by František Šorm, one of the most renowned Czechoslovak chemists and science mangers, and Phoebe Standart at the Institute of Fermentation Chemistry, headed by Arnošt Kleinzeller. Despite that, George Standart remained at the disposal of the chemical industry as required, for instance, when resolving issues with the gas supply system for the chemical plants in northwestern Bohemia.

At university, he first held lectures in chemical engineering, a discipline which was still in its early days in Czechoslovakia, and thermodynamics for staff working in the chemical industry. Commencing in the 1949/1950 term, he launched his own lectures for the 4th grade in Chemical Engineering, a new discipline. He managed to provide all his students with *The Principles of Chemical Engineering*, a book by the American chemists W. H. Walker, W. K. Lewis and W. H. McAdams.[29] In 1950, he helped to set up a new section at the university, the Institute of Chemical Engineering, which was allocated eight rooms, five assistants and one auxiliary staff member. Along with Jan Marek and Zdeněk Novosad, students originally assigned to Standart as academic assistants due to their excellent knowledge of the discipline as well as their good English skills, George Standart prepared the first Czech textbook for the discipline, *Chemical Engineering: Foundations of Equipment Computations*, published in 1951. When the University became independent, the Institute was renamed the Department of Chemical Technology Processes and Apparatuses of the Faculty of Inorganic Chemistry (headed by Hanuš Seidl), and built its technological hall and chemical engineering laboratory in 1955.[30] Chemical Engineering as a discipline made its way to being one of the basic subjects of all the disciplines studied at the VŠCHT at that time, although the journey was far from easy at the beginning. In one of his letters from the early 1950s, G. Standart complained that he was emphatically urged not to lecture on chemical engineering in Prague until a sufficient number of experts in the discipline had been trained, so that lectures could be given in a comparable quality in Pardubice and Bratislava, too. At the same time, he was referred to as a representative of "Prague imperialism".[31]

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[27] Linus Pauling, "My Efforts to Obtain a Passport", *Bulletin of the Atomic Scientists* 7 (1952): 253–256.

[28] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart.

[29] Magdalena Bendová, *Eduard Hála*, Praha: Institute of Chemical Process Fundamentals of the Academy of Sciences of the Czech Republic in Academia Publishers, Center of Administration and Operation AS CR, 2014: 5, 17.

[30] Oldřich Holeček and Jiří Vlček, *Staré pověsti chemicko-inženýrské* [Old Chemical Engineering Legends], Praha: Ústav chemického inženýrství VŠCHT, 2018: 4, 10.

[31] ABS, Svazky tajných spolupracovníků, sign. TS 741518 MV, George Standart, p. 44.

In 1956 the whole institution became part of the Faculty of Fuels Technology, and in 1960 of the newly established Faculty of Automation and Economics of Chemical Production, while lecturing in chemical engineering as a specialised subject was renewed. In the 1980s it was renamed the Department of Chemical Engineering. From the very beginnings, the scientific research at the Institute or Department was focused both on basic research of transport processes and reactions as well as on applied research. Several research groups took shape here in the 1960s, namely for mixing, fluidisation, matter sharing, reactors, and system engineering, all of them excelling with extensive publication work, successful solutions to many industrial problems as well as in international cooperation in Europe and beyond. Its central focus of research was non-linear dynamics – formulating the basic modern theory of this discipline, the construction and methods of computerised analysis of mathematical models of non-linear dynamic systems, and the theory of chaos. G. Standart himself was awarded the State Prize for Chemistry in 1959.[32] In the same year a short popularising film entitled *Chemické inženýrství – Proudění kapalin*, directed by Josef Plíva and with George Standart as expert advisor, received an award – a medal in the Technical Films category at the Rouen International Days of Technical, Industrial and Agricultural Films.[33]

In 1955 he was appointed an associate professor and in 1961 became a professor (Figure 1). His doctoral dissertation, *Transport Phenomena on a Phase Boundary*, was successfully defended in 1963 as the first Czechoslovak thesis of its type in chemical engineering. It was he that instigated the establishment of the Laboratory of Chemical Engineering as part of the Czechoslovak Academy of Sciences, with him becoming its first director, and at the end of the 1950s he also assisted in building up the Institute of Theoretical Foundations of Chemical Technology. He was also the chairman of the Chemical Engineering Section of the Czechoslovak Society for Chemistry. He held postgraduate courses in his field for many years for staff from the chemical industry, and was involved in organising the international CHISA congresses in chemical engineering, equipment design and automation.[34]

It is apparent from the above that George L. Standart's knowledge, at least when he arrived in Central Europe, went far beyond the Czechoslovak standards in his academic fields at the time. Especially his contribution to Czechoslovak chemical engineering must be seen as crucial as he moved the discipline forward by leaps and bounds and nurtured a number of promising students. The space he was provided with for his academic and pedagogical work was well in line with the general postwar preference for the natural and technical sciences. That was due to the global boom in science and technology as well as to local efforts to save the Czechoslovak economy, which was in decline as a result of incompetent changes dictated by the Soviet Union,[35] by means of natural and technical scientific advancements. The chemisation of industry was seen as one of the key factors driving the economic revival so much longed for after the war.[36] A US citizen bringing in new state-of-the-art and highly practicable scientific expertise was thus a welcome acquisition. The culture shock, intensified by the reality of the communist regime, however, lay heavily on him also in the academic and university milieu, such as in the difficulty of working with graduates from the so-called Labourers' Courses intended to pad out the numbers of university graduates of "working-class origin", not to mention the lack of interest shown by state industrial enterprises, with no market-based motivation, in innovations.[37]

While all of G. Standart's twenty years in Czechoslovakia were linked to science (at the VŠCHT and later the Czechoslovak Academy of Sciences), his wife took on several other jobs, such as at the International Students' Union or Czechoslovak Radio. This was due to her poor state of health, especially gynaecological issues, which also caused her three miscarriages at late stages of pregnancy. Undoubtedly, this was also down to the new cultural, material and, especially, political and ideological circumstances that put her under enormous pressure, something she had to adapt to after her arrival in Czechoslovakia. Several years later, however, the Standarts finally had two daughters, Nancy and Sally.

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[32] "Vědecké státní ceny K. G. a vyznamenání v květnu 1959" [K.G. State Scientific Awards and Honours in May 1959], *Věstník Československé akademie věd* 4 (1959): 503.

[33] "Filmy vyznamenané v roce 1959 na filmových přehlídkách a festivalech" [Award-winning Films in 1959 at Film Shows and Festivals]. *Zpravodajství Ústřední správy Československého filmu* 12 (1959): 14.

[34] Věra Dvořáčková, Ivana Lorencová, *Zaostřeno na chemii. Kapitoly z historie Vysoké školy chemicko-technologické v Praze* [Focused on Chemistry. Chapters on the History of the University of Chemistry and Technology in Prague] (Praha: Vydavatelství VŠCHT, 2022): 104.

[35] Namely a focus on heavy industry, the destruction of the free market, liquidation of private ownership, collectivisation of agriculture, etc.

[36] Cf. e.g. MÚA, A AV ČR, Řízení a správa ČSAV I [ČSAV Management and Administration I], Box 18, sign. 78, Records from the meeting of chairmen and secretaries of commissions for the preparation of "Key Directions of Scientific Research Work until 1960".

[37] Cf. e.g. MÚA, A AV ČR, ÚTAM ČSAV, uncatalogued, Myslivec, Alois: Comments on the Theses of the Central Committee of the Communist Party of Czechoslovakia Regarding the Development of Science and Technology, 1 July 1967.

THE INVOLVEMENT OF THE STANDART'S IN THE ACTIVITIES OF THE SECRET SERVICES

Even though George Standart was seen by his colleagues primarily as an excellent expert and, considering his somewhat unusual journey from the West to the East, as an ardent idealist and left-wing enthusiast,[38] whom no one would suspect of collaborating with the Czechoslovak intelligence services, the files lodged with the Security Services Archive give us a different account. Despite the fact that some of the documents pertaining to the Standarts were discarded in the 1980s, the items preserved give us a vivid picture of their involvement.

As of 22 May 1951, G. Standart was officially taken over by the Czechoslovak State Security "from a friendly service" by which he had earlier been "instructed". The "friendly service" was apparently the Soviet KGB, which had most likely contacted him while still in the USA as an intellectual in the left-wing university environment, a communist party member, not to mention an employee involved in military research. His collaboration with the KGB is implied both from the note about having been instructed in respect of intelligence work, and the fact that when he and his wife were poor and needy during their first years in Czechoslovakia, they always turned to the Soviet embassy. They were therefore here under the permanent surveillance of the Soviet intelligence services. The amounts paid to him by the Czechoslovak State Security for collaborating were, according to the preserved documents, approved at "higher positions", i.e., outside the competence of the Czechoslovak services.[39] Incidentally, an identical initial situation can also be seen with Morton Nadler, who had most likely been contacted by the KGB back when working at his US place, and his transfer to Europe was ordered by the Soviets, as was his subsequent self-establishment in Czechoslovakia.

The fact that the Czechoslovak State Police recruited the Standarts as informers "only" after they had spent three years in the country was due not only to the initial mistrust of an American citizen, but primarily also to the fact that the communist secret services were only starting to take shape during the first years after the 1948 February coup. The competences of three ministries (defence, interior, and national security) were just to be defined, as was the State Security's exact scope of work. The service was originally to carry out counter-intelligence tasks, i.e., to hinder foreign espionage in Czechoslovakia, but from the turn of the 1940s/1950s it began to be redesigned following the Soviet model, which, amongst other things, broadened its scope of activity, soon also to include foreign intelligence work. Hand in hand with this went a sharp rise in the number of informers, as well as the people and entities of interest. In 1953 the ministries of the interior and national security merged, following which a centralised and unified registry of operative cases, entities of interest and informers was implemented.[40] This is also evidenced by the fact that the definite memorandum of collaboration with George L. Standart and Phoebe A. Standart was signed at the time, in the first half of 1953.

Standart's code name was Vašek; his wife went by Anna. They were both assessed as diligent, intelligent, conscientious and disciplined people who believed in a people's democracy, who had no ambition to enter the party but regularly participated in May Day parades and other similar events, and who donated 1200 CZK in a public collection to support the starving people of Korea.

Initially they worked in foreign espionage as so-called tipsters who sought suitable foreign candidates to recruit as new agents. If their task was to establish contacts with a foreigner or another person, they always carried out their assignment well. According to the records, they also did intelligence work at the VŠCHT where they focused on particular professors and assistant professors. G. Standart's work was usually evaluated as follows: "One of his advantages is that he has opportunities to make contact with people at higher echelons and foreigners. His limitation is that he is overburdened with school and scientific work, and has considerably less time for collaboration. (…) He reports shortcom-

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[38] Interview with Prof. Miloš Marek by Věra Dvořáčková, 16 November 2021.

[39] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, The Informer "Vasek" – Periodical Assessment, 1958, p. 70.

[40] For details on the activities of intelligence and security services in postwar Czechoslovakia, cf. e.g. Jan Frolík, *Nástin organizačního vývoje státobezpečnostních složek SNB v letech 1948–1989* [Outline of the Organisational Development of State Security in 1948–1989]. Sborník archivních prací č. 2, 1991; František Koudelka, *Státní bezpečnost 1954–1968 (Základní údaje)* [State Security, 1954–1968 (Basic Facts)]. Sešity Ústavu pro soudobé dějiny AV ČR, Svazek 13, Praha: Ústav pro soudobé dějiny AV ČR, 1993; Karel Kaplan, *Nebezpečná bezpečnost (Státní bezpečnost 1948–1956)* [Dangerous Security (State Security in 1948–1956)], Brno: Doplněk, 1999; Jiřina Dvořáková, *Státní bezpečnost v letech 1945–1953 (Organizační vývoj zpravodajských a státně bezpečnostních složek)* [State Security in 1945–1953 (Organisational Development of the Intelligence and State Security Services)]. Praha: Úřad dokumentace a vyšetřování zločinů komunismu, 2007; Jan Kalous, *Štěpán Plaček. Život zpravodajského fanatika ve službách KSČ* [Štěpán Plaček. The Life of an Intelligence Fanatic in the Service of the Communist Party], Praha: ÚSTR, 2010; Jiřina Dvořáková, Zdeňka Jurová, Petr Kaňák, *Československá rozvědka a pražské jaro* [The Czechoslovak Foreign Intelligence and the Prague Spring], Praha: ÚSTR, 2016; Katherine Verdery, *Secrets and Truths: Ethnography in the Archive of Romania's Secret Police*, Budapest: Central European University Press, 2014.

ings he encounters in his occupation, which are quite serious. (…) He is focused on persons that have been granted asylum in Czechoslovakia, and besides this, he is tasked with gathering intelligence on foreigners arriving at Czechoslovakia. The intelligence gained from him turned out to be true after verification. However, for the most part it is of an informative and control nature." The meetings at which he passed information were held either at the Standarts' apartment or at various restaurants, and took place held regularly on a weekly or fortnightly basis.

It is not uninteresting to note that Morton Nadler was entrusted with the task of cross-checking the Standarts' reliability in 1955,[41] actually a very logical process, considering the fact that all the three figures shared characteristics that it would be hard to find in any other informer, i.e., that they were all members of the academic community, shared a focus on the natural and technical sciences, were of US origin, spoke English as their native language, and had arrived in Czechoslovakia under similar circumstances.

In November 1957, when it appeared apparent that the Standarts had firmly settled and had no intention of returning, George L. Standart was transferred to the competence of counter-intelligence, namely the section that dealt with the Anglo-Saxon world. He was tasked with monitoring contacts with foreign nationals who were arriving in Czechoslovakia and the relevant asylum seekers. Besides this, he should occasionally have been available to the science and technology intelligence section. However, after that, no "realisation" was made on the grounds of his intelligence reports; only once did he allegedly provide information about a certain person that involved seditious speech against the state, later verified as true. He was paid the so-called compensation fee for his work until 1960 (initially five thousand crowns per month, and one thousand after the currency reform) when the service came to the conclusion that he was becoming less useful. There were at least two principal reasons why Standart was not pushed into more efficient collaboration as an informer. One was his considerable scientific and pedagogical workload, which he was not prevented from doing because of the excellent renown he enjoyed both at home and abroad. As early as in 1952, the State Security stated the following in respect of his work for the VŠCHT: "He is deemed one of Europe's best mathematicians and thermodynamics experts. The university's prestige would be impaired considerably if he left, as for the time being there is no-one to match him in these disciplines."[42] The other reason was that in the 1960s the number of informers was declining as a result of the liberalising trends in society and a more rational approach to the State Security's work.[43]

It is moreover apparent from the periodical assessment of Standart's (and his wife's) intelligence work for the State Security that his activity was most probably not motivated by an effort to harm someone, but rather stemmed from his idealism and desire for peace when he, unfamiliar with this non-American environment, got tangled up in the work of authorities and organisations the substance of which was far beyond his understanding. His supervisors' growing dissatisfaction with his intelligence work would then have been merely the a logical conclusion of this premise.

However, their trust in his frank approach to the people's democratic regime showed no cracks during the whole of the Standarts' stay in Czechoslovakia. Moreover, his contribution to science and industry was all the more apparent, and the country needed experts of his type. In the 1960s, when increasing numbers of Czechoslovaks were travelling abroad, including to countries of the West, George Standart also journeyed out of the country several times. In 1963 and 1964, he spent a couple of months in Cuba where he helped to establish chemical engineering as a discipline at the University of Havana, and also visited some neighbouring countries, such as Germany.[44]

In 1967 he applied for a permit to leave, with his whole family and at their own expense, for a year-long stay in Great Britain at the invitation of Professor Peter Victor Danckwerts of the University of Cambridge, where he was supposed to stay for the first 3–4 months, and then at the Imperial College in London, where he was to stay for the remaining time. The authorities approved his journey easily; he was only provided with the usual defence instructions explaining extraordinary situations that he might encounter, as well as his model responses. He also mentioned that the USA could demand that he be extradited from Great Britain.

In the spring of 1968, however, the Standarts did not return to Czechoslovakia, nor did they give any news of themselves. Jaroslav Ulbrecht of the ČAS Institute of Chemical Processes (the former ČSAV Institute of Theoretical Foundations of Chemical Technology), who

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[41] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 442023 MV, Phoebe Anne Standart, p. 69.

[42] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Identification Statement, 7 August 1952, p. 18.

[43] František Koudelka, *Státní bezpečnost 1954–1968 (Základní údaje)*, Sešity Ústavu pro soudobé dějiny AV ČR, Svazek 13, Praha: Ústav pro soudobé dějiny AV ČR, 1993, p. 53.

[44] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Closing Report No. 9, 8 February 1963.

stayed in touch with G. Standart, wrote in 2010 that during their British stay, the position of head of the Department of Chemical Engineering at the University of Manchester had become vacant, and G. Standart was offered the post. He not only decided to stay in Britain but subsequently enhanced his excellent scientific renown in his native United States through his lecture tours, and was offered many jobs.[45] However, he was unable to take the offers, as he died in 1978. His wife Phoebe survived him by nine years and died in Stockport, UK in 1987. Their two daughters devoted their life in Great Britain to science. Nancy successfully established herself in biochemistry and Sally in medicine.

The above would imply that his final departure from the Eastern Bloc was probably motivated only by the better conditions, both academic and financial, that he was offered in Britain, and the whole story could be concluded by recognising Standart's successful career in the West and stating the probable ideological sobering of a left-wing intellectual – if it weren't for a report passed to the Czechoslovak Ministry of the Interior by the Hungarian intelligence services in 1980.

This report pertains to George Gerbner, a Hungarian-born American who was a professor of journalism in the USA and, according to information from the Hungarian intelligence services, worked for the US military intelligence at least in 1943–1947. He was therefore suspected, when visiting his native country in 1960, of improperly stating study and family reasons in his visa application only to disguise his continuing intelligence activity. In 1960–1961 he made three journeys to Czechoslovakia, and the Czechoslovak authorities were therefore alerted to stay watchful of his activities. Following this alert, it was found that his Czechoslovak contacts also included George Standart, which puts Standart's later departure for England in a different light.[46]

CONCLUSION

The phenomenon of emigration in connection with communist Czechoslovakia usually involves people leaving the Central European country for the West. The reasons that led to their decision were usually linked to the politically and ideologically motivated persecution they faced, and their unwillingness to reconcile themselves to the idea of living in a society that was not free. The Czechoslovak academic world was considerably weakened by this emigration, as asylum in the West was the ambition in life for dozens or even hundreds of scientists, the overwhelming majority of whom never returned to their homeland. At the same time, only a negligible few academics sought asylum in Czechoslovakia. They were educated, intelligent and highly capable people who made no secret of their sympathies with leftist ideals. Their political stances and opinions saw them face various forms of persecution in their home countries that eventually compelled them to emigrate. By far the most apparent political asylum seeker was Antonio Cordón García, who had played a prominent political and military role in the Spanish Civil War and sided with those who eventually lost it. Moreover, the winner, General Francisco Franco, established a clerical fascist regime in the country that defined itself as anti-communist but suppressed democratic and socialist trends and held on until Franco's death in 1975. Going into exile in an Eastern Bloc country was therefore the logical option for García. The Americans G. S. Wheeler, M. Nadler, G. L. Standart and P. A. Standart also did not feel safe in their homeland with the escalating witch hunt, sometimes fanatical, for communists and their sympathisers. Moreover, during the early stages of the Cold War, their leftist tendencies made them a target for the KGB, which did not hesitate to use them for espionage. The Standarts' and Nadler's collaboration with the Soviet KGB and Czechoslovak StB, now clearly proven, however, was likely not a purposeful effort to help the intelligence services of the Eastern Bloc and thus damage their country and its citizens. The experience they gained while on the eastern side of the Iron Curtain gradually led them to rethink their ideals and, especially in G. Standart's case, to realise that the Eastern Bloc was no less militant than the USA and other "capitalist" countries. The situation was probably different with just I. F. G. Milner, even though his actions may originally have also been based on pure idealism, but during his collaboration with the KGB he became fully aware of the consequences or potential impact that his operations could have.

Based on the documents pertaining to the Standarts lodged in the Security Services Archive, it is apparent that this pair was a tricky case for the Czechoslovak authorities throughout their entire stay in Czechoslovakia. On the one hand, there was unquestionable information that they had been taken over from the Soviet services, where they must have been involved in the late 1940s and early 1950s; on the other hand, there were no other reports that could soundly confirm the so-called reliability of the Standarts. On the one hand, the abso-

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[45] Jiří Hanika, ed., *Ústav chemických procesů AV ČR, v. v. i., Almanach 1960–2010* [CAS Institute of Chemical Processes. Anthology, 1960–2010], Praha: Ústav chemických procesů AV ČR 2010: 4–11, 38–39, 56.

[46] ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Informer Dispatch No. 4/Fx.

lute absence of any shortcoming, or suspicion of shortcoming, on their side was attractive for the Czechoslovak StB, as was Standarts' diligence and intelligence; on the other hand, there was considerable disappointment that the information from the code names Vašek and Anna turned out to be of little use for the State Security. George L. Standart was in fact an outstanding scientist, one of the best in his discipline and at that time, who made an unexpected and unprecedented contribution to the Czechoslovak chemical sciences and their practical industrial applications. Not only did he actually introduce chemical engineering to Czechoslovakia and began teaching it at universities; he also raised the standard of the discipline to a level that would have been unthinkable without him. Even though he never aspired to obtain Czechoslovak citizenship, his definite departure from the country in 1967 was another major blow to Czechoslovak science alongside the damage it had suffered by the emigration of its academic elite to the West. His heavy academic workload prevented him from being more extensively exploited by the Czechoslovak intelligence services, and one can therefore assume that even his potential collaboration with a western intelligence service, whether in connection with his meeting with George Gerbner or not, would have not been overly relevant.

BIBLIOGRAPHICAL REFERENCES

1. V. Jeřábek et al., *Českoslovenští uprchlíci ve studené válce* [Czechoslovak Émigrés in the Cold War], Brno: Stilus, 2005; P. Koura, „Svědomí mi nedovolí šířit takové lži". Český exil mezi únorem 1948 a srpnem 1968 ["Conscience would not Allow Me to Spread Such Lies". Exiled Czechs between February 1948 and August 1968]. In L. Wittlichová (ed.), *Vzkazy domů. Příběhy Čechů, kteří odešli do zahraničí (emigrace a exil 1848–1989)* [Messages to Their Homes. Stories of the Czechs who Left Abroad (Emigration and Exile 1848–1989)], Praha: Dny české státnosti, Labyrint, 2012, pp. 203–215; L. Paukertová, Několik základních údajů o odchodech z Československa, 1948–1991 [Some Basic Facts about Escapes from Czechoslovakia, 1948–1991]. In S. Brouček, K. Hrubý (eds.), *Češi za hranicemi na přelomu 20. a 21. století: Sympozium o českém vystěhovalectví, exulantství a vztazích zahraničních Čechů k domovu 29.–30. června 1998*, Praha: Karolinum – Etnologický ústav AV ČR, 2000, pp. 25–31.
2. S. Štrbáňová, A. Kostlán (eds.), *Sto českých vědců v exilu* [One Hundred Czech Scientists in Exile], Praha: Academia, 2011.
3. D. Olšáková, "V krajině za zrcadlem. Političtí emigranti v poúnorovém Československu a případ Aymonin" ["In the Landscape Behind the Mirror. Political Émigrés in Post-February Czechoslovakia and the Aymonin Case"], *Soudobé dějiny* 4 (2007), pp. 719–743 (here: 738).
4. P. Wright, *Iron Curtain: From Stage to Cold War*, Oxford: Oxford University Press, 2009, pp. 360–361.
5. "Prohlášení George Wheelera, bývalého amerického plukovníka" ["Statement by George Wheeler, a Former American Colonel"], *Rudé právo*, 8 April 1950: 2; Kathleen Geaney, "At Home among Strangers: The Extraordinary Year 1950 in the Life of an Ordinary American Family in Communist Czechoslovakia", *Comenius: Journal of Euro-American Civilization* 1 (2015), pp. 25–42.
6. J. Bašta, "Propagandistické využití kausy amerického emigranta G. S. Wheelera: Dezinformace StB ve sdělovacích prostředcích" ["Propagandistic Use of the Case of American Émigré G. S. Wheeler: Disinformation by the Secret Police in the Mass Media"], *Securitas Imperii* 7 (2001), pp. 224–251 (here: 225).
7. M. Eiroa, *Españoles tras el Telón de Acero: El exilio republicano y comunista en la Europa socialista*, Madrid: Marcial Pons Historia 2018.
8. J. Petráň, *Filozofové dělají revoluci* [Philosophers Doing the Revolution], Praha: Karolinum, 2015, p. 186.
9. Cf. e.g. S. I. Zhuk, *The KGB, Russian Academic Imperialism, Ukraine, and Western Academia, 1946–2024*, Lexington Books, 2024, pp. 71–90.
10. S. T. Usdin, *Engineering Communism: How Two Americans Spied for Stalin and Founded the Soviet Silicon Valley*, Yale University Press, 2005, p. 19.
11. According to Nadler's Recollections, they were Filipp Staros and Joe Berg, see M. Nadler, No regrets. In P. Golan, R. Kolliner (eds.), *Almanach historie Výzkumného ústavu matematických strojů 1950–1997, Díl V.: Osobnosti VÚMS a vzpomínky aktérů*, Praha, 2021, pp. 258–285 (here: 267).
12. M. Kuchment, "The American Connection to Soviet Microelectronics", *Physics Today*, 9 (1985), pp. 44–50 (here: 47).
13. Archiv bezpečnostních složek (Security Services Archive, ABS), Svazky tajných spolupracovníků [Informer Files], sign. TS 114876 MV, Morton Nadler.
14. Masaryk Institute and Archives of the Czech Academy of Sciences, Archiv Akademie věd ČR, Ústav matematických strojů ČSAV, box 11, inv. č. 24, Morton Nadler's Letter to Section I of the ČSAV of 28 March 1957.

15. H. Durnová, D. Olšáková, "Academic Asylum Seekers in the Communist Czechoslovakia". In Marco Stella, Soňa Štrbáňová, Antonín Kostlán (eds.), *Scholars in Exile and Dictatorship of the 20th Century*, Praha: Centre for the History of Sciences and Humanities of the Institute for Contemporary History of the Czech Academy of Sciences, 2011, pp. 90–103 (here: 95).
16. N. Menon, 'Fancy Calculating Machine': Computers and planning in independent India. *Modern Asian Studies* 2 (2018), pp. 421–457 (here: 422).
17. P. Deery, Cold War Victim or Rhodes Scholar Spy? Revisiting the Case of Ian Milner. *Overland* 47 (1997), pp. 9–12.
18. D. Ball, D. Horner, *Breaking the Codes: Australia's KGB Network, 1944–1950,* St Leonards, NSW: Allen & Unwin, 1998.
19. R. C. S. Trahair, R. L. Miller, *Encyclopaedia of Cold War, Espionage, Spies, and Secret Operations*, New York: Enigma Books, 2009, pp. 248–250.
20. A. R. Jackson, Socialism Tells its Own Story: Ian Milner and the Dream of a Redeemed Socialism in the Prague Spring, *New Zealand Journal of History* 2 (2021), pp. 3–31.
21. Cf. T. Heenan, "Milner, Ian Frank (1911–1991)". Melanie Nolan, Malcolm Allbrook (eds.), *Australian Dictionary of Bibliography*, Volume 19: 1991–1995, Canberra: ANU Press, 2021, pp. 587–588; P. Hrubý, *Nebezpeční snílci: Australská levice a Československo* [Dangerous Dreamers: Australian Left and Czechoslovakia], Brno: Stilus, 2007, pp. 123–228.
22. ABS, Svazky kontrarozvědného rozpracování [Operative Files], sign. KR 638164 MV, George Shaw Wheeler.
23. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 621743 MV, John Frank Milner.
24. I. Milner, *English Prose and Poetry: Reader for Language and Literature Courses*, Praha: SPN, 1974.
25. His publications include, for instance, M. Nadler, *Elektronkový oscilograf: instruktivní pomůcka technikům a zájemcům* [Valve Oscillograph: Instructive Guide for Technicians and Interested Parties], Praha: SNTL, 1954; M. Nadler, V. Nessel, *Elektronkový osciloskop* [Valve Oscilloscope], Praha: SNTL, 1960.
26. Unless otherwise noted, all biographical information for George L. Standart and his wife is quoted from ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart and sign. TS 442023 MV, Phoebe Standart.
27. L. Pauling, "My Efforts to Obtain a Passport", *Bulletin of the Atomic Scientists* 7 (1952), pp. 253–256.
28. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart.
29. M. Bendová, *Eduard Hála*, Praha: Institute of Chemical Process Fundamentals of the Academy of Sciences of the Czech Republic in Academia Publishers, Center of Administration and Operation AS CR, 2014, pp. 5, 17.
30. O. Holeček, J. Vlček, *Staré pověsti chemicko-inženýrské* [Old Chemical Engineering Legends], Praha: Ústav chemického inženýrství VŠCHT, 2018, pp. 4, 10.
31. ABS, Svazky tajných spolupracovníků, sign. TS 741518 MV, George Standart, p. 44.
32. "Vědecké státní ceny K. G. a vyznamenání v květnu 1959" [K.G. State Scientific Awards and Honours in May 1959], *Věstník Československé akademie věd* 4 (1959), p. 503.
33. "Filmy vyznamenané v roce 1959 na filmových přehlídkách a festivalech" [Award-winning Films in 1959 at Film Shows and Festivals]. *Zpravodajství Ústřední správy Československého filmu* 12 (1959), p. 14.
34. V. Dvořáčková, I. Lorencová, *Zaostřeno na chemii. Kapitoly z historie Vysoké školy chemicko-technologické v Praze* [Focused on Chemistry. Chapters on the History of the University of Chemistry and Technology in Prague], Praha: Vydavatelství VŠCHT, 2022, p. 104.
35. Namely a focus on heavy industry, the destruction of the free market, liquidation of private ownership, collectivisation of agriculture, etc.
36. Cf. e.g. MÚA, A AV ČR, Řízení a správa ČSAV I [ČSAV Management and Administration I], Box 18, sign. 78, Records from the meeting of chairmen and secretaries of commissions for the preparation of "Key Directions of Scientific Research Work until 1960".
37. Cf. e.g. MÚA, A AV ČR, ÚTAM ČSAV, uncatalogued, Myslivec, Alois: Comments on the Theses of the Central Committee of the Communist Party of Czechoslovakia Regarding the Development of Science and Technology, 1 July 1967.
38. Interview with Prof. Miloš Marek by Věra Dvořáčková, 16 November 2021.
39. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, The Informer "Vasek" – Periodical Assessment, 1958, p. 70.
40. For details on the activities of intelligence and security services in postwar Czechoslovakia, cf. e.g. J. Frolík, *Nástin organizačního vývoje státobezpečnostních složek SNB v letech 1948–1989* [Outline of the Organisational Development of State Security in 1948–1989]. Sborník archivních prací č. 2, 1991; F. Koudel-

ka, *Státní bezpečnost 1954–1968 (Základní údaje)* [State Security, 1954–1968 (Basic Facts)]. Sešity Ústavu pro soudobé dějiny AV ČR, Svazek 13, Praha: Ústav pro soudobé dějiny AV ČR, 1993; K. Kaplan, *Nebezpečná bezpečnost (Státní bezpečnost 1948–1956)* [Dangerous Security (State Security in 1948–1956)], Brno: Doplněk, 1999; J. Dvořáková, *Státní bezpečnost v letech 1945–1953 (Organizační vývoj zpravodajských a státně bezpečnostních složek)* [State Security in 1945–1953 (Organisational Development of the Intelligence and State Security Services)], Praha: Úřad dokumentace a vyšetřování zločinů komunismu, 2007; J. Kalous, *Štěpán Plaček. Život zpravodajského fanatika ve službách KSČ* [Štěpán Plaček. The Life of an Intelligence Fanatic in the Service of the Communist Party], ÚSTR, Praha 2010; J. Dvořáková, Z. Jurová, P. Kaňák, *Československá rozvědka a pražské jaro* [The Czechoslovak Foreign Intelligence and the Prague Spring], Praha: ÚSTR, 2016; K. Verdery, *Secrets and Truths: Ethnography in the Archive of Romania's Secret Police*, Budapest: Central European University Press, 2014.

41. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 442023 MV, Phoebe Anne Standart, p. 69.
42. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Identification Statement, 7 August 1952, p. 18.
43. F. Koudelka, *Státní bezpečnost 1954–1968 (Základní údaje)*. Sešity Ústavu pro soudobé dějiny AV ČR, Svazek 13, Praha: Ústav pro soudobé dějiny AV ČR, 1993, p. 53.
44. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Closing Report No. 9, 8 February 1963.
45. J. Hanika (ed.), *Ústav chemických procesů AV ČR, v. v. i., Almanach 1960–2010* [CAS Institute of Chemical Processes. Anthology, 1960–2010], Praha: Ústav chemických procesů AV ČR 2010, pp. 4–11, 38–39, 56.
46. ABS, Svazky tajných spolupracovníků [Informer Files], sign. TS 741518 MV, George Standart, Informer Dispatch No. 4/Fx.

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Research Article

The Early Development of the Casale Process for the Production of Synthetic Ammonia (1917-1922). The protagonists, the technology, and a link between Italy and the United States

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Abstract. The Haber-Bosch process for the industrial synthesis of ammonia, originally intended for use in manufacture of nitrogen fertilizers, was inaugurated by BASF of Germany in 1913. During the First World War the process proved to be of tremendous value to Germany for the production of munitions. This was appreciated by the Allied nations, but, despite great efforts, they were unable to replicate the process prior to the cessation of hostilities. Notwithstanding tremendous postwar demand for nitrogen products to ensure national security in both munitions and fertilizers, BASF refused to license its ammonia process. This ultimately forced inventors and firms elsewhere to innovate based on their wartime research efforts. This paper provides an account of the emergence of the first successful rival to the BASF Haber-Bosch synthetic ammonia process, that of the Italian inventor Luigi Casale. To accomplish this goal, Casale in 1917 gained the support of the wartime chemical manufacturer Idros, at Terni, north of Rome, headed by the Franco-American entrepreneur René Leprestre. Better facilities for development of a working process, including byproduct hydrogen, were available at the works of the Rumianca company. Casale moved there in 1919. Leprestre brought in representatives of a group of investors from the United States to observe Casale's process in action at the Rumianca works. However, disputes quickly emerged. Agreements were broken, followed by lengthy litigation, and the return of Casale to Idros at Terni. At Terni there were different problems. There, barriers to the supply of electricity were created, in part because the ammonia technology was perceived to be highly disruptive to another, well developed, local nitrogen process, that of calcium cyanamide. Compromises were reached. The outcome was the almost simultaneous foundation in 1921 of two companies, Ammonia Casale SA, in Lugano, Switzerland, which handled international licensing, and the Società Italiana per l'Ammoniaca Sintetica (SIAS) which absorbed the Idros works. In 1922 SIAS acquired a mothballed hydroelectric factory at Nera Montoro. It would serve as the industrial testing site for improvements in Casale's process. The widespread, and rapid, dispersal of Luigi Casale's highly successful synthetic ammonia process became an outstanding example of technology transfer from Italy during the early 1920s. This transfer included, through Leprestre, to the United States, as recently described in *Substantia*. The origins of the transatlantic interest can be discerned in the very earliest attempts by Casale to develop his novel ammonia technology, as described here.

Keywords: Luigi Casale, René Leprestre, Nitrogen fixation, Italian Chemistry, Terni, Casale Ammonia Process

INTRODUCTION

The three most important plant nutrients are nitrogen, phosphorus and potassium. While mineral sources of phosphorus and potassium are abundant and readily accessible, this is not the case for nitrogen even though it constitutes the greater part of the earth's atmosphere. In the nineteenth century the main source of nitrogen fertilizer was South America, first the native guano (accumulated bird droppings), and then sodium nitrate, extracted from rocks in the desolate Atacama Desert. Concerns in Europe that the nitrate might become depleted early in the twentieth century stimulated research into methods for fixing atmospheric nitrogen. Two processes requiring inexpensive electricity were introduced in 1905. However, they were restricted to regions where continuous sources of cheap hydropower were available. The situation changed in 1913 with the inauguration of the Haber-Bosch process for the production of synthetic ammonia by BASF in Germany. It is considered to be one of the greatest triumphs of twentieth century technology. However, the process was proved not through its ability to feed the worlds' population but through its use in the manufacture of munitions by Germany during the First World War. The Allied nations failed in their elusive quests for breakthroughs in replicating the German technology. After the Armistice, BASF refused to license the Haber-Bosch process, which was widely considered essential for maintaining national security in both agriculture and defence. This encouraged the development of wartime research in order to achieve success in the fixing of nitrogen as synthetic ammonia. As a result, by the early 1920s, three processes had been developed, by Georges Claude in France, and Luigi Casale and Giacomo Fauser in Italy. It is noteworthy that the Claude and Fauser processes feature more in the technical and historical literature than the Casale process*. Perhaps this is because they were associated with, and promoted by, large corporations (Air Liquide and Montecatini, respectively). Yet the Casale process, by completely overcoming problems of fractures in the outer walls of converters (as encountered in the Claude process), was the first to achieve a high level of sustained performance, and, moreover, through licensing arrangements and supply of the complex machinery (unlike Montecatini at least until the mid-1920s), to launch the global synthetic ammonia industry. This was no mean achievement, requiring safe operation under brute force conditions and the production of pure gases, nitrogen and hydrogen. How the lone-inventor Luigi Casale was enabled to achieve this pre-eminence is the remit of this paper. It involves Casale's engagement with two wartime startup companies, the influence of a Franco-American entrepreneur, who was also an influencer in gaining business from the Italian government, disputes over contracts, and litigation, and negotiations with a firm that saw the Casale process as a disruptive technology and prevented access to the critical hydroelectric power needed for driving machinery and for electrolysers that produced pure hydrogen. The outcome was the founding in 1921 of two companies that represented Casale interests, one to promote licensing outside Italy, the other to take up manufacture and onsite testing on a scale that overcame the limitations of Casale's workshop in the city of Terni, Umbria, central Italy.

THE NITROGEN PROBLEM WITH SPECIAL REFERENCE TO ITALY ON THE EVE OF THE FIRST WORLD WAR

From the 1840s, an important source of nitrogen as agricultural fertilizer was guano, vast amounts of bird droppings, imported mainly into Great Britain from the Chincha Islands off Peru, and marketed throughout Europe. The peak of imports into Europe was reached in 1870 (280,000 tons). The progressive depletion of the richest deposits of guano led to its replacement from around 1880 with sodium nitrate, also exported from South America, mainly to Great Britain, but now from Chile.[1] The nitrate was also exported to North America. A significant advantage of Chilean nitrate was its flexibility of use: in addition to being used as fertilizer for agriculture, it could be transformed into nitric acid for the manufacture of explosives and organic products such dyestuffs.[2]

Relevant here is the fact that in the early 1900s only a small fraction of nitrate, about 50,000 tons per year, were imported into Italy. This resulted in insufficient fertilization of Italian land; in fact, the average consumption of nitrate per hectare in Italy was significantly lower than in other European nations, just one twenty-fourth that of Belgium, one eighth that of Germany, and about half that of by France.[3] The deficit contributed to low agricultural productivity, with a grain yield of barely 20 quintals per hectare in some regions, a figure which, although similar to the German average, was still below that of Great Britain. As a result, dependence on foreign imports of wheat remained high, highlighting the need for more intensive agricultural activity and for the production of nitrogen fertilizers in Italy in order to ensure food self-sufficiency.

Another important fertilizer in Europe in the late nineteenth century was ammonium sulphate, obtained mainly from gas works where coal was distilled for the production of illuminating or coal gas. For the gas and coking plants, the recovery of ammonia and the production of the sulphate was considered a secondary activity compared to the more profitable production of gas. Nonetheless, in 1913, 433,000 tons of sulphate were produced in Britain, most of which was exported. Of this amount, only 30,000 tons was destined for Italy, where national production of sulphate was estimated at around 5,000 tons.[4]

Chile's sodium nitrate retained its role as the main source of nitrogen fertilizer for Italy and for much of the world. However, at the beginning of the twentieth century there was a widespread fear of the depletion of nitrate reserves within just a few decades. This created a nitrogen problem, that took on considerable urgency in industrialized countries, where projections of population growth suggested that they were heading towards a food crisis of potentially catastrophic dimensions. In 1898 Sir William Crookes, then president of the British Association for the Advancement of Science, in a highly publicised address before the association, made an urgent appeal for a solution to what he called the "Wheat Problem." There was, he opined, "a glimmer of light into darkness and despair," namely through the *fixation of atmospheric nitrogen*.[5]

The fixation of atmospheric nitrogen, especially its combination with other elements to create stable compounds suitable as fertilizers, captured the close attentions of several scientists and entrepreneurs. Though the reaction between nitrogen and hydrogen to produce ammonia is represented by a simple equation, it presented considerable difficulties. The first attempts at production of "synthetic nitrates," involved combination of nitrogen with oxygen rather than hydrogen and took place in electric arcs.

