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The role of chromosomal rearrangements, polyploidy, and genome size variation in the diversity and ecological distribution of *Asparagus* L. species: a landscape cytogenetics meta-analysis approach

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Abstract. The *Asparagus* genus includes a group of plants with economic and medicinal importance. Although numerous cytogenetic and genetic studies have been conducted on *Asparagus* species, there are no reports on landscape genetics, landscape cytogenetics, or *Asparagus* cultivation in response to climate change. Therefore, we designed this study to answer the above-mentioned objectives. We performed a meta-analysis involving landscape genetic studies based on available cytogenetic data and reported DNA C-values for several *Asparagus* species from different countries. Additionally, species distribution modeling (SDM) was performed on some selected *Asparagus* species. This combined study not only identifies the genetic fragmentation and genetic clines within plant species but also predicts their growth and distribution across different regions and in response to climate change. We used discriminant analysis of principal components (DAPC) to group *Asparagus* taxa based on karyotype and chromosome pairing data. We also performed random forest (RF) analysis to determine the contribution of cytogenetic traits to *Asparagus* speciation based on the Gini index. An association study was performed using redundancy analysis (RDA) of cytogenetic data with geographic variables (longitude and latitude). We used spatial principal component analysis (sPCA) to analyze the contribution of spatial variables to the cytogenetic structure of the studied *Asparagus* species. We used Bioclim and Maxent species distribution models (SDMs) to predict and identify areas suitable for selected *Asparagus* species in response to climate change by 2050. Results indicated that ploidy and chromosome size, the occurrence of heterozygote translocation, frequency and distribution of chiasmata, and genome size play role in *Asparagus* species diversification and adaptation. These cytogenetic characters are significantly associated with spatial variables and *Asparagus* species formed cytogenetic clines in response to local environmental conditions. SDM analyses showed that a combination of temperature and precipitation factors affect *Asparagus* species distribution and that in the coming future, some of these species may have a reduced cultivation area due to climate change which must be tackled by planning a proper conservation program worldwide.

Keywords: *Asparagus*, cytogenetic clines, genome size, Maxent, RDA.

INTRODUCTION

The genus *Asparagus* contains about 240 species, with *Asparagus officinalis* L., commonly known as the garden *Asparagus* as the most famous and economically important species. This edible vegetable crop is cultivated, grown, and used all over the world for its edible spear (Kanno et al. 2011). However, along with garden *Asparagus*, other species are also cultivated such as *Asparagus maritimus* L., or have been proposed as a genetic source for breeding programs like *Asparagus acutifolius* L.

Moreover, the young shoots of some wild *Asparagus* spp. namely, *A. acutifolius* L., *A. aphyllus* L., *A. acerosus* Thunb. ex Schult. & Schult. f., and *A. laricinus* Burch., are consumed fresh. Similarly, *A. verticillatus* L. is used for medicinal purposes and *A. densiflorus* (Kunth) Jessop as well as *A. plumosus* Baker have been grown as ornamental plants (Kubitzki et al. 1998).

Polyploidy has been reported in both the garden *Asparagus* (4x, and 8X), as well as in several species related to garden *Asparagus*, for example in *Asparagus falcatus* (2x, and 4x), *Asparagus racemosus* var. *javinica* (2x, and 4x), *Asparagus maritimus* (4x, and 6x) (see for example, Sheidai and Inamdar 2017; Mousavizadeh et al. 2021; Sala et al. 2023).

Although numerous cytogenetic and genetic studies have been conducted on *Asparagus* species, there are no reports on landscape genetics, landscape cytogenetics, or *Asparagus* cultivation in response to climate change. Therefore, we designed this study to answer the abovementioned objectives. We used our own cytogenetic data (Sheidai and Inadar 1997) and publicly available cytogenetic and nuclear DNA quantity data reports (C-value data) (Pires et al. 2006; Bouberta et al. 2017; Suma et al. 2017; Mousavizadeh et al.2021; Plath et al.2022), and performed a meta-analysis in a landscape genetic context. Additionally, species distribution modeling (SDM) was performed on some selected *Asparagus* species. This combined study not only identifies the genetic fragmentation and genetic clines within plant species but also predicts their growth and distribution across different regions and in response to climate change.

Cytogenetic studies, including chromosome morphology and chromosome pairing analysis, are essential for plant breeding, QTL hybridization and molecular mapping, and genetic transfer of beneficial genes between different species and cultivars. Moreover, the frequency and location of recombination through chiasmata formation can promote adaptation in a species by creating new genetic combinations (Rice 2002). Recombination can vary within chromosomes, between chromosomes, and between individuals, sexes, populations and species and its proportions can be influenced by environmental and demographic factors (Charlesworth 1976; Rice 2002), but are inherited, maintained, and selected for at specific genetic loci (Chinnici 1971). Moreover, studies across different taxonomic scales have shown that recombination frequency and occupying a particular landscape may be controlled by different mechanisms in different taxa (Ortiz-Barrientos et al. 2015; Johnston et al. 2016).

Landscape genetics considers the genetic basis of diversity in response to spatial variables such as geographic distance, altitude, and latitude (Provost et al. 2022). The frequency and distribution of chiasmata (cross-over) which are genetically controlled, as well as the occurrence of heterozygote translocations, result in genetic variation through chromosome genetic rearrangement. It is also known that polyploidy is one of the main genetic mechanisms in *Asparagus* genus speciation (Sheidai and Inamdar 2017). Therefore, landscape genetics and species distributions allow for determining the role of global and regional spatial variables as well as climatic variables in the genetic makeup of *Asparagus* species and potentially suitable growing regions. Species distribution models (SDMs) are methods that study the current geographic distribution of plant species and predict future events in the face of climate change. Through this, suitable habitats for the cultivation of the plant species of interest can be identified, and conservation measures can be proposed if there is a possibility that the cultivation area of the target plant species will decrease (Elite and Litwick 2009; Lee-Yaw et al. 2021).

MATERIAL AND METHODS

For landscape cytogenetic studies we used both karyotype and chromosome pairing data of 18 *Asparagus* taxon (Table 1). For genome size analysis we used the freely available published data of Plath et al (2022). For the species distribution modeling (SDM), we used the occurrence data points for the selected *Asparagus* species from GBIF (the Global Biodiversity Information Facility), as well as the published materials.

DATA ANALYSES

Cytogenetic grouping

We used discriminant analysis of principal components (DAPC), for grouping of *Asparagus* taxa for both karyotype and chromosome pairing data. An analysis of

Table 1. Karyotype data of *Asparagus* species used in present study.

Species	Country	Locality	Longitude	Latitude	2n	Ploidy	Total chromatin length	Mean chromatin length	Shortest chromosome	Longest chromosome	Ratio
A. racemosus var. *Javanica*-1	India	Orissa	84.27	20.23	40	4	66.5	3.325	1.00	2.60	2.60
A. racemosus var. *Javanica*-2	India	Pune-University	73.82	18.55	20	2	38.52	3.852	1.13	2.93	2.59
A. densiflorus cv. Myers	India	private nursery Pune	73.88	18.51	40	4	78.58	3.929	1.30	2.82	2.16
A. laevissimus	India	J.N.H-Pune	73.87	18.53	40	4	66.12	3.30	0.91	2.91	3.20
A. myriocladus	India	Pune-University botanical garden	73.05	18.03	40	4	92.2	4.61	1.00	3.66	3.66
A. racemusus subacerosa	India	Pune-law college hills	73.82	18.51	40	4	65.2	3.26	1.06	2.26	2.13
A. sprengeri	India	Agharkar Research Institute	73.50	18.31	40	4	58.82	2.94	1.06	2.47	0.78
A. virgatus	India	Fergussen college	73.50	18.31	40	4	67.26	3.363	1.12	2.49	2.22
A. gonoclados	India	Pune	73.05	18.03	60	6	87.38	4.369	1.96	5.30	2.70
A. adsendens	India	J.N.H-Pune	73.87	18.53	20	2	73.3	7.33	0.99	2.00	2.02
A. falcatus	India	private nursery Pune	73.88	18.51	20	2	28.48	2.85	1.00	1.99	1.99
A. Officinalis-1	India	Fergussen college	73.50	18.31	20	2	85.68	8.57	2.58	5.85	2.27
A. racemosus	Bangladesh	University of Dhaka	90.39	23.77	20	2	18.18	1.81	0.53	1.23	2.32
A. setaceus	Bangladesh	Dakha	90.39	23.77	20	2	18.50	1.85	0.48	1.47	3.06
A. albus	Algeria	Tipaza	2.27	36.25	20	2	44.47	4.44	3.19	6.25	1.95
A. acutifolius	Algeria	Senalba	3.10	34.38	20	2	52.07	5.2	4.21	6.61	1.57
A. horridus	Algeria	Emir Khaled	2.12	36.08	20	2	36.04	3.6	2.91	4.46	1.53
A. Officinalis-2	Algeria	Tessala El Merdja	2.54	36.37	20	2	51.18	5.11	3.15	6.52	2.06

variance (ANOVA) test was performed on the cytogenetic data to reveal a significant difference between *Asparagus* species in different countries. We also performed random forest (RF) analysis to reveal the contribution of cytogenetic characters in *Asparagus* species differentiation based on the Gini index. These were performed in the adegenet package of R. 4. 2.