In this method, introduced successfully in 1905, atmospheric nitrogen was reacted at temperatures of about 2,000-3000 °C with oxygen under the action of powerful electric discharges produced by a direct current ($N_2 + O_2 \rightarrow 2NO$). This process generated oxides of nitrogen which were further oxidized and then dissolved in water, forming a solution of nitric acid from which a wide range of nitrates could be derived ($3NO_2 + H_2O \rightarrow 2HNO_3 + NO$). Because the main disadvantage of the electric arc process was its high consumption of electricity, factories using this technology were predominantly located in Norway, where there was sufficient low cost hydropower. The main product was *Norgesalpeter*, a form of calcium nitrate $Ca(NO_3)_2$ that was widely exported (about 115,000 tons in 1913, of which 3,000 tons arrived in Italy) and that had an efficacy in soils similar to that of sodium nitrate.[6] However, because market prices were determined by the price of Chilean nitrate, this process was not successful elsewhere due to the prohibitive energy costs.

Another means for fixation of atmospheric nitrogen was by the electrothermal production of calcium cyanamide from calcium carbide in electric furnaces, a process invented in Germany by the chemists Adolph Frank and Nikodem Caro.

$$CaC_2 + N_2 \rightarrow CaCN_2 + C$$

Again, the main constraint was represented by the high energy consumption. However, this electrothermal process was important in Italy, which though it suffered from a scarcity of fossil fuels, particularly coal, possessed the water resources needed to produce hydroelectric power. The cyanamide process, significantly, required much less electricity than the arc process (12,000 kW per ton of nitrogen compared to 61,000 kW), and cyanamide was a useful nitrogen fertilizer. Also, treatment of calcium cyanamide with superheated steam gave ammonia, from which ammonium sulphate fertilizer could be produced.[7] The first large cyanamide factories were set up north of Rome in the industrial city of Terni, in the Conca Ternana region of southern Umbria, also in 1905.

At Terni, surrounded by hills and mountains, the Nera and Velino rivers were two of the main engines of Italian industrial development, favouring the construction of a weapons factory and the nation's first steelmaking factory, where rails for the expanding railway network were manufactured. Exploitation of hydraulic energy (hydropower), known as "white coal", for the production of cheap electricity, led to the establishment of the first plants for the production of calcium carbide by the Società Italiana per il Carburo di Calcio e Altri Gas (Carburo, established in 1896).

Carburo emerged as a leader in the Italian and international markets for calcium carbide and its derivatives, in particular acetylene, which was widely used for public lighting in cities. After the efficient transmission of electricity over great distances became available, the use of acetylene declined, and carbide took on a new role. Carburo acquired the patents of Frank and Caro for the conversion of carbide to calcium cyanamide. Frank and Caro, with mainly German backers, established the Società Generale per la Cianamide, in Rome, an organisation that licensed the cyanamide process worldwide. In 1914, 15,000 tons of cyanamide were produced in Italy.[8]

However, the main breakthrough in nitrogen fixation occurred in Germany, at the technical institute in Karslruhe. There, in 1909, the physical chemist Fritz Haber brought about the direct synthesis of ammonia from its elements on the laboratory bench.[9] Because the yield was low, the process was designed to be continuous, with recirculation of unreacted gases. The apparatus, including the reaction chamber (converter) was made of steel. BASF, of Ludwigshafen, the leading chemical company in Germany, took on the challenge of industrial scale up of Haber's invention. This included design of the necessary equipment to withstand the high pressures and temperatures of a reaction that took place in the presence of an active but inexpensive catalyst, and apparatus for production of pure hydrogen and nitrogen. A team of chemists and engineers under the direction of the engineer Carl Bosch overcame a slew of difficult technical problems, especially overcoming difficulties with the steels then available, and in the design of innovative gas compression machinery. The best catalyst was found to be iron, containing promoters. In September 1913 the full-size industrial plant, at Oppau, near Ludwigshafen, produced the first quantities of synthetic ammonia. The gas was cooled by refrigeration to give the anhydrous product. What became known as the Haber-Bosch process operated at a pressure of 250 atm and a temperature of about 500° C.

$$N_2 + 3H_2 \rightleftarrows 2NH_3.$$

The converter was the core of the Haber-Bosch process. The mixed nitrogen-hydrogen gas, under the high pressure, was introduced into its lower part and forced through a tubular heat exchanger where it was preheated by gas leaving the reaction area. The hot charge of gas then rose through a central tube to the top of the converter and was directed down into the catalyst chamber where the reaction took place. Here, around 8% of the gas mixture was transformed into ammonia. The mixture of gases passed through the preheat tube bundle, after which it was expelled at the base of the converter.[10] Ammonia was isolated and the unreacted gas mixture was recirculated.

Shortly after war broke out in 1914, Germany's supply of Chilean nitrate was cut off by a British naval embargo. Germany then turned mainly to the Haber-Bosch process for the nitrogen products required in manufacture of munitions. The Allies took note. With the urgent increase in nitrogen demand for the production of explosives and fertilizers during the war period, solving the "nitrogen problem" became critical among all major belligerents. It soon became apparent that in the production of synthetic ammonia the combined technological and chemical capabilities of the Allies was decidedly inferior to that of Germany. Shortages of nitrogen products meant shortages of both explosives and bread. In Italy, in particular, the conflict made imports of sodium nitrate difficult, while production of calcium cyanamide and ammonium sulphate was insufficient to meet the new requirements imposed by the war.[11] In addition, Italy suffered from a shortage of wheat from Russia and Romania following the closure of the Dardanelles.[12] These factors highlighted the need to develop an industry for the production of synthetic nitrogen compounds. But, as elsewhere, there was very little progress, even immediately after cessation of hostilities.

The possibility of adopting the Haber-Bosch process in Italy after 1918, as was the case elsewhere, encountered obstacles due to the policy of BASF in refusing to grant licenses. There was also the need of coal to produce hydrogen as well as nitrogen, according to the BASF processes. Coal was a scarce raw material in Italy. This would have meant dependence on the import of coal to meet nitrogen requirements. However, there was an alternative, production of hydrogen from water by electrolysis, even though electrolysis required a vast amount of electricity.[13] For Italy, for strategic reasons, the choice between the two processes, coal-based or electrolytic, was not difficult. Production of synthetic ammonia would draw on electrolytic hydrogen, ensuring that an Italian nitrogen industry would be independent of imports of fossil fuels. Also, the large-scale increase in the production of synthetic ammonia would lead to a significant reduction in unit costs of nitrogen as compared with the arc and cyanamide processes.[14]

As a result, several Italian chemists undertook research to develop a process for the synthesis of ammonia from electrolytic hydrogen. Among them, Luigi Casale emerged as the most prominent figure.

LUIGI CASALE AND THE PILOT PLANTS IN TERNI AND PIEVE VERGONTE

Luigi Casale was born on 22 November 1882 in Langosco Lomellina (fig. 1).[15] He graduated in chemistry at the University of Turin, where he took the Advanced Course of Physics, Chemistry and Electrotechnics, under Professor Arturo Miolati. Subsequently, he began his academic career as an assistant at Turin, and later at the Institute of General Chemistry and Electrochemistry. During 1912-1913 he undertook research in Berlin under the guidance of physical chemist Walther Nernst, famous for his studies on physical chemistry, including the equilibria in gas reactions when carried out under various conditions, and to a great extent paralleling the studies of Fritz Haber.[16] In Berlin, Luigi was joined by his wife Maria Sacchi, also a chemist.

After Italy joined the Allies in 1915. Luigi Casale moved to Naples and joined the university, where he collaborated with Oscar Scarpa, who taught physical chemistry, and had a strong interest in nitrogen fixation. Scarpa was also director of the Electrochemical Laboratory of the Royal Polytechnic High School of Naples. In addition to their joint interests, Casale engaged in research into asphyxiating gases and protective measures for the war effort at the university's Institute of Pharmaceutical Chemistry and Toxicology, directed by Arnaldo Piutti.

Figure 1. Luigi Casale as a young man. (Collection of Francesco Casale)

While undertaking this research, Casale became the victim of severe poisoning from manipulating a toxic gaseous mercury compound. This incident, which took place in 1917 (and would lead to his untimely death in 1927), forced him to abandon the study of asphyxiating gases. He decided to devote himself to the fixation of atmospheric nitrogen as synthetic ammonia, at first working closely with Scarpa. Both Miolati and Scarpa had taken up an interest in nitrogen fixation even before the war at a time when chemistry departments in Italian universities and technical institutes were increasingly dealing with industrial problem solving. This new direction for Casale was probably suggested by his mentor, Miolati, a prominent figure in the commissions established during the war to solve the urgent needs related to war materials of various kinds, including explosives and products for the chemical industry and agriculture. But even more significant was the fact that Casale's time in Berlin had introduced him to the physical chemistry of gas reactions, including between hydrogen and nitrogen.

Casale soon moved to the city of Terni, where, no doubt on the recommendation of Miolati, he joined Idros, an important chemical company that by electrolysis of water produced hydrogen for the Italian navy for use in dirigible and other aerial balloons, and oxygen for oxyacetylene welding. Società Idros (its name derived from idrogeno and ossigeno) was founded in Bologna on 19 February 1916.[17] The objects of establishment of Idros included, in addition to production of hydrogen and oxygen, the extraction of nitrogen from the atmosphere and generation of electricity. Prominent members of the board of directors were René Leprestre, as president, the lawyer Mario Santangelo, as CEO, and Carlo Andreucci, as technical expert.[18] Leprestre, a Franco-American entrepreneur, based in New York City, had played a key role in the creation of Idros specifically for production of certain industrial and military chemicals (fig. 2).

Figure 2. Portrait of René Leprestre, in 1916. (Elizabeth Burrows [family] Collection)

Leprestre profited from various business activities conducted during the war. He appears to have travelled abroad frequently. In the summer of 1916, he visited France, Great Britain and Italy to organise and manage the sale of horses and mules from the United States to European armies. Aware of the facilities offered to industrial enterprises engaged in the war effort by the special war legislation, Idros in November 1916 signed a contract with the Italian Military Administration for the supply of 36,000 240 mm bombs at the price of 123,6 lire each, for a total order amount of £4,490,000[19], and another contract for the supply of 237.75 cubic metres of oxygen per day at the price of 1.1 lire per cubic metre.[20] Around the same time, Idros began receiving orders from the Italian Air Force for production of electrolytic hydrogen at an auxiliary plant. The need for nitrogen products, essential both as fertilizers and for the production of explosives, was particularly critical. Through the inventor Carlo Andreucci, Idros became interested in synthetic ammonia.

Towards the end of 1916 the Idros facility in Terni was enlarged. Shortly after, Carlo Andreucci obtained a patent for the extraction of atmospheric nitrogen by burning a mixture of air and hydrogen.[21] It is therefore likely that Casale moved to Terni in 1917 in order to collaborate with Andreucci on a process for synthetic ammonia that drew on Casale's studies in Berlin and followed the basic chemistry and workings of Haber's method. By July 1917, with the support of Santangelo, who was interested in the technological and economic developments of nitrogen fixation, Casale and Andreucci had developed a method for the continuous production of ammonia in a closed apparatus through recirculation of unreacted gas.[22] Their small experimental apparatus produced a few kilograms of ammonia. The reaction was carried out at temperatures between 350 and 600° C in the presence of a catalyst, as documented in the patents filed in Italy on 18 July 1917 and in France on 19 October 1917 on behalf of Casale, Andreucci and Santangelo.[23] Although the piping of the ammonia unit was declared suitable for resisting high-pressure gases, the pressure range used was not specified, presumably to prevent possible imitations. Only later would it be revealed that Casale employed a considerably greater working pressure than Haber, with as, Casale calculated, a much greater yield. The nearby Terni steelworks provided a suitable steel pressure vessel, as converter, which according to later accounts was the redundant barrel of a naval cannon from the battleship *Dante Alighieri*.

However, under the wartime conditions, Idros, could not spare the hydrogen necessary for Casale's pilot plant research. This was because rights to practically all the local water resources were controlled by Carburo. Idros had access to sufficient water from the Nera to enable production of gases required by the military. Apart from this consideration, Idros might have considered the ammonia process a somewhat speculative venture.

As a result, Casale, on 16 July 1918, entered into a collaborative arrangement for further development with the engineer Alfonso Vitale, the CEO of the chemical manufacturer Rumianca. Rumianca, founded in 1915, was engaged in the production of alkali (caustic soda) and chlorine using the electrolytic Kastner-Kellner process, as well as the manufacture of war gases, including phosgene. Substantial quantities of hydrogen, for which apparently there was no demand, were produced as a by-product of the electrolysis process. Also, the Rumianca factory at Pieve Vergonte, in the Piedmont region, included a suitable working area, with idle equipment, including compressors and pumps, and vessels capable of withstanding high pressures. Casale probably already had in mind development of the ammonia process alone (without Andreucci) but he needed adequate equipment and in particular hydrogen. Therefore, the interests of both parties merged: Rumianca's wartime plant included machinery that Casale needed as well as pure hydrogen; and Rumianca could possibly benefit from the discoveries of Casale.

The facilities seemed ideal for demonstrating the industrial feasibility of Casale's ammonia process. According to the agreement, Rumianca undertook to provide Casale with the necessary resources to register relevant patents. Casale agreed to keep confidential the practical details of the production process, and Rumianca would cover the related expenses.

Towards the end of 1918, Casale moved to the Rumianca factory, where he undertook the construction of his first 100 kg per day ammonia unit. By May 1919, the plant was operational, albeit with intermittent stoppages. Casale filed his first patent relevant to the process on 12 July 1919 (granted on 12 July 1920) for an apparatus suited to the production of mixtures of hydrogen and nitrogen for the ammonia synthesis by burning air in a hydrogen atmosphere. The burning took place inside a combustion chamber, from which the resulting mixture (of water, hydrogen and pure nitrogen) was passed through a refrigerant, enabling ready isolation of the reactant mixture of nitrogen and hydrogen.[24]

The success with the 100-kg unit led to the drawing up of a formal agreement between Casale and Vitale on 17 May 1919. According to this agreement, valid for a period of five years, Rumianca would provide suitable premises, technical and scientific equipment, and qualified personnel for the construction and operation of a semi-industrial synthetic ammonia plant capable of producing 1,070 kg per day. To this end Rumianca would supply 3,000 cubic metres of hydrogen to match up with the requirements of the intended ammonia production. Casale would receive a royalty of 0.1 lire per kg of ammonia produced, reduced to 0.08 lire per kg if the production exceeded 1,500 kg per day. In addition to the royalty, Casale was guaranteed a monthly allowance of 1,500 lire, valid until 31 March 1920.

This agreement took place at a time when Italian factories were going through a major, and expensive, crisis in converting and reverting to peacetime needs, which included seeking opportunities to exploit byproducts. It was anticipated that the market for nitrogen fertilizers, especially in Italy, would be dominated by calcium cyanamide. Ammonia was not considered a major contender for satisfying the nitrogen fertilizer demand. Synthetic ammonia was a gamble for Rumianca, and probably considered suitable for distribution to the alkali industry and for refrigeration, and certainly not for large-scale production as would be required in the manufacture of fertilizer. Rumianca in any case had to restrict production of ammonia to the maximum amount of byproduct hydrogen (3,000 cubic metres per day). There were no plans to increase production of alkali which would have enabled production of more ammonia. These were difficult times. Because of the post-war crisis, Rumianca, in common with other Italian chemical companies, was facing financial difficulties.

This is perhaps why, in exchange for his services and commitments to Rumianca, Casale was granted the right to bring in other parties prepared to invest in Rumianca's ammonia. There was, however, an important caveat in the agreement, regarding certain business conditions: It had to be ratified by 30 September, at which time Rumianca would need to have provided all the machinery and made available the necessary daily supply of hydrogen to enable production of ammonia at the rate of a calculated 1,070 kilograms per day.

Notwithstanding the confidentiality agreement with Rumianca, Casale maintained close relations with Idros, especially with Leprestre, who had been busy developing business networks with political and banking representatives in Italy and the United States. In 1917, during his stay in Italy, Leprestre had established important political and military relations, in particular with the Minister of Trade Francesco Saverio Nitti.[25] Their negotiations focused mainly on agreements aimed at accelerating the process of Italian industrialization to support the war effort, especially through the use of hydroelectric energy, and to ensure the supply of war material from the United States. This no doubt brought in more business for Idros and may have led to Leprestre's meeting with the Italian-American banker Vincent Handley. Handley was company secretary of the American Italian Commercial Corporation, established at the behest of Banca Commerciale Italiana in New York in 1917 to facilitate trade in steel and ships destined for Italy.

By May 1919, Leprestre, from whatever information he had gleaned, understood that the ammonia method developed by Casale was proving successful and could potentially constitute a means to challenge the monopoly of the BASF Haber-Bosch process for production of synthetic ammonia. On 11 July 1919, Casale and Leprestre signed an informal agreement, within the terms that Casale had agreed in May with Rumianca, giving Leprestre the right to visit the ammonia plant at Rumianca. Three days later, on 14 July, Leprestre accompanied a group of American investors interested in Casale's process on a visit to the Rumianca plant. The participants were a Mr Scheffey, a Dr Fred Chamier and Vincent Handley. With them was Giuseppe Bruni, professor at the Royal Polytechnic of Milan and the Royal University of Milan, who was charged with providing testimony and advice concerning Casale's process as an independent expert. On a second visit a week later, the same American group was impressed by the claim that an average rate of production of 1,070 kg per day for three months had been achieved (which was not the case).

Consequently, on 2 August 1919, Casale and Leprestre signed a formal, secret agreement for the commercial exploitation of the Casale process for the production of synthetic ammonia, before the notary Agatocle Mollaioli of Rome.[26] In the contract, Casale clearly specified that he excluded Carlo Andreucci and Mario Santangelo from any involvement, reserving exclusively for himself and Leprestre ownership of the two Casale inventions through their patents, one for the production of synthetic ammonia and the other for production of a mixture of hydrogen and nitrogen, for both Italy and abroad. Casale also declared in the agreement with Leprestre that he had entered into an agreement with Rumianca, conditional on ratification within sixty days, specifying the quantity of ammonia produced, and accepting the clause covering protection of industrial secrecy. However, Casale retained the right to ask third parties to examine the existing or future facilities he developed at Rumianca, with a view to investing in Rumianca's ammonia business.

In addition, according to the agreement with Leprestre, Casale would allow Bruni, the technical expert, in accord with the Casale-Rumianca agreement, to check data from the plant until late September 1919, in order to determine the industrial cost for each kg of synthetic ammonia produced. On the basis of a satisfactory report prepared by Bruni, Leprestre would be committed to pay Casale the sum of one hundred thousand lire as compensation, in addition to an additional four hundred thousand as the initial payment for the acquisition of processes and related patents, for use both in Italy and abroad. It was then reiterated that the agreement with Rumianca would remain in place subject to its meeting the conditions of its contract.

The sums specified by Leprestre as payable to Casale would be handed over only after Casale had explained, in front of Leprestre and two expert chemists designated by the American group, the recipe for the composition of the catalyst, demonstrated the process of its preparation, and provided a satisfactory estimate for the cost of operation. The demonstration would involve placing the catalyst in a sealed tube, under the supervision of those present, and its insertion into the converter, followed by verification, by observation, of its industrial performance. Once the industrial cost per kg of anhydrous ammonia and the performance of the catalyst had been demonstrated on a semi-industrial scale, the American group would undertake to: acquire and protect the patent; guarantee to invest ten million lire in Rumianca for developing a process capable of manufacturing ammonia at the rate of 20 tons per day; and also promote the process in the United States.[27] The American investors would invest in the Casale process by selling licenses and generating royalties in the United States. Rumianca would receive an injection of funds in order to scale up production to 20 tons of synthetic ammonia per day, and perhaps the investors would acquire a controlling interest in Rumianca, because ten million lire was four times the share capital of Rumianca. In these circumstances, Rumianca would quickly gain the synthetic ammonia monopoly in the Italian market.

If, however, Rumianca was unable to fulfil its part of the agreement, that is, by 30 September, Casale would declare the prior agreements void, and move to Idros at Terni to build an industrial ammonia unit. There, Leprestre would provide all the necessary means in order to allow Bruni to gather data on the spot. In addition to the total compensation of five hundred thousand lire, Leprestre would pay Casale 20 % of the profits deriving from the commercialization of patents. In addition, Leprestre would provide Casale with all the necessary means to establish a chemical laboratory in Terni for the industrial development of the catalyst, and supply the recipe to the companies that adopt the Casale process. The laboratory would also conduct further studies and research under the direction of Casale, who would receive a monthly allowance of 1,500 lire and 20 % of the net profits, in addition to the foregoing amounts, on income arising from innovations made at the laboratory.

It was probably in August 1919 that relations between Casale and Vitale began to deteriorate, probably as a result of the CEO of Rumianca learning of the secret contract between Casale and Leprestre. This would explain why Vitale started getting in the way of Casale's efforts. As a result, in a letter dated 25 August 1919, Casale reminded Vitale that the conditions agreed in May had not yet been realised. The earlier agreement was based on the strict caveat: the deadline for the finalisation of the agreement, based on the working of the process, was 30 September. Casale stated that in the absence of the agreement being concluded he would cease all cooperation with Rumianca.[28] Vitale's response, on 13 September 1919, emphasized the need to make some amendments to the convention, since, in his opinion, the industrial feasibility of the Casale process had not yet been demonstrated.[29]

In October, Casale replied, informing Vitale that he had appointed his legal counsel, the lawyer Mario Santangelo, to negotiate with the vice president of Rumianca, the prominent engineer Lorenzo Allievi, to permit only those changes deemed fair and reasonable. Santangelo was confronted with an outright rejection by Vitale. The parties could not come to an arrangement for permitting the requested changes. Rumianca filed a lawsuit against Casale, claiming that he had reneged on their contract. Casale submitted to the court a detailed expert report drawn up by Professor Bruni. The litigation would continue for a decade and is described later. Casale moved to Terni.

THE RETURN TO TERNI AND THE AGREEMENT BETWEEN CASALE AND CARBURO

In contrast to the experimental plant at Rumianca, for which less power was available, and where operation was intermittent, and whose converter under the high-pressure conditions was subject to cracks when exposed to temperatures above 400° C (as a result of hydrogen attacking ordinary steel), the Terni plant operated with regularity and stability thanks to the design of an internally heated catalyst tube, according to the specification patented in September 1920.[30]

Luigi Casale's converter, as perfected after his return to Idros, became the standard model.[31] It was comprised of a number of concentric tubes. By flushing the inner wall of the outer tube, or shell, of the converter with cold synthesis gas, it was maintained at a temperature which greatly reduced the stress on the tube. This was the feature which permitted the use of ordinary steel for the outer wall and overcame the problem of fracture, caused by hydrogen. The innermost tube was the catalyst bed. It surrounded the electrical heating element. Heat from the catalyst bed warmed up the incoming gas mixture. "The synthesis tube [converter] is constructed so as to ensure efficient heat interchange, and the pressure container [outer tube] itself never attains a temperature higher than 200 °C. This low temperature enables special steels to be dispensed with in the design of the catalyst bomb".[32]

The compressed nitrogen-hydrogen gas-feed entered at the bottom of the converter, and flowed up, in the annular space between the inner wall of the converter shell and the corrugated partition. At the top, the gas reversed its direction and flowed down the next annular space, where it came into contact with the outer wall of the inner catalyst tube. Here it absorbed heat from the exothermic reaction. At the bottom the gas again changed direction, this time entering into the catalyst tube where, within the catalyst bed, the ammonia was formed (fig. 3).

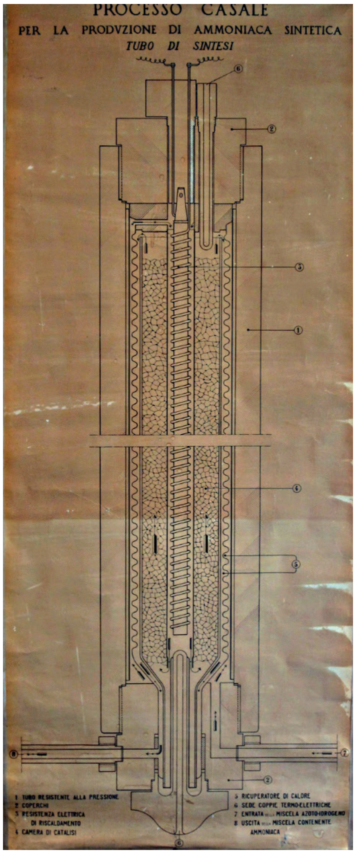

Figure 3. Casale ammonia converter, 1920s. (Casale SA)

As in the Haber-Bosch process, a gas recirculation pump, for return of the unreacted gas mixture to the converter, was incorporated into the early synthesis loop.

Another important advantage of the use of very high pressures, as Casale had calculated, was the increased yield, 15-18%, compared with around 5% for the Haber-Bosch process. The yield, in fact, was greater than expected by theory. This meant that less gas was pumped and circulated to produce the same volume of ammonia as in the Haber-Bosch process. In the synthetic ammonia processes at the lower (Haber-Bosch) pressure range, at 200 to 350 atm, the iron (magnetite) catalyst had to be specially formulated with the addition of promoters, or activators, which were oxides of certain metals, to ensure high activity. At very high pressures, a less active catalyst, though still incorporating promoters, was used, as was the case in the Casale process, which required around one-seventh the amount of catalyst as compared with the Haber-Bosch process. At Casale's working pressure of 800-850 atm, it was possible to separate out anhydrous liquid ammonia from the gas mixture by simply cooling with water. Thus, because of the high working pressure, there was no need for a refrigeration compressor for separating out liquid ammonia; the condenser operated at the water-cooling temperature. This was a major advantage over the Haber-Bosch process, for which expensive refrigeration equipment was necessary in order to produce anhydrous, liquid ammonia from the gaseous product mixture. Moreover, direct production of liquid ammonia under Casale's conditions involved more compact units than were required in the Haber-Bosch process.

Leprestre showed full satisfaction and provided additional funding to Casale for the study and erection of a plant capable of taking 3,000 cubic metres of hydrogen daily, equivalent to the design capacity of the Rumianca facility. For this purpose, a limited liability company, Nitram, was established, which appears to have been intended as a syndicate involving a group of investors brought together by Leprestre and Handley in order to provide liquidity for Casale's work at Idros. Leprestre's enthusiasm was such that he went to the United States in order to promote the Casale ammonia process, with the intention of attracting investors.

There was one major stumbling block remaining that prevented industrial scale production. In order to generate the necessary electricity for the electrolysers and for driving compressors and other machinery, Idros was required to negotiate with the Municipal Electricity Company of Terni. However, the electricity company was forbidden from supplying energy to any chemical company competing with the cyanamide manufacturer

Carburo, according to a 1910 agreement between the Municipality of Terni and Carburo. As a result, Carburo invoked the agreement, thus preventing the supply of electricity to Idros.[33] This move most likely followed information provided by Lorenzo Allievi, of Rumianca, to Carburo, following the falling out with Casale. In addition, the Casale process was now seen as a strong competitor to the cyanamide process. Delicate negotiations were required to bring about a change at Carburo. If Carburo became involved with the Casale process, it was argued, Carburo would have a cost and energy saving alternative to the production of ammonium sulphate, as compared with calcium cyanamide. Another consideration was that the Frank-Caro cyanamide patents were about to expire.

The outcome of the deliberations was a compromise. For Idros, which desperately needed the energy to operate the semi-industrial plant of Casale, it became necessary to meet certain demands of Carburo. On 3 January 1921, Carburo and Idros signed an agreement that allowed the former to inspect the Idros unit for the production of synthetic ammonia. Once convinced of the effectiveness of the Casale process, Carburo acquired exclusive rights to the Casale patents, valid for Italy, for the countries of the former Austro-Hungarian Empire, and for Romania. On 23 April 1921, Carburo and Idros together established a new company.[34] This was based on reorganisation of Idros as a new entity, the Società Italiana per l'Ammoniaca Sintetica (SIAS), in which Carburo would direct the industrial programme, while Casale would assume the role of scientist-entrepreneur. Nitram was absorbed into SIAS. Idros would increase its capital to 23 million lire, divided into 12 million invested by Carburo (4 million of which would be used to pay off Idros's debts, and 8 million remaining available for investment in new plants); 5 million as compensation for the Casale patent; and 6 million as the previous capital of Idros.[35] The Board of Directors of SIAS was composed of President Riccardo Bianchi, engineer and former head of the Italian State Railways, Arturo Bocciardo, engineer and CEO of the Terni – Società per l'Industria e l'Elettricità, Luigi Casale, and Pietro Fenoglio, engineer and board member of the Banca Commerciale Italiana; the CEO was Vincent Handley, the secretary was Ranieri Pontecorvi, and the technical director was engineer Osvino Ranieri Tenti.

A few days later, on 27 April 1921, Casale and Leprestre founded Casale Ammonia SA in Lugano (Switzerland) with the aim of acquiring, exploiting and protecting patents abroad for the production of synthetic ammonia according to the processes devised by Casale. Of a total of two hundred 500-franc shares issued, Casale held seventy-eight and Leprestre ninety. The Board of Directors was composed of three members, namely Leprestre (president), Giuseppe Albisetti (colonel and merchant of Massagno), and Tommaso Quadri (architect of Lugano).

Table 1. Events in the Development of the Casale Ammonia Process

Year	*Main events*
1915	Establishment of Rumianca, at Rumianca (Pieve Vergonte).
1916	Establishment of Idros at Terni, for production of hydrogen and oxygen for the military. The president is René Leprestre.
1917	Casale joins Idros, on recommendation of Miolati. Casale collaborates with Carlo Andreucci on synthetic ammonia.
1918	Casale moves from Idros to Rumianca because Terni does not produce surplus hydrogen. For ammonia studies. Rumianca makes available redundant equipment, and by-product hydrogen from its alkali plant.
1919	In May, Casale demonstrates synthesis of ammonia at the rate of 100 kg per day. Casale and Rumianca draw up a contract, signed on 17 May, specifying production by 30 September 1919 of 1,070-kg per day. Casale still owns his patents but agreed not to reveal confidential technical information for working the process developed at the Rumianca factory. Casale is allowed to deal with outside investors. In July, Leprestre visits Rumianca with Giuseppe Bruni, his expert, and representatives of a US group prepared to invest in Rumianca's ammonia, and promote the process outside Italy, subject to satisfaction of the conditions laid down in the Casale-Rumianca contract. Things fall apart because on 11 July Leprestre and Casale came to a separate agreement over control of the ammonia patents, and this was discovered by Rumianca; Rumianca in any case could not achieve the agreed output. The Casale-Rumianca agreement was declared void. Late in 1919 Casale returns to the now closed Terni works where he now has sufficient working space, and a supply of hydrogen from the electrolysers there.
1920	In April, Casale's converter produces about 200 kg of ammonia daily. In June Casale completes the Terni ammonia unit and in July and September he obtains two patents for the ammonia process.
1921	Ammonia Casale SA and SIAS are established. SIAS is created from Idros and calcium cyanamide manufacturer Carburo. Casale receives his first order for industrial scale synthetic ammonia machinery from the Japanese cyanamide manufacturer Shitagau Noguchi.
1922	A 2-metric ton per day converter is installed at Terni (September). SIAS acquires access to larger premises at Nera Montoro for testing and development in collaboration with Ammonia Casale.

The official constitution of SIAS took place on 22 May 1921, with headquarters in via Due Macelli 66 in Rome, the headquarters of Carburo.[36] On 29 September 1921, the SIAS factory in Terni was visited by ministers Alberto Beneduce (Minister of Labour) and Luigi Gasparotto (Minister of War), who paid tribute to Casale for his hard work and dedication and gave great praise to Leprestre for recognising the potential of the Casale process and for investing considerable sums for its development.[37] Around the same time, Casale was negotiating a licensing agreement with Japanese engineer Shitagau Noguchi, an entrepreneur active in the production of calcium carbide and calcium cyanamide on the island of Kyushu. This agreement was finalized in December 1921 with the payment of 10 million lire to Ammonia Casale.[38] Noguchi had been introduced to Casale by Enrico Cairo of Carburo.

In September 1922, a 2-metric ton per day Casale ammonia converter was installed at Terni. This proved of tremendous value in demonstrating the capabilities, and reliability, of the Casale process. To develop and test larger converters it became necessary to gain access to additional space. Carburo had already given full backing to the expansion of SIAS activities. In 1922, SIAS leased the substantial Nera Montoro electrochemical plant that had been inactive for some years.[39] This would enable development work on ammonia converters, in 1923 of 7.5-tons per day capacity, equivalent to 24,000 tons per year of nitrogen, and, within three years, to converters of 20-tons per day capacity.

The selection of the Nera Montoro site was not random. The Montorese plain is characterized by the availability of hydropower, ideal for the production of electrolytic hydrogen, the most important raw material for the synthesis of synthetic ammonia, and of electricity for operating the machinery, including electrolysers. The factory site, owned by Società Idroelettrica di Villeneuve e Borgofranco (Villeneuve), was specifically designed for electrochemical production, such as chlorine and aluminium. Therefore, much of its existing machinery could be readily reused or adapted for production of hydrogen. In addition, the local staff of peasant origin had accumulated considerable experience in chemical processing during the war and were a valuable source of cheap and semi-skilled labour. Nera Montoro was the ideal location for expanding and developing the electrochemical process associated with Luigi's Casale's ammonia process. Within a short time, orders were placed for the additional equipment needed in the synthesis of ammonia at Nera Montoro.

THE LITIGATION BETWEEN RUMIANCA AND LUIGI CASALE

Following the inability of Rumianca to meet the 30 September 1919 production target, and Casale's refusal to allow extra time, Rumianca built up a legal case for claiming compensation for loss of business from Casale. The resultant litigation went as follows: Casale challenged Rumianca on two main issues. The first concerned an estimate of the potential production of synthetic ammonia at Rumianca from September 1919 to May 1934, taking into account both the existing equipment and what could have been developed using around 3,000 cubic metres of hydrogen per day. At the same time, it was necessary to determine the percentage of royalties owed to Casale, based on this higher production. The second issue concerned the impediment placed by Rumianca towards Bruni, in charge of verifying the industrial effectiveness of the Casale process on behalf of an American group. Moreover, it was argued, Rumianca's inability to satisfy the conditions of the contract was contested because it had led to delays in the implementation of new plants and in entering into licensing agreements based on the Casale patents. Consequently, the crucial aspect was to determine whether, and for what specific reasons, Casale had lost the competitive advantage in cornering the production and consumption of ammonia in different countries when compared with other similar and competing processes, and if this had harmed Casale's interests by causing financial loss.[40] Rumianca's lawyers took Casale to trial in order to contest his claims.

The case was first tried before the Court of Pallanza, and then before the Court of Appeal of Turin. The Court of Pallanza, in elaborating on its verdict, declared that Casale had provided exhaustive evidence of the efficiency of his process.[41] The two court rulings, delivered on 30 December 1922 and on appeal on 2 July 1923, respectively, established that Rumianca was obliged to compensate Casale on the basis established by the agreement of 17 May 1919 for ammonia that could have been produced, as well as for damages due to the failure to conclude the contract, amounting in all to 179 thousand lire for unpaid consultancies and a further 660 thousand lire for unpaid royalties. The final judgement was issued by the Court of Appeal in April 1929, over two years after the death of Luigi Casale, and after review of a series of statements and investigations requested by a panel of experts appointed by the court the previous year. This panel of experts was composed of professors Angelo Menozzi and Felice Garelli, as well as the engineer Eduardo Ferrua.

In the defence memorial presented by Rumianca, and signed by the then CEO Ostilio Severini, it was explained that the measure of success of the plant could be determined only by the prospects for marketing ammonia. It was also argued that it would be easy to replicate the same experimental plant in Terni, stressing that Casale could make further improvements by virtue of the specifications of his 1920 patent, which was earlier than those of Fauser and Claude. Therefore, the defence for Rumianca argued, the alleged losses complained of by Casale were totally non-existent.[42]

In response, Casale interests, represented by Bruni, argued that the failure to comply with the agreements had caused a delay in the development and expansion of the Casale process both in Italy and abroad, thus nullifying the technological advantage. In fact, it was already clear in May 1919 at the Rumianca plant that the industrial efficiency of the process could then have been demonstrated, and this was well before the time required for the completion of the Terni plant in June 1920 (started in November 1919). The need to regain Leprestre's trust, and the agreements with the companies that led to the establishment of SIAS in July 1921 to exploit a daily production of one ton, resulted in a delay of two years in the industrial application of the Casale process. Bruni observed that Claude (whose patent was filed in March 1917, before that of Casale) had already started a plant in Montereau and on 2 February 1920, had demonstrated a working unit before the French Academy of Sciences with production of 6-7 litres of liquid ammonia per hour. Claude had exploited very high pressures, around 1,000 atm, to obtain higher concentrations of ammonia but confronted considerable difficulties in constructing the necessary equipment. On 26 November of the same year, by means of a different catalytic tube system, he was able to produce 60-70 litres per hour. Finally, on 21 April 1921 a plant was under construction for production of 5 tons of anhydrous ammonia per day. Fauser, in the meantime, had obtained his patent and was working to persuade his potential backers. Consequently, if the Casale process had been properly consolidated two years earlier at Rumianca, it could have established itself considerably earlier as a predominant method worldwide by virtue of its technical merits.[43]

According to Bruni, the construction of a plant similar to that of Rumianca in Terni was initially delayed due to the lack of hydrogen and adequate equipment (compressors and refrigeration pumps), but especially the converter, which had already been ordered by Rumianca from the engineering firm Ansaldo.[44] The panel of experts recognized that the installation of the catalytic tube (converter) would have made the process operational by the end of October 1919, thus allowing an effective production of synthetic ammonia from 1 November. The main plant, designed to absorb the 3,000 cubic metres of hydrogen, if made available, could have operated at full capacity by 1 November 1920. As a result, it was decided that from this date onwards full production capacity based on the amount of hydrogen available should be taken into consideration. With Casale's estimate that 2.6 cubic metres of hydrogen were needed to produce one kg of ammonia, a total daily production of about 1,150 kg of ammonia was calculated. It was estimated that in the fifteen-year period 1919-1934 about 5,400 tons of ammonia could have been produced. This production would have generated for Casale an estimated income of 580,000 lire, calculated on the basis of 0.1 lire per kg, and rounded up to include interest payable.[45]

In 1929, the panel of experts also recognized delays forced on Casale in developing his invention and the resulting loss of pre-eminence to Claude and Fauser. This situation led to considerable difficulties in acquiring the markets for production and consumption, both because of the emerging competition and the discredit that Casale suffered by the actions of Vitale, in addition to the widespread defamation in judicial, scientific and industrial circles. Moreover, the obstacles encountered by Casale following the legal dispute—and taking into account Claude's possible claim on priority—may have influenced Montecatini's decision to opt for Fauser. Guido Donegani, Montecatini's CEO, was an advisor to Carburo and therefore had an intimate knowledge of the Casale process.[46]

However, determining precisely the extent of the material and non-material damage proved complex, except for the loss of the income relating to the 20-ton plant, which was to have been financed by American investors but which, because of the complications introduced by Rumianca, was never realized. Consequently, the experts, with a majority of two to one, came to the conclusion that Rumianca was responsible for the non-fulfilment of the conditions of its agreement with Casale and therefore Rumianca was ordered to pay the sum of two million lire to Casale. In total, it was estimated that the company would have to compensate Casale for an approximate sum of two and a half million lire. According to the ISTAT converter,[47] two and a half million lire in 1929 would correspond to more than two and a half billion euros today (to be precise, 2,635,649.99 euros).

CONCLUSION

According to heroic narratives, mixed with a little pure mythology, in the history of science and technology, several individuals are credited with breakthrough inventions. While this may be true, bringing an innovation to market requires multiple actors. Typically, to accomplish this goal the inventor has to convince potential investors and those who place resources at his/her disposal for development of a viable process. The path to success is, invariably, not smooth. There might be disputes over contractual arrangements. Some companies, at least initially, might have little or no incentive to invest in new technologies, especially if they might constitute a disruptive influence. They may even place barriers in the way of potential competitors that are engaged in the same or similar markets. In these circumstances, diplomacy must come into play to overcome prejudices. All these elements are prominent in the development of the synthetic ammonia process of Luigi Casale. The arrangement of his steel converter tube, packed with other tubes, one containing a catalyst, and others that heated and cooled gases—preventing rupture of the outer wall of the converter—and including a heating element, was the basis of a technology that rivalled BASF's Haber-Bosch process. A major incentive for the development of the Casale process was the fact that BASF tightly guarded the secrets of its technology, and its conditions for licensing were so onerous that the Haber-Bosch process was not taken up elsewhere. Casale in contrast, licensed his process globally, and was the first to do so, with considerable success. In that sense he benefitted greatly from the BASF strategy. At the same time, with René Leprestre's encouragement and backing, he was enabled to garner international interest at a time when the Claude process was facing operational difficulties and the Fauser process was serving the needs of Montecatini alone, and within the borders of Italy.