Redundancy analysis (RDA)

Association studies were performed using redundancy analysis (RDA) of cytogenetic data with geographic variables (longitude and latitude). RDA which is a constrained ordination method models the linear relationships between environmental predictors and genetic variation (Capblancq and Forester, 2021). This analysis was performed through 999 permutations in PAST ver. 4.

Spatial principal components analysis (sPCA)

We used the Spatial principal components analysis (sPCA), to analyse the spatial variables' contribution to the studied *Asparagus* species cytogenetic structuring. The sPCA is a multivariate method that is independent of Hardy Weinberg expectations and produces estimates summarizing both genetic variation and spatial structure between individuals (or populations) (Jombart et al.2008). Global structures (patches, wedges, and intermediate junctions) are statistically comparable to local structures (strong genetic differences between neighbors) and random noise. The sPCA also performs Moran's I test and IBD (isolation by distance). The sPCA analyses were performed with the adegenet package version R. 4. 2.

Species Distribution Modeling (SDM)

We used species distribution modeling (SDM) to predict and identify suitable regions for selected *Asparagus* species in response to climate change by 2050. In species distribution modeling, we used layers of forecast climate data for the current period (~1950-2000) and 2050 (2050-2061 average) based on 19 bioclimatic variables at a 5-minute resolution. Data was loaded from the WorldClim database. To represent the impact of climate change, future climate variables in 2050 were projected

according to the "representative concentration trajectory" (RCP, 2. 6).

We used bioclimatic variables derived from the monthly temperature and rainfall values. These bioclimatic variables are coded as follows:
BIO1 = Annual Mean Temperature
BIO2 = Mean Diurnal Range (Mean of monthly (max temp - min temp))
BIO3 = Isothermality (BIO2/BIO7) (×100)
BIO4 = Temperature Seasonality (standard deviation ×100)
BIO5 = Max Temperature of Warmest Month
BIO6 = Min Temperature of Coldest Month
BIO7 = Temperature Annual Range (BIO5-BIO6)
BIO8 = Mean Temperature of Wettest Quarter
BIO9 = Mean Temperature of Driest Quarter
BIO10 = Mean Temperature of Warmest Quarter
BIO11 = Mean Temperature of Coldest Quarter
BIO12 = Annual Precipitation
BIO13 = Precipitation of Wettest Month
BIO14 = Precipitation of Driest Month
BIO15 = Precipitation Seasonality (Coefficient of Variation)
BIO16 = Precipitation of Wettest Quarter
BIO17 = Precipitation of Driest Quarter
BIO18 = Precipitation of Warmest Quarter
BIO19 = Precipitation of Coldest Quarter

To build the SDM model, we used the Bioclim and maximum entropy methods implemented in the Dismo package in R 4. 2 and the Maxent program. SDMs require the occurrence data points on which the pseudo-absences (PAs) points are estimated, followed by model prediction. All the models were constructed with 80% training and 20% testing of occurrence data. The model evaluation was performed by both the threshold method and AUC determination (ROC curve).

Bioclim models the species distributions in relation to climatic variables and thus assumes that species occurrence is influenced by climate at the scale of climate variables and that these variables are normally distributed. Similarly, Maxent (Maximum Entropy Modeling) predicts the occurrence of a species by finding the one closest to the most common or uniform distribution, taking into account the limits of environmental variables in a known location (Phillips et al.2004). I n this method, the fit is measured as gain, which is basically a likelihood statistic that maximizes the probability of being present for background data adjusted for the case where all pixels have an equal (uniform) probability. The final probability distribution becomes the basis for fitted predictor variable coefficients. (Phillips et al.2004). The importance of bioclimatic variables influencing the distribution of *Asparagus* species was assessed by the Jackknife incremental method and the AUC value.

RESULTS

Cytogenetic data based on the country of origin of the *Asparagus* species used in this study are shown in Tables 2 and 3. An analysis of variance (ANOVA) test performed on the cytogenetic data revealed a significant difference between these countries in karyotype data ($p<0.01$, Fig. 1). These results demonstrate that genetic variation accompanies *Asparagus* species diversity in different regions of the world. A similar analysis for chiasma frequency and chromosome pairing could be performed on the species studied and not among the countries, but a significant result ($p<0.01$), indicated the species cytogenetic differences even within a particular country i. e. India.

Cytogenetic grouping of the studied *Asparagus* species based on karyotype data is presented in the DAPC plot (Fig. 2). These species are scattered in three distinct groups based on their country of origin.

Association analyses performed by CCA and RDA for karyotype data produced significant results ($p<0.01$, Fig.

Table 2. The mean value for chiasma frequency and distribution, and chromosome pairing in *Asparagus* species studied.