The entrepreneur Leprestre enters the story as the head of the Italian company Idros at Terni founded during wartime for the express purpose of producing gases for the Italian military. Leprestre was a wily businessman who drew on his contacts with prominent Italian politicians to gain contracts for the supply of bombs. He was intrigued by Luigi Casale's ammonia invention and provided support for its development at Terni. However, at Terni the facilities there were inadequate for Casale's work. So Casale moved to Rumianca, which had idle equipment suited to ammonia research, and by-product hydrogen that was available. Casale was semi-independent and he did not break his links with Leprestre. Casale's 1919 contract with Rumianca, subject to the firm fulfilling its part of the bargain, gave Rumianca certain exclusive rights, but it allowed Casale to bring in outsiders who would invest in and promote and license the process elsewhere. Casale and Leprestre drew up a separate, and secret, contract for control of Casale's ammonia patents. Leprestre gathered together a small group of American investors who were enabled to observe the Casale process at work. As soon as Rumianca found out about the Leprestre-Casale agreement, it put a stop to the visits. Casale returned to Idros where Leprestre was now in a better position to back the ammonia studies. At Terni, Casale overcame the several teething problems that he had experienced at Rumianca. However, as soon as Idros planned to manufacture ammonia it was confronted with a stumbling block put up by the cyanamide manufacture Carburo, which denied Idros access to hydroelectricity. This difficulty was overcome after Carburo was brought into a partnership with Idros, to exploit synthetic ammonia, through the newly created SIAS.

SIAS was founded in 1921 at almost the same time as the establishment of Ammonia Casale, by Leprestre and Casale, and whose main objective was licensing the ammonia process abroad. Because the Terni site was inadequate for industrial scale development of the Casale process SIAS took over the Nera Montoro power station and its associated chemical works which had been out of action since the end of the war. Much of the machinery could be readily adapted to the needs of the Casale process. Additional machinery, including special compressors and apparatus for burning air in hydrogen to produce nitrogen was purchased and installed. In 1925, Nera Montoro began producing ammonia. From this time Ammonia Casale and SIAS collaborated closely in the further improvements in the process, especially the enlargement of converters capable of producing 20 tons per day, and more, of synthetic anhydrous ammonia. Further trials and tribulations followed. As for Rumianca, the company claimed compensation from Casale according to the agreement of 17 May 1919.

According to the May 1919 agreement between Casale and Vitale, of Rumianca, Casale would increase production from the 100 kg experimental plant to an industrial unit of 1 ton per day, using byproduct hydrogen made available by Rumianca at the rate of 3,000 cubic metres per day. In the event, Ruminaca was unable to provide all the necessary hydrogen, and thus was unable to demonstrate the scaled-up process, according to its agreement with Casale. This is evidenced by the fact that when Casale and Leprestre signed their secret agreement among the clauses it was specified that the American group would increase the share capital of Rumianca only if the specified higher daily capacity was attained.

Casale claimed that Rumianca had held up development of the Casale process by not fulfilling the conditions of the May 1919 contract. Most especially Rumianca's resources were shown to be insufficient to meet the targeted production. Casale's expert witnesses argued in reports placed before the court in litigation that Casale had lost a two-year headstart against the main rivals, Fauser and Claude, and deserved compensation for loss of business. On appeal, the court found in Casale's favour. Had Rumianca not prevented Casale from demonstrating his apparatus before outside interested parties in 1919, then both sides would have benefited. Significantly the absence of competition outside Italy from Fauser and Claude at the time of the lawsuit was not raised in the court case between Rumianca and Casale.

There were also the American investors. Though they would appear to have disappeared from the scene not long after Casale departed from Rumianca, it is highly likely that Leprestre, having gained their trust, was able to raise funds for development of the Casale process in Terni from the United States. Vincent Handley, one of the observers in 1919, and who had worked at an Italian bank in New York, was appointed the secretary of Ammonia Casale and with Leprestre arranged funding. In addition, Leprestre through his war-related businesses in the United States and Italy probably gained considerable wealth, enabling him to be the principal shareholder of Ammonia Casale in its early years. His reputation was such that in mid-1923, as a resident of New York City, he raised considerable capital from American investors for an enterprise based on manufacture in the United States and international commercialisation of the Casale process. However, the venture did not succeed. More success was achieved in Japan, from 1923, where converters of 7.5-tons daily capacity were installed. This gave a tremendous boost to the Casale process. Several licenses were acquired by French and other chemical companies during 1924. The licensees used Italian-made converters or converters based on the Casale design. By just after the mid-1920s, Terni and Nero Montoro engineers were testing new 20-ton converters under the high-pressure conditions. A novel feature invented in 1923 and introduced in 1927, the year of Casale's death, was the ejector, which did away with a number of moving parts. The simplicity and efficiency of the Casale process was such that the basic model devised by the late 1920s was, with few changes, widely taken up until the 1950s. Ammonia Casale fell behind in the 1970s but was revived in the 1980s through introduction of a novel converter that incorporated both radial and axial flows of gas. Rebranded as Casale SA in 2014, the firm today is a global leader in ammonia and related technologies.

* In recent years, the authors have contributed towards unravelling the hidden history of the Casale process, so far rarely mentioned in the historiography of technology. In particular, Anthony S. Travis has examined the development of the Casale process in the United States, which was adopted by Du Pont in the late 1920s (A. S. Travis, *Substantia*, **2021**, *5*, 55-77), in addition to describing the technological innovations that accompanied the expansion of Ammonia Casale SA, now Casale SA (A. S. Travis, *Catalysis Today*, **2022**, *387*, 4-8).

Lorenzo Francisci has concentrated on the development of the Casale process in Italy, with particular reference to its impact on the industrial policies of Terni - Società per l'Industria e l'Elettricità (TERNI). Francisci has highlighted the role of the Nera Montoro plant, which at the time of its construction in 1923 was the world's largest facility for the production of synthetic ammonia (L. Francisci, *Proposte e Ricerche*, **2020**, *84*, 121-132). Francisci has examined the industrial dynamics and the competition between TERNI and Montecatini over control of the Italian nitrogen industry, leading to the primacy of the Fauser process (L. Francisci, *Memoria Storica*, **2023**, *61*, 103-130). Ammonia Casale pioneered, in connection with the Italian autarchy programme, the use of ammonia as an automobile fuel (R. Covino, L. Francisci, *Ricerche Storiche*, **2020**, *1*, 41-57).

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REFERENCES

1. A. S. Travis, *The Synthetic Nitrogen Industry in World War I. Its Emergence and Expansion*, Springer, Cham, **2015**, p. 9.
2. V. Villavecchia, *Dizionario di merceologia e di chimica applicata*, vol. III, Hoepli, Milano, **1923**, p. 58.
3. O. Scarpa, *L'industria elettrochimica dell'azoto atmosferico,* Tipografia Editrice Italia, Roma, **1917**, p. 8.
4. L. F. Haber, *The Chemical Industry (1900-1930). International Growth and Technological Change,* Oxford University Press, London, **1971**, pp. 100-101.

5. W. Crookes, *Science*, New Series, **1898**, *8*, no. 20 (28 October), 561-575.
6. See ref. 2, p. 62.
7. E. Molinari, *Trattato di chimica generale applicata all'industria*, vol. I "Chimica Inorganica", Hoepli, Milano, **1943**, p. 825.
8. See ref. 1, p. 42.
9. On Haber, see, D. Stoltzenberg, *Fritz Haber: Chemiker, Nobelpreisträger, Deutscher, Jude: Eine Biographie*, VCH, Weinheim, **1994**, and the English version, *Fritz Haber: Chemist, Nobel Laureate, German, Jew,* Chemical Heritage Press, Philadelphia, **2004**; M. Szöllösi-Janze, *Fritz Haber 1868-1934: Eine Biographie,* VCH-Beck, Munich, **1998**; B. Johnson, *Making Ammonia: Fritz Haber, Walther Nernst, and the Nature of Scientific Discovery*, Springer, Cham, **2022,** https://doi.org/10.1007/978-3-030-85532-1; D. Sheppard, *Robert Le Rossignol: Engineer of the Haber Process,* Springer, Cham, **2020**, pp. 109-207.
10. D. Maveri, *La storia dell'azoto,* Ipotesi, Milano, **1981**, p. 82.
11. In all, about 100,000 tons of nitrogen fertilizers were consumed in Italy (about 17,500 tons of nitrogen) in 1913, falling progressively to 88,000 tons in 1914, to 75,000 tons in 1915, 62,000 tons in 1916, and 47,000 tons in 1917 and 1918. However, the need for nitrogen for the approximately 15 million hectares of Italian arable land would have require around 300,000 tons of nitrogen fertilizers (about 52,000 tons of nitrogen). See F. Zago, *Le concimazioni chimiche in Italia,* Firenze, **1923**, pp. 21-25.
12. A. S. Travis, *Nitrogen Capture. The Growth of an International Industry (1900-1940),* Springer, Cham, **2018**, p. 229.
13. A. Zambianchi, *Giornale di Chimica Industriale e Applicata*, **1923**, *4*, 171-176.
14. With the Haber-Bosch process, coal consumption was one tenth of the arc process and one third of the calcium cyanamide process, while electricity consumption was fifty times lower and twelve times lower, respectively. See C. Rossi, *Giornale di Chimica Industriale e Applicata*, **1920**, *6*, 301-307.
15. On Luigi Casale, see G. Marchese, *Casale Luigi*, Dizionario Biografico degli Italiani, Roma, **1978**, https://www.treccani.it/enciclopedia/luigi-casale_res-263d981d-87ea-11dc-8e9d-0016357eee51_(Dizionario-Biografico)/ (accessed on 05 June 2024).
16. D. K. Barkan, *Walther Nernst and the Transition to Modern Physical Science,* Cambridge University Press, Cambridge, **1999**; P. Coffey, *Cathedrals of Science: The Personalities and Rivalries that Made Modern Chemistry,* Oxford University Press, Oxford, **2008**.
17. Acciai Speciali Terni Historical Archive, *II deposit*, folder 86, dossier 11, copy of the deed of incorporation of Idros di Terni by deed of the notary Umberto Rimini, 19 February **1916**.
18. Ibid, List of members of the Board of Directors of Idros in **1916**.
19. Central State Archives, *Ministry of Arms and Ammunition Fund, Original Contracts*, folder 9, dossier 778, copy of the contract between Idros and the Military Administration for the supply of 36,000 bombs, 29 November **1916**.
20. Ibid, copy of the contract between Idros and the Military Administration for the supply of 237.75 cubic metres of oxygen, 29 Novembre **1916**.
21. Acciai Speciali Terni Historical Archive, *II deposit*, folder 103, dossier 7, minutes of the meeting of the Executive Committee of Carburo, 22 September 1915. See also "Andreucci Carlo's Patent 483-227", cited in *Annals of Applied Chemistry*, **1917**, *7-8*, 98.
22. As for the catalyst, it was a compound of inorganic and organic matter (salts, oxides, metals, alkali metals, earth alkaline metals, earth metals, carbon and metalloid oxides). See Accademia Nazionale delle Scienze detta dei XL Historical Archive, *Nicola Parravano Fund*, folder 22, dossier 221, copy of the patent "Metodo industriale ed apparecchio per la produzione sintetica dell'ammoniaca, mediante un nuovo catalitico di grande rendimento" of Luigi Casale, Carlo Andreucci, Mario Santangelo, 18 luglio **1917**.
23. Ibid.
24. Ibid, copy of the patent "Metodo ed apparecchio per produrre da idrogeno miscele di azoto, idrogeno od azoto" of Luigi Casale, 12 July **1920**.
25. A. M. Falchero, *Italia contemporanea*, **1982**, *146-147*, 67-92.
26. See ref. 22, copy of the agreement between Casale and Leprestre, 2 August **1919**.
27. Ibid, copy of the judgement of the Turin Court of Appeal in the Rumianca-Casale case, April **1929**, p. 2.
28. State Archive of Terni, *Siri Fund*, folder 3699, letter from Luigi Casale to Alfonso Vitale, 25 August **1919**.
29. Ibid, Letter from Luigi Casale to Alfonso Vitale, 13 September **1919**.
30. See ref. 22, copy of the patent "Apparecchio di catalisi per la sintesi dell'ammoniaca" of Luigi Casale, René Leprestre, 21 September **1920**.
31. Ibid. See also copy of the patent "Perfezionamento nell'apparecchio catalitico per la sintesi dell'ammoniaca" of Luigi Casale, René Leprestre, 15 February **1921**.
32. B. Waeser, *The Atmospheric Nitrogen Industry: With Special Consideration of the Production of Ammo-*

nia and Nitric Acid (transl. Ernest Fyleman), vol. I, P. Blakiston's Son & Co., Philadelphia, **1926**, p. XVII, with an introduction by J. F. Crowley. Translation of B. Waeser, *Die Luftstickstoff-Industrie, mit besonderer Berücksichtigung der gewinnung von ammoniak und salpetersaure*, Otto Spamer, Leipzig, 1922. Crowley and Partners represented Ammonia Casale in the UK.

33. Acciai Speciali Terni Historical Archive, *Archives Acquired Fund*, *Carburo*, register 64, minutes of the meeting of the Board of Directors of Carburo, 2 May **1921**.
34. Ibid, minutes of the meeting of the Board of Directors of Carburo, 27 April **1921**.
35. Ibid.
36. Acciai Speciali Terni Historical Archive, *Archives Acquired Fund*, *SIAS*, register 1, minutes of the meeting of the Board of Directors of SIAS, 6 February **1922**.
37. Ibid, minutes of the meeting of the Board of Directors of SIAS, 8 October **1921**.
38. B. Molony, *Technology and Investment: The Prewar Japanese Chemical Industry*, Council on East Asian Studies/Harvard University Press, Cambridge, Mass, **1990**. See also https://www.treccani.it/enciclopedia/l-industria-dei-composti-azotati_(Il-Contributo-italiano-alla-storia-del-Pensiero:-Tecnica)/ (accessed on 10/05/2024).
39. See ref. 36, minutes of the meeting of the Board of Directors of SIAS, 27 June **1922**.
40. See ref. 22, copy of the judgement of the Turin Court of Appeal in the Rumianca-Casale case, April **1929**, p. 2.
41. Ibid, pp. 8-12.
42. Ibid, pp. 14-16.
43. Ibid, pp. 16-18.
44. Ibid, pp. 59-64.
45. Ibid, pp. 104-106.
46. Ibid, pp. 101-103.
47. https:///inflationhistory.com/it-IT/?currency=ITL&amount=2500000&year=1929 (accessed 25 May 2024).

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Research Article

Friedlieb Ferdinand Runge (1794-1867) – An Unusual Chemist

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Abstract. This paper presents life and work of Friedlieb Ferdinand Runge, an unusual chemist, ignored for a long time. There are discussed his researches in natural products and coal tar, the discovery of caffeine and priority claims on the discovery of quinine. Important industrial activities at the Oranienburg factory are described, including the 'German guano'. Important issues are the discussion of alternative chemical theories by the anti-atomist Runge, as well as his occupation with the relation Chemistry-Art, expressed in his *Musterbilder*, also a forerunner of chromatography.

Keywords: History of Chemistry – Friedlieb F. Runge – Phytochemistry – Coal tar – Chemical Industry – Alternative chemical theories - Chemistry and Arts

> *Chemistry is the science of the components of our Earth*
> (F. F. Runge)

INTRODUCTION

Friedlieb Ferdinand Runge (1794-1867) was certainly one of the chemists with the most informal and unusual personality and behaviour of the 19th century, which gained him great popularity. Coming from Medicine to Chemistry, he worked and researched in several areas, from phytochemistry and tar, a productive industrial activity, the first data on what we now call chromatography. He wrote books in a simple but chemically correct language, to reach a wider audience and disseminate chemical knowledge, an activity that contributed to his popularity during his lifetime. Interestingly, soon after his death in 1867, he quickly fell into oblivion, despite the efforts of August Wilhelm Hofmann (1818-1892), then president of the German Chemical Society, to preserve his memory. What could have led to such a rapid decline? Informality and unorthodox behaviour for a member of the Academy certainly contributed, but I believe that his adherence to *Naturphilosophie*, useful in phytochemistry but detrimental to the development of Organic Chemistry as a whole, and his staunch anti-atomism, visible in his presentation of General Chemistry and Inorganic Chemistry, made it difficult to work on an equal footing with the academic community of the time, represented for example by Liebig, Wöhler or Bunsen.

Figure 1 Friedlieb Ferdinand Runge (1794-1867), holding a glass of wine (perhaps his 'synthetic wine'). Photo by an unknown author, the only real portrait of Runge, ca.1860. (Stiftung Preussischer Kulturbesitz, image in public domain).

In any case, given the important contributions to phytochemistry and the study of tar, the forgetfulness is surprising. Runge had few collaborators, and according to August Wilhelm Hofmann there seemed to be "something that put people off" about him, perhaps his idiosyncratic behaviour. In the international context, silence was established around Runge. For example, James R. Partington (1886-1965), in volume IV of his "History of Chemistry" (1962), dedicated a few lines to him, which we transcribe here:

"F. F. Runge was at first a pharmacist, then associate professor in Breslau (after a long residence in Paris), then in the Prussian Marine Service in Berlin and Oranienburg. He published several technological and other papers, also on the motion of electrically polarized mercury, and books. Runge rediscovered aniline in coal tar oil and called it cyanol, since it gave a blue color with bleaching powder, he found that it stained pine wood and elder pith yellow, and gave a greenish-black color when oxidized by a cupric salt (aniline black). In the same research he discovered in coal tar oil another base leukol (quinoline), an acidic substance which he called carbolic acid (*Karbolsäure*, phenol), pyrrol (*pyrró* = fiery red), also rosolic acid, and three other bases. He did not analise any of these substances"[1].

Aaron Ihde (1909-2000), in "Development of Modern Chemistry" (1984), gives him some space, related to research with atropine, tar and as a remote precursor of chromatography. More recent historians, or historians belonging to the Anglo-Saxon cultural context, such as William Brock (*1936), in "The Fontana History of Chemistry" (London, 1992), or Eduard Farber (1892-1969), in "Great Chemists" (New York, 1961), don't even mention him.

Figure 2 Plaque allusive to Runge's birthplace, 1936, on the Rectory building (Courtesy Billwärder Verein and Katja Haack, Hamburg)

Finally in 1994, at the bicentenary of Runge's birth, the city of Oranienburg, where Runge carried out much of his activity as a chemist and where he lived and died, decided to pay him a double tribute. In front of the building attached to the Oranienburg Palace, where Runge had his laboratory, was erected a bronze statue, designed by Stephan Möller (*1954); and the *Stiftung Preussischer Seehandel* established the Friedlieb Ferdinand Runge Prize, for "Unconventional Forms of Art Transmission" (very much to Runge's taste...). In fact, in 1863, through August Wilhelm Hofmann, the London Industrial Exhibition awarded Runge a medal. To the messenger who brought it to his home, Runge responded with Friedrich Schiller's quote, "You arrive late – but you arrive!" (from the play "Wallenstein").

Runge's basic biographies are those by Hermann Schelenz (1848-1922), pharmacy historian, "Friedlieb Ferdinand Runge" (1907); by Max Rehberg (1882-1945), "Friedlieb Ferdinand Runge, Entdecker der Teerfarben" (1935), both hagiographic, to be consulted *cum grano salis*, and by Berthold Anft, "Friedlieb Ferdinand Runge – sein Leben und Werk" (1937, reprint 1977). A recent biography is "Friedlieb Ferdinand Runge, sein Leben, sein Werk und die Chemische Produkten-Fabrik Oranienburg", by Christa Niedobitek and Fred Niedobitek (2011).

Figure 3 Parish Rectory in Billwärder, Hamburg, erected in 1840, in substitution of the old Rectory, where Runge was born in 1794 (Courtesy Billwärder Verein and Katja Haack, Hamburg)

ORIGIN AND STUDIES

Friedlieb Ferdinand Runge was born on February 8, 1794 (not 1795, as it is sometimes read; the origin of the confusion is the baptismal certificate) in Billwerder, a village southeast of Hamburg, the third of the seven children of the Lutheran pastor Johann Georg Runge. Billwerder was an ancient rural community, known since 1150; incorporated into Hamburg, it maintained its rural character. During the Napoleonic Wars, Hamburg was occupied by the French (1806/1811), and during the administration of Marshal Louis-Nicolas Davout (1770-1823) the city's population decreased from 100,000 to 55,000 residents. Pastor Runge's family also experienced difficulties, and Friedlieb was unable to attend a renowned school, such as the famous *Johanneum* in Hamburg, founded in 1529 by the reformer Johannes Bugenhagen (1485-1558). So, he went to study at Schiffbeck elementary school. The Billwerder rectory was demolished in 1840 and replaced by a more solid building, where in 1936 a bronze plaque was placed commemorating the birth there of the chemist Runge (figures 2 and 3).

From 1810 to 1812 he learned pharmacy from an uncle at the *Ratsapotheke* in Lübeck, the oldest municipal pharmacy in Germany (founded in 1412). There he carried out his first experiments and had his first contact with henbane (*Hyoscyamus niger L.*), which among other alkaloids contains atropine, so important to him in his future activities. The obvious path for Friedlieb was the study of Medicine, from 1816 to 1818 at the University of Berlin. In 1818 he transferred to the University of Göttingen, where Friedrich Stromeyer (1776-1836), professor of Chemistry, convinced him to study Chemistry. A new transfer, still in 1818, to the University of Jena, continued his interest in Chemistry, taught there by Johann Wolfgang Döbereiner (1780-1849), Goethe's scientific advisor. He also attended lectures on *Naturphilosophie* by Lorenz Oken (1779-1851), a disciple of Friedrich Wilhelm von Schelling (1775-1854). He received his doctorate in 1819 and returned to Berlin for a second doctorate, aiming to work as a *Privatdozent* (1822), being approved by a panel composed of Hermbstädt, Weiss and Hegel.

After a short stay at university, he traveled in Europe from 1823 to 1826: in Paris he met Alexander von Humboldt and Liebig, in France and Switzerland he visited industrial facilities, in England and the Netherlands agricultural activities. Coming back from the trip, his *peregrinatio academica*, he returned to Breslau (since 1945 the Polish city of Wroclaw), where he had already settled in 1823. He served there as *Privatdozent* , and in 1828 was appointed permanent professor of Technological Chemistry at the University of Breslau, where he remained until 1831, when he returned to Berlin. In Breslau he met again his friend, the poet and professor August Heinrich Hoffmann von Fallersleben (1798-1874), who had founded the "*Zwecklose Gesellschaft*" (Society without Purpose), and joined the "*Schlesische Gesellschaft für Vaterländische Kultur*" (Silesian Society for National Culture), founded in 1803, where he gave lectures on chemical and technological topics for pharmacists, students and workers. In the spirit of the intellectual formation in force in the 18th and early 19th centuries, Runge had his formal education at the university and his *peregrinatio academica*.

RUNGE AND PHYTOCHEMISTRY

In 1820 Runge published the book "*Neueste phytochemische Entdeckungen zur Begründung einer wissenschaftlichen Phytochemie*"[2] (Newest phytochemical discoveries for a scientific Phytochemistry), in which he presented the observations (but not yet analyses) on alkaloids, that he had made in Jena, and in which he also presented the criteria that he believed were necessary for a “scientific phytochemistry” (*phytos* = plant, the chemistry of plant constituents). It is difficult to say who was the first phytochemist, it is a subjective question that depends on who makes the choice, and on the criteria chosen for defining “phytochemistry”.There were Nicolas de Saussure (1767-1846), from Geneva, for the discovery of photosynthesis, the “mother reaction” of the entire plant universe; Friedrich Wilhelm Sertürner (1783-1841), for the first isolation of an alkaloid (morphine, in 1805); Pierre Joseph Pelletier (1788-1842), for the introduction of new solvent extraction methods, for the analysis of the “green matter” of plants (1817), and for the isolation, with Jean Bienaimé Caventou (1795-1877), of quinine (1820). Others prefer to wait for analyses and studies of groups of plant substances to be carried out, as those done by Friedrich Rochleder (1819-1874), a student of Liebig, or by Heinrich Hlasivetz (1825-1875). But there is also the earlier study of extracts from medicinal plants, such as those by Johann Friedrich Cartheuser (1704-1777) at the University of Frankfurt/Oder, and even the isolation of thymol (1719) and camphor (1725) by Caspar Neumann (1683-1737).

Let’s go back to Runge in Jena. In Döbereiner’s laboratory he studied the extracts of the “three narcotics”[3], the henbane (*Hyoscyamus niger L.*), the native *Bilsenkraut* (a variety of *Hyoscyamus);* the *Tollkirsche* (nightshade), the *Erba Belladonna* of the Venetians or the *Atropa belladonna L.* of Carl von Linné (1707-1778), whose toxic effect was already known by botanist Leonhart Fuchs (1501-1566), the reformer of Botanics during the Scientific Revolution; and the thorn apple, devil’s trumpet, or “witch fig”, *Datura stramonium L.*, the *Gemeiner Stechapfel*. [the three plants contain, among others, the alkaloids atropine and hyoscyamine]. Henbane was already familiar to Runge, from the Lübeck pharmacy: when preparing a medicine, part of the plant’s extract splashed into Runge’s eyes, causing intense and long-lasting mydriasis, a dilation of the pupil caused by some drugs. In Jena, he repeated these experiments, now including *Belladonna*. Mydriasis caused by the extracts was tested on Runge’s cat. (The curious effect has even had forensic use: if a suspicious liquid causes mydriasis in an animal, it contains the toxic alkaloid). Runge dealt with toxic substances so much that he was nicknamed *Dr. Gift*, poison doctor, a source of pride for him.

Goethe, who was interested in chemistry and science in general, had heard about mydriasis, and through Döbereiner, invited Runge to demonstrate the experiment at his home, in what would be one of the most delicious anecdotes in the history of Chemistry, mainly as narrated by Runge himself[4]. So, on October 3, 1819, wearing borrowed clothes a little too big for him (which later became fashionable in Jena), with the cat under his arm, he set out on his way to Goethe’s house (Goethe lived in Weimar, capital of the grand duchy, but in September and October 1819, returning from Karlsbad, he remained for some time in Jena). The demonstration and conversation were quite informal, and at the end Goethe gave the young chemist a box of coffee beans, perhaps he could find there the principle responsible for his insomnia. In fact, Runge isolated caffeine from coffee beans in 1819. Nervous when leaving Goethe’s house, Runge forgot the cat: Goethe called him back, saying “you forgot your assistant”[5]. The visit was important for both: Goethe began to become interested in alkaloids, and Runge began his important and largely forgotten contributions to Chemistry. But the alkaloids atropine and hyoscyamine were not isolated by Runge, but, still in 1819, by the pharmacist Rudolph Brandes (1795-1842), also active in Döbereiner’s laboratory.

From coffee beans, Runge extracted in 1819 the “*Kaffeebase*”, the “base of coffee”, that is, the alkaloid caffeine[6], a discovery also claimed (1821), without reason, by Pelletier, Caventou and Pierre Jean Robiquet (1780-1840). Runge published the discovery of caffeine in 1819, in Lorenz Oken’s (1779-1851) journal *Isis*[7], and in 1820 in his own book on phytochemistry[8]. There is no doubt about the priority of Runge’s discovery of caffeine. The formula for caffeine, $C_8 H_{10} N_4 O_2$, [in our notation] was only established in 1833 by Christoph Heinrich Pfaff (1773-1852) and Justus von Liebig (1803-1873).

Still in Jena, and still in Döbereiner’s laboratory, Runge continued his phytochemical research, now with the bark of *Cinchona* from South America, *Cortex Peruvianum*, from which he extracted “*Chinabase*”, the “base of cinchona” (“base” is a substance of a basic, or alkaline, nature), that is, the alkaloid quinine, in 1819 (actually a mixture of alkaloids). The results were published in the same article in the journal *Isis,* still in 1819[9]. *Isis* was a respected scientific journal at the time, published from 1816 to 1848, first in Jena, then in Rudolstadt; the high standard of the journal is demonstrated by its collaborators, like Alexander von Humboldt, Georges Cuvier, Mme. De Staël; the excuse that Runge published in unknown or less qualified journals does not apply. The histories of Chemistry are practically unanimous in attributing the discovery of quinine to Pelletier and Caventou, in 1820[10]. Runge’s *Chinabase* and the compound isolated by the French were later analysed and compared, finding that they were the same substance, and therefore, the priority in the discovery of quinine clearly lies with Runge, whose work had been published a year earlier than that of Pelletier and Caventou[11]. Historiographical revisionism? No, just the consistent application of a lesson of the great historian Marc Bloch (1886-1944): “The past is, by definition, a given that nothing can change. But knowledge of the past is something in progress, which is continually transformed and perfected”[12]. Is Runge undoubtedly the ignored discoverer of quinine? The priority discussion about the discovery of quinine is indeed complex. Runge was one year in advance in the research on china bark. Using acetic acid as solvent, he obtained a basic extract, which was actually a mixture of alkaloids, which he was unable to separate, so he could not study its properties. He considered the extract useless for medical purposes, and went to search for the ‘true’ effective component of the bark. These results were published in Runge’s *Isis* paper (1819) and in his book on Phytochemistry (1820). At the same time, unaware of Runge’s work, Pelletier and Caventou obtained from Peruvian bark the same alkaloid, in a more pure form, the quinine. They were able to study its properties and its medical uses, and published their results in 1820. Pelletier was unaware of Runge’s researches, but historians from later times were not, and several authors suggested a more rigorous investigation about the priority of the discovery of quinine. In 1970, Wolfgang Schneider (1912-2007), professor in Braunschweig, and his coworker Horst Real, repeated Runge’s experiments, following exactly Runge’s recipe, obtaining the same results[13], so that the priority question is not yet solved. In my opinion, Runge deserves the priority, using as criterium the first (later confirmed) published results. This is in accordance with Marc Bloch’s proposal mentioned above.

Another matter of priorities is the isolation of the dye glitter (*Krapp-purpur*), a less common component of madder (*Rubia tinctorium L.*), a discovery generally attributed to Robiquet and Jean-Jacques Colin (1784-1865), as *purpurin*, in 1826, along with alizarin[14]. Runge had already dealt with madder before. Unfortunately for him – academically and financially – the discovery of alizarin escaped him, but there is no doubt about the discovery of glitter, for which he applied for a patent in 1822, granted in 1823. But the official historiography is irreducible.

In Anft’s opinion, Runge’s experiments on natural products strictly followed the precepts of *Naturphilosophie,* but Runge also considered the purely empirical data of his experiments. Adherence to *Naturphilosophie*, the “pest of the sciences” in Liebig’s words, contributed to Runge’s oblivion. Another issue to determine is the almost systematic refusal to recognize its proven priority in some discoveries, such as quinine and purpurin. Many of his articles received heavy criticism at the time of their publication.

At the beginning of his treatise on phytochemistry as a new discipline[15] Runge initially addresses mineral chemistry and divides it into an empirical part (essentially analysis), a mathematical part (stoichiometry) and a speculative part, yet to be studied in detail. He compares phytochemistry with mineral chemistry, saying that phytochemistry, as it deals with living matter, is mineral chemistry at a higher level. Until now, there was little knowledge of phytochemistry; mineral chemistry is taught in universities, and textbooks also deal with mineral chemistry. Phytochemistry is mentioned at a glance, mentioning extracts of some plants, generally still problematic. For its evolution, the publication of the work “The Development of Vegetable Substances” by Nees von Esenbeck [Theodor Nees von Esenbeck (1787-1837), professor in Bonn] was important. Next, considering the genesis of the plant as also the genesis of phytochemistry, it sets out a series of rules and principles to arrive at a “scientific” phytochemistry, discussing obtaining (extraction with solvents, precipitation), properties, reactions, etc.

For my part, I add that Organic Chemistry at the end of the 18th century and beginning of the 19th century is essentially chemistry of vegetable extracts, that is, phytochemistry: see for example the obtaining of carboxylic acids by extraction or precipitation, by Carl Wilhelm Scheele (1742-1786), or the quantity of compounds obtained from alcohol, such as the obtaining of ether described by Valerius Cordus (1515-1544), perhaps the first organic chemist.

RUNGE AND THE COAL TAR

After leaving his studies and Jena, Runge dedicated himself to a new field of research, in Berlin, Breslau and Oranienburg: coal tar. Some of the discoveries are pioneering, others are expansions of what was already known. The increasing use of mineral coal from 1830 onwards, for coking and obtaining lighting gas, left ammoniacal waters and coal tar as by-products, both sources of obtaining various substances. Several researchers began to focus on tar, including Runge, and this simultaneity would lead to controversies and disputes over priorities. It is discussed, for example, whether we can consider Runge as the first chemist to obtain artificial dyes, as early as 1834, which in the light of the writings we have is not the case[16], although it cannot be denied that some of his pioneering experiments with tar provided elements for future dye syntheses. Judging by Runge’s own later writings (1866), he thought that with good will a dye industry based on his data would have been possible. Biographer B. Anft (1957) is also of the opinion that Runge should be credited with starting an artificial coloring industry[17]. The obtaining by Runge, from coal tar, of phenol (carbolic acid, 1833), aniline (*Kyanol*, 1834), pyrrole (1834) and quinoline (*Leukol*, 1834) are undisputed facts. Runge's experimental procedures were systematic, involving, in addition to distillations, also extractions with acidic and basic solutions. Directly attacking the tar with solvents (water, alcohol, ethyl ether) and acid and basic solutions did not produce any results, which led Runge to move on to more drastic methods[18]. Dry distillation in a sand bath provided a distillate, which was then broken down into two fractions: a volatile liquid and a thick brown oil. The volatile liquid was subjected to new procedures: with $Ca(OH)_2$ it forms a clear solution (a sign that the liquid was acidic), which when treated with HCl forms "carbolic acid" (our phenol); heating the clear solution formed by the addition of $Ca(OH)_2$ gives rise to a dark red powder, from which Runge extracted rosolic acid (a synthetic dye, methylaurine) and "brunolic acid", a substance that even today chemists were unable to identify. Treating the volatile liquid itself with H_2SO_4 and then with NaOH leads to obtaining "*leukol*" (our quinoline, actually a mixture that Runge was unable to separate); The addition of calcium chloride ($CaCl_2$) to the volatile liquid gives rise to a blue solution, which with chlorinated lime forms "*Kyanol*" (our aniline), and by oxidation with dichromate a black compound (aniline black). The above description is very simplified, Runge's experimental procedure is extremely complicated, which in the opinion of August Wilhelm Hofmann, who subsequently dealt with this subject, discouraged other chemists from dealing with this topic. Runge did not analyse these compounds, which is why he is criticized, but at the time no one analysed the compounds he obtained, as analysis and laboratory methods in general were still poorly developed. The analysis of organic compounds became routine after the techniques introduced by Jean-Baptiste Dumas (1800-1884) and mainly by Justus von Liebig (1803-1873). The chemist Carl von Reichenbach (1788-1869), discoverer of creosote (1833) and paraffin (1830), harshly criticized Runge's discoveries, denying the existence of the four announced substances, to which Runge responded with elegance and conviction.

The four compounds obtained by Runge from coal tar were of great importance in the evolution of Organic Chemistry. In coal tar Runge rediscovered aniline, discovered in 1826 by Otto Unverdorben (1806-1873), a pharmacist in Dahme, near Berlin, by sublimation of indigo. Unverdorben called the compound "*Kristallin*"[19]. Tar was a much more abundant raw material, a reason why Runge's rediscovery is important. Other chemists obtained aniline: Carl Julius von Fritzsche (1798-1871) in 1840, by heating indigo with alkali, naming the product "aniline" (from the Portuguese "*anil*"). In 1843, Russian chemist Nikolai Zinin (1812-1880) obtained aniline (which he called "*benzidam*") by reduction of nitrobenzene, a compound obtained by nitration of benzene, by Eilhard Mitscherlich (1796-1863) in 1835.

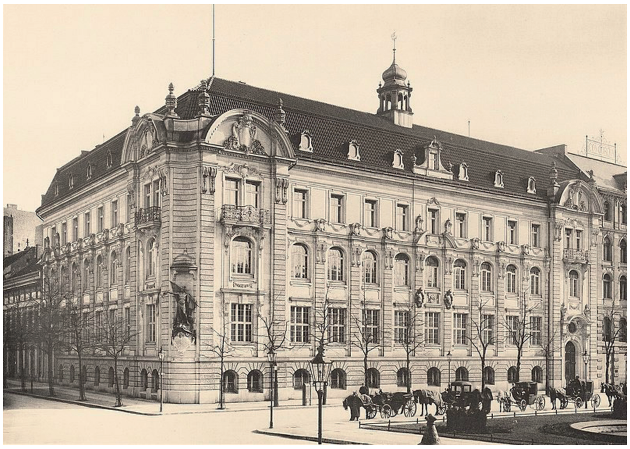


Figure 4 New building of the Preussische Seehandlung, 1904, on the Gendarmenmarkt,Berlin, by an unknown photographer. From "Atlas zur Zeitschrift für Bauwesen", 1904 (image in public domain)

Figure 5 Oranienburg Palace, in Oranienburg, built in the 17th century for the Great Elector's wife, Louise Henriette. In a lateral wing was installed Runge's chemical industry (Courtesy City Archive and Municipality of Oranienburg)

This whole subject was studied again in more detail by August Wilhelm von Hofmann (1818-1892), initially in his doctoral thesis (1841) with Justus von Liebig (1893-1873) in Giessen, a thesis whose theme was precisely aniline. As Liebig's assistant in 1843, Hofmann found that Unverdorben's *Kristallin*, Fritzsche's aniline and Zinin's *benzidam* were the same compound, for which Hofmann chose the name "aniline". Hofmann himself developed a more effective method of obtaining aniline, by distillation. He also discovered that Runge's "*leukol*" was a mixture of quinoline, isoquinoline and quinaldine. In 1845 Hofmann published all these details in an article in the *Annalen*[20]. From aniline he prepared some coloring principles, which leads most historians to consider Hofmann, and not Runge, as the precursor of the chemistry of synthetic dyes. It was Hofmann, then at the *Royal College of Chemistry* in London, who suggested to his student William Perkin (1838-1907) to try to synthesize quinine from aniline: the result was mauvein, frequently considered as the first artificial dye (1856).

In 1866, ten years after Perkin's discovery, Runge commented on the discovery of artificial dyes: "Finally Dr. A. W. Hofmann arrived and showed in his article [...] that all my data on this dye were absolutely true. With that, I once again dedicated myself to this almost forgotten subject..." Runge proposed in 1847 to the state factory in Oranienburg (see below) the manufacture of artificial dyes, "but my efforts failed because of the opinion of an official unfamiliar with the subject"[21] (financial director E. Cochius).

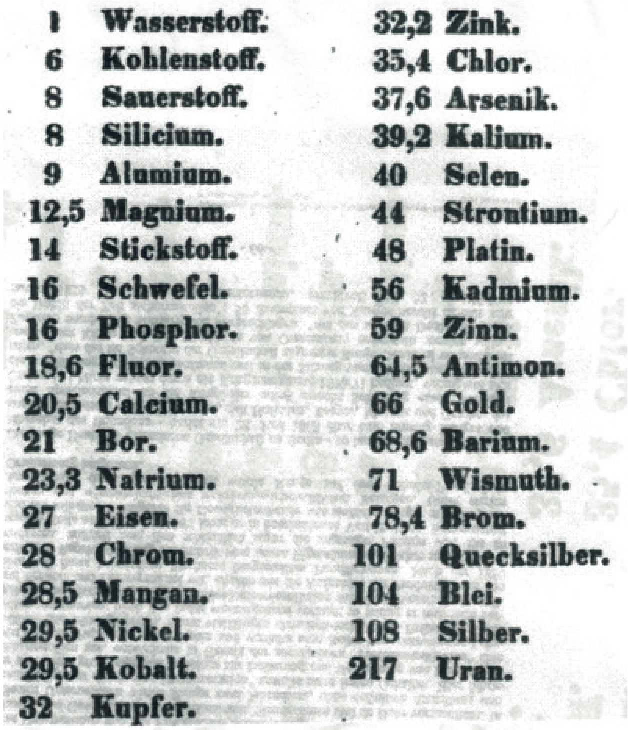
RUNGE AND THE CHEMICAL INDUSTRY

Runge carried out his experiments on coal tar while he was chemical director (1832/1852) of the chemical factory in Oranienburg, a city located northeast of Berlin. The factory was part of the *Preussische Seehandlung* (figure 4), a state-owned company founded in 1772 by King Frederick II the Great (1712-1786), to develop industrial and commercial activities. The chemical factory was housed in Oranienburg Palace (figure 5), built in 1651 by Frederick William the Great Elector (1620-1688) for his wife Louise Henriette (1627-1667), a princess of the house of Orange-Nassau (hence Oranienburg), expanded by King Frederick I (1657-1713), but later decommissioned as a royal residence and sold in 1802 to pharmacist Johann Gottfried Hempel (1752-1817). Hempel and his son Georg established a weaving factory there and in 1814 the chemical factory, producing mainly sulfuric acid using the lead chamber process, transferred in 1848 to a new location[22]. In the 19th century, the palace was partially restored as a royal residence by King Frederick William IV (1795-1861) and is now a museum. The poet and novelist Theodor Fontane (1819-1898) wrote about the factory and the palace: "The vapors of sulfuric acid corroded and deteriorated and swept away the last traces of the past beauty. I remember when I was a child, when I came this way, and from the square and the bridge I looked with fear at the old and terrifying building, immersed in smoke and ash"[23].

The *Preussische Seehandlung* went through difficulties in the Napoleonic period, but recovered with the efficient administration of Christian von Rother (1778-1848), maintaining textile factories (Breslau, Glatz), metallurgical factories (Berlin, Breslau, Dirschau), paper factories (Bromberg), zinc (Ohlau), and since 1841 the Oranienburg factory; the *Seehandlung* was one of the first European companies to trade with independent South America: in 1822 a ship left for Rio de Janeiro, taking textile products and returning with coffee, sugar and cotton.

The period of Rother's administration corresponds to the period in which Runge was chemical director, from 1832 to 1852. The factory began to produce, in addition to sulfuric acid and *oleum*, stearin (1833) and paraffin (1834) candles, soaps, soda, ammonium salts, Glauber's salt, Prussian blue. Runge's proposal to attempt the manufacture of dyes (1847) was not accepted by the factory management. For Andreas, the factory did not have the necessary equipment. In the opinion of Richard Anschütz (1852-1937), Kekulé's successor and biographer, the initiative would have been possible[24].

Table 1 Runge's table of 'Mischgewichte'. Illustration from 'Einleitung in die Technische Chemie füt Jedermann', Berlin, 1836, on page 16



1	Wasserstoff.	32,2	Zink.
6	Kohlenstoff.	35,4	Chlor.
8	Sauerstoff.	37,6	Arsenik.
8	Silicium.	39,2	Kalium.
9	Alumium.	40	Selen.
12,5	Magnium.	44	Strontium.
14	Stickstoff.	48	Platin.
16	Schwefel.	56	Kadmium.
16	Phosphor.	59	Zinn.
18,6	Fluor.	64,5	Antimon.
20,5	Calcium.	66	Gold.
21	Bor.	68,6	Barium.
23,3	Natrium.	71	Wismuth.
27	Eisen.	78,4	Brom.
28	Chrom.	101	Quecksilber.
28,5	Mangan.	104	Blei.
29,5	Nickel.	108	Silber.
29,5	Kobalt.	217	Uran.
32	Kupfer.		

From 1844 onwards the company experienced financial difficulties, and Runge feared the privatization of the factory in the course of the liberal movements of 1848. In fact, the *Seehandlung* sold the factory in 1850 to Ernst Eduard Cochius, who dismissed Runge in 1852, but was unable to avoid the bankruptcy of the enterprise. Cochius committed suicide in 1856. Even without an official connection with the factory, Runge continued to provide his assistance, maintaining his salary and a modest home.

In 1853/1855 Runge invented a substitute for saltpeter from Chile and guano from Peru, the "German guano", rejected "because it was not natural" (in the words of Cochius, it is "a criminal arrogance to want to produce bird excrement in the laboratory")[25] which led Runge to sell the process to the ministry of war. Runge explicitly refers to competition with England in the trade of Peruvian guano. He used recycled products to obtain the fertilizer, which in essence was ammonium phosphate combined with ammonium sulfate. A friend of Runge said: "In Oranienburg there must be a guano island!"[26] Offended by Runge's initiative, Cochius's widow expelled him from his palace residence and cut off the pension he received. Runge went to live in Oranienburg in a more than modest way.

RUNGE AND THEORY[27]

It is a widespread opinion that Runge had no theoretical interests, which is not correct. We will look at his publications later, for now let us limit ourselves to analysing his theoretical points of view expressed in the "*Einleitung in die Technische Chemie*", from 1836 (Introduction to Technological Chemistry) (figures 6 and 9). It is likely that Runge's theoretical work did not have greater repercussion because of his anti-atomism and his adherence to *Naturphilosophie*, already rejected by the majority of chemists. Runge does not consider his conceptions as "theory", but as the result of his experiments. He prefers a "dynamic chemistry" to atomism, and chemical compounds would be formed by mutual "interpenetration" (*Durchdringung)* of substances with opposite properties, which presupposes the divisibility of matter "to infinity". From this *Durchdringung* emerges a "third compound".

Runge knew 54 elements (Table 1), which he classified into three groups: the elements of the "oxygen series" (O, S, Se, Te, Cl, Br, I, P), roughly our non-metals; the metals; the metalloids (H, C, B, N). In compounds, metals can be replaced by others, which is a criterion for including them in a group. Runge classifies metals as light (Na, K, Mg, Ca, Sr, Ba, Al, etc.) and heavy (Fe, Ni, Co, Ag, Au, Tt, Mn, U etc.) and observes that in many aspects H is close to metals. It is also true in Chemistry that differents attract each other and similars repel each other, but this is in the presence of a metal. Without the metal, elements from the oxygen group can also combine with each other. The concept of metalloid used by Runge differs from the concept of metalloid common at his time[28].

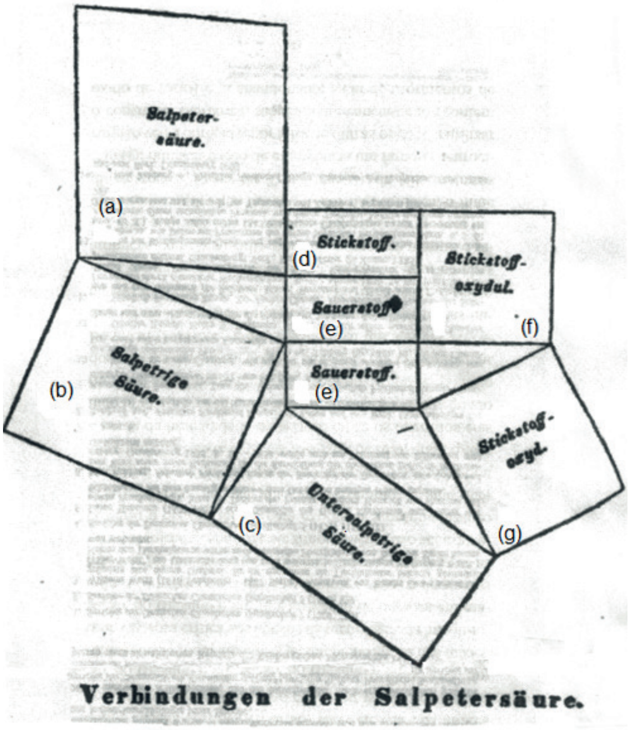
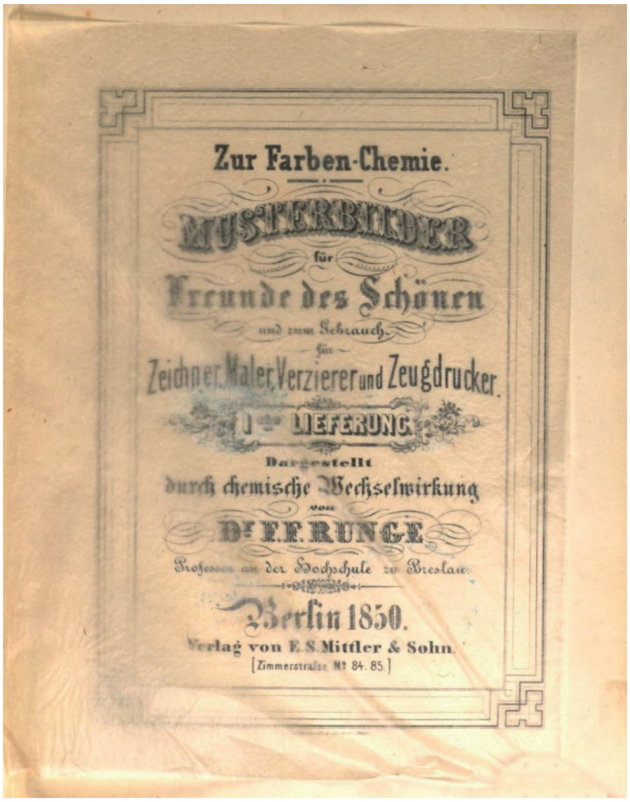


Figure 6 Compounds derived from Nitric Acid. Illustration from "Einleitung in die Technische Chemie", Berlin, 1836, on page 338. (a) nitric acid; (b) nitrous acid; (c) hyponitrous acid $H_2N_2O_2$: (d) nitrogen (e) oxygen; (f) N_2O ; (g) NO_2.

Klaus Röker observes that with his "dynamic chemistry" Runge independently arrived at the laws of constant proportions (Proust) and multiple proportions. To obtain a chemical compound by the interpenetration of substances with opposite properties, a certain "*quantum*" [term used by Runge] of properties of one of the substances must be canceled by an equal "*quantum*" of opposite properties of the other substance. Substances therefore combine according to numerical proportions determined by their "chemical activity" (*Chemische Wirksamkeit*). The greater the "chemical activity", the smaller the numerical value involved in these proportions, and the weaker the activity, the greater the value of this number: hydrogen, the lightest substance, therefore has stronger chemical activity (*Chemische Wirksamkeit*), uranium, the heaviest substance then known, the weakest activity.



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[Zimmerstraſse No 84. 85.]

Figure 7 F. F. Runge, "Musterbilder", cover, F. Mittler & Son, Berlin, 1850. (image in public domain).

Thus, to reach balance in the interpenetration of two substances, numerical values must be assigned, the *Mischungsgewichte* (= something like interaction weights), or *Chemischer Wert* (= chemical value), and Runge assigned numerical values to the 54 simple substances (= elements) then known. Hydrogen has the value 1, uranium has the value 217. In this system, the values 6 were assigned to carbon, 8 to oxygen, 14 to nitrogen. Therefore, to completely cancel out the opposing proportions of hydrogen and oxygen, 8 pounds [unit used by Runge] of oxygen interact with 1 pound of hydrogen, forming 9 pounds of water or ice[29]. Runge created a table of *Mischunsgewichte*[30], and, if he were an atomist, we would say that he created a table of atomic weights (see Table 1). He wrote about combinations (Runge used the expressions 'mixture' and 'combination' interchangeably): "In words, this relationship means nothing more than saying that the number that precedes each substance represents the numerical relationship with which it participates in a chemical combination. It is unalterable and can only vary in the sense of doubling, tripling, etc. Because of this, all oxygenated substances contain oxygen in proportions of 8 or 16 or 24 or 32 or 40 and so on"[31]. A comment not only historiographical, but even epistemological is in order here: being anti-atomist, Runge arrived, based only on empirical data and concepts consistent with *Naturphilosophie*, and without making use of the concept of quantitative atom, to conclusions equal to those reached by Dalton and others with the application of atomic theory and its consequences. The evolution of the theory is later than Dalton's, and much more complicated than it would be with the use of the concept of 'atom'. In any case, there are often several conceptually different ways to explain certain theoretical situations. In Runge's case, as the specific situations to be explained become more complicated, the theory cannot satisfactorily explain what happens, which led to its abandonment and oblivion. But its role in the evolution of theories involving formulas, equivalents, atomic weights, 'chemical value' and so on, remains important. I believe it can be said that there is a certain analogy with the abandonment of the phlogiston theory: there was a need for more and more *ad hoc* hypotheses, so that the system became impractical, and it was abandoned. In any case, with respect to atoms, molecules, equivalents etc., there would only be some order after the Karlsruhe Congress in 1860.

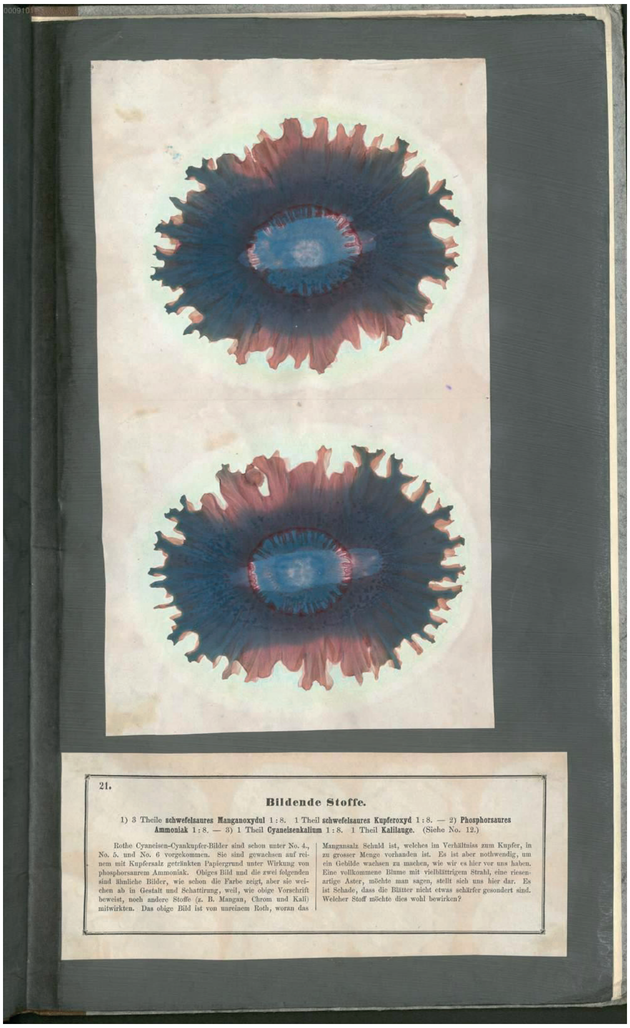
Runge also presented some other theoretical ideas. Combinations can be of various "orders". "First order" combinations involve only two substances, but there can be different stoichiometries, as in the combinations between N and O (N_2O, NO, NO_2 , N_2O_3 , N_2O_5). In "higher order" combinations, with three or more substances, the 'construction' of substances occurs in stages[32].

He also distinguishes three classes of combined compounds[33]:

- true acids, which always behave like acids (sulfuric acid, nitric acid, hydrochloric acid).
- true bases, which always behave like bases (soda, potash, ammonia).
- "acidic bases" (our amphoteric acids), which behave either as an acid, or as a base, depending on the other substance present (alumina, zinc oxide, tin oxide, lead oxide).

With an ingenious graphical representation method Runge represented the "oxygenation stages". Figure 6 shows the case of nitrogen[34], and Runge developed such graphical representations of the "oxygenation states" of several other elements, like, manganese, iron, lead, copper, tin and others. Substances (or elements) are represented by rectangles on the right of the diagram, compounds by rectangles arranged at an angle, around internal triangles.

Other theoretical subjects were addressed by Runge, such as Elective Affinities, but nothing new or different was added.



21.

Bildende Stoffe.

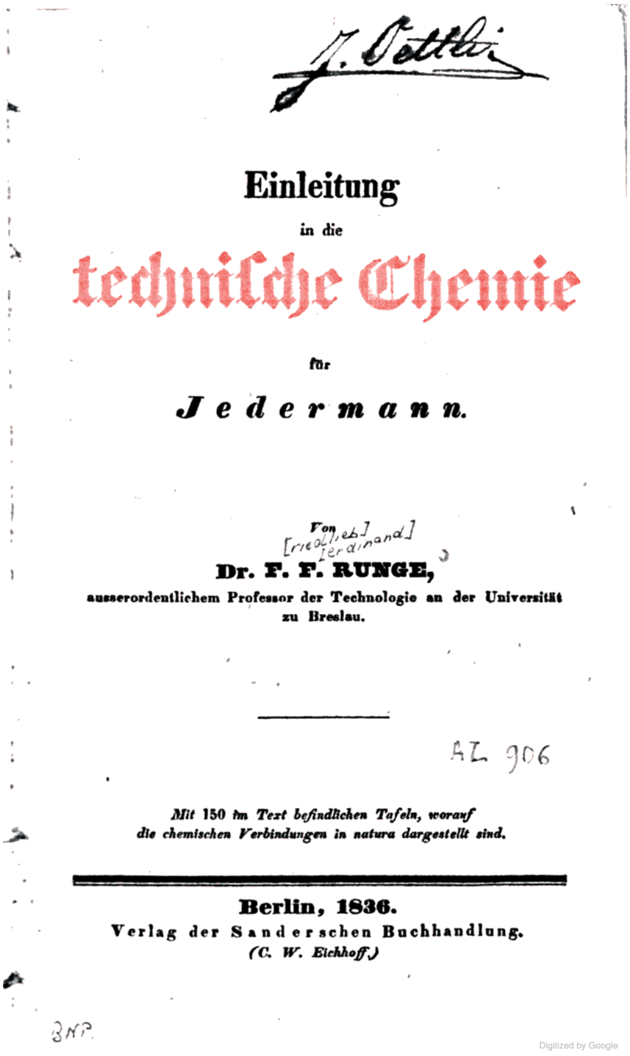
1) 3 Theile **schwefelsaures Manganoxydul** 1 : 8. 1 Theil **schwefelsaures Kupferoxyd** 1 : 8. — 2) **Phosphorsaures Ammoniak** 1 : 8. — 3) 1 Theil **Cyaneisenkalium** 1 : 8. 1 Theil **Kalilauge.** (Siehe No. 12.)

Rothe Cyaneisen-Cyankupfer-Bilder sind schon unter No. 4., No. 5. und No. 6 vorgekommen. Sie sind gewachsen auf reinem mit Kupfersalz getränkten Papiergrund unter Wirkung von phosphorsaurem Ammoniak. Obiges Bild und die zwei folgenden sind ähnliche Bilder, wie schon die Farbe zeigt, aber sie weichen ab in Gestalt und Schattirung, weil, wie obige Vorschrift beweist, noch andere Stoffe (z. B. Mangan, Chrom und Kali) mitwirkten. Das obige Bild ist von unreinem Roth, woran das Mangansalz Schuld ist, welches im Verhältniss zum Kupfer, in zu grosser Menge vorhanden ist. Es ist aber nothwendig, um ein Gebilde wachsen zu machen, wie wir es hier vor uns haben. Eine vollkommene Blume mit vielblättrigem Strahl, eine riesenartige Aster, möchte man sagen, stellt sich uns hier dar. Es ist Schade, dass die Blätter nicht etwas schärfer gesondert sind. Welcher Stoff möchte dies wohl bewirken?

Figure 8 An example of a 'Musterbild', page 21. (1) reaction of manganese sulfate with copper sulfate. (2) reaction of ammonium phosphate with potassium ferrocyanide (State Library of Bavaria, Munich) (image in public domain)

RUNGE, THE *MUSTERBILDER*, AND THE BEGINNINGS OF CHROMATOGRAPHY

That Science creates Art is nothing new, and Runge's "*Musterbilder*" or *Professorenklexe* are an eloquent example. The absorption on paper of the product of two reacting solutions forms "self-forming" designs, which resulted in Runge's famous book (figure 8), and the procedure can be considered a precursor to chromatography. In an 1836 book by the Englishman George Field (1777-1854) the word "chromatography" already appears. And there is a record from 1731, in "A treatise on colors", by a certain Taylor (1685-1731), watercolourist, which contains the word "chromatography", from the Latinized Greek words '*chromato*' (= color) and '*graphein*' (= to write).



J. Oettli

Einleitung
in die
techniſche Chemie
für
Jedermann.

von
[Friedlieb]
[Ferdinand]
Dr. F. F. RUNGE,
ausserordentlichem Professor der Technologie an der Universität
zu Breslau.

AL 906

Mit 150 im Text befindlichen Tafeln, worauf
die chemischen Verbindungen in natura dargestellt sind.

Berlin, 1836.
Verlag der Sanderschen Buchhandlung.
(C. W. Eichhoff.)

3NP.

Figure 9 Cover of Runge's "Einleitung zur Technischen Chemie für Jedermann", published by Sandersche Buchhandlung, Berlin, 1836. (public domain)

There is more or less consensus that the invention of absorption chromatography should be credited (there are dissenting voices) to the Russian botanist Michail Tswett (1872-1919). For many historians there are precursors, such as the Englishman Lester Reed (1860-1926), in Beneke's opinion, or the American David Talbot Day (1889-1925), for Aaron Ihde[35]. But both Lester and Day limited themselves to describing some simple cases of separation, they never worried about the theoretical aspects of this separation technique. The 'capillary analysis' of the Swiss Friedrich Goppelsröder (1837-1919) can also be considered a precursor to chromatography. And there are historians who go back even further, considering that the *spot tests* that Runge carried out in 1822 on fabrics and on paper, with the purpose of observing the behavior of dyes, would be the beginnings of empirical chromatography[36].

Figure 10 Runge's house in Oranienburg. He lived there from 1856 until his death. The house was demolished in the thirties (Courtesy City Archives and the Municipality of Oranienburg)

Runge's "natural creations" and his "*Musterbilder*" resemble a chromatogram *avant la lettre*, they had nothing to do with processes of separation, but rather with an incursion into aesthetics, into works of art "created by nature", spontaneously, and by the self-organizing force of Nature. Runge wrote about it: "What, then, are these figures? They are natural creations, which arise as a result of chemical reactions. The simple narrative of how I arrived at them will clarify to the reader the story of their creation"[37]. These spontaneous "natural creations" are formed as follows: paper impregnated with copper sulfate (blue) and potassium ferrocyanide solution ($K_4[Fe(CN)_6]$, mixed with $(NH_3)H_2PO_4$ as a developer, form a stain with a red core and green edges; a solution of manganese sulfate (pink) reacting with a solution of potassium chromate ($K_2Cr_2O_4$), and ammonia as a developer, give rise to a blue stain with a brown border[38]. Runge explains that by repeating these experiments exactly, using the same type of paper, the same salts in the same concentrations, and dripping the solutions at the same speed and in the same sequence, any researcher will obtain as many images as he wants. "Natural creation" is really science: the different speed of adsorption of the different colored solutions by the paper corresponds to what happens in chromatography, in the solid stationary phase.

Incredible as it may seem, Runge's interest in the spontaneous formation of these figures is not the chemical aspects involved, but the chemical-aesthetic aspects (although the experiments originate from his occupation with dyes). Something difficult to imagine at a time when Chemistry more and more takes on the characteristics of an exact science, but Runge's personality was eccentric enough to get involved with such experiments that for him were more aesthetic than chemical. In the year 1850, Runge chose 126 of these colorful figures and published the book "*Zur Farbenchemie. Musterbilder für Freunde des Schönen und zum Gebrauch für Zeichner, Maler, Verzierer und Zeugdrucker*", (figure 7) dedicated to the King of Prussia, Frederick William IV (1795-1861). The king personally thanked his somewhat exotic subject. In 1855, Runge published a second volume on the subject, "*Der Bildungstrieb der Stoffe. Veranschaulicht in selbständig gewachsenen Bildern*"[39].

Runge highlights the aesthetic aspect of these figures, their importance for the arts. He was convinced that his *Musterbilder* would be useful to painters and designers, for example, with new colors and new color combinations. Runge himself said: "Would we condemn Chemistry, if it, with more pride than Michelangelo, would exclaim: also *io sono pittore*!, why Chemistry is a painter, without a brush?". Today's idea of self-organization was expressed early on by Runge, when he stated that his still wet drawings are alive, as they still grow![40]

Let's see what Runge himself writes about his *Musterbilder*: "Whoever carefully observes the various figures in this book will soon clearly perceive that they could not have been painted with a brush. The so peculiar blurring and shading show that there cannot be here the arbitrariness practiced by the brush. The same goes for different colors, which can be produced by some arbitrary combination [...] The colors here are separate and not separate; they interpenetrate equally in separation and interpenetration. Something like this can only develop naturally and from within. What are these figures? They are natural formations, which are formed by chemical interactions"[41]. And further on: "Here a new world of formations, structures and color combinations suddenly appears, which I would never have imagined and which were not predictable, which surprised me even more. I soon learned the conditions for reproducing them in any quantity. Determining these aspects was particularly important for me, because this discovery, in addition to its chemical value, acquires importance for the plastic arts [...]"[42]. Without a doubt, a subject suitable for an eccentric and unorthodox chemist.

Figure 11 Friedlieb Ferdinand Runge (1794-1867), bronze statue by Stephan Möller(*1956), erected 1994 in Oranienburg, in front of Runge's laboratory, near the Palace (Courtesy City Archives and the Municipality of Oranienburg)

This item cannot be concluded without referring to an analogy between the *Bildungstrieb*, the driving force for the formation of substances, proposed by Runge, and the *Od* proposed by Karl von Reichenbach (1799-1869), a "hypothetical force that permeates all of Nature", responsible for phenomena such as hypnotism, magnetism, light and others. This very little known analogy was suggested by Leslie Ettre (1922-2010) and H. Bussemas, historians of chromatography. Runge wrote on the subject "*Das Od als Bildungstrieb der Stoffe*" (Oranienburg, 1866), a copy of which survives in the Yale University Library[43]. The peculiarity of this book also suggests a philosophical basis common to Runge and Reichenbach, in this case, linked to *Naturphilosophie*. Interestingly, Runge and Reichenbach had a strong personal friction in the 1830s, when both were researching coal tar, with dissenting points of view[44]. The Yale book may contain other secrets.

The subject of the *Musterbilder*, however, soon fell into oblivion, but recently, a more open view of Chemistry has returned to value these curious experiments by an unique chemist. Runge already valued non-chemical subjects from the beginning of his professional activity. In 1826, in Breslau, where he was a professor, he participated in the *Zwecklose Gesellschaft*, 'Society without Purpose', with his friend the poet August Heinrich Hoffmann von Fallersleben (1798-1874), who visited him twice in Oranienburg, to which also belonged painters (Carl Bräuer [1798-1874]), musicians (Immanuel Sauermann, Carl Schwindt), sculptors (Mächtig), the philologist Wilhelm Wackernagel (1806-1869), the industrialist Karl August Milde (1805-1851), in whose fabrics factory in Breslau Runge tested his dyes, and with whom he traveled through France, Switzerland and Great Britain, to visit industrial facilities. The Society met on Saturday nights, to read poems and other texts, to discuss cultural aspects, and ephemerides[45].

PUBLICATIONS

Unlike most chemists of his time, Runge published little in the scientific journals then in vogue, mainly his research on phytochemistry and coal tar derivatives, in *Isis*, in Liebig's *Annalen, in Poggendorffs Annalen,* all of them journals with wide circulation at the time. It cannot, therefore, be said that Runge was and is little cited and remembered because he published in little-known journals.

On the other hand, Runge published many books on Chemistry, both theoretical (as we have seen, without considering atomic theory) and practical. They were aimed not at the scientist, but rather at the common reader, the industrialist, the merchant, the practical chemist, which is why the practical chemist, which is why the most important series of these texts was called " *Jedermann-Chemie* ", something like "Chemistry for everyone" (the translation of the German word *Jedermann*, literally 'anyone', presents many subtleties). Runge himself explained his objective as follows: "[it is] a Chemistry of general understanding, but rigorously scientific, intended mainly for the trader, the manufacturer, the practical chemist, to be at their disposal as a faithful advisor". He intentionally gives up many chemical facts, which are less cited, but which can be found in Chemistry treatises. He also finds it unnecessary to present the physical properties of chemical compounds in great detail[46].

The first book published by Runge with this conception was "*Grundlagen der Chemie für Jedermann*" (Fundamentals of Chemistry for everyone), in 1830. We have already seen the theoretical bases on which Runge based his book. This was followed by "*Einleitung in die Technische Chemie für Jedermann*" (Introduction to the Technical Chemistry for everyone) in 1836, and "*Technische Chemie der Nützlichen Metalle für Jedermann*" (Technical Chemistry of Useful Metals for everyone), in 1838/1839. Unlike the first, these last two books are abundantly illustrated. In 1844, the crown prince and future King Maximilian II of Bavaria (1811-1864) ordered from Runge more than 7000 copies of a book in the "*Jedermann*" style, the "*Grundriss der Chemie für Jedermann*" (1846, Outline of Chemistry for everyone), to be distributed in all schools in Bavaria. He also published, in 1834, "*Farbenchemie. Die Kunst zu Färben*" (Chemistry of Colors. The Art of dyeing), supplemented in 1842 by "*Farbenchemie. Die Kunst zu Drucken*" (Chemistry of colors; The Art of printing). In 1839, he published the translation into German of "Conversations on Chemistry", by Jane Marcet (1769-1858), a popular text much to his liking[47].

In addition to the aforementioned "*Zur Farbenchemie. Musterbilder für Freunde des Schönen und zum Gebrauch für Zeichner, Maler, Verzierer und Zeugdrucker*" (1850), and a second volume on the subject, "*Der Bildungstrieb der Stoffe*" (1855), and "*Deutscher Guano*" (1858), Runge published in 1866 "*Hauswirtschaftliche Briefe*" (Domestic Letters), in which he addressed several issues, including those relating to his personal life (such as the visit made to Goethe on October 3, 1819, commented above) and the recipe for making his "synthetic wine". This "synthetic wine" was an alcoholic solution of citric acid, with the addition of various flavoring ingredients, which he kept secret.

With his *Jedermann* books, we can conclude today, Runge had in mind a new educational proposal. In the Preface on his "*Einleitung in die Technische Chemie*" (1836) he wrote: "The reglements of the *Schulmeister* (school teacher) are past. Even this submission no more exists in our times, which became free. To be educated is no more the same as to know the old classics. Finally we all are convinced that the knowledge of God's creations is more important than the knowledge of the human creations; that it is better to learn about Nature and all its connections with life, than to learn the dead languages of former times, related to our times only through the thread of History"[48]. Thus, Runge proposes an education system in which Mathematics and Sciences exert a central role, and should be the nucleus of an educational system; even at the universities, Medicine and Law students should be confronted with physical and chemical lectures. Almost every activity in modern times has a physical and chemical basis. The variety and quantity of chemical discoveries and researches call the attention of lay people, so that "confusion turns to clarity and fright turns into surprise and joy". Doubtless Runge's educational ideas contributed to his oblivion.

FINALE IN ORANIENBURG

After Runge's friction with the managers of the Oranienburg factory, Cochius's widow expelled him in 1856 from his lodgings in Oranienburg Palace. He then rented a more modest one-story house from a friend (figure 10), compatible with his income, which no longer exists (demolished in the 1930s). Also the commemorative plaque placed there in 1923 no longer exists. Single, he lived there writing the "*Hauswirtschaftliche Briefe*", or notices for newspapers and journals, but without carrying out any formal activity: King Frederick William IV granted him an annual pension. He met up with some friends, demonstrating his skills as a cook and enjoying his "synthetic wine". He also manufactured other "wines" and prepared various types of preserves. He lived simply, advised professionals in all areas, and also the common people who showed an interest in certain subjects. He died at his home on March 26, 1867, and was buried at the *Nikolaifriedhof*, where his tomb was a victim of vandals in 1923, but restored shortly afterwards. In 1949 the cemetery was closed, and the tomb was transferred to the municipal cemetery[49].

As we saw, belatedly, in 1994, a bronze statue of Runge was erected in front of his laboratory, attached to the Oranienburg Palace, designed by Stephan Möller (figure 11). The statue, at ground level, shows Runge at his work table, with various equipments. In 2017, on the 150th anniversary of his death, there were several celebrations in Oranienburg.

CONCLUSION

Runge was certainly an unusual, unorthodox chemist, which brought him a certain popularity during his lifetime, but not recognition among his peers. Why did he not find recognition from his colleagues, despite some undoubtedly important discoveries in the field of phytochemistry and coal tar? Perhaps for leaving academic activities and research to dedicate himself to industry, where his industrialization proposals were also not well received? Or perhaps because of his exotic and unusual behaviour? Perhaps for remaining faithful to *Naturphilosophie,* and because he did not accept the atomic theory? If Runge enjoyed some popularity during his lifetime, after his death he fell into almost total oblivion. A first writing lamenting the neglect of Runge, "Contribution to the history of scientific discoveries", is anonymous, and was published in 1863 in the popular (382.000 copies in 1875) weekly magazine *Die Gartenlaube*, founded by Ernst Keil (1816-1878) and published in Leipzig since 1853[50]. In 1869 the German Chemical Society created a commission (Hofmann, Baeyer, Magnus, Graebe) to honor Runge with a representative tomb, which only occurred in 1872[51]. The biographies of Schelenz (1907) and Rehberg (1936) clearly exaggerate the qualities attributed to Runge and his achievements, and the presentation of a "typical German researcher", in science and in personal life, was a big deal for Nazi-fascist ideology from the 1930s, even distorting Runge's real personality. All of this did not contribute to keeping Runge's name visible in historiography. The novelist Karl Aloys Schenzinger (1886-1962), a supporter of Nazi ideology but without being a member of the party, found in Runge, by disfiguring his character and personality, the first "hero" of his novel "Anilin" (1936), the "novel of a dye", nationalist and full of errors and omissions, but very popular (almost a million copies sold until 1945)[52]. In the 1950s, after the compromising passages from the first editions had been expunged, new versions of the novel were published (last edition in 1973)[53]. Clearly, the idealized chemist in "Anilin" is not Runge. In any case, one should not confuse character and author/work: with the growing importance of chromatography, Runge and the "*Musterbilder*" gained prominence, and with the interaction that exists today between Chemistry and other human activities, Runge's role draws attention, for example, in the Runge Prize for unconventional artistic creations, awarded since 1994. A new Runge emerges, closer to historical reality.

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REFERENCES

1 Partington, J., "History of Chemistry", Macmillan, London, vol. IV, pp. 183-184 (1964). Pechenkin, A., "The History of Research on Chemical Periodic Processes", Springer, p. 4 (2018),

2 Runge, F., "Neueste phytochemische Entdeckungen zur Begründung einer wissenschaftlichen Phytochemie", G. Reimer, Berlin (1820).

3 Runge, F., *op. cit.*, (1820), pp. 101-144.

4 Runge, F., "Mein Besuch bei Goethe im Jahre 1819", in "Hauswirtschaftliche Briefe", 36th Letter, p. 153, Oranienburg (1867) (reprint, Weinheim, 1988).

5 Schwedt, G., "Goethe als Chemiker", John Wiley/Verlag Chemie, New York/Weinheim, (1998).

6 Runge, F., "Hauswirtschaftliche Briefe", p. 153 (1866)

7 Runge, F., "Über Pflanzenchemie", *Isis*, **2** (I8), pp. 317-321, 1320 (1819). See also Anft, B., "Friedlieb Ferdinand Runge: a Forgotten Chemist of the 19th Century", *J. Chem. Educ.*, **32**, 566-574 (1955)

8 Runge, F., *op. cit.*, (1820), pp. 122-160.

9 Runge, F., "Uber Pflanzenchemie", *Isis*, **2** (8), pp. 317-321 (1819)

10 Pelletier, P., Caventou, J., "Recherches Chimiques sur les quinquinas", *Ann. Chim. Phys*, **15**, 289-318, 337-365 (1820). See also Délepine, M., "Joseph Pelletier and Joseph Caventou", *J. Chem. Educ.*, **28**, 454-461 (1951).

11 Anft, B., *op. cit.* p. 567.

12 Bloch, M., "Apologia da História", Zahar, Rio de Janeiro (2002). (1949)

13 Real, H., Schneider, W., "Wer entdeckte Chinin und Cinchonin?", *Beiträge zur Geschichte der Pharmazie*, **22**, 17-19 (1970).

14 Runge, F., "Monographie des Krapps", Pietsch, Berlin, p. 1 (1835). See also 'Annual Report of the Progress of Chemistry and the allied Sciences', edited by J. von Liebig and H. Kopp, volume IV, Walton and Maberly, London, p. 354 (1853).

15 Runge, F., *op. cit.* (1820).

16 Andreas, H., "Friedlieb Ferdinand Runge: Wegbereiter der Teerfarbenchemie?, *Mitteilungen der Fachgruppe Geschichte der Chemie, GDCh.,* **20**, 9-22 (2008).

17 Anft, B., *op. cit.*, p. 571.

18 Runge, F., "Über einige Produkte der Steinkohlenteerdestillation", *Poggendorffs Annalen*, **31**, 65-67; 513-524 (1834). 32, 308-327; 328-332 (1834).

19 Schelenz, H., "Otto Unverdorben, der Anilinentdecker", *Zeitschrift f. Angewandte Chemie*, **34**, 51-52 (1921).

20 Hofmann, A. W., "Chemische Untersuchungen der organischen Basen im Steinkohlen-Theröl". *Annalen*, **47**, 37-87 (1843).

21 Andreas, H., *Mitteilungen der Fachgruppe Geschichte der Chemie, GDCh*, **20**, 9-22 (2009).

22 Lärmer, S., "Schwefelsäure aus Oranienburg", *Berlinische Monatsschrift*, pp. 11-17 (1999).

23 Fontane, T., "Wanderungen durch die Mark Brandenburg", part III, p. 148, Munich, (1994).

24 Anschütz, "Friedlieb Ferdinand Runge", in Allgemeine Deutsche Biographie, 29, 686-686 (1889). Asche, W., "Erste Teerfarben aus der Berliner Region", *Berlinische Monatsschrift*, pp. 52-54 (1996)

25 Anft, B., *op*. cit., p. 573.

26 Runge, F., "Der Deutsche Guano in Oranienburg", F. Duncker, Berlin (1858).

27 Röker, K., "Die Jedermann-Chemie des Friedlieb Ferdinand Runge", *Mitteilungen Fachgruppe Geschichte der Chemie, GDCh*, **23**, 52-70 (2013).

28 Runge, F., "Einleitung in die Technische Chemie für Jedermann", pp. 5-12, Sandersche Buchhandlung, Berlin (1838).

29 Röker, K., *op. cit*., (2013), p. 57.

30 Runge, F., *op. cit*. (1836), pp. 13-16.

31 Runge, F., *op. cit*. (1836), pp. 21-22.

32 Röker, K., *op. cit*., pp. 59-60.

33 Runge, F., *op. cit*. (1836), pp. 34-37.

34 Runge, F., *op. cit*. (1836), p. 338. See also Röker, K. *op. cit*., p. 59

35 Ihde, A., "The Development of Modern Chemistry", Dover Publications, N. York, p. 571 (1984).

36 Anft, B., *op. cit*., p. 572.

37 Runge, F ., "Zur Farbenchemie. Musterbilder für die Freunde des Schönen und zum Gebrauch von Zeichner, Maler, Verzierer und Zeugdrucker", Berlin (1850).

38 Schwenk, E., "Friedlieb Runge and his Capillary Designs", *Bull. Hist. Chem*., **30**, 30-34 (2005).

39 Runge, F., "Der Bildungstrieb der Stoffe", Oranienburg (1855).

40 Rex, S. "Friedlieb Ferdinand Runge und seine Musterbilder", *Chimia*, **48**, 11-15 (1994).

41 Schweitzer, S., "Natur zwischen Ästhetik und Selbstorganisation." in "Zum Naturbild der Gegenwart", vol. II, Stuttgart, pp. 93-119 (1994), on p. 2 (quoting Runge).

42 Schweitzer, S., *op. cit*., on p. 3 (quoting Runge).

43 Bussemas, H., Ettre, L., "Forerunners of Chromatography: Runge's Self-Grown Pictures", *LGCG America*, **32**, 262-270 (2004).

44 Engel, M., "Karl Ludwig Freiherr von Reichenbach", in Neue Deutsche Biographie, 21, 305-307 (2003).

45 Hoffmann von Fallersleben, A., "Mein Leben", part I, volume II, Berlin (1894).

46 Röker, K., *op. cit*., p. 55.

47 Röker, L., *op. cit*., p. 67.

48 Runge, F., "Einleitung in die Technische Chemie", Preface, pp. V-X , Sandersche Verlagsbuchhandlung, Berlin (1836).

49 Engel, M., Teichmann, H., "Zum 200. Geburtstag Friedlieb Ferdinand Runge's: Gedenkveranstaltungen in Oranienburg", *Mitteilungen der Fachgruppe Geschichte der Chemie, GDCh*, *11*, 66-69 (1995).

50 Anonimous, "Beitrag zur Geschichte der wissenschaftlichen Entdeckungen", *Die Gartenlaube*, n° 4, p. 63, Leipzig (1867).

51 Engel, M., Teichmann, H., *op. cit*., p. 66

52 Keas, M., "Karl Aloys Schenzinger's Novel 'Anilin': Chemistry and Chemical Technology in Nazi Literaturpolitik", *Ambix*, **39,** 127-140 (1992).

53 Hoeft, L.. "Karl Aloys Schenzingers 'Anilin' als durchgesehene und ergänzte Neuauflage", University of Halle-Wittenberg (2014).

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Implementing ChatGPT as Tutor, Tutee, and Tool in Physics and Chemistry

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Abstract. In the age of modern technology, generative artificial intelligience-powered chatbots offer a variety of uses for different purposes. Undoubtedly, ChatGPT is one of the most widely used chatbots in science education. In this paper, we review the implementations of chatbots, focusing particularly in teaching and learning physics and chemistry. Their roles in the context of science education are classified as tutee, tutor, and tool. We found the development of ChatGPT to be quite impressive. As a tutee, the latest version of ChatGPT is a fast learner, capable of passing standard tests and providing accurate scientific answers using approaches like Chain-of-Thought and Socratic-style dialogue. As a tutor, it can help students learn through classroom teaching techniques such as scaffolding and enhance critical thinking by acting as a personal tutor that offers instantaneous feedback. As a tool, ChatGPT can assist in reviewing students' handwritten homework, drafting scientific writing, and generating code for science programming. Although ChatGPT offers many benefits, it can sometimes provide inaccurate information, necessitating human oversight in science education. Importantly, students should be taught to critically assess the responses provided by ChatGPT and understand its ethical use to ensure effective utilization.

Keywords: Artificial intelligence, ChatGPT, Physics, Chemistry, Science education

1. INTRODUCTION

Chatbots and other artificial intelligence (AI) tools are shaping the future across various domains, including education [1-3]. The successful integration of AI into education depends on collaboration among educators and policymakers. In science education, ChatGPT has the potential to enhance student learning and improve educational outcomes [4]. Educators can use ChatGPT to supplement traditional teaching methods, providing students with additional resources to support their learning and engage them further in the subject matter. However, ChatGPT can sometimes provide misleading information and be susceptible to negative and unethical outcomes [1-6]. Therefore, the challenge for educators is to leverage chatbots and other AI tools to maximize student learning efficiency and prepare them for their future professional lives [4]. The effectiveness of chatbots depends on several factors, including the academic discipline and specific domain [6,7].

This article reviews the implementation of ChatGPT in physics and chemistry education. To provide context, the development of chatbot technology is briefly summarized in the following section. The subsequent sections (3 and 4) are structured around the three roles of chatbots: tutor, tutee, and tool. The roles of ChatGPT as a tutee and tutor in physics and chemistry are particularly intriguing as they reflect the chatbot's ability to comprehend concepts and deliver accurate information. Given the importance of reliability in education, ChatGPT must be capable of providing precise and trustworthy knowledge that can be effectively utilized in teaching physics and chemistry. Following this discussion, ChatGPT's performance is compared with other AI chatbots to offer broader insights. The article concludes by addressing ethical considerations and providing an outlook on the future of AI in education.

2. BRIEF HISTORY OF CHATBOT DEVELOPMENT

Chatbots, or conversational agents, are programs designed to interact with humans via text- or voice-based interfaces. These systems are created and trained to comprehend user input, identify the purpose of the dialogue, and generate human-like responses. Chatbot development can be broadly categorized based on either pattern-matching algorithms or machine learning models, the latter of which is foundational to modern AI-driven chatbots. The history of chatbots dates back to 1966 with Eliza, one of the first conversational agents created by Joseph Weizenbaum [8]. Eliza simulated a psychotherapist by engaging in simple conversations based on pattern matching. In 1972, Parry was developed as a more advanced chatbot, employing a system of assumptions and simulated human reactions to replicate the thought patterns of someone with paranoid schizophrenia [9]. Both chatbots, while rudimentary by today's standards, laid the groundwork for future developments in AI dialogue systems.

Schobel et al. summarized the evolution of chatbot technology into five waves: the zero-hour wave, the explore wave, the kick-off wave, the hype wave, and the AI wave [10]. During the explore wave, the integration of Natural Language Processing (NLP) became a key focus. NLP enabled chatbots to not only understand but also analyze and interpret natural human language, vastly improving their conversational capabilities. Significant advancements in this period included Jabberwacky in 1988 and A.L.I.C.E in 1995, and the term AI was used firstly used for the chatbots with the former [9]. The kick-off wave was marked by the debut of IBM Watson in 2006, introducing AI-driven chatbot technology into the mainstream with real-world applications. This demonstrated the potential of chatbots to handle large-scale knowledge queries and complex problem-solving tasks. The hype wave saw the mass adoption of chatbots for a variety of consumer-facing roles. This wave introduced some of the most recognizable AI-driven assistants, including Apple's Siri (2011), Amazon Alexa (2014), Microsoft Cortana (2014), and Google Assistant (2016). These systems moved beyond simple Q&A and evolved into fully integrated personal assistants. They could understand complex voice commands and perform tasks ranging from setting reminders to controlling smart home devices. By this time, chatbots were widely implemented across sectors such as marketing, customer support, healthcare, education, and entertainment.

The launch of ChatGPT by OpenAI in late 2022 initiated the AI wave, which has sparked unprecedented global interest in generative AI technologies [10]. Unlike its predecessors, ChatGPT was built using a large language model (LLM), specifically GPT-3.5, and later GPT-4, allowing it to generate coherent and contextually relevant text responses across a wide array of topics. In response, major technology firms accelerated their AI chatbot development, leading to the introduction of Bard by Google and Bing Chat by Microsoft in 2023. These new generative AI chatbots offer capabilities such as answering complex questions, explaining scientific principles, summarizing texts, and even generating academic essays or code, positioning them as indispensable tools for both casual users and professionals alike. As AI continues to evolve, the role of chatbots in education, research, and everyday life will likely expand, with generative models like ChatGPT serving as critical companions in learning, problem-solving, and content creation.

3. CHATGPT IN PHYSICS EDUCATION

3.1 Tutee

Many researchers and educators investigate and train ChatGPT to test its knowledge of physics and train it to perform better. In the following paragraphs, we will briefly describe several studies considering ChatGPT to be a tutee.

One such investigation by Wang scrutinizes ChatGPT's proficiency in solving physics problems [11]. Initially tasked with resolving the motion of a body on a frictionless incline, ChatGPT adeptly interprets the query and correctly identifies physical variables such as angle, gravitational acceleration, force, and mass. However, the acceleration sign is incorrect, which could cause coordinate system confusion. The author then requests that ChatGPT simulate a model of the situation. The results are extremely incredible. ChatGPT can generate a simulation to resolve this problem correctly. The author challenges ChatGPT with a more difficult question about the Stern-Gerlach experiment. The results showed that ChatGPT was unable to answer the question accurately. Consequently, while ChatGPT appears capable of solving simple physics problems, such conceptual topics remain challenging to tackle.

Interestingly, Kortemeyer delves into whether ChatGPT could pass an introductory physics course [12]. Employing a multifaceted assessment approach that includes multiple-choice questions, homework assignments, clicker questions, programming exercises, and exams, ChatGPT exhibits varying degrees of competence. The results found that ChatGPT scored 18 out of 30 points on the Force Concept Inventory (FCI) [13]. For homework, this allowed it to make 5 attempts on topics including trajectory motion, friction, thermodynamics, capacitance, and special relativity, covering a total of 76 homework problems. It was found that ChatGPT frequently made numerical errors and was unable to correct these mistakes even after they were pointed out. Despite this, ChatGPT solved 55% of the homework problems using an average of 1.88 attempts per problem. For clicker questions, ChatGPT correctly answered 10 out of 12 questions, a score better than most students in the actual course. Additionally, ChatGPT was also assigned to write Python code related to an anharmonic oscillator for programming exercises. The author noted that ChatGPT performed much better than many students in the course. ChatGPT scored 14 out of 30 points on the midterm and final exams. The results showed that five of the incorrect answers were due to numerical calculation errors. Although ChatGPT could answer correctly after reverse token verification, full credit was not awarded. Considering the grading policies (20% homework, 5% clicker questions, 5% programming exercises, and 70% exams), the author determined that ChatGPT would receive a course grade of 1.5. However, if it had been more accurate in numerical operations, the course grade could have been 2, reflecting a 60% performance. Hence, ChatGPT can pass an introductory physics course.

Another perspective to evaluate the performance of ChatGPT is to use it to write short essays. Yeadon et al. examined ChatGPT's ability to write essays on physics concepts and historical and philosophical themes, such as "Is physics based on facts that follow from observations?" and "How did natural philosophers' understanding of electricity change during the 18th or 19th centuries?" [14]. The exam comprised five short-form essay questions, each limited to 300 words, with a maximum score of 100 per question. These essays were graded by five different markers, and ChatGPT achieved an average score of 71±2%, which is high enough to qualify for a First Class grade, the highest distinction available at UK universities. The plagiarism rates, checked by both Grammarly and Turnitin, were found to be 2±1% and 7±2%, respectively. The authors pointed out that ChatGPT poses a significant threat to the integrity of short-form essays as an assessment method in physics courses.

In term of educational learning objectives, López-Simó and Rezende explored ChatGPT's capability to solve five types of physics questions related to Bloom's taxonomy: dictionary definitions, simple calculations, multistep calculations, reasoning problems, and Fermi problems [15]. Using GPT-3 for its broader accessibility, each question was asked ten times in separate windows. ChatGPT performed well on dictionary definitions (e.g., Newton's second law) and simple calculations, answering correctly 7 out of 10 times. However, it failed to solve multistep calculations correctly even once and showed a preference for certain options in reasoning problems. For Fermi problems, which require interpretation and informed reasoning, ChatGPT provided answers closer to the expected order of magnitude but with inconsistency. The authors concluded that ChatGPT is still unreliable as a self-help tool for introductory physics. However, the authors suggested leveraging its limitations to engage students in critical discussions, enhancing their understanding of complex physics problems.

The subsequent version of ChatGPT, ChatGPT-4, demonstrates a remarkable ability to tackle advanced physics problems. This capability was thoroughly investigated across various physics domains. Dazhen et al. investigated the performance of ChatGPT-4 in solving physics conceptual understanding and reasoning problems, encompassing mechanics and electromagnetism [16]. The research utilized two primary problem sets in physics education, namely the FCI and the Conceptual Survey of Electricity and Magnetism (CSEM) [17]. The authors selected sixteen multiple-choice questions to assess ChatGPT's abilities. Impressively, ChatGPT answered all questions correctly, whereas the average score for the FCI test among 415 university students in 2018 was 56.3%. For CSEM, the average score among 9,905 students who completed an electromagnetic course was 44.6%.

The author conducted another test that utilized primitive physics problems (PPPs). This evaluates various knowledge representations such as abstraction, assignment, image, physics, methodology, and mathematics, as proposed by Xing et al. [18]. However, ChatGPT struggled with image representation, which was subsequently excluded from the evaluation. In this assessment, the authors compared ChatGPT's performance with that of 388 middle school students across four questions following PPPs. Three researchers independently scored the physics reasoning problems based on the aforementioned representations. The results revealed that ChatGPT achieved a significantly higher score of 87.5% overall, compared to the middle school students' average score of 23.32%. A similar study conducted by West compared the performance of ChatGPT-3.5 and ChatGPT-4 on a modified version of the FCI, consisting of 23 usable questions [19]. The results showed that ChatGPT-3.5 answered fifteen questions correctly (65%), performing just below or slightly above the average student. Impressively, ChatGPT-4 answered 22 out of the 23 questions correctly. The authors noted that ChatGPT-4's ability to engage in metaphor and utilize multiple representations distinguishes it from novices and aligns more closely with expert-level understanding.

In addition, Kumar and Kats conducted a comparative analysis of ChatGPT's performance in solving 13 electromagnetism problems of differing complexities from an introductory course at the Department of Electrical and Computer Engineering, University of Wisconsin-Madison [20]. The study included different versions of ChatGPT, namely ChatGPT-3.5, ChatGPT-4, and ChatGPT-4 with a Code Interpreter (4/CI). ChatGPT-4 introduces a range of plugins, such as WolframAlpha and Code Interpreter, enabling it to execute calculations and generate Python code for plotting graphs. The study found that ChatGPT-4 with Code Interpreter (4/CI) outperformed its predecessors in resolving all 13 electromagnetic problems. ChatGPT-4/CI consistently demonstrated high accuracy in solving these problems, including tasks like integrating charge density in Cartesian coordinates and calculating electric fields in dielectrics. However, it should be noted that ChatGPT-4/CI occasionally identified its own errors when prompted for explanations. Furthermore, it exhibited a stochastic nature when solving vector calculus problems, occasionally yielding different answers, both correct and incorrect.

In contrast to studies with a small number of questions, Yeadon and Halliday assessed ChatGPT's performance on Durham University physics exams [21]. They analyzed 42 exam papers from 10 different physics courses, spanning 2018 to 2022, with 593 questions, including traditional and COVID-era adaptive formats. During the COVID period (2021-2022), open-book exams were allowed. ChatGPT-4 outperformed ChatGPT-3.5, achieving average scores of 49.4% compared to 38.6%. Pre-COVID, ChatGPT-4 scored 50.8% while ChatGPT-3.5 scored 41.6%, and post-COVID scores dropped to 47.5% and 33.6%, respectively. ChatGPT-4's performance showed minimal variation between the two periods. This demonstrates ChatGPT-4's improved performance.

The ability to interpret graphs is also crucial in physics. ChatGPT-4 was enhanced to process image data, a feature examined by Polverini and Gregorcic [22]. The authors tested this capability using the Test of Understanding Graphs in Kinematics (TUG-K), a multiple-choice assessment widely used to evaluate students' comprehension of one-dimensional motion graphs. Each of the 26 survey items was uploaded as a "png" screenshot, and the test was submitted to ChatGPT 60 times across 1,560 separate chats. The average score was 10.85 points (41.7%), a performance comparable to that of high school students. The authors noted ChatGPT's tendency to answer certain items correctly or incorrectly consistently. Additionally, the authors cautioned against relying on ChatGPT for tutoring students with typical learning difficulties, highlighting the need for careful consideration in its educational application.

Furthermore, Polverini and Gregorcic demonstrated techniques for improving ChatGPT's conceptual physics task performance [23]. They highlighted the Chain-of-Thought (CoT) strategy, which involves prompting the AI to think step-by-step. For example, asking, "If two bodies with different masses have the same kinetic energy, which one has the largest momentum?" without CoT led to incorrect answers four out of eight times. However, framing the question to include reasoning improved accuracy, for example, "If two bodies with different mass have the same kinetic energy, which one has the largest momentum? Provide your reasoning first and only then provide the answer," with correct answers seven out of eight times. This illustrates the potential of CoT prompts in enhancing AI performance. The authors recommended dialogue-based approaches, such as asking, "Isn't there another way to do it?" to train ChatGPT and help students use AI chatbots effectively in physics education.

3.2 Tutor

As ChatGPT and other chatbots are anticipated to become indispensable tools for academic training and assessment in future education, the role of ChatGPT as a physics tutor is discussed. Traditionally, students do not receive instantaneous instructor feedback, but modern technology like ChatGPT can significantly enhance student learning. In this section, we summarize methods for using ChatGPT in the classroom.

Liang et al. investigated ChatGPT's potential in physics education [24]. They found that ChatGPT can provide scaffolding by generating step-by-step guidance. For instance, ChatGPT can explain projectile motion by breaking down complex movements into components and offering detailed explanations. It can also generate questions and hints to assess students' understanding and summarize variables from questions in a table, making problem-solving easier. However, it sometimes makes errors when judging vector directions and calculations. ChatGPT can permute variables to create new computation problems, helping instructors prepare homework materials with commands like "Permute the physics variables and give me another problem: [input problem]." Additionally, the authors suggest using commands to enhance physics study and motivation, such as "Please tell me some stories about physicist [name]" and "Physics concepts are applied in various real-world scenarios. Please provide examples of how [concept name] is applied." Despite these benefits, the study's limitation is that it has not yet been applied in real classrooms and lacks empirical support.

Another study claimed that ChatGPT can encourage critical thinking. This experiment was conducted by using high school students as samples. Bitzenbauer uses ChatGPT to teach quantum physics to 53 secondary school students [25]. The author first instructs students to ask ChatGPT questions such as "What is a photon?" and "What kind of particle is a photon?". After obtaining a response from ChatGPT, students can double-check the answer by looking for relevant answers to compare errors provided by ChatGPT, which they have already studied in class. This allows students to think about and become aware of the ChatGPT answer. This refers to critical thinking ideas such as generating conclusions based on facts, making decisions, or forming perceptions about something. Students were assigned to discuss ChatGPT-generated statements in pairs. In this phase, students could look through textbooks and scientific papers to check and revise their ChatGPT answers before debating them. This helps to cultivate the habit of verifying and sharing discoveries. In the last stage, the author utilizes ChatGPT to generate conceptual questions to assess students. This will allow students to review the topics they discussed with their friends. Finally, the authors compare a questionnaire from before and after the course regarding perspectives on learning with ChatGPT. Overall, the average student's rate of agreement after the class is greater than before the class. The question "we should all learn to incorporate ChatGPT in our lives" has a substantial favorable impact on student perceptions of ChatGPT. However, "We can use ChatGPT even if we do not understand how it works" lowers after the class, indicating that students are more aware of ChatGPT results.

Alneyadi and Wardat utilized ChatGPT to teach the concrete idea of electromagnetism [26]. The authors employ ChatGPT to teach electromagnetics to eleventh-grade students, using 58 students for experimental and 64 students for control. In their experiment, the authors placed the students into two groups: those who used ChatGPT and those who did not. This experiment lasts four weeks. The data in this study is separated into two categories: quantitative and qualitative data. The electromagnetism exam is a part of quantitative data that contains a pretest and a post-test. It consists of 25 multiple-choice questions based on Bloom's taxonomy, including recall, comprehend, apply, and analyze. Following data collection, the authors apply the t-test to determine whether there is a significant difference in average pretest and posttest scores for both the experimental and control groups. The results revealed that the experimental group's mean post-test score was higher than that of the control group.

Open-ended survey questions by interviewing record audio were used to gather qualitative data. Authors conducted interviews covering six themes related to students' perceptions of ChatGPT: (i) ChatGPT as a Learning Tool: Students noted its usefulness in providing instant answers and explanations, particularly aiding in overcoming language barriers. (ii) Impact on Performance: While students appreciated its assistance with visual aids and homework problem-solving, some expressed concern over the lack of grading feedback. (iii) Comfort with Using ChatGPT: Overall, students reported feeling comfortable, though a few voiced occasional hesitations or frustrations with the technology. (iv) Differences in ChatGPT Use: Male students tended to use ChatGPT for quick answer confirmations, whereas females utilized it for deeper understanding, investing more time in its use. (v) Suggestions for Improvement: Students suggested enhancements such as accent and dialect recognition, more visual aids and animations, and faster response times to questions. (vi) Recommendation to Other Students: Students indicated they would recommend ChatGPT to peers, underscoring its perceived value in aiding learning.

In higher education, ChatGPT is still useful for assessing students' understanding of many physics ideas. Dahlkemper et al. conducted an experiment to assess first-year students' understanding of physics principles and their attitudes regarding AI [27]. This contains students from two university physics courses totaling 95 people. The authors initially collected general data on students' perceptions of using ChatGPT; the findings revealed that the majority of students (84%) had heard of ChatGPT, but only half had used the chatbot. Furthermore, 74% of respondents say they would never employ ChatGPT in physics. Following that, the formal experiment was performed. The author tasked students with evaluating the performance of four physics responses across three topics: rolling motion, waves, and fluid dynamics, generated by ChatGPT. Notably, one response in each topic was provided by experts. The familiarity level varied among the topics: rolling motion, being the easiest, was previously assigned as homework; waves were studied a few weeks prior, while fluid dynamics, being the least familiar, required application of knowledge from various fields and was considered the most challenging.

Initially, students self-assessed their performance on a scale of 0 to 6 without attempting the problems. It was found that a decrease in average score was observed across the less familiar topics. The next step involved assessing the answers labeled as both ChatGPT and expert responses, followed by students evaluating both the scientific accuracy and linguistic quality. A two-way repeated-measures analysis of variance was conducted to compare the average scores between two groups: scientific accuracy and linguistic quality. The analysis revealed significant differences in assessing ChatGPT responses across the topics. In the case of rolling motion and waves, students rated the expert answer highest for both scientific accuracy and linguistic quality. In contrast, for fluid dynamics, the expert solution was rated significantly higher in scientific accuracy, while linguistic quality did not differ significantly. This study sheds light on student perceptions of ChatGPT responses and the factors influencing their assessments.

Ding et al. also conducted a study involving 40 college-level students to explore their perceptions of using ChatGPT [28]. The students were tasked with regaining lost exam credits by interacting with ChatGPT on topics related to light and radioactivity. Over 1.5 weeks, students posed questions to ChatGPT, reviewed its responses, and assessed their validity, providing reasons for their decisions. If they disagreed with ChatGPT's answers, they had to argue their points and repeat the process. Following this activity, the students were surveyed about their experience. During the task, out of 362 questions asked, ChatGPT answered 85% correctly. However, upon further questioning, it revised its responses, correcting itself from incorrect to correct answers in 7 instances but also changing from correct to incorrect in 34 instances. Additionally, the authors employed K-means clustering to categorize students into three groups based on their level of trust in ChatGPT's responses: the trust group, partial trust group, and distrust group. ANOVA and MANOVA analyses were used to examine the statistical significance among these groups and assess differences in perception. The findings revealed that nearly half of the students trusted ChatGPT's answers regardless of their accuracy, perceiving it as a knowledgeable machine. These students found ChatGPT easy to use and expressed a greater likelihood of using it in the future than partial trust and distrust groups.

3.3 Tool

The literature discussed presents diverse viewpoints regarding the effectiveness of ChatGPT as both a physics tutee and tutor. In this section, we introduce additional literature advocating ChatGPT as a valuable tool for educators.

Gregorcic and Pendrill conducted an experiment wherein basic physics questions were posed to ChatGPT [29]. For instance, the question "A teddy bear is thrown into the air. What is its acceleration at the highest point?" yielded inconsistent and sometimes incorrect responses from ChatGPT. Despite providing some correct information, its answers often contradicted themselves, indicating a lack of coherence in understanding concepts like net force. Despite its linguistic prowess, ChatGPT struggled to recognize and rectify its own contradictions. However, the authors suggest potential applications in education, particularly in teacher training as a tool, where it could aid in recognizing and interpreting problematic argumentation. Subsequently, Gregorcic et al. reported that ChatGPT-4 demonstrated improved performance through repeated questioning, accurately answering all queries [30]. When tasked with describing graphs, ChatGPT4 exhibited proficiency, albeit with occasional unclear expressions regarding graph slopes. Notably, this version of ChatGPT showed the ability to detect inconsistencies in its responses and could provide correct answers with prompting. The authors concluded that ChatGPT-4 holds promise as a training tool for teaching physics through Socratic dialogue, based on both their direct experiences and insights from their pilot study.

In addition, one of the intensive tasks for physics teachers is grading students' work. When it comes to physics problems that require derivations, this can become a significant workload, demanding both time and effort. Fortunately, technology nowadays can help address these challenges. Kortemeyer examines an AI-assisted workflow to grade handwritten physics derivations using MathPix and GPT-4 [31]. The process begins with scanning handwritten papers into PDF files using the smartphone app Scanner Pro, which are then transcribed into LaTeX using MathPix. The output from MathPix is subsequently refined using GPT-4. The author evaluates the effectiveness of ChatGPT in grading electricity problems compared to human graders. The scores range from 0 (worst) to 4 (best) based on a rubric assessing the correctness of approach, symbolic derivations, numerical results, and straightforwardness. ChatGPT has demonstrated considerable potential for grading student work. While AI-assigned grades show a strong correlation to manually assigned grades ($R^2 = 0.84$), they are currently unreliable enough for summative assessments with limits and errors when conducting symbolic and numerical computations. However, it is reliable enough to help human graders by sorting or grouping answers and offering preliminary grades.

4. CHATGPT IN CHEMISTRY EDUCATION

4.1 Tutee

Much like those in physics, chemistry educators and researchers have investigated ChatGPT's understanding and accuracy in responding to various topics. Clark et al. discovered that ChatGPT's accuracy significantly varied across topics [32]. It performed well in pH calculations for strong acids and bases but struggled with more complex problems like titrations and aqueous salts. Unlike students, the chatbot avoided heuristic errors but made uncommon mathematical mistakes. Leon and Vidhani noted that while ChatGPT can provide correct answers within a given context, it often has difficulty verifying the computational or analytical accuracy of those answers [33]. Fergus et al. reported that ChatGPT's responses to chemistry assessments were generally well-written but varied in quality [34]. The chatbot had difficulties with application and interpretation questions, especially those involving non-text information. This observation was consistent with Clark, who found that ChatGPT could identify concepts in closed-response questions with significant chemical symbolism but performed below the class average in problem-solving [35]. For open-response questions, ChatGPT demonstrated strong language processing abilities, performing better on questions that could be solved with generalizable information rather than specific skills taught in lectures. However, incorrect responses and flawed explanations often seemed logically sound and persuasive to students.

Nascimento and Pimentel highlighted ChatGPT's low accuracy in converting SMILES representations into compound names and vice versa, with errors such as missing or adding methyl groups, including non-existent atoms, confusing regular cyclic and aromatic compounds, or misunderstanding isomers [36]. The chatbot also had difficulty with the most current and robust string representation. Daher et al. used the theoretical framework encompassing transfer, depth, predict/explain, problem-solving, and translation to evaluate ChatGPT's conceptual understanding in the material science domain [37]. They found significant difficulties in conceptual knowledge across various categories, particularly in representations and depth, hindering effective knowledge transfer.

4.2 Tutor

While chatbots should not be solely relied upon for providing answers or explanations to students due to their shortcomings noted in sections 2.1 and 3.1, they have significant potential as teaching assistants to complement educators' efforts. When financial limitations prevent institutions from hiring multiple teachers, chatbots provide a cost-effective solution to meet the individual needs of students. According to Alasadi and Baiz [38], AI-assisted teaching benefits students by providing additional support and personalized attention. Chatbots can function as virtual co-teachers, assisting in evaluating student progress, offering tailored feedback, and delivering targeted interventions. This allows human educators to focus on fostering meaningful interactions and deepening students' understanding of the subject. Guo and Lee observed significant improvements in students' confidence to ask insightful questions, analyze information, and understand complex concepts, thanks to ChatGPT's ability to present diverse perspectives and challenge existing thought processes [39]. Students also reported using ChatGPT more frequently to enhance their critical thinking skills and expressed a willingness to recommend it to others. Exintaris et al. described a classroom activity combining metacognitive scaffolding, problem-solving practice, and critiquing ChatGPT-generated solutions [40]. This approach showed that students engaged with metacognition as a key part of their problem-solving toolkit and appreciated the collaborative nature of the exercise. They also identified errors and flaws in the provided incorrect solutions, although to varying degrees.

4.3 Tool

Chatbots have gained popularity as tools for academic writing due to their abilities to generate ideas, draft content, edit, and proofread. West et al. analyzed the strengths and weaknesses of laboratory reports generated by ChatGPT [41]. Rojas et al. noted that although students found ChatGPT helpful for scientific writing, they were reluctant to use it to generate entire texts [42]. Clark et al. discovered that students could distinguish between essays on sustainability written by ChatGPT and those written by humans [43]. While students' essays contained more scientific reasons and chemistry concepts, they were impressed by ChatGPT's ability to discuss sustainability solutions, policies, and practices. Desaire et al. pointed out that manuscripts generated by ChatGPT are likely to be detected by chemistry journals [44].

Beyond writing assistance, chemistry researchers recognize the potential of chatbots in laboratory research and design. Araujo and Saude showed that ChatGPT could conceptualize problems and laboratory activities accessible to chemistry students, although the accuracy and safety of these activities require human oversight [45]. Scoggin and Smith examined ChatGPT's ability to help students generate experimental designs based on general chemistry textbook questions, noting that success depended on the clarity of the questions [46]. Chatbots also show promise in designing chemical reactions. Zheng et al. used ChemPrompt Engineering to train ChatGPT to text-mine peer-reviewed articles on Metal-Organic Frameworks (MOFs) [47]. ChatGPT could then answer questions about synthesis procedures, identify critical factors in MOF crystallization, and predict experimental outcomes. Mahjour et al. demonstrated that ChatGPT could formulate reaction arrays for common pharmaceutical reactions, with these results usable as inputs for management software like Phactor, enabling automated execution and analysis of assays [48]. Kong et al. employed ChatGPT to create interactive learning environments and simulate real-world engineering thinking processes in distillation column design for undergraduate mass-transfer courses [49]. Hasrod et al. used human prompts and an error message feedback loop with ChatGPT to generate working code for a graphic user interface (GUI) to predict sulfate levels in acid mine drainage [50]. This template allows students to create their GUIs for codes or models developed during their studies, demonstrating the potential for augmenting analytical data to infer or approximate non-directly analyzable parameters.

5. COMPARISON OF THE PERFORMANCE OF VARIOUS CHATBOTS IN SCIENCE, ENGINEERING, AND MEDICAL EDUCATION

Chatbots offer valuable educational information across various fields, serving as tutee, tutor, and tool—key roles in advancing educational technology. Apart from reviewing ChatGPT, we also compare its performance with other chatbots. To offer diverse perspectives, we include literature from various fields such as science, engineering, and medical education, as there are few comparative studies on chatbots in physics and chemistry.

In a study by Dos Santos [51], ChatGPT-3.5, ChatGPT-4, Bing Chat, and Bard were compared to investigate their effectiveness. The author posed questions related to motion and energy concepts to these chatbots across three sessions: analyzing the acceleration of a teddy bear at its highest point (similar to references [29,30]), understanding the speed of a ball at half height, and discussing a roller coaster loop. ChatGPT-4 emerged as particularly notable for its accurate application of physics concepts. Nikolic et al. [52] investigated the performance of ChatGPT, Copilot, Gemini, SciSpace, and Wolfram across assessment tasks in 10 engineering subjects, including quizzes, numerical problems, oral, visual, programming, and writing tasks. The results showed that chatbots are generally unlikely to pass these assessments, with visual, project-based written, and research-based written assessments being secure. In addition, the authors note that ChatGPT-4 is particularly reliable for most engineering applications, suggesting the benefits of the paid version.

In chemistry, Watts et al. compared writing-to-learn assignments produced by ChatGPT-3.5, ChatGPT-4, and Bard [53]. They found that while the responses varied, the chatbots seldom discussed electron movement, a critical component of mechanistic reasoning. As a result, the chatbots did not engage in reasoning to the same extent as students. Regarding structural notations, Hallal et al. compared ChatGPT and Bard's understanding of condensed structures, InChi, and SMILES, and their ability to answer organic chemistry-related questions [54]. They assessed the chatbots' abilities to convert IUPAC names, InChi, and SMILES notations into condensed forms and vice versa, identify functional groups, generate molecular formulas, and predict resonance patterns. Leite [55] also compared the performance of ChatGPT, Gemini, and Copilot in defining five basic chemistry concepts (atom, electron, mole, molecule, and chemical substance) against IUPAC definitions. The author found that Copilot was the only chatbot to cite sources for its generated text, while Gemini provided references. Additionally, the author used GPTZero, Plagium, Smodim, and AI Content Detector to determine if the texts were AI-generated. The results showed that GPTZero, Plagium, and Smodim detected AI-generated text 33.3% of the time, while AI Content Detector detected it only

6.7% of the time. Overall, the author noted that Gemini delivered the most satisfactory responses, followed by Copilot and ChatGPT. Nascimento Jr et al. [56] compare ChatGPT 3.5, ChatGPT 4.0, Google Bard, Bing Chat, Adobe Firefly, Leonardo.AI, and DALL-E, focusing on both textual and imagery content. These AIs are classified as Free or Paid (ChatGPT 4.0 and DALL-E). For textual content, the chatbots perform well in chemical bonding, aligning with scientific consensus (Ct3). In imagery, only the paid ChatGPT 4.0 effectively identifies chemical content, Lewis structures, and arrow orientations, generating mostly accurate responses with minor errors.

In biophysical phenomena, liquid–liquid phase separation (LLPS) was presented to ChatGPT-4 and Gemini to evaluate their explanations and understanding [57]. The authors analyzed accuracy, response time, response length, and cosine similarity index (CSI) of the responses. Gemini consistently provided more accurate answers than ChatGPT, though neither model answered all questions correctly. The CSI was 0.62, indicating moderate similarity between the models.

In medicine, Rossettini et al. [58] investigated the performance of ChatGPT-4, Microsoft Copilot, and Google Gemini in passing the Italian entrance exam for healthcare science degrees (CINECA test). The results show that ChatGPT-4 and Microsoft Copilot outperformed Google Gemini. The authors note that differences in neural network architecture impact accuracy, with ChatGPT-4 and Microsoft Copilot using GPT (Generative Pre-trained Transformer) architecture, while Google Gemini employs LaMDA (Language Model for Dialogue Application) and later PaLM 2 (Pathways Language Model) combined with web search. Meyer et al. [59] compared responses from ChatGPT, Gemini, and Le Chat in interpreting complete blood counts in an online health forum. The results showed inaccuracies in the chatbots' interpretations, particularly with complex patient questions. This research highlights the need for patient caution when using chatbots for self-diagnosis. Saeedi and Aghajanzadeh [60] used ChatGPT and Perplexity to assess dysphonia's perceptual level by analyzing voice self-assessments and acoustic data. Chatbots were asked to classify the severity of voice disorders (vocally healthy, mild, moderate, or severe dysphonia). The authors found that while chatbots occasionally made correct assessments, their reliability was inconsistent for clinical use. Kaba et al. [61] tested ChatGPT-3.5, ChatGPT-4, Gemini, and Perplexity with MRI safety-related questions, finding that ChatGPT-4 outperformed the others with an accuracy of 93.3%, followed by Gemini, Perplexity, and ChatGPT-3.5. The authors suggest that these chatbots could potentially assist healthcare professionals in the future.

Throughout our review of comparison studies on chatbots used as tutees, tutors, and tools across various fields, many studies suggest that the paid version of ChatGPT-4 outperforms other chatbots. However, we believe that more analysis is still needed to compare chatbots, as paid versions should provide greater accuracy in all chatbots, not just ChatGPT. It is noted that the paid versions of ChatGPT, Gemini, and Perplexity, which are popular chatbots, are priced at 20 USD per month. This advantage raises concerns about educational inequality, particularly for those with financial constraints. Different chatbots offer various benefits and perspectives. For instance, ChatGPT relies on its dataset to create responses based on trends discovered during training, while Gemini retrieves and analyzes data from Google Search in real-time for more realistic queries. However, both of them have drawbacks related to transparency, unlike Perplexity AI, which allows for the citation of its information sources [62]. In terms of research tools and education, we believe Perplexity is more suitable for providing scientific citations. However, its overall performance requires further exploration, as this is just the beginning of the chatbot era, where training biases and user interactions impact accuracy. Furthermore, we believe that a comparison of paid chatbots is needed, as much of the literature compares free versions of chatbots with paid versions of ChatGPT, which could introduce bias in assessing the accuracy of chatbot performance.

6. ETHICAL CONSIDERATIONS AND OUTLOOK

This literature review offers insights into the emerging roles of ChatGPT in physics and chemistry education. ChatGPT demonstrates impressive capabilities as a tutee, effectively providing scientific descriptions, solving conceptual problems, and passing standard tests. However, obtaining effective answers often requires an understanding of ChatGPT's functioning and specific approaches. Utilizing strategies like Chain-of-Thought and Socratic-style dialogue can significantly enhance its efficacy and utility [23,63,64]. Advanced versions of ChatGPT and other chatbots exhibit improved abilities in creating simulations and reading graphs. Nonetheless, chatbots are inherently limited in their in-depth knowledge and advanced analysis, occasionally resulting in misleading information. It follows that teaching should not rely solely on current chatbot technology. The literature reveals varying levels of effectiveness among current chatbots as tutors, suggesting that they can be helpful as teaching assistants. ChatGPT-4 is particularly notable for its accurate application of scientific concepts, demonstrating a deep understanding of the learning process by exhibiting excellent facilitation skills, delivering content knowledge, and encouraging student engagement. The literature also highlights substantial advancements in chatbots' roles as facilitators for academic writing and assessment, showcasing their versatility as tools for science education. Interestingly, chemistry educators and researchers have explored the potential of ChatGPT in laboratory and research design. Such chatbot assistance in idea development will be extremely useful across various other science disciplines.

Given the massive impact of generative AI, the focus has shifted to supporting educators in integrating the technology effectively and raising student awareness of its ethical use. Ethical considerations regarding the use of AI, as highlighted by several authors [2,65-68], are summarized as follows. Over-reliance on technology, bypassing critical thinking, writing, or problem-solving processes, risks diminishing human creativity, ingenuity, and intellectual development in both teaching and learning. Misuse of AI can lead to academic dishonesty, plagiarism, and reduced engagement in the learning process. Transparency and accountability in AI decision-making, as well as copyright issues, remain areas of concern. Educators and students must verify AI-generated outputs through trusted academic sources to ensure accuracy, as AI systems can produce incorrect or misleading information. Generative AI, trained on vast datasets, may inadvertently amplify biases related to gender, race, or socioeconomic status, and without fully understanding the context, AI can produce inappropriate outputs for specific age groups or academic levels. Additionally, concerns about security and privacy arise, as personal data and interactions with AI systems may be stored and analyzed without explicit consent. The introduction of AI in education could also exacerbate the digital divide, disadvantaging students without access to high-quality technology and resources. Furthermore, integrating AI in education may reduce human-to-human interaction, which is crucial for developing social and emotional skills. Overcoming these challenges is a moral obligation for users of the technology. Jobin et al. identified eleven key ethical principles in AI implementation [69], while Petricini emphasized the importance of truthfulness, temperance, prudence, and courage—four specific Aristotelian virtues—to guide ethical AI practices [70]. While the emergence of chatbots and other AI tools should indeed be cherished as opportunities for progress, their implementation must remain anchored in human intelligence and moral virtue.

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REFERENCE

[1] M. Ivanova, G. Grosseck, C. Holotescu, Unveiling insights: A bibliometric analysis of artificial intelligence in teaching. *Informatics* **2024**, 11, 10. DOI 10.3390/informatics11010010

[2] M. R. Zheltukhina, O. V. Sergeeva, A. R. Masalimova, R. L. Budkevich, N. N. Kosarenko, G. V. Nesterov, A bibliometric analysis of publications on ChatGPT in education: Research patterns and topics. *Online J. Commun. Media Technol.* **2024**, 14(1), e202405. DOI 10.30935/ojcmt/14103

[3] E. Dimitriadou, A. Lanitis, A critical evaluation, challenges, and future perspectives of using artificial intelligence and emerging technologies in smart classrooms. *Smart Learn. Environ.* **2023**, 10, 12. DOI 10.1186/s40561-023-00231-3

[4] F. Jia, D. Sun, C.-K. Looi, Artificial intelligence in science education (2013–2023): Research trends in ten years. *J. Sci. Educ. Technol.* **2024**, 33, 94-117. DOI 10.1007/s10956-023-10077-6

[5] P. Lo Nostro, Artificial intelligence vs. Natural stupidity. *Substantia* **2023**, 7, 5-6, DOI 10.36253/Substantia-2250

[6] L. S. Balhorn, J. M. Weber, S. Buijsman, J. R. Hildebrandt, M. Ziefle, A. M. Schweidtmann, Empirical assessment of ChatGPT's answering capabilities in natural science and engineering. *Sci. Rep.* **2024**, 14, 4998. DOI 10.1038/s41598-024-54936-7

[7] C. K. Lo, What is the impact of ChatGPT on education? A rapid review of the literature. *Educ. Sci.* **2023,** 13, 410. DOI 10.3390/educsci13040410

[8] J. Weizenbaum, Eliza—a computer program for the study of natural language communication between man and machine. *Commun. ACM* **1966**, 9, 36-45.

[9] E. Adamopoulou, L. Moussiades, Chatbots: History, technology, and applications. *Machine Learn. Appl.* **2020**, 2, 100006. DOI 10.1016/j.mlwa.2020.100006

[10] S. Schobel, A. Schmitt, D. Benner, M. Saqr, A. Janson, J. M. Leimeister, Charting the evolution and future of conversational agents: A research agenda along five waves and new frontiers. *Inform. Sys.*

Front. **2024**, 26, 729-754. DOI 10.1007/s10796-023-10375-9

[11] J. Wang, ChatGPT: A test drive. *Am. J. Phys.* **2023**, 91, 255-256. DOI 10.1119/5.0145897

[12] G. Kortemeyer, Could an artificial-intelligence agent pass an introductory physics course? *Phys. Rev. Phys. Educ. Res.* **2023**, 19, 010132. DOI 10.1103/PhysRevPhysEducRes.19.020163

[13] D. Hestenes, I. Halloun, Interpreting the force concept inventory: A response to March 1995 critique by Huffman and Heller. *Phys. Teach.* **1995**, 33, 502-506. DOI 10.1119/1.2344278

[14] W. Yeadon, O. Inyang, A. Mizouri, A. Peach, C. Testrow, The death of the short-form physics essay in the coming AI revolution. *Phys. Educ.* **2023**, 58, 035027. DOI 10.1088/1361-6552/acc5cf

[15] V. López, M. Rezende, Challenging ChatGPT with different types of physics education questions. *Phys. Teach.* **2024**, 62, 290-294. DOI 10.1119/5.0160160

[16] D. Tong, Y. Tao, K. Zhang, X. Dong, Y. Hu, S. Pan, Investigating ChatGPT-4's performance in solving physics problems and its potential implications for education. *Asia Pac. Educ. Rev.* **2024**, 25, 1379-1389. DOI 10.1007/s12564-023-09913-6

[17] D. Maloney, T. O'Kuma, C. Hieggelke, A. Heuvelen, Surveying students' conceptual knowledge of electricity & magnetism. *Am. J. Phys.* **2001**, 69, S12-S23. DOI 10.1119/1.1371296.

[18] H. Xing, W. Gong, P. Wang, Y. Zhai, Y. Zhao, The measuring instrument of primitive physics problem for junior high school students: Compilation and research. *J Baltic Sci Educ.* **2022**, 21, 305-324. DOI 10.33225/jbse/22.21.305

[19] C. West, AI and the FCI: Can ChatGPT project an understanding of introductory physics? *Arxiv* **2023**, DOI 10.48550/arXiv.2303.01067

[20] T. Kumar, M. A. Kats; ChatGPT-4 with Code Interpreter can be used to solve introductory college-level vector calculus and electromagnetism problems. *Am. J. Phys.* **2023**, 91, 955-956. DOI 10.1119/5.0182627

[21] W. Yeadon, D. P. Halliday, Exploring Durham University physics exams with large language models. *Arxiv* **2023**, DOI 10.48550/arXiv.2306.15609

[22] G. Polverini, B. Gregorcic, Performance of ChatGPT on the test of understanding graphs in kinematics. *Phys. Rev. Phys. Educ. Res.* **2024**, 20, 010109. DOI 10.1103/PhysRevPhysEducRes.20.010109

[23] G. Polverini, B. Gregorcic, How understanding large language models can inform the use of ChatGPT in physics education. *Eur. J. Phys.* **2024**, 45, 025701. DOI 10.1088/1361-6404/ad1420

[24] Y. Liang, D. Zou, H. Xie, L. Wang, Exploring the potential of using ChatGPT in physics education. *Smart Learn. Environ.* **2024**, 10, 52. DOI 10.1186/s40561-023-00273-7

[25] P. Bitzenbauer, ChatGPT in physics education: A pilot study on easy-to-implement activities. *Contemp. Educ. Technol.* **2023**, 15, ep430. DOI 10.30935/cedtech/13176

[26] S. Alneyadi, Y. Wardat, ChatGPT: Revolutionizing student achievement in the electronic magnetism unit for eleventh-grade students in Emirates schools. *Contemp. Educ. Technol.* **2023**, 15, ep448. DOI 10.30935/cedtech/13417

[27] M. Dahlkemper, S. Lahme, P. Klein, How do physics students evaluate artificial intelligence responses on comprehension questions? A study on the perceived scientific accuracy and linguistic quality of ChatGPT. *Phys. Rev. Phys. Educ. Res.* **2023**, 19, 010142. DOI 10.1103/PhysRevPhysEducRes.19.010142.

[28] L. Ding, T. Li, S. Jiang, A. Gapud, Students' perceptions of using ChatGPT in a physics class as a virtual tutor. *Int. J. Educ. Technol. Higher Educ.* **2023**, 20, 63. DOI 10.1186/s41239-023-00434-1

[29] B. Gregorcic, A.-M. Pendrill, ChatGPT and the frustrated Socrates. *Phys. Educ.* **2023**, 58, 035021. DOI 10.1088/1361-6552/acc299

[30] B. Gregorcic, G. Polverini, A. Sarlah, ChatGPT as a tool for honing teachers' Socratic dialogue skills. *Phys. Educ.* **2024**, 59, 045005. DOI 10.1088/1361-6552/ad3d21

[31] G. Kortemeyer, Toward AI grading of student problem solutions in introductory physics: A feasibility study. *Phys. Rev. Phys. Educ. Res.* **2023**, 19, 020163. DOI 10.1103/PhysRevPhysEducRes.19.010132

[32] T. M. Clark, E. Anderson, N. M. Dickson-Karn, C. Soltanirad, N. Tafini, Comparing the performance of college chemistry students with ChatGPT for calculations involving acids and bases. *J. Chem. Educ.* **2023**, 100, 3934-3944. DOI 10.1021/acs.jchemed.3c00500

[33] A. J. Leon, D. Vidhani, ChatGPT needs a chemistry tutor too. *J. Chem. Educ.* **2023**, 100, 3859-3865. DOI 10.1021/acs.jchemed.3c00288

[34] S. Fergus, M. Botha, M. Ostovar, Evaluating academic answers generated using ChatGPT. *J. Chem. Educ.* **2023**, 100, 1672-1675. DOI 10.1021/acs.jchemed.3c00087

[35] T. M. Clark, Investigating the use of an artificial intelligence chatbot with general chemistry exam question. *J. Chem. Educ.* **2023**, 100, 1905-1916. DOI 10.1021/acs.jchemed.3c00027

[36] C. M. C. Nascimento, A. S. Pimentel, Do large language models understand chemistry? A conversation with ChatGPT. *J. Chem. Inform. Model.* **2023**, 63, 1649-1655. DOI 10.1021/acs.jcim.3c00285

[37] W. Daher, H. Diab, A. Rayan, Artificial intelligence generative tools and conceptual knowledge in problem solving in chemistry. *Information* **2023**, 14, 409. DOI 10.3390/info14070409

[38] E. A. Alasadi, C. R. Baiz, Generative AI in education and research: Opportunities, concerns, and solutions. *J. Chem. Educ.* **2023**, 100, 2965-2971. DOI 10.1021/acs.jchemed.3c00323

[39] Y. Guo, D. Lee, Leveraging ChatGPT for enhancing critical thinking skills. *J. Chem. Educ.* **2023**, 100, 4876-4883. DOI 10.1021/acs.jchemed.3c00505

[40] B. Exintaris, N. Karunaratne, E. Yuriev, Metacognition and critical thinking: Using ChatGPT-generated responses as prompts for critique in a problem-solving workshop (SMARTCHEMPer). *J. Chem. Educ.* **2023**, 100, 2972-2980. DOI 10.1021/acs.jchemed.3c00481

[41] J. K. West, J. L. Franz, S. M. Hein, H. R. Leverentz-Culp, J. F. Mauser, E. F. Ruff, J. M. Zemke, An analysis of AI-generated laboratory reports across the chemistry curriculum and student perceptions of ChatGPT. *J. Chem. Educ.* **2023**, 100, 4351-4359. DOI 10.1021/acs.jchemed.3c00581

[42] A. J. Rojas, An investigation into ChatGPT's application for a scientific writing assignment. *J. Chem. Educ.* **2024**, 101(5), 1959-1965. DOI 10.1021/acs.jchemed.4c00034

[43] M. J. Clark, M. Reynders, T. A. Holme, Students' experience of a ChatGPT enabled final examina non majors chemistry course. *J. Chem. Educ.* **2024**, 101(5), 1983-1991. DOI 10.1021/acs.jchemed.4c00161

[44] H. Desaire, A. E. Chua, M.-G. Kim, D. Hua, Accurately detecting AI text when ChatGPT is told to write like a chemist. *Cell Rep. Phys. Sci.* **2023**, 4, 101672. DOI 10.1016/j.xcrp.2023.10167

[45] J. L. Araujo, I. Saude, Can ChatGPT enhance chemistry laboratory teaching? using prompt engineering to enable AI in generating laboratory activities. *J. Chem. Educ.* **2024**, 101(5), 1858-1864. DOI 10.1021/acs.jchemed.3c00745

[46] J. Scoggin, K. C. Smith, Enabling general chemistry students to take part in experimental design activities. *Chem. Educ. Res. Pract.* **2023**, 24, 1229. DOI 10.1039/d3rp00088e

[47] Z. Zheng, O. Zhang, C. Borgs, J. T. Chayes, O. M. Yaghi, ChatGPT chemistry assistant for text mining and the prediction of MOF synthesis. *J. Am. Chem. Soc.* **2023**, 145, 18048-18062. DOI 10.1021/jacs.3c05819

[48] B. Mahjour, J. Hoffstadt, T. Cernak, Designing chemical reaction arrays using Phactor and ChatGPT. *Org. Process Res. Dev.* **2023**, 27, 1510-1516. DOI 10.1021/acs.oprd.3c00186

[49] Z. Y. Kong, V. S. K. Adi, J. Sunarso, J. G. Segovia-Hernandez, Complementary role of large language models in educating undergraduate design of distillation column: Methodology development. *Digit. Chem. Eng.* **2023**, 9, 100126. DOI 10.1016/j.dche.2023.100126

[50] T. Hasrod, Y. B. Nuapia, H. Tutu, ChatGPT helped me build a chemistry App, and here's how you can make one also. *J. Chem. Educ.* **2024**, 101, 653-660. DOI 10.1021/acs.jchemed.3c01170

[51] R. Dos Santos, Enhancing physics learning with ChatGPT, Bing Chat, and Bard as agents-to-think-with: A comparative case study. *arXiv* **2023**, DOI 10.48550/arXiv.2306.00724

[52] S. Nikolic, C. Sandison, R. Haque, S. Daniel, S. Grundy, M. Belkina, S. Lyden, G. M. Hassan, P. Neal. ChatGPT, Copilot, Gemini, SciSpace and Wolfram versus higher education assessments: An updated multi-institutional study of the academic integrity impacts of Generative Artificial Intelligence (GenAI) on assessment, teaching and learning in engineering. *Australasian J. Eng. Educ.* **2024**, 29(2), 126-153. DOI 10.1080/22054952.2024.2372154.

[53] F. M. Watts, A. J. Dood, G. V. Shultz, J.-M. G. Rodriguez, Comparing student and generative artificial intelligence chatbot responses to organic chemistry writing-to-learn assignments. *J. Chem. Educ.* **2023**, 100, 3806-3817. DOI 10.1021/acs.jchemed.3c00664

[54] K. Hallal, R. Hamdan, S. Tlais, Exploring the potential of AI-Chatbots in organic chemistry: An assessment of ChatGPT and Bard. *Comput. Educ.: Artif. Intel.* **2023**, 5, 100170. DOI 10.1016/j.caeai.2023.100170

[55] B. Leite, Generative Artificial Intelligence in chemistry teaching: ChatGPT, Gemini, and Copilot's content responses. *J. Appl. Learn. Teach.* **2024**, 7(2), 1-15. DOI 10.37074/jalt.2024.7.2.13

[56] W. J. D. Nascimento Jr, C. Morais, G. Girotto Jr, Enhancing AI responses in chemistry: integrating text generation, image creation, and image interpretation through different levels of prompts. *J. Chem. Educ.* **2024,** 101, 3767-3779. DOI 10.1021/acs.jchemed.4c00230

[57] N. Rana, N. Katoch, AI for biophysical phenomena: A comparative study of ChatGPT and Gemini in explaining liquid–liquid phase separation. *Appl. Sci.* **2024**, 14, 5065. DOI 10.3390/app14125065

[58] G. Rossettini, L. Rodeghiero, F. Corradi, C. Cook, P. Pillastrini, A. Turolla, G. Castellini, S. Chiappinotto, S. Gianola, A. Palese, Comparative accuracy of ChatGPT-4, Microsoft Copilot and Google Gemini in the Italian entrance test for healthcare sciences degrees:

Across-sectional study. *BMC Med. Educ.* **2024**, 24, 694. DOI 10.1186/s12909-024-05630-9

[59] A. Meyer, A. Soleman, J. Riese, T. Streichert, Comparison of ChatGPT, Gemini, and Le Chat with physician interpretations of medical laboratory questions from an online health forum. *Clin. Chem. Lab. Med.* **2024**, 62(12), 2425-2434. DOI 10.1515/cclm-2024-0246

[60] S. Saeedi, M. Aghajanzadeh, Investigating the role of artificial intelligence in predicting perceived dysphonia level. *Eur. Arch. Otorhinolaryngol.* **2024**, , 281, 6093-6097. DOI 10.1007/s00405-024-08868-7

[61] E. Kaba, H. M. Bülbül, G. Burakgazi, A. T. Varlık, N. Hürsoy, M. Solak, S. Tabakoğlu, F. B. Çeliker. Large language models on magnetic resonance imaging safety-related questions: Accuracy of ChatGPT-3.5, ChatGPT-4, Gemini, and Perplexity. *Curr. Res. MRI* **2024**, 3, 16-19. DOI 10.5152/CurrResMRI.2024.24095

[62] M. Shukla, I. Goyal, B. Gupta, J. Sharma, A comparative study of ChatGPT, Gemini, and Perplexity. *Int. J. Innov. Res. Comput. Sci. Technol.* **2024**, 12(4), 10-15. DOI 10.55524/ijircst.2024.12.4.2

[63] C. Sirisathitkul, Slow writing with ChatGPT: Turning the hype into a right way forward. *Postdigit. Sci. Educ.* **2024**, 6, 431-438. DOI 10.1007/s42438-023-00441-5

[64] A. Brailas, Postdigital duoethnography: An inquiry into human-artificial intelligence synergies. *Postdigit. Sci. Educ.* **2024**, 6, 486-515. DOI 10.1007/s42438-024-00455-7

[65] F. F.-H. Nah, R. Zheng, J. Cai, K. Siau, L. Chen, Generative AI and ChatGPT: Applications, challenges, and AI-human collaboration, *J. Inform. Technol. Case Appl. Res.* **2023**, 25, 277-304. DOI 10.1080/15228053.2023.2233814

[66] T. Adiguzel, M. H. Kaya, F. K. Cansu, Revolutionizing education with AI: Exploring the transformative potential of ChatGPT. *Contemp. Educ. Technol.* **2023**, 15, ep429. DOI 10.30935/cedtech/13152

[67] H. Crompton, D. Burke, The educational affordances and challenges of ChatGPT: State of the field. *TechTrends* **2024**, 68, 380-392. DOI 10.1007/s11528-024-00939-0

[68] A. O. Ifelebuegu, P. Kulume, P. Cherukut, Chatbots and AI in Education (AIEd) tools: The good, the bad, and the ugly. *J. Appl. Learn. Teach.* **2023**, 6, 332-345. DOI 10.37074/jalt.2023.6.2.29

[69] A. Jobin, M. Ienca, E. Vayena, The global landscape of AI ethics guidelines. *Nature Machine Intell.* **2019**, 1, 389-399. DOI 10.1038/s42256-019-0088-2

[70] T. Petricini, What would Aristotle do? Navigating generative artificial intelligence in higher education. *Postdigit. Sci. Educ.* **2024**, 6, 439-445. DOI 10.1007/s42438-024-00463-7

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Research Article

Institutional Echoes: the *Laboratório Químico Municipal do Porto* (1884-1907) as a Model for the Dynamics of Scientific Institutionalization

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Abstract. This article explores the institutionalization process of scientific organizations, with a focus on the case-study of the *Laboratório Químico Municipal do Porto* (LQMP). Within the context of Institutionalization Theory (IT) and framed by the history of chemistry, the LQMP's lifecycle is examined to propose a new model for the institutionalization of scientific entities. The article reveals the dynamic interplay between historical developments in chemical science, micro-level scientific practices and macro-level societal changes, offering a comprehensive understanding of how scientific institutions influence public policy and societal norms. Through the lens of IT, the article provides a nuanced exploration of the LQMP's inception, development, and closure, shedding light on the broader implications of scientific institutionalization. This research addresses a gap in LQMP's institutional understanding as a historical peripheral landmark in the emergence of science-based legislation. Its findings contribute to the theoretical discourse on the institutionalization of scientific practices, with implications for policy formulation on public health and food regulation.

Keywords: Institutionalization Theory; Scientific Institutions; Public Policy; Laboratório Químico Municipal do Porto; Science-based legislation

INTRODUCTION

Institutionalization is a transformative movement that influences human interactions through the dynamics of rules involving both individuals and organizations.[1] The Institutionalization Theory (IT) provides a comprehensive understanding of how societal norms, along with individual and collective actions, shape the institutionalization process. It emphasizes the importance of analyzing both micro and macro-levels of social events to understand the complexities of institutionalization, highlighting the interplay between individuals and societal structures.

Studying institutionalization is crucial for clarifying the emergence of science-based legislation and the dynamics of science communication. IT helps to address how practices, initially developed within scientific communities, shape social values, policies, and legislative frameworks.[2] By analyzing the role played by various stakeholders, IT explores how their interactions affect the public's understanding of scientific advancements. But IT also studies the establishment of scientific standards, regulations, and norms, clarifying their contribution to building public trust in scientific institutions. Naming the challenges encountered during the institutionalization process helps this theory design strategies to enhance the relationship of scientific research with significant policies. It offers a framework to decipher the intricate relationships in the science-society-policy triad, providing insights into how values, norms, and power dynamics influence the embracement of scientific innovations.

While IT offers a robust theoretical framework, it lacks a specific method to incorporate scientific institutions. This is significant when considering the theory's potential to substantiate the implication of laboratories in shaping new science policy. Although IT explains stakeholders' roles and the evolution of practices within scientific communities,[3] it does not provide a simple method to trace how these laboratories, as institutions, transition from sites of knowledge production to influential policymaking entities. A robust theoretical apparatus can help fill this gap by offering a structured way to laying out the pathways through which scientific practices become embedded within regulatory frameworks. This is especially important for understanding how scientific institutions influence public policy and how, in turn, these institutions are shaped by social and political forces. The model provides a lens to examine how scientific institutions gain legitimacy, influence decision-making, and contribute to broader societal change.

The importance of such model is further amplified when considering scientific institutions located in peripheral or historically marginalized regions. Often, studies on the institutionalization of science focus on central or dominant institutions in major urban or academic hubs, leaving peripheric institutions underexamined. A theoretical model tailored to this process allows for the analysis of case studies from such institutions, offering insights into how they contribute to the broader scientific landscape. As these institutions often face unique barriers in gaining legitimacy, a structured model can highlight how they navigate these challenges and interact with national and international policy. Understanding these dynamics is critical for fostering equitable science policy, as it ensures that contributions from all regions are integrated into broader policy discussions.

This article aims to develop a comprehensive IT model tailored to scientific institutions, using an historic chemistry laboratory as a case study. It hypothesizes that the Portuguese *Laboratório Químico Municipal do Porto* (LQMP), opened in 1884 and closed in 1907, serves as a model to unravel the complex dynamics of institutionalization within scientific practices. This interdisciplinary approach is relevant for a history of chemistry, as it delves into how scientific laboratories shaped legislative frameworks, particularly in areas like public health and food regulation. It posits an essential question: to what extent do the processes and practices cultivated within scientific laboratories influence the development of laws and societal norms? By using the LQMP's historical case study, the article seeks to build a theoretical framework to analyze the broader impact of scientific practices on institutional evolution. This approach not only enriches the understanding of chemistry's role in societal development but also highlights how chemistry laboratories were instrumental in standardizing practices that shaped public policy. This novel exploration is crucial for historians of chemistry, as it provides a detailed account of how laboratory advancements translate into legislative changes.

INSTITUTIONALIZATION THEORY AND SCIENTIFIC PRACTICE

One can define institutionalization as a transformative movement that aims to influence the dynamics of human interactions. Max Weber[4] defined the logic of institutionalization given three regulatory actions: a phenomenon of regulation; adapting existing rules; and replacement by new rules. It is human activity that installs rules and procedures, involving people (individual and collective) and organizations (social or political), making work a set of norms that establish the social contract.[5] Although it is not this research's goal to analyze the context of institutionalization and its historical development, it highlights the dynamics of how practices become shared rules, formalized around all society's actors.

There are several theories to analyze how institutional movements can occur. Amongst them, there is a confluence of opinions on key three conceptualizations.[6] First, (i) a cultural movement, process of constitutive institutionalization that emerges based on the values and social norms which interconnect people and institutions. Building stakeholders' mutual trust is essential for the effective adoption of regulations, key to understand behavior change. Second, (ii) in a movement of rational choice, institutional momentum comes as a pragmatic urgency of the common good, overlapping actors' individual decisions. Perhaps the most cited explanation for the analysis of society policies, this approach allows a rational observation by assuming that the preferences of the individual are 'stable' and their dynamics of interest manifest in society within pre-established rules. Therefore, it must substantiate certain caution when the model falls into the assumption of more dynamic societies. Finally, (iii) a crisis movement explains that institutions emerge and change because of external shocks that disrupt their normal functioning. The shift in the institutional landscape's *status quo* is a response to these changes, understood as a framework for resolving a generalized crisis. Most times, this type of thinking comes from the understanding of radical changes in social regimes, limiting movement explanations to these moments of tension. Such definition illustrates a sophisticated event, which is comprehensible considering the interdependence between the three referenced movements of community-based norms. Therefore, I intend to highlight the positive aspects that an institutional analysis brings to the compression of historiographical movements. Not in comparison to the advances highlighted by political science, but as a contribution to the integration of actors' actions within society.

The goal of the proposed model is to provide, upon a investigation into different ways of interpreting IT, an overarching potential meta-theory[7] of a scientific organization's institutionalization process. This would be a model that monitors change between levels of explanation and recognizes practice diversity. To extend IT's connection to the main quandaries of scientific knowledge, namely the link between macro or micro-level social events, the focus is on the sequence by which organizations gain institutional status. In new institutionalism, such as most theories of social systems, individual actors represent the micro-level and structures uphold individuals' actions portrays the macro-level.[8] As social truth rises from many individual-level actions, a simple combination model lacks sufficient detail to describe the practice heading towards social standing.

Coleman emphasizes the necessity for social theory to delimit between the micro and macro-levels of social events,[9] insisting that understanding system segments dynamics is crucial for this clarification. It is vital to avoid merely amalgamating individual system components, as the interplay between levels is a complex phenomenon that may not always be predictable. Addressing the challenge of capturing the nuanced interrelations between these levels is of pivotal concern for theories focused on institutional activities.[10] This typically involves situating the macro narrative within the context of an emerging organizational field. And this will be the backbone of the proposed model: it incorporates three development stages, starting with a macro-level trigger towards micro-level influences,[11] culminating in a new, unified macro-level framework. The process, initiated by a social trigger and mediated through individual and group actions, leads to the adoption of new practices and the establishment of a cohesive scientific field. Several factors are incorporated for a comprehensive understanding of the institutionalization process, as they include identifying the triggers that drive the process and recognizing the role of agency throughout. It is also crucial to identify the mechanisms that embed institutional norms: analyze the factors influencing the spread of the process, the level of uniformity within the organizational field, and pinpoint the conditions that unify various practices and beliefs.

Applying this model to study scientific institutions also provides a structured approach to examining how scientific practices evolve. From individual actions within a laboratory setting to influential factors shaping societal norms, they help understand the institutionalization of scientific practices as a dynamic between micro-level innovations and macro-level structural shifts.[12] It can be thus understood how scientific institutions contribute to the development of science-based policy. This approach aligns with Coleman's suggestion[13] that 'serendipity' in scientific discoveries can catalyze new theoretical understandings, emphasizing the significance of IT in framing the role of scientific institutions.

THE *LABORATÓRIO QUÍMICO MUNICIPAL DO PORTO* (1884-1907) – AN INSTITUTIONAL CASE-STUDY

At the end of the 20th century, a clear scientific movement emerged in the social sciences on the study of laboratories as institutions. Perhaps the most impactful studies originate in European sociological schools, as Bruno Latour's works provided a new look at the true extent and influence of activity of scientific laboratories.[14] The new conception on the construction of the scientific argument launched a profound debate on the actual weight of laboratories, as institutional social elements. It did not take long for the historiographical debate focused on this theme, as that influence will be evident in this section.[15]

In the development of scientific policies in historical contexts of laboratories as institutions,[16] most of the research deals especially with the period between wars and post-World War II.[17] In line with a great literature impulse in scientific communication, historiography was concerned with the role of these actors as engines of a scientific agenda, uneasy with society's technological literacy and the adaptation of states' perception to the 20th century scientific revolution.[18] This was later recovered by contemporary science historians interested with the process of science and technology development in the 20th century.

The lifecycle of the Laboratório Químico Municipal do Porto, (1884-1907)

The LQMP was a chemical laboratory established in Porto, Portugal, in the late 19th century.[19] Its creation was part of the city's initiative plan to improve public health, urban sanitation, and food safety standards.[20] The laboratory was set up to conduct chemical analyses, primarily focusing on food and water safety to combat adulteration. Under the leadership of notable chemists such as António Joaquim Ferreira da Silva (1853-1923), the LQMP played a crucial role in the city's scientific advancements, contributing to the establishment of standards and regulations in Portuguese food safety and hygiene.[21] It became a model for integrating scientific practices into governance and public policy, reflecting the broader trend of institutionalizing science towards societal needs.

In discussing the genesis and evolution of the *Laboratório*, it's pivotal to delve into two distinct, yet intertwining contexts: the academic and political realms of late 19th century Porto. The growth of the *Academia Politécnica do Porto* ("Polytechnic Academy of Porto", APP) and the ambitious "City Improvement Plan" marked key milestones that drove LQMP's establishment. However, the interconnection between these two catalysts – each with its unique origin – raises intriguing questions about academic advancement in nurturing scientific institutions.

António Ferreira da Silva emerges as a key figure in this narrative, bridging the academic pursuits of the *Academia* and the city's development aspirations outlined in the "Improvement Plan". While the former's influence has been explored,[22] the latter warrants a closer examination. The "Improvement Plan"[23] unveiled by the Municipality of Porto in 1881 championed a vision of progress, aiming to transform the city into a desirable destination for tourists and a European commercial hub. Rooted in the aftermath of the 1876 economic and banking crisis, addressing the perceived isolation of the historic city center[24], the plan proposed an integrated approach to urban renewal beyond architectural rejuvenation. The plan's advocates articulated a straightforward vision: "The only remedy now," as stated in the prologue, "was to progress, and improve in order to excite the desire to be visited, and to offer visitors such comforts". Much like historians of science have argued that peripheral European cities played an essential role in shaping scientific modernity,[25] despite often being perceived as "backward" compared to major scientific centers.

Central to this vision was the imperative of public health and sanitation, considered fundamental for revitalizing the city's economic vigor, aligning with the standards of prominent European centers. The plan's scope encompassed citywide hygiene improvements, extended to individual homes' integration into the public sanitation network, reflecting a holistic approach to urban development. Acknowledging the legislative efforts made abroad to inspect health standards of consumable goods (like meat, fish, milk, wine, and olive oil), the plan scrutinized the "pernicious influence on the public health of foodstuffs". Until that time, Portuguese food regulation was largely a legal matter without a robust legislative scientific foundation.[26] The report recognized the emerging need for "special technicians" trained in detecting food adulteration, highlighting an international trend toward establishing specialized laboratories that offer analyses for individuals concerned about the safety of their food. In this context, the proposal suggested that establishing such a laboratory would be a valuable service to Porto's citizens. By providing scientific expertise in the areas of toxicology, food safety, and sanitation, the LQMP would become central to these efforts, much like its counterparts in cities like Paris, Barcelona and Madrid.[27] By exemplifying how peripheral cities could localize scientific models to address their own urban needs, I argue that LQMP's establishment was not merely a scientific endeavor, but a strategic component of a broader initiative to elevate Porto's stature, blending scientific innovation with urban progress.

For this purpose, in 1882 the municipality of Porto began installing the LQMP. Eminent chemist António Ferreira da Silva is personally invited by the Mayor to direct this process, later becoming its first (and only) Director.[28] But Ferreira da Silva's choice is not only justified by his scientific curriculum. He had previously taken part as an expert on the city's water distribution assessments. His research on how water from surrounding rivers were supplying the city, proved to be important in the city's determination to pursue sustainable sanitation programmatic lines. On November 9th, 1882, a municipal council session approved LQMP's establishment, installing it near the city's Town Hall on *Rua do Laranjal*, a street that no longer exists today (due to the opening of the current *Avenida dos Aliados*). In January 1883, City Council sworn in Ferreira da Silva as LQMP's director, which opened on June 2nd, 1884.

Figure 1. Photograph of the front of the *Laboratório Químico Municipal do Porto* (c. 1900).[29]

The institution was active until 1907, when it was compulsorily closed by the Municipality of Porto. Alves and Alves' research vividly captures the tension between the Porto City Council and the LQMP's director, amidst discussions on the depletion of competencies and the "consequences of political tactics."[30] While the definitive closure of the LQMP became apparent only a decade later in 1917, the laboratory never resumed its activities. This prompted disappointment within the scientific community, with many lamenting the laboratory's disappearance as a significant setback for chemical science. One of the most poignant reflections came from chemist Alberto de Aguiar, who mourned the closure of what he coined as a "sanctuary of chemistry."[31]

International Models: a Blueprint for Porto's LQMP

Before LQMP's establishment, Ferreira da Silva would visit the facilities of a model laboratory, selected among those who "around the world tackle the falsification of foodstuffs": the Municipal Chemistry Laboratory of Paris.[32] This laboratory, established in 1878 and following the well-achieved model of the city of Brussels, aimed to enhance public health and hygiene by monitoring adulterated food products. Initially serving only police requests, it became a pivotal institution in the modernization of urban health systems, setting a precedent for municipal laboratories across Europe. But when under pressure from the city's food merchants, it began "hybrid" operations[33] in 1880, catering to both public and private entities.[34] As such, it was the ideal candidate to emulate a new Municipal laboratory in Europe's periphery.

The influence of the Parisian laboratory on LQMP's establishment in 1884 is particularly significant. Founded to address the challenges posed by urbanization, pollution, and the need for public health standards, the laboratory became a trailblazer in chemical analysis. It set new analytical standards, but it also served as a model for other European cities. It inspired the adoption of similar practices in places like Lisbon and Porto in Portugal, as well as major Spanish cities such as Barcelona and Madrid.[35] Unlike Paris', which restricted private service initially, Porto's laboratory was accessible to the city's populace from the beginning, highlighting its commitment to community engagement. This closeness to society is a defining feature of the laboratory's institutionalization, demonstrating how it was essential in shaping its operational framework and contribution to IT.

The Paris Municipal Laboratory's integration into the fabric of municipal governance set it apart as a model of scientific institutionalization. It was not just a research facility. It was embedded within the city's administrative framework, as Parisian chemists played a central role in investigating public health and food safety risks. This model of governance was appealing to other European cities which were eager to modernize a more scientific approach to urban issues.[36] In Porto, the LQMP was similarly integrated into the municipal structure. This close relationship between the laboratory and municipal governance echoed the Parisian example, showing how science could be institutionalized from a local administration.

The adaptation of the Parisian model involved not only mimicking its structures, but also tailoring the laboratory's functions to the specific Portuguese challenges. Porto was an industrializing city with rising public health challenges, but the local context of wine production, food adulteration, and sanitation standards created unique demands for the LQMP. While the Paris laboratory dealt with a broad spectrum of urban health issues, the LQMP placed particular emphasis on food safety, especially in regulating wine production, central to the regional economy. Wine adulteration posed a significant health risk, and international trade required the enforcement of safety standards to maintain foreign markets' credibility.[37] The Paris laboratory had set a precedent for addressing food adulteration, particularly in detecting harmful compounds, such as sulfur dioxide used in wines. The LQMP adopted similar methods, applying rigorous scientific analysis to ensure that Porto's wine production adhered to local health standards and international trade regulations. This effort to ensure wine safety can be seen in the LQMP's focus in detecting common adulterants like saccharine and sulfites.[38] As noted in the laboratory's activity reports, work on wine adulteration was critical to maintaining public health and the city's economic standing, by following the Paris laboratory's model of chemical analysis.

Beyond Portugal, the Parisian model could also be seen in Spanish laboratories, particularly those in Barcelona and Madrid. Like Porto, these Spanish cities were grappling with the consequences of industrialization and urban growth, which brought about public health crises.[39] Municipal laboratories in Spain developed similar frameworks for food regulation through scientific analysis. For example, the customs laboratory in Madrid and the municipal laboratory in Barcelona took on roles like LQMP in addressing food adulteration and toxic risks in public health. However, the Spanish context differed in that these laboratories often operated under more centralized national frameworks, particularly in customs regulations.[40] The Spanish laboratories focused heavily on the regulation of imported goods, ensuring that food products entering the country met national health standards. This focus on customs was less pronounced in the LQMP, which had a more local emphasis, though the "Lisbon Customs Laboratory" filled this role for Portugal.[41] The LQMP's emphasis on local issues made it distinct from its Spanish counterparts, which were more closely tied to national regulatory frameworks.

Additionally, LQMP was at the forefront of forensic toxicology in Portugal, becoming involved in high-profile legal cases that required scientific expertise in detecting poisons and toxic substances. While this forensic function was already established in Paris and other European cities, it became a hallmark of LQMP's innovative contributions at the local level, ultimately establishing the laboratory as a national reference.

LQMP's institutionalization can thus be traced back to the practices imported from the Municipal Chemistry Laboratory of Paris. This relationship can be categorized though features that were fully (or partially) adopted or not implemented. Concerning adopted features, the LQMP emulated the Paris laboratory's mission of combating commercial fraud and food adulteration, enhancing public health and hygiene. More than just a local scientific entity, the LQMP grew into a recognized national institution, pivotal in the fight against food fraud.[42] Its analyses gained legal credibility, often used as forensic evidence, underscoring its institutional legitimacy.[43] This recognition extended further as the LQMP played a crucial role in standardizing laboratory procedures, illustrating its established institutional stature.[44] This is documented in the amassed reports of the first Portuguese commission to "Standardize Chemical Analysis" for food inspection purposes, where many of the LQMP's procedures are adopted, mainly on wine, olive oils and water analysis.[45]

Under Ferreira da Silva's leadership, the LQMP spearheaded a committee for procedural unification, but also became a beacon of excellence, influencing laboratory practices beyond its walls.[46] This pivotal role, deeply intertwined with the academic community in Porto, signifies the LQMP's impact on the institutionalization of scientific practices, drawing inspiration from, and expanding upon, the foundational model established by the Paris laboratory. However, it is difficult to envision Ferreira da Silva as more than just a brilliant academic without considering the significant contributions he made through his work at the LQMP. There is the possibility that both him and the laboratory have together distilled their legitimacy in the social and academic field. Without this symbiosis, perhaps the conditions for Ferreira da Silva to have chaired the committee for the study and unification of the methods of scientific analysis of wines, olive oils and vinegars in 1895 had not been met.[47]

But LQMP's institutionalization also highlights aspects that were not incorporated. Unlike in the beginning of its Parisian counterpart, as discussed, the LQMP embraced a more inclusive approach, offering its services to both public and private sectors from its inception. Other major difference was the LQMP's initial lack of national legislative certification.[48] This absence is notable given the laboratory's aim to serve as a scientific institution, but it can be explained because of the lack of a national legal framework defining food regulation standards at the time of LQMP's inception. Indeed, it was the LQMP's director that supervised the scientific effort in designing such regulations. The supervisory role and the need for scientific credibility by other power entities were significant challenges during the laboratory's existence, until the consolidation of Portuguese food regulation and public health act between 1902 and 1905.

LQMP's scientific credibility was bolstered by another pivotal moment for its institutionalization: its involvement in the *Urbino de Freitas Case*, 1890-1893, a high-profile alleged poisoning case in Porto. The laboratory's findings were scrutinized in court, especially when contrasted with diverging results from other institutions and international experts.[49] This case exemplified LQMP's struggle to establish its legitimacy and authority in the forensic domain. The case, often regarded as the first major forensic case in Portugal, brought to the forefront the role of scientific evidence in legal proceedings. Urbino de Freitas, a respected doctor, was accused of poisoning his nephews to claim their inheritance. The case became infamous not only for the crime but also for the public debate it sparked around the legitimacy of scientific evidence versus authority-based arguments in court. At the heart of the trial was the forensic analysis conducted on the LQMP led by Ferreira da Silva, who applied advanced analytical chemistry techniques to detect toxic substances in the victims. The defense, however, questioned Ferreira da Silva's findings, leveraging arguments of authority: that more senior scientists and international experts should be trusted over the younger, local chemists involved. But Ferreira da Silva's interventions were crucial in establishing the credibility of LQMP's scientific analysis as forensic evidence. The trial demonstrated the increasing importance of scientific methodologies in legal decision-making, challenging the traditional reliance on authority figures and subjective judgments, marking a shift towards evidence-based legal practices. Having LQMP's analysis decisively impacted the conviction of Urbino de Freitas, it shaped public opinion on laboratory's status as a credible institution in the development of analytical chemistry and its institutionalization of science in legal frameworks. This blending of science and law was less pronounced in other peripheral city laboratories, where the focus was on broader public health. LQMP's evolution reflects its transition from a purely scientific entity, showing how scientific institutions must secure legitimacy within their broader societal environments.

These efforts were halted with the abrupt closure of the LQMP. Highlighted as a personal crusade against its director and not merely an administrative decision, it was a move that rippled the scientific community, municipal governance, and Porto's society.[50] Especially when the *Laboratório* had garnered international recognition for its scientific contributions.[51] In 1907, the Porto City Council closed the facility influenced by a report[52] suggesting the laboratory had strayed from its core mission of analyzing foodstuffs. The report, coupled with the argument that the laboratory's functions were being absorbed by newer institutions (accompanied from the Portuguese food regulation act) provided the official rationale for its closure. However, this surface reasoning obscured deeper layers of academic rivalry, political maneuvering, and economic considerations.[53] Indeed, the laboratory had broadened its scope of work, which included analyses beyond its original remit, but this expansion enabled the laboratory to tackle an array of public health issues beyond food safety.[54] This was not seen as a scientific advantage from the institution, as critics argued it had diverted from its foundational purpose. Despite several protests, the laboratory was shuttered, a move that some perceived as a dismissal of science's value in societal progress. Although Ferreira da Silva challenged the decision, the laboratory's fate had already been sealed, marking the end of an era in Porto's scientific history.

A case for LQMP's institutional analysis

Introducing IT in the context of the LQMP and its significance involves understanding the interplay between micro-level actions and macro-level social structures.[55] The LQMP serves as a pivotal case study illustrating how scientific practices become institutionalized, influencing broader legislative frameworks, culminating in the emergence of food regulation. Initially, the laboratory's creation was a response to a macro-level social need for improved public health standards, particularly on food safety. This need acted as a trigger, starting a process where micro-level actions (specifically, the scientific analyses and methodologies developed by the LQMP) began to influence and shape social structures.

At the micro-level, individual actors within the LQMP, led by figures like Ferreira da Silva, engaged in scientific practices that laid the groundwork for standardized methods of food regulation. These practices contributed to a shift in how foodstuffs were regulated at a broader societal level. The transition from micro-level actions to macro-level institutionalization occurred as the practices and findings of the LQMP gained recognition and were eventually incorporated in chemical standards. The laboratory's work helped to establish a mutual trust between the scientific community, the public, and policymakers, demonstrating the effectiveness and necessity of science-based approaches to food regulation. This trust facilitated the adoption of the LQMP's practices, leading to their integration into legislative frameworks.[56] Furthermore, the LQMP's experience aligns with the crisis movement of institutionalization. Societal shocks, such as public health crises related to food safety, prompted a reevaluation of existing practices and the adoption of new validated approaches. The laboratory's success represented a response to these pressures, leading to a transformation in how food safety was regulated.

THE INSTITUTIONALIZATION THEORY MODEL

In creating the institutionalization process within the context of the LQMP, I propose the following framework based on IT towards scientific laboratories. This model emphasizes the centrality of cognitive and innovation processes in the scientific laboratories' institutional development. It highlights the importance of legitimation, routinization, and the appropriation of these practices in establishing norms to fit new contexts. In parallel, adaptability within the scientific field is compatible with institutional pressures, reflecting the ongoing need for scientific interpretation and public understanding of science. This intends to be a foundation for empirical research aimed at describing the specific causal relationships in the process of institutionalization in scientific laboratories. In this proposal, the original IT model is split into three complementary macro-level phases, as depicted in Figure 2. Each phase has its own micro level environment, which is now introduced.

Figure 2. Macro-level description of the proposed IT model.

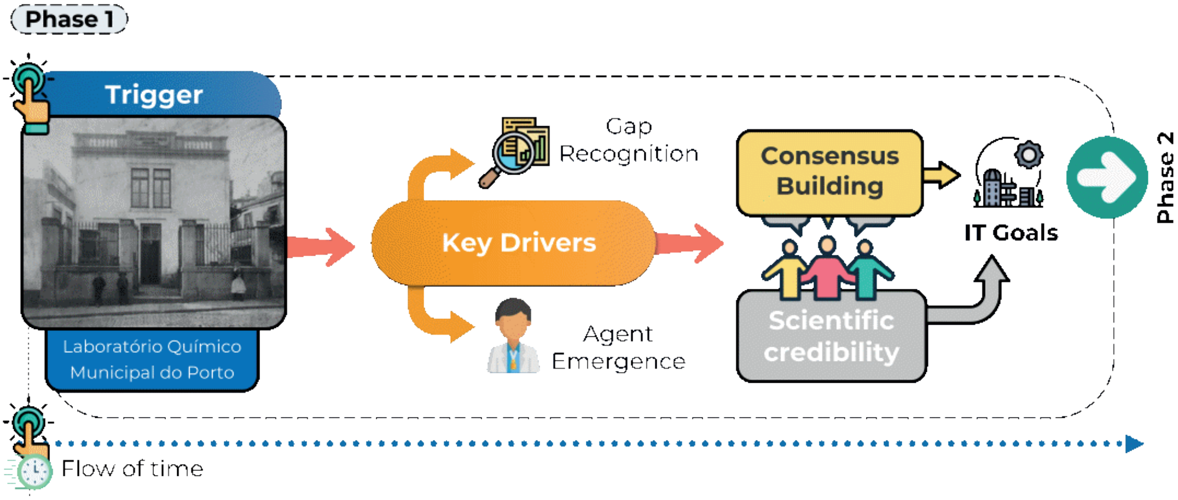

Figure 3. Micro-level description of the proposed Phase One of the IT model.

Phase One – Trigger: launch of scientific laboratories' institutionalization

This phase begins with the recognition of the material and socio-cognitive conditions within laboratories that call for institutionalization – dubbed as "the trigger". These could be, for example, breakthrough discoveries, paradigm shifts, policy changes or crises in scientific credibility. Next, the key agents (or drivers) that influence institutionalization initiation must be identified. Influential scientists at the helm of scientific institutions could play a pivotal role as well as funding bodies or regulatory agencies. There should be an explicit gap recognition that institutionalization drivers aim to address. It might involve advancing scientific knowledge, enhancing scientific credibility, or addressing societal concerns. And finally, initial consensus building aggregates previous findings by establishing a direction for the institutionalization process. Even if not fully detailed, there should be an early agreement on what the institutionalization aims to achieve within the laboratory context. Figure 3 represents the micro-level structure of Phase 1.

Phase Two – Legitimation: structuring institutional practices

In the second phase, a novel method, technology, or organizational structure emerges as a response to the "trigger". Various trends can influence the acceptance of new practices within the scientific community, influenced by its structure. Institutionalization occurs when these new methodologies become a central part of the laboratories' *modus operandi*, establishing through habits and incorporating formalities. This phase is crucial for process materialization, where abstract concepts turn into tangible practices. It starts with the development of new practices, involving the introduction of novel methodologies (technologies or processes) in response to the identified needs. Next, these new practices are integrated into existing laboratory frameworks. Such incorporation requires adaptation, where practices change or become tailored to fit the unique contexts of different laboratories. The concept of agency, where individuals or groups within laboratories take action to implement new practices, often signals the emergence of these innovations. This can result from leadership driving change, collaborative efforts, or institutional mandates.

But the goal of these new practices is also to gain legitimacy within the scientific community. Demon-

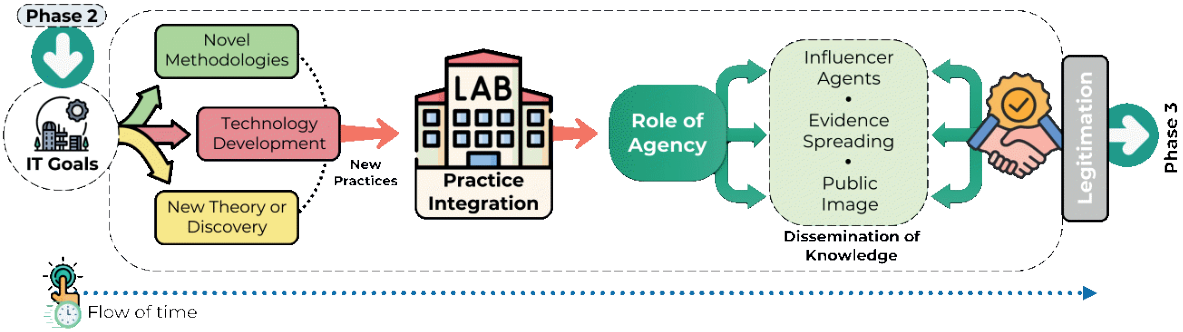

Figure 4. Micro level description of the proposed Phase Two of the IT model.

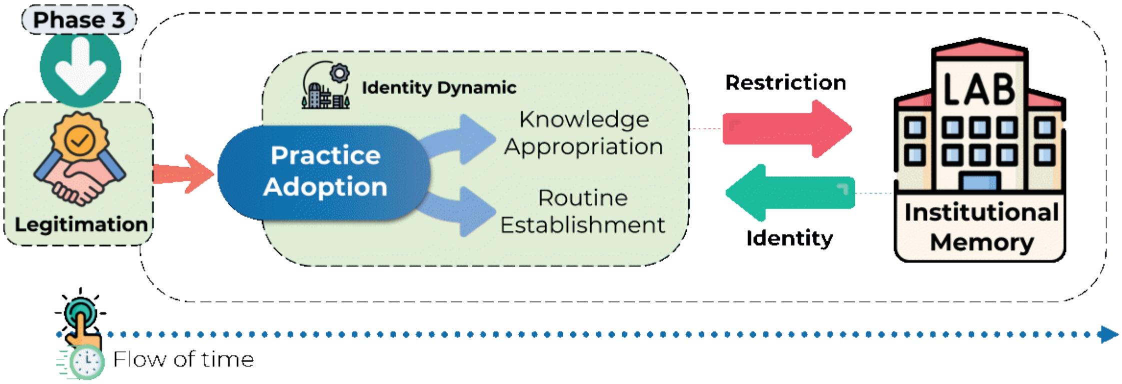

Figure 5. Micro level description of the proposed Phase Three of the IT model.

strating reliability with scientific values can be achieved through successful new practices' routinization and endorsements from influential figures. Through practice consistent monitoring, this feedback is crucial for refining and solidifying the institutionalization process. Thus, Phase Two ensures that new practices are developed, integrated, and legitimized within the scientific laboratories, paving the way for their transition to Phase Three. Figure 4 represents the micro-level structure of Phase Two.

Phase Three – Memory: diffusion and homogenization of scientific laboratories

The last phase depicts the establishment of new practices. It signifies the maturation of institutionalization, with the eventual appropriation of their norms or standards, adapted to new regulation contexts for policymakers.

Practice adoption is a hallmark of successful institutionalization, showing that they have gained acceptance within the scientific community. Once they become part of a routine, they are no longer seen as new or optional but essential components of scientific work. They are further embedded in the institutional memory of the community, preserved through documentation and transmission of knowledge. In shaping future research beyond cultural integration movements, they now become part of collective knowledge. Evaluating the long-term impact of these institutionalized practices by identifying cultural movements can either expand or restrict their development. This evaluation helps understand the consequences of the institutionalization process. Figure 5 represents the micro-level structure of Phase Three.

INSTITUTIONAL ANALYSIS OF THE *LABORATÓRIO QUÍMICO MUNICIPAL DO PORTO*, (1884-1907)

LQMP's Phase One

By examining the LQMP through the lens of Phase One, its inception aligns with the theoretical framework that emphasizes the importance of socio-cognitive conditions, key drivers and consensus building in the early stages of institutionalization. The laboratory appears in response to Porto's public health concerns, particularly related to food adulteration and water quality.[57] This idea materialized in Porto's urban improvement plan, which highlighted the role of science and technology in advancing the city's welfare, establishing a scientific institution dedicated to addressing these public health issues.

The approval of Porto's urban improvement plan, which included the establishment of the LQMP, was the trigger event in this context. The LQMP was a response to a recognized need for scientific approaches to address food safety and public health, reflecting a socio-cognitive shift towards trust in science for societal improvement. It's focus on bromatology, public health and chemistry directly addressed the city's concerns, demonstrating a clear link between the institution's needs and objectives.[58] However, the renewal of the APP cannot be ignored in this context. Only through structural reforms in higher education that the critical mass needed to staff this laboratory could be generated.[59]

Porto's challenges with food adulteration, which threatened public health, led to the establishment of the LQMP to address food safety through scientific analysis. This move was also a reflection of a broader trend, aligning Porto with other European cities that had already recognized the importance of scientific laboratories in enhancing urban health standards.[60] The LQMP was envisioned as a pivotal institution that would not only bridge the existing gaps in public health management, but also as a beacon of scientific advancement within the city.

Initial consensus around the creation of the LQMP involved support from various stakeholders, including the city council, the academic community, and the public.[61] By positioning the LQMP at this intersection, stakeholders collectively acknowledged the laboratory's role in fostering a safer urban environment. Such consensus was crucial in ensuring the laboratory's successful integration into the city's public health strategy, solidifying the commitment from various sectors to use the LQMP's services. Also, the laboratory's alignment with the city's improvement plan and its support from prominent figures like Ferreira da Silva would have facilitated this consensus-building process.[62] With his expertise and reputation, Ferreira da Silva played a pivotal role in the laboratory's establishment. His international recognition and scientific acumen provided the credibility and authority to drive the LQMP's creation and its subsequent scientific endeavors.

LQMP's Phase Two

In Phase Two of institutionalization, the focus shifts towards the development, integration, and legitimation of new practices within the institution. This involves the operationalization of its scientific practices within the framework of Porto's public health agenda.

Upon its establishment, the LQMP embarked on developing and refining scientific practices, particularly in analytical chemistry. These practices align with the theoretical aspect of Phase Two, where an institution operationalizes its foundational objectives. The laboratory pioneered methods for detecting food adulteration, a practice that was not only novel for Porto's context but also set a new standard beyond it. Methodologies were developed in response to relevant societal quarrels, depicted in two major events: the *Salicilagem dos Vinhos do Porto*, (1885-1905) (to be discussed further) and the *Caso Urbino de Freitas,* (1890-1893).[63] Such practice integration into existing laboratory frameworks required a dynamic adaptation process, where traditional methods were reconfigured to accommodate the innovative approaches introduced by the LQMP. Therefore, the municipal laboratory eventually collaborated with other national services to enhance public health.[64] This integration showcases the laboratory's fitness within the broader municipal context, reflecting IT's emphasis on practice integration within the existing framework.

Under the leadership of Ferreira da Silva, the LQMP showed agency in advancing the field of food safety and public health policies.[65] Along with the collective efforts of the laboratory staff, his action ensured that the new practices were not only developed but also effectively adopted and integrated into the laboratory's routine operations. His role in research, analysis, and public engagement illustrates the concept of agency in institutionalization, where his scientific contributions in addressing public health concerns, helped legitimize practices both within the scientific community and the public.[66] The laboratory's international recognition underscore its legitimation, a key aspect of Phase Two where the new practices gain acceptance.

As described in its statutes, the LQMP performed bromatological and food analyses at the service of the population. Its financing, however, came from complementary analytical work outside the LQMP's main area of operation. In this context, attention is drawn to the analysis that Alves and Alves made about LQMP's activity in the period 1884-1906:

> *According to the Regulation, the analyses requested by individuals or other authorities would be repaid according to an accessible tariff: 500 réis for any qualitative analysis; 2250 réis for quantitative analyses of water, vinegar, sugar, coffee and cheese; 4500 réis of beer, wine, milk, chocolate, flour, butter and bread; According to the objective of its creation, the Laboratory developed its action mainly in the analysis of food substances and their counterfeits, because between 1884-1906, of the 10487 samples counted, 7726 were related to food examinations.*[67]

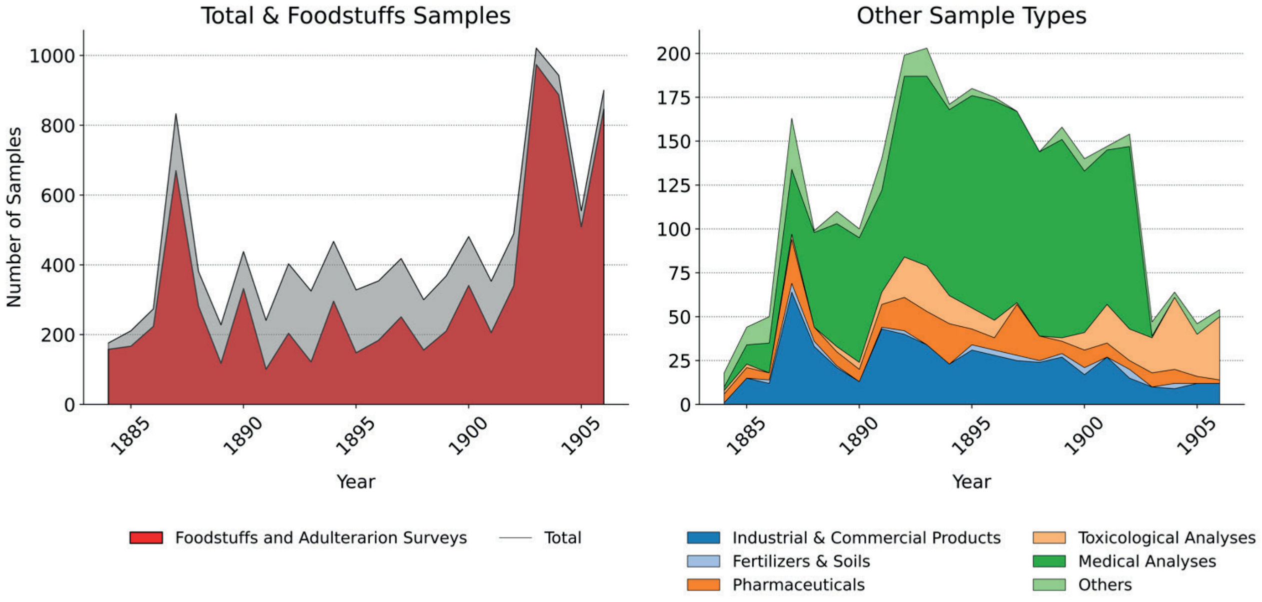

Figure 6. Graph distribution of samples for analysis, submitted to the LQMP from 1884 to 1906, divided by internal category classification. Data extracted from Ferreira da Silva, A Questão do Laboratório Municipal de Chimica do Porto, pp. 66 & 68.

In that period, the LQMP provided society with an analytical inspection service, executed by experienced chemists, on diverse items like wine, bread, milk or butter (and other reports of coffees and vinegars). Simultaneously, the only foodstuffs whose analyses are centrally regulated are wines, olive oils and vinegars. Not only has the Municipality of Porto founded a specific laboratory institution to analyze foodstuffs, but it has also adopted systematic, reproducible methods, making them freely available on the market. Here, its early action fulfilled the city's "Improvement Plan", positioning it as a key player in the emerging food security standards seen across Europe. The laboratory's role as an agent of change, spreading evidence-based practices, helped legitimize its efforts despite the apparent legislative delays from the Portuguese government.[68] To account for this, Figure 6 depicts the sample types analyzed in early years by the LQMP.

Figure 6 shows a significant increase in the number of samples analyzed, especially in the early years, followed by sustained high levels of activity. The peak around the late 1880s shows a heightened public concern about food adulteration during this period, leading to more frequent analysis requests. This represents Phase Two's period of consolidation and expansion of the LQMP's role in food safety, marked by the systematic application of analytical chemistry to inspect foodstuffs. Also, Figure 6's right chart shows a diversification of sample types beyond food, especially after 1890. The large green section corresponds to medical analyses, which became a significant part of the LQMP's work, as this diversification reflects the broader role the laboratory played in municipal health efforts. The increase in non-food-related analyses like medical, industrial and toxicological testing, particularly in the 1890s, shows how the laboratory adapted to additional needs while maintaining its focus on food safety.

On food analysis, the largest part of the LQMP's activities (shown by the dominant red area in the left chart), reflects the core mission of the laboratory during Phase Two: to protect public health by identifying fraudulent practices in food production. The graph demonstrates that the lab responded to the growing awareness and concern regarding food adulteration, a crucial public health issue at the time. As the laboratory's infrastructure and methods matured, more samples were processed, and reproducible methods became standard. This increased testing volume can also be linked to improved institutional practices. According to the regulatory framework, the LQMP offered its services at accessible tariffs, making it easier for individuals to request tests. Reasonable fees likely contributed to the increasing number of samples for analysis as these practices became more established. To achieve this, the LQMP had adequate resources, including a specialized library and state-of-the-art equipment.[69] The increased testing volume can also be linked to these resources, as routinization of successful practices at the LQMP involved transforming experiments to standard procedures.

As elements of consolidation, we can mention the three most relevant. First, the LQMP's role in the legislative sphere in preparing official laboratory documents, in the figure of its director, confers a relevant status for the science carried out in this institution.[70] Second, society recognizes the public image depicting the laboratory's actions. The decisive role in the analysis of high-profile cases conferred a reputation that would lead to the credibility of the science established in the laboratory. Key example of this is the case of *Salicilagem dos Vinhos do Porto* that rose in the late 19th century when Brazilian authorities banned the import of Port wines, citing the presence of salicylic acid, a preservative used to extend shelf life. This threatened a key export market for Port wine, prompting Porto's merchants to ask the LQMP, under Ferreira da Silva, to investigate. Research revealed that the traces of salicylic acid in Porto wine were in fact false positives, not the result of adulteration. By providing scientific evidence, he contested the Brazilian claims and demonstrated that regulatory decisions should account for the quantitative presence of substances, evolving from the binary qualitative system. This scientific intervention not only protected a critical commercial relationship but also underscored the importance of analytical chemistry in distinguishing natural from artificial substances. The LQMP's work influenced the development of more nuanced food safety standards, moving towards decisions based on empirical evidence, setting a precedent for the use of science in resolving trade disputes and shaping future regulations.

Finally, this consolidation is driven by the promotion of scientific education, by being the editorial sponsor of the first Portuguese chemistry journal: the *Revista de Química Pura e Aplicada* ("Journal of Pure and Applied Chemistry") in 1905.[71] This publication consolidated the laboratory in the academy by bridging science popularization between chemists and laypeople in early 20th century. In fact, this publication was later adopted by the *Sociedade Portuguesa de Química* (the "Portuguese Chemistry Society") as its official bulletin, a tradition that still lives up to this day.[72] It focused on reporting news from European science academies, spreading word on new methods and legislation of interest to chemists. It also engaged in communicating LQMP's findings, as emphasis on food safety analysis in noteworthy in its first years of publication.[73]

Phase Two culminates with LQMP's legitimization, embedding a multilayered phenomenon that stresses the laboratory's establishment as a credible entity. First (i), the LQMP's commitment to scientific rigor played a fundamental role. By adhering to stringent scientific methods and producing reliable, reproducible results, the laboratory established a reputation for trustworthiness. This scientific rigor ensured that the methodologies developed by the LQMP were dependable. The systematic implementation of food safety measures, reflected in the sharp rise in food sample testing between 1885 and 1905, also marked the laboratory's ability to regularly analyze essential foodstuffs, providing reliable, science-based data on food quality. Furthermore, the laboratory expanded its scope to include medical and industrial analyses, contributing to a more holistic public health approach.

Second (ii), the recognition that followed further amplified the LQMP's legitimacy. On public engagement, the laboratory built a strong rapport with the community it served. Examples of this legitimacy are prompted by its repeated reports in contesting incorrect accusations of adulteration from Porto's food merchants. Ferreira da Silva amassed interesting accounts on various analyses made in "his" laboratory, that disputed some specialists' claims of food adulterated products. He goes as far as claiming that "some authorities' analysis and conclusions lacked scientific rigor". This incompatibility amongst reports is the by-product of a bifurcated Portuguese regulatory apparatus, headed by two different institutions, which had some dissimilarities.[74] Such actions built a reputation for the LQMP's as a "defender" of the "small Porto's merchants", not by partisanship, but by conferring specific foodstuff criteria instead of broad, often blind, ungeneralizable parameters.[75]

Third (iii), collaborations and endorsements from key figures within the scientific community also played a crucial role in legitimizing the LQMP. These not only expanded the laboratory's influence but also served as a testament to its standing in the scientific domain, such as in establishing the Commission for food analysis standards. It showed the practical applicability and relevance of the LQMP's work, as its legitimation is a testament to the multifaceted nature of gaining credibility in the scientific and societal spheres.

LQMP's Phase Three

In Phase Three of the institutionalization process, focus shifts to the embedding of new practices established in Phase Two, leading to their widespread adoption. For the LQMP, this phase would involve the acceptance of its scientific practices to Porto's public health context and beyond.[76] As described previously, practices developed by the LQMP would become standard procedures. Not

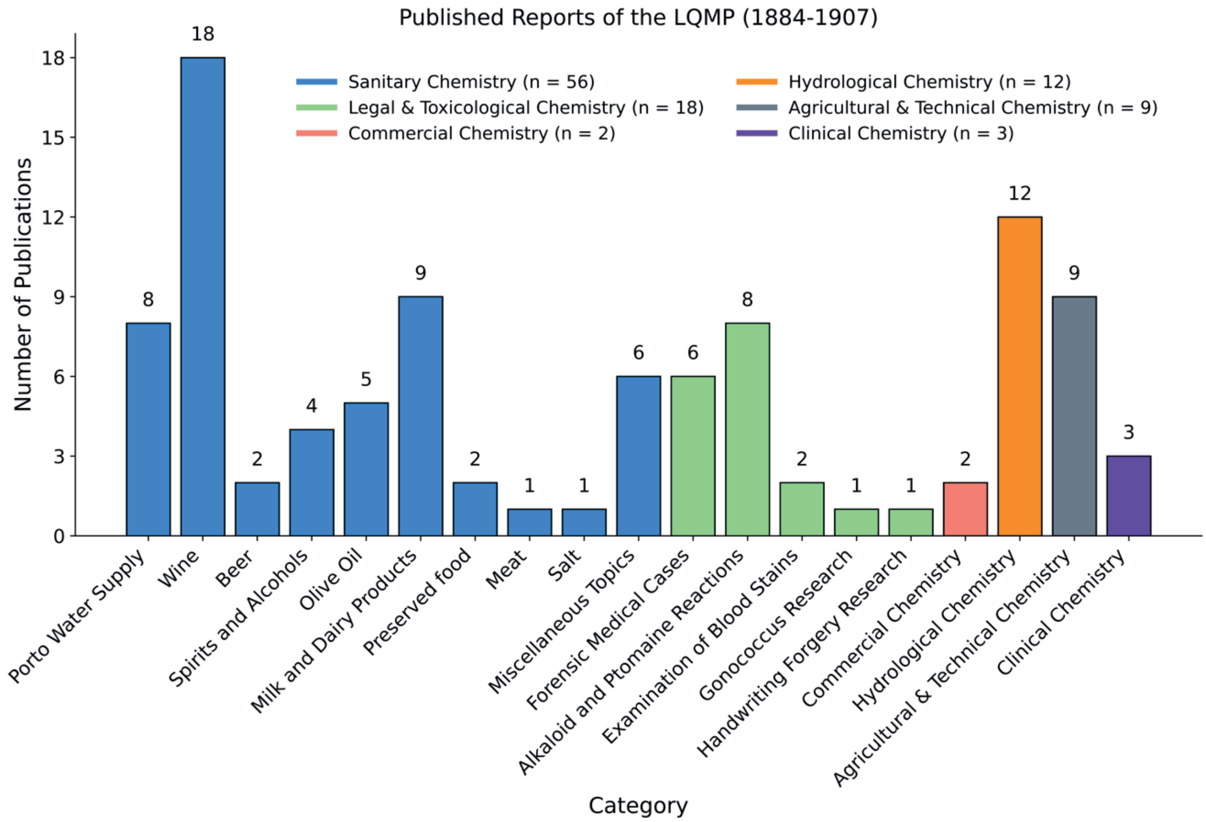

Figure 7. Main works carried out at the LQMP between 1884 and 1907, that were subject to publication, divided by internal category classification. Information extracted from Ferreira da Silva, A Questão do Laboratório Municipal de Chimica do Porto, Section 1: "Main Works Carried out at the Laboratório Chimico Municipal do Porto", pp. 1-34.

just within Porto, but in the country, reflecting a broader acceptance, a hallmark of Phase Three, showing that the institution's practices have become embedded.

The *Laboratório's* analytical methods and interventions would become routine components of Porto's public health infrastructure, no longer viewed as novel, but as standard practice. This routinization signifies that the institution's practices became institutionalized, part of the everyday fabric of the city's efforts. Moreover, the LQMP's eventual designation as a photometric post further attests to the confidence placed in the institution's public service. Values, beliefs, and practices promoted by the LQMP would permeate its status, reflecting a deeper integration of the laboratory's scientific ethos into the city's approach to public service,[77] as this cultural integration is a key aspect of Phase Three.

The LQMP played a role in the development of the food regulation act in Portugal, demonstrating a significant incorporation of institutional memory.[78] The laboratory's pioneering work in combating food adulteration laid the groundwork, serving as a crucial repository of knowledge that perceived the legislation. Figure 7 illustrates this amassed expertise with the broad range of published reports by the LQMP from 1884 to 1907, showcasing laboratory's diverse scientific contributions.

Figure 7 reflects how the institution's work on sanitary chemistry, particularly in areas such as the analysis of wine, water supply, and preserved food, became integral to shaping public health policy. The overwhelming focus on food-related reports, especially in wine (18 reports) and water supply (8 reports), underscores the laboratory's commitment to addressing key issues to Porto's economic and health landscape. This highlights the LQMP's continuous effort to tackle adulteration in this sector, offering vital data that influenced regulatory measures. It's work on milk and dairy products (5 reports) and spirits and alcohols (4 reports) further illustrates how its expertise extended across various food categories, each of which was prone to adulteration in late 19th and early 20th century Portugal.

This systematic approach to analyzing foodstuffs provided a solid foundation for the regulatory frame-

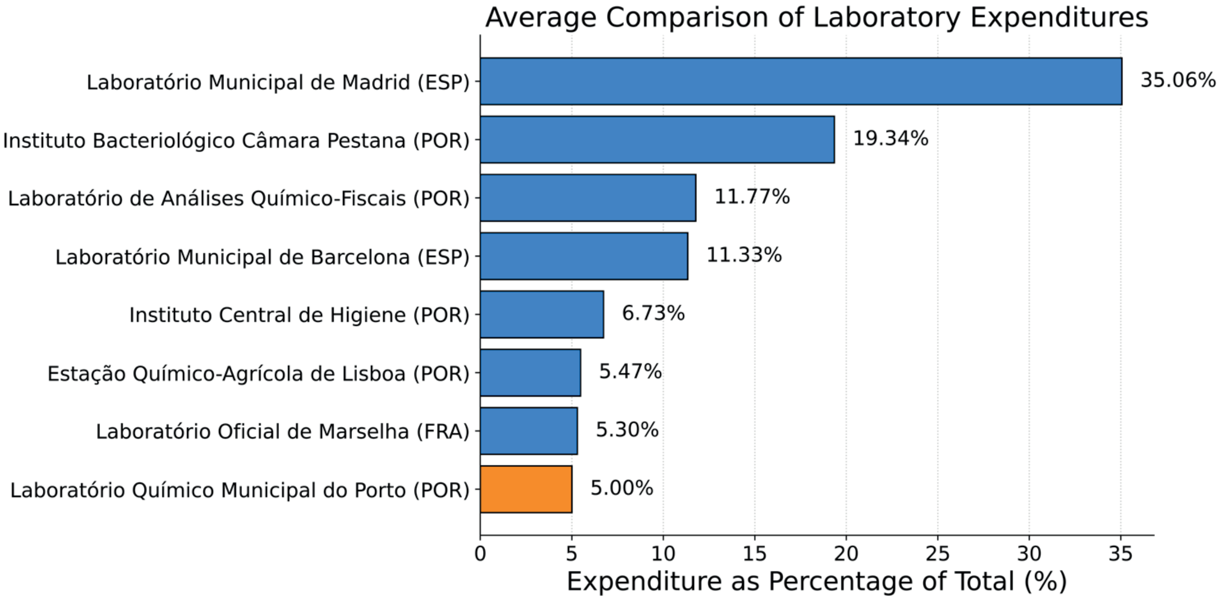

Figure 8. Comparison of estimated yearly expenditure of several Portuguese, Spanish and French laboratories in the period 1899-1907, displayed as percentage of total amassed expenditure. Data extracted from Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto*, pp. 100-101. The data for the LQMP and the Estação Químico-Agrícola de Lisboa report to the median average expenditure between 1899 and 1901. The remaining figures account for the years 1906 and 1907

work. By contributing its institutional memory to the legislative process, the LQMP ensured that the food regulation act was not only reflective of the latest scientific understanding[79] but also tailored to the specific challenges identified through the laboratory's research. It is noteworthy how much of the foodstuffs analyzed are related to Porto's economic fabric, essential to both the local economy and the national food supply.[80] This integration of institutional memory into the legislative process exemplifies how the LQMP's accumulated knowledge was not confined within its walls, ensuring that the laboratory's contributions have a lasting impact. Indeed the reports related to legal and toxicological chemistry (18 reports) and hydrological chemistry (12 reports) reveal how the LQMP's expertise contributed not only to food safety but also to forensic regulations, playing a multifaceted role in public policies.

While Phase Three emphasizes practice embedding, it allows room for feedback and evolution. The *Laboratório's* methodologies could be subject to continuous refinement and adaptation in response to new challenges, ensuring their relevance and efficacy. In the third moment of analysis, let us consider the elements of identity and restriction in the addressed timeline (1884-1907). The lack of clear legislation on issues closely tied to laboratory work, such as teaching and practice, reflects weak oversight at the central level. However, the renewal of higher and technical education in Porto supplied scientific resources beyond teaching. Laboratories at the Polytechnic Academy and the Industrial Institute were commonly recognized as "accredited" for food analysis. The decentralization of resources along with other economic factors lead to an "emptying" of LQMP competencies with implementing the new food regulation act.[81] During this period, legislation regarding the analysis of goods at Porto's customs named the "Chemical-Fiscal Laboratories" to empirically assess substances deemed dangerous. With regulatory branches now accredited under the new act, these developments mark moments of restriction for LQMP's operations.[82]

On LQMP's limitations, the installation of new central institutions was a determining factor. It created new entities that ensured, under national scrutiny, tasks within the competence of the LQMP.[83] At the beginning of the 20th century, Portuguese legislation included "first-line" laboratories in charge of performing food and economic analysis. Most of these under the tutelage of the *Ministério do Reino* that, until this time, had been little mentioned in the legislation for official purposes. This dynamic here describe is further highlighted by the data showed by Figure 8.

Figure 8 compares laboratory expenditures across several European institutions that had overlapping

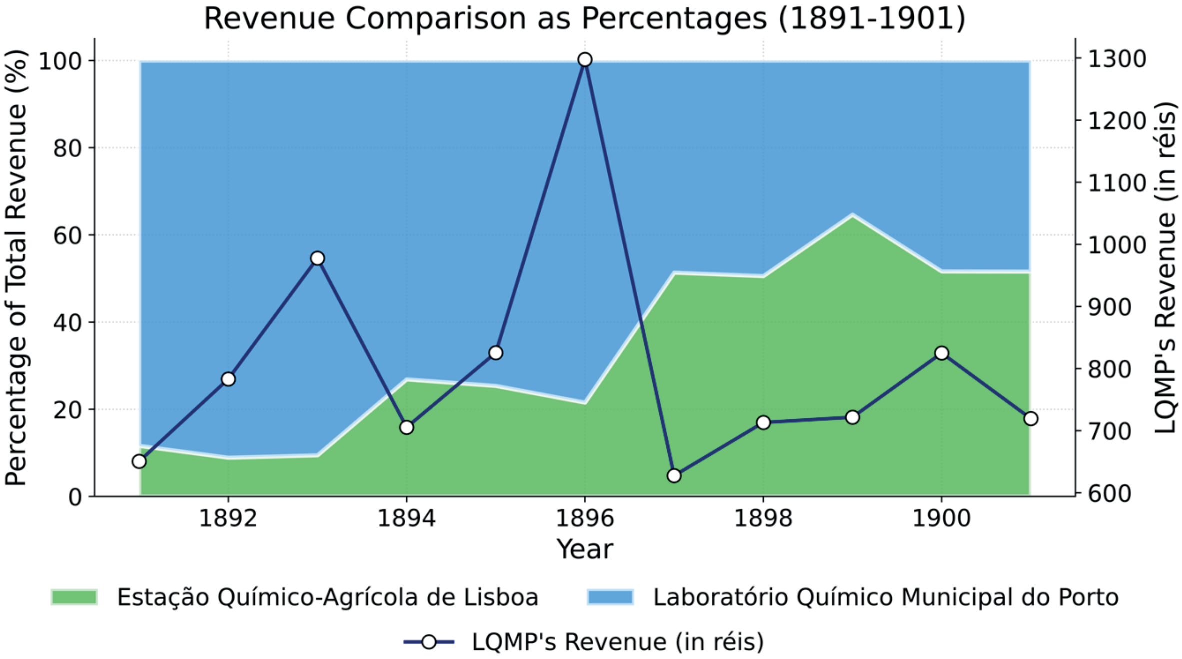

Figure 9. Comparison of percentage of revenue of the LQMP and the *Estação Químico-Agrícola de Lisboa* in the period 1891-1901. The blue line represents LQMP's numeric revenue (secondary yy axis). Data extracted from Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto*, pp. 72.

directives. In the context of the LQMP, it shows a relatively modest percentage of total expenditures (5.00 %) compared to institutions like the Municipal Laboratory of Madrid (*Laboratório Municipal de Madrid*, 35.06 %) or the *Instituto Bacteriológico Câmara Pestana* (19.34%) could illustrate the shift in resources and centralization of public health responsibilities. The increased expenditures in other Portuguese laboratories may reflect the centralization of laboratory functions under national authority, which diminished the LQMP's prominence and redirected resources to newly centralized entities.

Building on this, Figure 9 further emphasizes the LQMP's moments of restriction by comparing the revenue generated by the LQMP and the *Estação Químico-Agrícola de Lisboa*[84] between 1891 and 1901. The figure visually illustrates the economic decline of the LQMP as new national-sponsored laboratories, like the *Estação*, began to dominate the analytical landscape in Portugal. As shown in the graph, the LQMP's revenue, indicated by the blue line, fluctuated significantly over the years, reflecting its increasingly marginal role in the country's public infrastructure.

Meanwhile, the steady rise of the *Estação's* revenue, particularly after 1895, coincides with the "centralized regulation of analytical methods and the creation of "first-line" laboratories", which were granted priority by the *Ministério do Reino*. This marks a shift where more resources were allocated to central institutions, which were now seen as the authority for conducting chemical analysis. The percentage comparison highlights that Lisbon's laboratory progressively gained a greater share of total revenue, consolidating its role as a player in national regulatory functions. This demonstrates how the decentralization of responsibilities away from the LQMP was accompanied by a corresponding shift in financial resources, leading to a reduction in the LQMP's influence. It complements the vision of restriction and identity loss during Phase Three, where Porto's laboratory increasingly became a peripheral institution as Portugal established more centralized institutions.

Concurringly, a key element of restriction is the unification of analytical methods decreed by Portugal from 1895, to harmonize scientific standards and practices. This element must be regarded not only as a restriction, but also of the identity of the LQMP itself. For example, the fact that its director, Ferreira da Silva, was called to chair the official committee for the elaboration of legislation on scientific practice, should be seen as an element that confers identity and purpose to the Laboratory of Porto.[85] For these reasons, it is clear that these sources of restriction/identity may have a certain social (and also political) value, which largely depends on their use by public opinion. The ease with which these factors can be generators of a climate of conflict between academic peers, political guardianship, and benefactors, can be strong enough to generate moments of instability. It may be the genesis of a new "trigger" that can initiate a one-way restriction process, whose sources may not be sufficient to address.

Besides the unification of analytical methods, other equally important sources of identity must be considered. The absence of a previous clarifying status on "who" and "how" is authorized to carry out analyses for supervisory purposes, led to the laboratory taking for itself a status of "specialist". With the projection of the research performed by Ferreira da Silva at the LQMP, especially in international scientific journals[86], the scientific contributions emerging from Porto gained significant recognition. And finally, the foundation of the *Revista de Química Pura e Aplicada*, considered by peers as a reference in the scientific Portuguese landscape, can be viewed as a product of the laboratories' identity.

But from all these events, the abrupt closure of the LQMP in 1907 stands as the pivotal incident that highlights the complexities of institutionalization in its Third Phase. The laboratory had firmly established itself as a key player in food regulation, yet political and economic pressures led to its shutdown. Despite its widespread contributions to society, the LQMP's fate illustrates how institutionalized scientific practices can be fragile when confronted with conflicting priorities, resource limitations, and shifting institutional identities.

The decision to close the LQMP was primarily justified by accusations of deviation from its original mission. According to the city council's report, the laboratory had "transformed into a school" and was conducting too many analyses outside its core purpose of food safety.[87] The council noted that, by 1906, the LQMP had conducted 900 analyses, but only 483 were food related. The rest involved substances such as "gasoline, sulfur, and medico-legal analyses like bloodstains and human viscera". This diversification of services was a departure from its original mandate to safeguard public health through food safety analysis. Additionally, the council highlighted the laboratory's "financial inefficiency", stating that it had been running at a loss. With annual expenses exceeding *5600 réis* and minimal revenue from paid analyses, the council argued the LQMP was no longer economically sustainable (as seen in Figure 9's decrease in LQMP's revenue). They also pointed to the availability of other governmental laboratories that could perform similar functions, making the LQMP redundant. Ultimately, the council asserted that "the laboratory had fulfilled its mission" and that its continued operation was unnecessary in these institutional concerns.

In response, the LQMP's director launched a powerful defense, positioning the laboratory as a key contributor to scientific knowledge in Portugal.[88] Ferreira da Silva argued that the laboratory's role extended far beyond mere food analysis. He emphasized that "the laboratory studied old and new water supplies," helping to secure drinking water purity. He stressed the LQMP had contributed to national and international scientific developments. One notable example was the laboratory's work on the salicylation of Port wines, which averted a major trade dispute with Brazil. This defense shows that, while the laboratory may have expanded beyond its original purpose, it had done so in ways that enhanced its scientific reputation, aligning with the Third Phase of institutionalization where institutions adapt and expand to remain relevant.

The closure of the LQMP illustrates the restrictions that institutions often face during the third stage of institutionalization. As scientific institutions expand their influence, they often encounter financial and political constraints that limit their ability to sustain operations. The laboratory's growing influence and expansion into other scientific realms, such as toxicology and industrial analyses, ran counter to the council's more restricted view of the LQMP's mandate. This tension between institutional growth and political restriction is a common feature of the third phase of institutionalization. While the LQMP had developed a broader identity, its expanding role came at a cost that the local government was unwilling to support. The political decision showcases the limitations that institutions often face in balancing their growing scope with the political realities of the environment in which they operate.

The debates over the LQMP's closure also highlight the shifting identity of the laboratory. Initially established as a municipal institution focused on public health, the LQMP had evolved into a multifaceted scientific organization conducting a wide range of analyses. This expansion was partly driven by the laboratory's growing increasing demands placed upon it by both the local government and the scientific community. However, this evolving institutional identity also contributed to its downfall. The city council's argument that the LQMP had strayed too far from its original purpose reflects the challenges that institutions face in the Third Phase of institutionalization. As the LQMP expanded its functions, it began to occupy a more ambiguous space between its original mandate and its new role as a scientific research institution. The council's decision to close the laboratory can be seen as an attempt to rein in this expanding identity and return the focus of municipal resources to more clearly defined public functions.

One of the key elements of the Third Phase of institutionalization is the preservation of institutional memory. In defending the laboratory, Ferreira da Silva underscored the LQMP's longstanding contributions to Porto's public health infrastructure and its role in shaping national food safety standards. He pointed out that the laboratory had been officially recognized by the Portuguese government in 1904 as an "official establishment for the analysis of foodstuffs" to assist in regulating food safety. This acknowledgment by the national government reinforced the LQMP's institutional memory as a critical player in safeguarding public health. The decision to close the LQMP, despite these historical contributions, reflects a break in the institutional memory of scientific practices. Its closure signaled the loss of this accumulated knowledge and expertise, a key aspect of de-institutionalization where institutions lose their historical continuity. Ensuing legal battles highlight how deeply the LQMP had become embedded in the social fabric of Porto and how its loss was felt as a blow to both the city and the scientific community. This serves as a case study of how scientific institutions, despite their contributions, must continually navigate the tensions between their expanding roles and the restrictive forces of politics to maintain their institutional legacy.

Influence of institutionalization in adopted legislation

The institutionalization dynamic exemplified by the LQMP plays a pivotal role in the emergence of food regulation. As the first Portuguese laboratory critically specialized in analyzing foodstuffs, the LQMP not only set a precedent for scientific intervention in Portugal's public health, but also laid the foundational framework for integrating scientific expertise into regulatory practices. By developing methodologies for detecting adulteration and contamination, the laboratory provided a scientific basis for establishing standards into legislation. This transition from *ad hoc* inspection to standardized, scientifically validated procedures is crucial for effective food regulation. Its work showed how scientific insights could directly influence policy, ensuring that regulations were not just descriptive but also based on demonstrable risks and interventions.

The *Laboratório's* integration into Porto's public health system highlighted the importance of institutional collaboration for effective food safety governance. By linking scientific research with its role in ensuring food safety, it helped build public trust in both the scientific community and the government's regulatory efforts.[89] It also served an educational role, raising awareness among producers, consumers, and regulators about the importance of food safety and the scientific principles underlying it.

The LQMP's contributions to the food regulation legislation in Portugal are further exemplified by the data on published reports and revenue trends from the laboratory during the institutionalization period. Figure 7, which categorizes the LQMP's reports between 1884 and 1907, underscores the institution's extensive research output in critical areas of food safety, instrumental in providing scientific data that directly influenced legislative frameworks. The focus on sanitary chemistry (56 reports) and legal and toxicological chemistry (18 reports) shows that the LQMP was not only addressing immediate public health concerns but also shaping the scientific basis for broader regulatory policies in Portugal.

Moreover, Figure 9, which compares revenue between the LQMP and the *Estação Químico-Agrícola de Lisboa*, reflects how the economic landscape shifted as regulatory responsibilities became centralized. The LQMP's fluctuating revenue throughout the 1890s and early 1900s can be interpreted because of this centralization, which gradually transferred authority to Lisbon's institutions. The combination of these elements – the LQMP's scientific output and the shifts in its financial stability – reinforces the argument that the laboratory's institutionalization was not only foundational to the development of food regulation but also reflective of broader political dynamics that shaped public governance. As the LQMP laid the groundwork for legislative action, its eventual decline and closure illustrate the complex relationship between scientific institutions, policy-making, and economic centralization, all of which played crucial roles in shaping Portugal's food regulation landscape.

The LQMP's closure and its interaction with the broader themes of institutionalization provide a nuanced understanding of the challenges in embedding scientific practice within public policy. Its establishment in 1884 under Ferreira da Silva's leadership was an embodiment of institutionalization in its early phase, where an identified need (food adulteration), was addressed by a new institution. However, the controversial decision to close the LMQP in 1907, illustrates the challenges inherent in the third phase of institutionalization – diffusion and homogenization. The laboratory's closure amidst political, economic, and institutional pressures showcases the fragile nature of institutional permanence and the influence of external factors on the sustainability of institutionalized practices. The legal battles and public outcry following the decision highlight the societal recognition of the LMQP's value.[90] This resistance against the laboratory's closure can be seen as a societal pushback against de-institutionalization, emphasizing the laboratory's integration within the local and the scientific community.

FINAL REMARKS

This article intended a comprehensive analysis of the LQMP within the framework of IT, providing a nuanced understanding of how scientific institutions undergo the process of institutionalization. Impacting public policy and societal norms, the LQMP's inception, operations, and eventual discontinuation encapsulate a spectrum of institutionalization dynamics.

The emergence of the LQMP was not an isolated event. It was a response to the burgeoning needs of a rapidly industrializing Porto, grappling with the challenges of public health and food safety. This response was emblematic of the First Phase of institutionalization, where a societal need catalyzes the formation of an institution. The laboratory's establishment under the guidance of Ferreira da Silva was a pivotal moment, marking the city's commitment to leveraging scientific expertise to address pressing public health issues. The LQMP was envisioned as a bridge between scientific inquiry and public welfare, tasked with the critical mission of ensuring food safety and public health through rigorous chemical analysis, as highlighted in its second phase.

But the institutionalization journey of the LQMP was not linear. The laboratory's closure in 1907, prompted by a combination of political, economic, and institutional factors, underscores the challenges inherent in the diffusion and homogenization phase of institutionalization. As hallmarks of its third phase of institutionalization, along with the public and scientific community's reaction, the LQMP's closure reflects the deep-rooted impact of the laboratory on Porto's societal and scientific landscape.

The detailed examination of the LQMP through a proposed multi-layered model of institutionalization – initiation, structuring, and homogenization – provides a rich narrative that encapsulates the role of scientific institutions in societal advancement. The model helps tracking LQMP's journey throughout its lifecycle, offering insights into the dynamics of institutionalization and on how scientific practices are influenced by societal and policy frameworks. This detailed analysis not only contributes to the understanding of the LQMP's historical significance, but also offers a broader replicable model to understand other laboratories on their intricate institutional interplay between science, society, and institutionalization.

LIST OF BIBLIOGRAPHY

Ana Simões, "From Capital City to Scientific Capital: Science, Technology, and Medicine in Lisbon as Seen through the Press, 1900–1910," in *Urban Histories of Science*, eds. Oliver Hochadel and Agustí Nieto-Galan (New York: Routledge, 2018), pp. 141-163.

Andreas W. Daum, "Varieties of Popular Science and the Transformations of Public Knowledge: Some Historical Reflections," *Isis* 100 (2009): 319-332.

António M. Basto, *Memória Histórica da Academia Politécnica do Porto* (Porto: Universidade do Porto, 1987).

Bruno Latour, *Laboratory Life: The Construction of Scientific Facts* (Princeton University Press, 1979).

Bruno Latour, *Science in Action: How to Follow Scientists and Engineers through Society* (Open University Press, 1987).

Christine Oliver, "Strategic Responses to Institutional Processes," *The Academy of Management Review* 16, no. 1 (1991): 145-179

Christoph Stadtfeld, "The Micro–Macro Link in Social Networks," *Emerging Trends in the Social and Behavioral Sciences* (2018): 1-15,

Graeme Gooday, "Placing or Replacing the Laboratory in The History of Science?" *Isis* 9 (2008): 783-795.

I. Suay-Matallana, "Customs laboratories, chemistry and excise: An historical introduction," *World Customs Organization News* 77 (2015): 34-37.

I. Suay-Matallana, "The material culture of the customs laboratory of Lisbon," *Conservar Património* 30 (2019): 131–139.

I. Suay-Matallana and X. Guillem-Llobat, "Poisoned Wine: Regulation, Chemical Analyses, and Spanish-French Trade in the 1930s," *Ambix* 65, no. 2 (2018): 99-121.

I. Suay-Matallana, "The Customs Laboratory of Lisbon from the 1880s to the 1930s: Chemistry, trade and scientific spaces," in *Science, Technology and Medicine in the Making of Lisbon 1840–1940*, eds. Ana Simões and Maria Paula Diogo (Leiden: Brill, 2022), pp. 179–202.

J. R. Bertomeu-Sánchez, "Chemistry, microscopy and smell: bloodstains and nineteenth-century legal medicine," *Annals of Science* 72, no. 4 (2015): 490-516.

James A. Secord, "Knowledge in Transit," *Isis* 94 (2004): 654-672.

James Coleman, *Foundations of Social Theory* (Cambridge: Harvard University Press, 1990), namely in the interconnection between micro and macro levels.

Jorge Alves and Rita Alves, "Ferreira da Silva e o Laboratório Químico Municipal do Porto (1884-1917)," *Estudos do Século XX* 12 (2012): 14-30.

Jorge Alves and Rita Alves, *Nos Caminhos da Química, A. J. Ferreira da Silva* (U. Porto Edições, 2013).

José Ferraz-Caetano and Bruno D. A. Pinheiro, "Legislators' Plague: How History of Science Can Explain the Struggles of Universal Pandemic Responses," in

Handbook of Research on Historical Pandemic Analysis and the Social Implications of COVID-19, eds. Antonio Cortijo Ocaña and Vicent Martines (Hershey, PA (USA): IGI Global, 2021), pp. 47-59.

José Ferraz-Caetano, "As raízes da regulação alimentar em Portugal: leis e práticas baseadas em ciência, 1875-1905," *Ler História* 82 (2023): 171-194.

"O caso da "salicilagem dos vinhos do Porto": ecos de visão social, económica e científica das relações Portugal-Brasil (1885-1905)" *Revista Brasileira de História da Ciência* 15, no. 1 (2022): 108-119.

José Ferraz-Caetano, *As Artes Químicas na Academia do Porto e o Surgimento da Regulação Alimentar em Portugal (1875-1905).* (Coimbra: University of Coimbra, Master's thesis, 2021).

José Ferraz-Caetano, João Paiva, and Francisco M. Romeiras, "António Ferreira da Silva and the Teaching of Chemistry at the Academia Politécnica do Porto (1877-1910)," *História da Ciência e Ensino: construindo interfaces* 20 (2019): 213-221.

M.W. Bauer, "The Evolution of Public Understanding of Science - Discourse and Comparative Evidence," *Science, Technology and Society* 14, no. 2 (2009): 221-240.

Maria Fernanda Rollo, Tiago Brandão, and Maria Inês Queiroz, "Revising the Institutionalization of Science Policies: Historical Contexts and Competing Models," *Portuguese Journal of Social Science* 17, no. 1 (Mar 2018): 37-61.

Max Weber, *Economy and Society: An Outline of Interpretive Sociology* (University of California Press, 1978).

Oliver Hochadel and Agustí Nieto-Galan, "How to Tell the Tale," in *Urban Histories of Science*, eds. Oliver Hochadel and Agustí Nieto-Galan (New York: Routledge, 2018), pp. 1-15.

P.S. Tolbert and L.G. Zucker, "The Institutionalization of Institutional Theory," in S. Clegg, C. Hardy, and W. Nord (Eds.), *Handbook of Organization Studies* (London: SAGE, 1996).

Paulo Augusto, "Teoria Institucional: Qual o Lugar da Agência," presented at the 30º Encontro da Associação Nacional dos Programas de Pós-graduação em Administração, Rio de Janeiro, 2007 (ANPAD, 2007).

Peter J. Atkins and A. Stanziani, "From Laboratory Expertise to Litigation: The Municipal Laboratory of Paris and the Inland Revenue Laboratory in London, 1870-1914: A Comparative Analysis," in *Fields of Expertise: A Comparative History of Expert Procedures in Paris and London, 1600 to Present,* (Cambridge Scholars Press, 2008), pp. 317-338.

R. J. Dinis-Oliveira, "Portugal's First Major Forensic Case and the Genesis of Forensic Toxicology: 10 Years of Research to Reconstruct the Event," *Forensic Sciences Research* 4, no. 1 (2019): 69-81.

Richard Harvey Brown, "Modern Science: Institutionalization of Knowledge and Rationalization of Power," *The Sociological Quarterly* 34, no. 1 (1993): 153-168.

Richard Whittington, "Putting Giddens into Action: Social Systems and Managerial Agency," *Journal of Management Studies* 29, no. 6 (1992).

Stephen Barley and Pamela Tolbert, "Institutionalization and Structuration: Studying the Links between Action and Institution," Organization Studies 18, no. 1 (1997).

T. Felin and N. Foss, "Microfoundations for Institutional Theory?" in P. Haack, J. Sieweke, and L. Wessel (Eds.), *Microfoundations of Institutions*, Research in the Sociology of Organizations, Vol. 65B (Emerald Publishing Limited, Leeds, 2019), pp. 393-408

Tanja Kuchenmüller, Laura Boeira, Sandy Oliver, Kaelan Moat, Fadi El-Jardali, Jorge Barreto, and John Lavis, "Domains and Processes for Institutionalizing Evidence-Informed Health Policy-Making: A Critical Interpretive Synthesis," *Health Res Policy Sys* 20, 27 (2022).

Walter W. Powell and Patricia Bromley, "New Institutionalism in the Analysis of Complex Organizations," in *International Encyclopedia of the Social & Behavioral Sciences,* Second Edition, ed. J.D. Wright (Elsevier, 2015), pp. 764-769.

Zuoyue Wang and Naomi Oreskes, "History of Science and American Science Policy," *Focus - Isis* 99 (2008): 365-373.

LIST OF SOURCES

António Ferreira da Silva, *A Fiscalisação Dos Generos Alimenticios No Porto: (Vinhos)*, Papelaria La Bécarre, Porto, 1903.

António Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto, (1907-1908)*, s.n., Porto, 1909.

António Ferreira da Silva, *Documentos sobre os trabalhos de química aplicada à higiene do Laboratório Municipal de Química do Porto*, Imprensa Portugueza, Porto, 1904.

António Ferreira da Silva, *Os Vinhos Portuguezes Genuínos Condenados como Falsificados: Notas Sobre os Trabalhos a Que Deu Origem a Questão dos Vinhos Suppostos Salicylados no Brasil com a Apreciação do Snr. H. Pellet e Vários Documentos*, Off. do Comércio do Porto, Porto, 1902.

António Ferreira da Silva, *Recursos e Peritos na Fiscalisação Sanitaria no Porto em 1903: (O Reverso da Medalha)*, Imprensa Portugueza, Porto, 1905.

António Ferreira da Silva, *Relatórios do Laboratório Municipal de Chimica do Porto no período de 1884 a 1896*, Typ. A. da Fonseca Vasconcellos, 1897.

António Ferreira da Silva, *Documentos sobre os trabalhos de química aplicada à higiene do Laboratório Municipal de Química do Porto: 1884-1906*, s.n., 1910.

António La-Grange, *Audiências de julgamento do Dr. Urbino de Freitas*, A.J. de Sousa, Porto, 1893.

Amando Seabra *et al.*, *Documentos Scientificos da Commissão Technica dos Methodos Chimico-Analyticos* (Coimbra: Imprensa da Universidade, 1910).

Arquivo Digital da Universidade do Porto, Fundo APP, 1837-1911, Livros de cópia dos relatórios químico-legais do Laboratório Químico Municipal do Porto (1900-1907), available at https://repositorio-tematico.up.pt/handle/10405/47964, last accessed on [20/03/2024].

Augusto de Aguiar, "Dr. António Joaquim Ferreira da Silva," *Revista de Química Pura e Aplicada*, 1924, 16, 7-11. 11-16. Diário do Governo, nos. 284, 24, 268, 50, 99, 90 (dates: 14 de dezembro de 1895, 7 de outubro de 1901, 26 de novembro de 1902, 4 de março de 1902, 6 de maio de 1903, 25 de abril 1904).

Diário do Governo, no. 284 (14 de dezembro de 1895).

Diário do Governo, no. 24 (7 de outubro de 1901).

Diário do Governo, no. 268 (26 de novembro de 1902).

Diário do Governo, no. 50 (4 de março de 1902).

Diário do Governo, no. 99, (06 maio de 1903).

Diário do Governo, no. 90 (25 de abril 1904).

Direção Geral dos Serviços Agrícolas, *Methodos Officiaes para as Analyses dos Vinhos, Vinagres e Azeites: Seguidos da Indicção Summaria dos Processos Empregados nos Documents sur les falsifications des matières alimentaires et sur les travaux du Laboratoire municipal: Deuxième rapport*, République Française, Préfecture de Police (Paris: G. Masson, Librairie de l'Académie de Médecine, 1885).

Laboratorios do Estado para o Exame do Leite e Lacticinios; Alcooes e Aguardentes, Typ. da Encyclopedia Portugueza, Lisboa, 1910.

Henri Pellet, *O Ácido Salicylico e a Questão dos Vinhos Portuguezes no Brazil em 1900*, Imprensa da Universidade de Coimbra, 1906.

O Laboratório Chimico Municipal do Porto: Relatório da Commissão de Inquérito, Aprovado em Sessão de 31-XII-1907, Typ. Emp. Guedes, Porto, 1909.

Plano de Melhoramentos da Cidade do Porto apresentado à Câmara Municipal em sessão extraordinária de 26 de setembro de 1881, Typ. José da Silva Teixeira, Porto, 1881.

NOTES

1 A broad introduction to institutionalization theory is given by P.S. Tolbert and L.G. Zucker, "The Institutionalization of Institutional Theory," in S. Clegg, C. Hardy, and W. Nord (Eds.), *Handbook of Organization Studies* (London: SAGE, 1996), pp. 175-190.

2 In the same framework where Kuchenmüller argued for the case of health-related policies: Tanja Kuchenmüller, Laura Boeira, Sandy Oliver, Kaelan Moat, Fadi El-Jardali, Jorge Barreto, and John Lavis, "Domains and Processes for Institutionalizing Evidence-Informed Health Policy-Making: A Critical Interpretive Synthesis," *Health Res Policy Sys* 20, 27 (2022).

3 As in the seminal example of institutionalization of knowledge in Richard Harvey Brown, "Modern Science: Institutionalization of Knowledge and Rationalization of Power," *The Sociological Quarterly* 34, no. 1 (1993): 153-168.

4 Namely in the sections on "legitimate domination" in Max Weber, *Economy and Society: An Outline of Interpretive Sociology* (University of California Press, 1978).

5 The exploration of Weber's initial theory is expanded in Christine Oliver, "Strategic Responses to Institutional Processes," *The Academy of Management Review* 16, no. 1 (1991): 145-179 and Stephen Barley and Pamela Tolbert, "Institutionalization and Structuration: Studying the Links between Action and Institution," Organization Studies 18, no. 1 (1997). We will use these references in framing the concept of IT into scientific institutions.

6 The basis for the threefold conceptual vison is first introduced in Richard Whittington, "Putting Giddens into Action: Social Systems and Managerial Agency," *Journal of Management Studies* 29, no. 6 (1992) and it is later operationalized with an example in Paulo Augusto, "Teoria Institucional: Qual o Lugar da Agência," presented at the 30º Encontro da Associação Nacional dos Programas de Pós-graduação em Administração, Rio de Janeiro, 2007 (ANPAD, 2007).

7 The proposed model will follow closely the article by James Coleman, *Foundations of Social Theory* (Cambridge: Harvard University Press, 1990), namely in the interconnection between micro and macro levels.

8 As defined in Walter W. Powell and Patricia Bromley, "New Institutionalism in the Analysis of Complex Organizations," in *International Encyclopedia of the Social & Behavioral Sciences,* Second Edition, ed. J.D. Wright (Elsevier, 2015), pp. 764-769.

9 Coleman, *Foundations of Social Theory.*

10 The book chapter T. Felin and N. Foss, “Microfoundations for Institutional Theory?” in P. Haack, J. Sieweke, and L. Wessel (Eds.), *Microfoundations of Institutions*, Research in the Sociology of Organizations, Vol. 65B (Emerald Publishing Limited, Leeds, 2019), pp. 393-408 gives a detailed description on how level dynamics can change how institutionalization is perceived for a particular organization.

11 In Christoph Stadtfeld, “The Micro–Macro Link in Social Networks,” *Emerging Trends in the Social and Behavioral Sciences* (2018): 1-15, this is explored between agents in institutionalization.

12 One similar example in the Portuguese context that poses a similar proposition was presented by Maria Fernanda Rollo, Tiago Brandão, and Maria Inês Queiroz, “Revising the Institutionalization of Science Policies: Historical Contexts and Competing Models,” *Portuguese Journal of Social Science* 17, no. 1 (Mar 2018): 37-61.

13 Coleman, *Foundations of Social Theory*.

14 For a general understanding of Bruno Latour’s works: Bruno Latour, *Science in Action: How to Follow Scientists and Engineers through Society* (Open University Press, 1987); and Bruno Latour, *Laboratory Life: The Construction of Scientific Facts* (Princeton University Press, 1979).

15 In historiographic debate upon this subject see especially Graeme Gooday, “Placing or Replacing The Laboratory in The History of Science?” *Isis* 9 (2008): 783-795.

16 On the circulation of scientific knowledge, James A. Secord, “Knowledge in Transit,” *Isis* 94 (2004): 654-672; Andreas W. Daum, “Varieties of Popular Science and the Transformations of Public Knowledge: Some Historical Reflections,” *Isis* 100 (2009): 319-332.

17 In the field of science communication, M.W. Bauer, “The Evolution of Public Understanding of Science - Discourse and Comparative Evidence,” *Science, Technology and Society* 14, no. 2 (2009): 221-240.

18 Zuoyue Wang and Naomi Oreskes, “History of Science and American Science Policy,” *Focus - Isis* 99 (2008): 365-373.

19 For a descriptive history of the laboratory, see Jorge Alves and Rita Alves, “Ferreira da Silva e o Laboratório Químico Municipal do Porto (1884-1917),” *Estudos do Século XX* 12 (2012): 14-30.

20 The Porto City Council, recognizing the central value of chemistry in the city’s development, highlights its importance in the *Plano de Melhoramentos da Cidade do Porto apresentado à Câmara Municipal em sessão extraordinária de 26 de setembro de 1881* (Porto: Typ. José da Silva Teixeira, 1881).

21 The LQMP’s food safety contributions are documented in José Ferraz-Caetano, “As raízes da regulação alimentar em Portugal: leis e práticas baseadas em ciência, 1875-1905,” *Ler História* 82 (2023): 171-194.

22 Ferreira da Silva’s role in the consolidation of Portuguese chemistry teaching and in the emergence of a new discipline, Analytical Chemistry, is explored in José Ferraz-Caetano, João Paiva, and Francisco M. Romeiras, “António Ferreira da Silva and the Teaching of Chemistry at the Academia Politécnica do Porto (1877-1910),” *História da Ciência e Ensino: construindo interfaces* 20 (2019): 213-221. For a wider view on the APP’s educational context, see António M. Basto, *Memória Histórica da Academia Politécnica do Porto* (Porto: Universidade do Porto, 1987).

23 *Plano de Melhoramentos da Cidade do Porto,* (1881).

24 A description of Porto’s social and economic landscape at the end of the 19th century can be found in Alves and Alves, “Ferreira da Silva e o Laboratório Químico.”

25 This is argued by depicting Lisbon as a case study in Ana Simões, “From Capital City to Scientific Capital: Science, Technology, and Medicine in Lisbon as Seen through the Press, 1900–1910,” in *Urban Histories of Science*, eds. Oliver Hochadel and Agustí Nieto-Galan (New York: Routledge, 1st ed., 2018), pp. 141-163.

26 The early inception of Portuguese food regulation is described in detail in the section “Origens da regulação alimentar portuguesa” in Ferraz-Caetano, “As raízes da regulação alimentar em Portugal.”

27 The dichotomy with the Iberian cases beyond food safety is reported by J. R. Bertomeu-Sánchez, “Chemistry, microscopy and smell: bloodstains and nineteenth-century legal medicine,” *Annals of Science* 72, no. 4 (2015): 490-516.

28 Jorge Alves and Rita Alves, *Nos Caminhos da Química, A. J. Ferreira da Silva* (U. Porto Edições, 2013), pp. 177-214.

29 Unknown author, *A entrada do Laboratório Químico Municipal, no n.º 41 da Rua do Laranjal,* c. 1900, MHNC-UP Archive.

30 The ensuing political tensions were personalized in Ferreira da Silva, whose “fervent catholic faith” clashed with the rising republican movements in early 20th century Portugal. Alves and Alves, “Ferreira da Silva e o Laboratório Químico”.

31 Augusto de Aguiar, “Dr. António Joaquim Ferreira da Silva,” *Revista de Química Pura e Aplicada* 16 (1924): 7-11.

32 Alves and Alves, *Nos Caminhos da Química*, pp. 131-176.

33 As a basis for the description of the Laboratory,

see P.J. Atkins and A. Stanziani, "From Laboratory Expertise to Litigation: The Municipal Laboratory of Paris and the Inland Revenue Laboratory in London, 1870-1914: A Comparative Analysis," in *Fields of Expertise: A Comparative History of Expert Procedures in Paris and London, 1600 to Present,* (Cambridge Scholars Press, 2008), pp. 317-338.

34 A practice that the LQMP would adopt more extensively.

35 Suay-Matallana has extensively written on this impact on Spanish institutions, namely customs laboratories in I. Suay-Matallana, "Customs laboratories, chemistry and excise: An historical introduction," *World Customs Organization News* 77 (2015): 34-37.

36 Oliver Hochadel and Agustí Nieto-Galan, "How to Tell the Tale," in *Urban Histories of Science*, eds. Oliver Hochadel and Agustí Nieto-Galan (New York: Routledge, 1st ed., 2018), pp. 1-15.

37 Reports on the Paris Laboratory activities are depicted in *Documents sur les falsifications des matières alimentaires et sur les travaux du Laboratoire municipal: Deuxième rapport*, République Française, Préfecture de Police (Paris: G. Masson, Librairie de l'Académie de Médecine, 1885).

38 Trade relationships with countries like Brazil, which had stricter regulations on sulfites and preservatives in imported wines.

39 I. Suay-Matallana and X. Guillem-Llobat, "Poisoned Wine: Regulation, Chemical Analyses, and Spanish-French Trade in the 1930s," *Ambix* 65, no. 2 (2018): 99-121.

40 A piece on customs laboratories can be found on I. Suay-Matallana, "Customs Laboratories, Chemistry and Excise: An Historical Introduction," *World Customs Organization News,* 77 (2015): 34-37.

41 I. Suay-Matallana, "The Customs Laboratory of Lisbon from the 1880s to the 1930s: Chemistry, Trade and Scientific Spaces," in *Science, Technology and Medicine in the Making of Lisbon 1840–1940*, eds. Ana Simões and Maria Paula Diogo (Leiden: Brill, 2022), pp. 179–202.

42 Both following contributions are well documented in the article of Alves and Alves, "Ferreira da Silva e o Laboratório Químico."

43 The key official legislative recognition of the laboratory's expertise is shown in *Diário do Governo*, no. 90 (25 de abril 1904), pp. 163.

44 There are three sources that document this. Mainly, both cited works of Ferreira da Silva are extremely detailed on the LQMP's activities, including technical details and collaboration reports. A. Ferreira da Silva, *Relatórios do Laboratório Municipal de Chimica do Porto no período de 1884 a 1896* (Typ. A. da Fonseca Vasconcellos, 1897) and A. Ferreira da Silva, *Documentos sobre os trabalhos de química aplicada à higiene do Laboratório Municipal de Química do Porto: 1884-1906* ([S.l.: s.n.], 1910).

45 On Seabra's book, that lists the Commission's activities, we find multiple citations of Ferreira da Silva's collaborators that devise these methods. Moreover, there are reports of a chemist, Hugo Mastbaum, who quotes practices imported from the Paris Municipal Laboratory, later changed (or partially changed) by the LQMP. Amando Seabra et al., *Documentos Scientificos da Commissão Technica dos Methodos Chimico-Analyticos* (Coimbra: Imprensa da Universidade, 1910). A key example is shown in the technical notes for cheese analysis.

46 The laboratory's influence in Portuguese food regulation implementation is seen in Ferraz-Caetano, "As raízes da regulação alimentar em Portugal."

47 This is strongly supported by the composition of the committee, where many of Ferreira da Silva's collaborators (and students) in the LQMP are nominated. See *Diário do Governo*, no. 284 (14 de dezembro de 1895).

48 An argument for the reasons behind this can be found in section 2 of the article Ferraz-Caetano, "As raízes da regulação alimentar em Portugal.", while a formal certification of LQMP's works is found in *Diário do Governo*, no. 90 (25 de abril 1904), pp. 163.

49 To understand the impact of Ferreira da Silva and the LQMP in resolving this case, it is essential to read the article by R.J. Dinis-Oliveira, "Portugal's First Major Forensic Case and the Genesis of Forensic Toxicology: 10 Years of Research to Reconstruct the Event," *Forensic Sciences Research* 4, no. 1 (2019): 69-81 and consultation of the court hearings of the trial in the compilation of António La-Grange, *Audiências de julgamento do Dr. Urbino de Freitas* (Porto: A.J. de Sousa, 1893).

50 Based on the City Council's report that sustained shutting down the LQMP in *O Laboratório Chimico Municipal do Porto: Relatório da Commissão de Inquerito, Aprovado em Sessão de 31-XII-1907* (Porto: Typ. Emp. Guedes, 1909) and its aftermath on Section 6 of Alves and Alves, "Ferreira da Silva e o Laboratório Químico."

51 Mostly due to Ferreira da Silva's work on the famous cases of the "'Salycilation" (see José Ferraz-Caetano, "O caso da "salicilagem dos vinhos do Porto": ecos de visão social, económica e científica das relações Portugal-Brasil (1885-1905)" *Revista Brasileira de História da Ciência* 15, no. 1 (2022): 108-119) and

"Urbino de Freitas" (Dinis-Oliveira, "Portugal's First Major Forensic Case").

52 *O Laboratório Chimico Municipal do Porto: Relatório da Commissão de Inquerito, Aprovado em Sessão de 31-XII-1907* (Porto: Typ. Emp. Guedes, 1909).

53 Section 6 of Alves and Alves, "Ferreira da Silva e o Laboratório Químico".

54 Ferreira da Silva's public defense is amassed in his book A. J. Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto, (1907-1908)* (Porto: [s.n.], 1909). It also includes echoes from peers and external scientific institutions.

55 Following the example of Stadtfeld, "The Micro–Macro Link in Social Networks".

56 The legislative directives that marked this integration are in *Diário do Governo*, no. 268 (26 de novembro de 1902) and *Diário do Governo*, no. 50 (4 de março de 1902).

57 As explained within the *Plano de Melhoramentos da Cidade do Porto* and Alves and Alves, "Ferreira da Silva e o Laboratório Químico".

58 Beyond the historical description in the previous section, the record of the laboratory's analysis found in the recent online repository, cross-reference these assertions with LQMP's activities. Arquivo Digital da Universidade do Porto, Fundo Academia Politécnica do Porto, 1837-1911, Livros de cópia dos relatórios químico-legais do Laboratório Químico Municipal do Porto (1900-1907), available at https://repositorio-tematico.up.pt/handle/10405/47964.

59 The impact is described in detail on Chapter 3 of the following dissertation: José Ferraz-Caetano, *As Artes Químicas na Academia do Porto e o Surgimento da Regulação Alimentar em Portugal (1875-1905).* (Coimbra: University of Coimbra, Master's thesis, 2021).

60 Porto's alignment with other major European city on Public Health was significantly increased with the outbreak of Bubonic Plague in early 20th century: José Ferraz-Caetano and Bruno D. A. Pinheiro, "Legislators' Plague: How History of Science Can Explain the Struggles of Universal Pandemic Responses," in *Handbook of Research on Historical Pandemic Analysis and the Social Implications of COVID-19,* eds. Antonio Cortijo Ocaña and Vicent Martines (Hershey, PA (USA): IGI Global, 2021), pp. 47-59.

61 As introduced in note 34, where the laboratory's works are cited amongst peers for official deliberation of food analysis procedures.

62 More on Ferreira da Silva's biography: Alves and Alves, *Nos Caminhos da Química*, pp. 177-214.

63 See note 50.

64 See note 44.

65 Ferraz-Caetano, *As Artes Químicas na Academia do Porto*, Chapter 3.

66 As discussed in Alves and Alves, "Ferreira da Silva e o Laboratório Químico".

67 Alves and Alves, "Ferreira da Silva e o Laboratório Químico", pp. 7.

68 As described in note 26.

69 The LQMP's specialized library was considered by Portuguese chemist's to be the most up-to-date compendium of chemistry books. Most of its equipment was based on the apparatus' used in the Paris Municipal Laboratory.

70 As the Laboratory's expertise was previously discussed in notes 31 and 32, including the official legislative remarks.

71 The aims and vision for the *Revista* are written by Ferreira da Silva himself in its debut edition A. Ferreira da Silva, "O Nosso Programa," *Revista de Química Pura e Aplicada* 1, no. 1 (1905): 1-3.

72 The journal is now published under the name *Química* (https://b-quimica.spq.pt/).

73 Specially in its first five years of publication. The complete historic archive of the journal is freely available at: https://www.spq.pt/magazines/rcpapplicada.

74 Experimental guidelines were not clear about the methodology on analytical procedures by the competent laboratories, as per Section 2 of Ferraz-Caetano, "As raízes da regulação alimentar em Portugal."

75 Ferreira da Silva makes a strong argument about this by citing several works of other scientists of the LQMP, on his book A. Ferreira da Silva, *Recursos e Peritos na Fiscalisação Sanitaria no Porto em 1903: (O Reverso da Medalha)* (Porto: Imprensa Portugueza, 1905). For example, he quotes a situation where a detection of "wine fortification", a traditional and common practice for producing certain organoleptic qualities, was argued by certain hygiene inspection as adulteration. However, a LQMP scientist's report was ignored in this decision.

76 See notes 44 to 46.

77 This is documented in the "unofficial" report of the LQMP's work, regarding public health, on a book published by its director, A. Ferreira da Silva, *Documentos sobre os trabalhos de química aplicada à higiene do Laboratório Municipal de Química do Porto* (Porto: Imprensa Portugueza, 1904).

78 As reported by the shared expertise and circulation of actors cited in the previous section.

79 The full disclosure of these regulations is compiled in Direção Geral dos Serviços Agrícolas, *Methodos Officiaes para as Analyses dos Vinhos, Vinagres e*

Azeites: Seguidos da Indicção Summaria dos Processos Empregados nos Laboratorios do Estado para o Exame do Leite e Lacticinios; Alcooes e Aguardentes (Lisboa: Typ. da Encyclopedia Portugueza, 1910).

80 As reported by Ferreira da Silva himself about the impact on Port Wine analysis in A. Ferreira da Silva, *Os Vinhos Portuguezes Genuínos Condenados como Falsificados: Notas Sobre os Trabalhos a Que Deu Origem a Questão dos Vinhos Suppostos Salicylados no Brasil com a Apreciação do Snr. H. Pellet e Vários Documentos* (Porto: Off. do Comércio do Porto, 1902) and its generalization in A. Ferreira da Silva, *A Fiscalisação Dos Generos Alimenticios No Porto: (Vinhos)* (Porto: Papelaria La Bécarre, 1903).

81 Specially in the period 1902 to 1905.

82 According to *Diário do Governo*, no. 99, (06 maio de 1903), pp. 3.

83 These entities were under the orientation from two ministries: the *Ministério do Reino* and *Ministério das Obras Públicas*, as revealed in Ferraz-Caetano, "As raízes da regulação alimentar em Portugal".

84 The *Estação Químico-Agrícola de Lisboa* (Lisbon Chemical-Agricultural Station) was a scientific institution founded in late 19th century Portugal. Its main objective was to support agricultural development by providing chemical analyses of soils, fertilizers, water, and agricultural products.

85 According to *Diário do Governo*, no. 24 (7 de outubro de 1901).

86 A key example is the recognition of famous scientist Henri Pellet, who invited Ferreira da Silva to present an updated version of his method to quantify salicylic acid in wines, at the French Academy of Sciences. A good account on this is on Ferreira da Silva's biography and in a publication by Pellet himself: H. Pellet, *O Ácido Salicylico e a Questão dos Vinhos Portuguezes no Brazil em 1900* (Coimbra: Imprensa da Universidade de Coimbra, 1906).

87 The full report can be found on Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto*, sction "Relatório do Vereador do Pelouro sobre a Extincção do Laboratório Chimico Municipal do Porto", pp. 35-63.

88 Ferreira da Silva condensed the outcomes (and his defense) on this quarrel in Ferreira da Silva, *A Questão do Laboratório Municipal de Chimica do Porto.* His formal statements are found on pp. 63 onwards.

89 See notes 44 to 46.

90 As in Alves and Alves, *Nos Caminhos da Química*, pp. 177-214.

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In Memoriam

Professor Marc Henry (1958-2024)

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Professor Marc Henry passed away on November 2024 aged 66 after a long fight against cardio-vascular disease. He was born in Avignon in an educated but unconventional family. His father was an engineer at the French atomic energy agency and his mother a pharmacist interested in homeopathy.

He was as young as ten years old when he decided that he had to learn chemistry, and he soon began to study in his father's pharmaceutical courses. At school, he always wanted to go into the unexplained details of what he was taught, and as the deep understanding always came after the exams, his marks were pretty low ! So when he finished the secondary school, the math teacher advised him not to go to university. A new university had just opened in Avignon lacking students and he was accepted. The class was small (four students) and he was the best, very far above the others. Then he passed the entrance exam of Ecole Supérieure de Physique et de Chimie in Paris, where he was one of the best students. His thesis, under the direction of Professor Livage was about condensation and hydrolysis in inorganic chemistry and involved the knowledge of quantum physics. During his work he understood the key role of hydration of the chemical reactions (1). His thesis earned him multiple prizes and quotations. It was the beginning of his lifelong interest on water.

In 1992, Marc Henry decided to become professor at the University of Strasbourg. However, he had to pass an examination called "Habilitation à Diriger les Recherches" to be allowed to compete. This exam consisted of a second, self-directed thesis. However, the university's director of theses refused to register him, as his doctoral thesis was too recent - four years instead of five. Marc got approval and the authorization to register arrived three days before the deadline for submission of the manuscript. The work Marc had accumulated over the past four years was enough and in three days he wrote his three-hundred page dissertation and submitted it before the deadline. He brilliantly obtained his habilitation in 1993, and the job in Strasbourg a couple of months later.

Marc Henry, aged 35, moved to teach chemistry at the university of Strasbourg which was under the direction of Professor Ourisson. Although Marc received a warm verbal welcome from the director of the laboratory, Marc Drillon. The work conditions were in total contradiction with the promises. He was given a desk in the hallway. The office he had been promised belonged to an older colleague, who was over eighty years old and who visited the laboratory, two or three times a term. No one felt able to ask this great man to share this place with a younger one.

A fellow professor and colleague, Waïs Osséini, who was working in Strasbourg under the direction of Jean-Marie Lehn, winner of shared Nobel Prize for Chemistry in 1987, was looking to become independent and set up his own unit. He invited Marc to come and stay with him, promising him an office, a lab and the scientific independence that Marc Henry needed!

Marc Henry only working with young students developed a high level of academic research. Because of administrative hurdles, he was mostly recognized abroad. He achieved the feat of publishing 192 articles, with 10,340 citations and a very high readership index

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(2). Marc dedicated his life to research. He was very affable with colleagues and students and was well liked. At the same time, he was extremely bad at public relations, as he never payed allegiance. His life was his fundamental research that he performed on a shoestring. At the Institut Le Bel, he was most of the time alone at his desk with limited contact with his immediate colleagues. At the end of his academic career, the prolongation of his emeritus status was denied which hurted him.

His academic career focused on the physical nature of water. This is of particular importance in the chemistry of gel-sol compounds, and quantum physics was essential to his understanding of this phenomenon. In the later part of life he focused on the role of water in life.

With his collaborators, the conversations were never futile but deeply rooted in the key problems of life. He could share and explain, in simple terms, chemistry or physics for hours on end. As his interest on the chemical complexity of water was not shared by his colleagues, he reached to the public with short and concise books or conferences on topics as varied as life, water, homeopathy and quantum physics.

For him the very reason of life was the laws of physics. Entropy was the driving force for the origin and complexity of life. He demonstrated that the characteristic property of any living entity was that its energy balance can be expressed by a very simple and elegant formula: Food = Biomass + Heat + Waste. In other terms, life is a steady, non-equilibrium process feeding on energy rich nutrients and resulting in the synthesis of biomass, the release of infrared photons and the release of material waste. This seemingly trivial equation leads to a new understanding of diseases. In every disease, from cancer to inflammation, there is a decreased mitochondrial activity and decreased release of heat resulting in the synthesis of macromolecules like membranes and DNA in cancer cells, amyloid plaques in Alzheimer's disease or cytokines and lymphokines in inflammation. In other words diseases as diverse as cancer, Alzheimer's or inflammation have a common root, the decrease of entropy release in the form of heat and a corresponding increase in the release of entropy carried away by molecules.

Molecules effective in the treatment of diseases had to increase the mitochondrial efficacy and the release of entropy in the form of heat. This was a novel way to classify and understand both the pathogenesis and the treatment (6).

Marc and his collaborators were convinced of the simplicity of life. He thought that the energy flux released by the sun was enough to synthesize the precursors of life. The rest was the straightforward consequences of the laws of thermodynamics.

Marc was also looking for practical applications of his knowledge. At the beginning of the Covid-19 epidemics, he became aware of the protective value of an old drug: Methylene Blue. He was able to retrace its history and the fact that it is a precursor of Chloroquine. With the help of his co-workers, he wrote a paper in 48 hours (7).

Marc had been working on chemotherapy at the Faculté de Chimie de Strasbourg but had limited interest in the discovery of new molecules. He was more interested in the repurposing of old drugs. He participated in the rewriting of cancer as an electronic disease, where the proliferation of cells is viewed as the direct consequence of an electronic short circuit. The electrons do not target oxygen like in catabolism but NAD or NADP which result in the activation of anabolism. Marc was at the crux of this key discovery: the electronic flux is the very reason not only for cancer but for every disease.

Shortly before his death, he expanded his vision to new pathologies such as macular degeneration. Here again there is an interplay between water and electronics (8). The leakage of choroidal vessels increase osmotic pressure which in turn shuts down the mitochondria and the electrons are rerouted toward synthesis. Anabolism results in Drüsen and alteration of the retina (9). When he worked with his colleagues, all felt secure that he had and will have an answer for every difficult question, especially those that lie between biology, chemistry, and physics. What a scientist!

In 2005, Marc experienced a near death experience while undergoing cardiac surgery. Since then, he knows that conscience survive death. And as usual with Marc, water was at the center of his understanding of life and death. He was convinced that a novel reading of life should be drawn. 99% of our molecules are water. The rest being mostly ions and some lipids and proteins. On all surfaces, water molecules interact with virtual photons in the vacuum, oscillating together to form a new entity: the water coherence domains.

These coherence domains carry bits of information. He claimed that on the surface of all the body's cell membranes, the entire life of a human being could be encoded. For him, all the books ever written could be encoded in a few hundred liters of morphogenic water. In a far reaching hypothesis, he claimed that the information encoded in water originated in the quantum vacuum. Marc was convinced of the key role of water in consciousness, memory and homeopathy.

Most scientist consider that first and most important is matter. First the organism, then consciousness. For these scientists, information and the consciousness are a consequence of brain activity. Marc claimed the opposite: information predates the organism and the brain. The brain is simply capable of retrieving the information. Time will tell (10).

Like all visionaries in science, Marc was often misunderstood by his peers but he left us with a surprisingly simple and universal message: IN AQUA VERITAS ! His leitmotiv was that the most modest and ubiquitous molecule of our own body conceals the deep secrets of life and death. The collective properties of water *in vivo* are amazingly complex at all length scales and should be explored further if we really want to understand the origin of life and the intimate functioning of any living being.

Marc was one of the few academic researchers capable of understanding and collaborating scientifically with colleagues from seemingly disjointed disciplines: physicists, doctors, biologists, botanists, agronomists, musicians... Marc was so eclectic that specialists in his field found it difficult to keep up with him. Thanks to his rare teaching skills, he found an enthusiastic audience among the general public. He restored the reputation of scientific research, dented by the many frauds associated with the management of COVID, and awakened new vocations. In the last year of his life, he gave half a dozen talks in schools to children aged 7 to 10, explaining the importance of water for life and good health. The person who organized these meetings has fond memories of his simplicity, kindness and clarity, as his talk was incredibly rich.

REFERENCES

1. Livage, J., Henry, M., & Sanchez, C. (1988). Sol-gel chemistry of transition metal oxides. *Progress in solid state chemistry*, *18*(4), 259-341.
2. https://www.researchgate.net/profile/Marc-Henry-2#research-items
3. Gigant, K., Rammal, A., & Henry, M. (2001). Synthesis and molecular structures of some new titanium (IV) aryloxides. *Journal of the American Chemical Society*, *123*(47), 11632-11637.
4. Alam, T. M., & Henry, M. (2000). Empirical calculations of 29 Si NMR chemical shielding tensors: A partial charge model investigation of hydrolysis in organically modified alkoxy silanes. *Physical Chemistry Chemical Physics*, *2*(1), 23-28.
5. Henry, M. (2021). Thermodynamics of Life. *Substantia*, *5*(1).
6. Schwartz, L., Henry, M., Alfarouk, K. O., Reshkin, S. J., & Radman, M. (2021). Metabolic shifts as the hallmark of most common diseases: the quest for the underlying unity. *International Journal of Molecular Sciences*, *22*(8), 3972.
7. Schwartz, L., Devin, A., Bouillaud, F., & Henry, M. (2020). Entropy as the driving force of pathogenesis: an attempt of diseases classification based on the laws of physics. *Substantia*, *4*(2).
8. Henry, M., Summa, M., Patrick, L., & Schwartz, L. (2020). A cohort of cancer patients with no reported cases of SARS-CoV-2 infection: the possible preventive role of Methylene Blue. *Substantia*, 888-888.
9. Attal, R., Bakkar, A., Bouillaud, F., Devin, A., Henry, M., Pontié, M., Radman, M. & Schwartz, L. (2024). From electrons to cancer: Redox shift as a driving force of tumorigenesis. *Advances in Redox Research*, *10*, 100087.
10. Schwartz, L., Schwartz, J., Henry, M., & Bakkar, A. (2024). Metabolic Shift and Hyperosmolarity Underlie Age-Related Macular Degeneration. *Life*, *14*(9), 1189.
11. Henry, M. (2024). Water and the Origin of Life. *Water*, *16*(19), 2854.

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