Asparagus species	Terminal chiasmata	Intercalary chiasmata	Total chiasmata	Ring bivalents	Rod bivalents
A. racemosus javanica	14.2	1.3	16	7.3	1.37
A. densiflorus cv. Myers	34.3	2.6	37	17.2	2.4
A. laevissimus	15.8	4	20	8.5	1.48
A. racemusus subacerosa	35.62	0.47	36.1	16.78	2.81
A. sprengeri	39	0.347	39.347	19.26	0.74
A. virgatus	36.1	1.3	37.4	18	1.7
A. gonoclados	55.28	5	60.23	26.84	2.46
A. adsendens	16.8	4.425	21.225	9.8	0.325
A. officinalis	13.8	1.36	14.54	5.54	3.36

Table 3. The genome size (1C-value) of *Asparagus* species used in the landscape cytogenetic analyses (data obtained from freely available published paper (Plath et al., 2022)).

Species	Country	Country-code	Longitude	Latitude	Ploidy (X)	C-value
A. acutifolius	Italy, Vittoria	1	14.53	36.95	2	1.35
A. aethiopicus	Spain, Malaga	2	4.42	36.71	6	0.86
A. albus	Portugal	3	8.22	39.39	2	1.23
A. amarus	Italy	1	12.56	41.87	6	1.37
A. arborescens	Canary	4	16.62	28.29	2	1.32
A. maritimus 1	Italy	1	12.56	41.87	6	1.33
A. maritimus 2	Italy	1	12.56	41.87	6	1.30
A. maritimus 3	Italy	1	12.56	41.87	6	1.28
A. maritimus 4	Italy, Vign	1	13.04	43.37	6	1.29
A. officinalis 'Darlise'	France	6	2.21	46.22	2	1.47
A. officinalis 'Ravel'	Germany	7	10.45	51.16	2	1.53
A. officinalis 'Steiners Violetta'	Germany	7	10.45	51.16	4	1.59
A. pastorianus	Macaronesia	8	16.84	28.23	4	1.40
A. plumosus	Cuba	9	82.36	23.11	2	0.42
A. plocamoides	Canary	4	16.62	28.29	2	0.71
A. prostratus 1	France, Ploemever	6	3.25	47.44	4	1.48
A. prostratus 2	France, Gavres	6	3.35	47.69	4	1.53
A. prostratus 3	France, Damgan	6	2.57	47.51	4	1.52
A. prostratus 4	France, Houat Is.	6	2.95	47.39	4	1.54
A. pseudoscaber	Italy	1	12.56	41.87	6	1.28
A. ramosissimus	Angola	10	17.87	11.20	2	1.15
A. scoparius	Africa, Cape Verde	11	23.04	16.53	2	0.73
A. stipularis 1	Ibiza	2	1.42	38.90	2	0.81
A. stipularis 2	Ibiza	2	1.42	38.90	2	1.13
A. stipularis 3	Ibiza	2	1.42	38.90	2	1.21
A. stipularis 4	Cyprus	12	33.42	35.12	2	1.19
A. albus	Portugal	2	8.22	39.39	2	1.23
A. ramosissimus	Angola	2	17.87	11.20	2	1.35
A. stipularis 4	Cyprus	2	33.42	35.12	2	0.86
A. scoparius	Africa, Cape Verde	12	23.04	16.53	2	1.23

3, A). These results showed an association between ploidy level, the ratio of the longest to the shortest chromosome the somatic chromosome number (2n). Moreover, the random forest result (Fig. 3, B) identified the ploidy level and somatic chromosome number as the main karyotype characters that differentiate the studied taxa.

A similar analysis performed on chiasma frequency and chromosome pairing of *Asparagus* species did not produce significant association ($p > 0.1$). This may be due to the fact that we obtained and used only the meiotic data of *Asparagus* species from India.

Spatial principal components analysis (sPCA)

The results of sPCA are presented in Fig. 4, A-F. The preliminary analysis of sPCA Eigenvalues showed the presence of strong positive and global spatial constraints over the cytogenetic characteristics of the studied *Asparagus* species (Fig. 4, A). This was supported by a significant global test obtained ($p = 0.01$, Fig. 4, B).

Similarly, the isolation by distance test (IBD), produced a significant result ($p = 0.01$, Fig. 4, D), indicating that cytogenetic differences among *Asparagus* species increased with increasing geographic distance.

The connection network (Fig. 4, E), showed a closer relationship (common shared cytogenetic features) between species from India and Bangladesh species, and the cytogenetic clines plot (Fig. 4, F), showed that the species studied in all three countries formed cytogenetic clines probably due to their spatial adaptation. Moreover, Moran's I test was not significant ($p > 0.1$), indicating that the similar spatial and geographical regions have similar effects on the studied cytogenetic features.

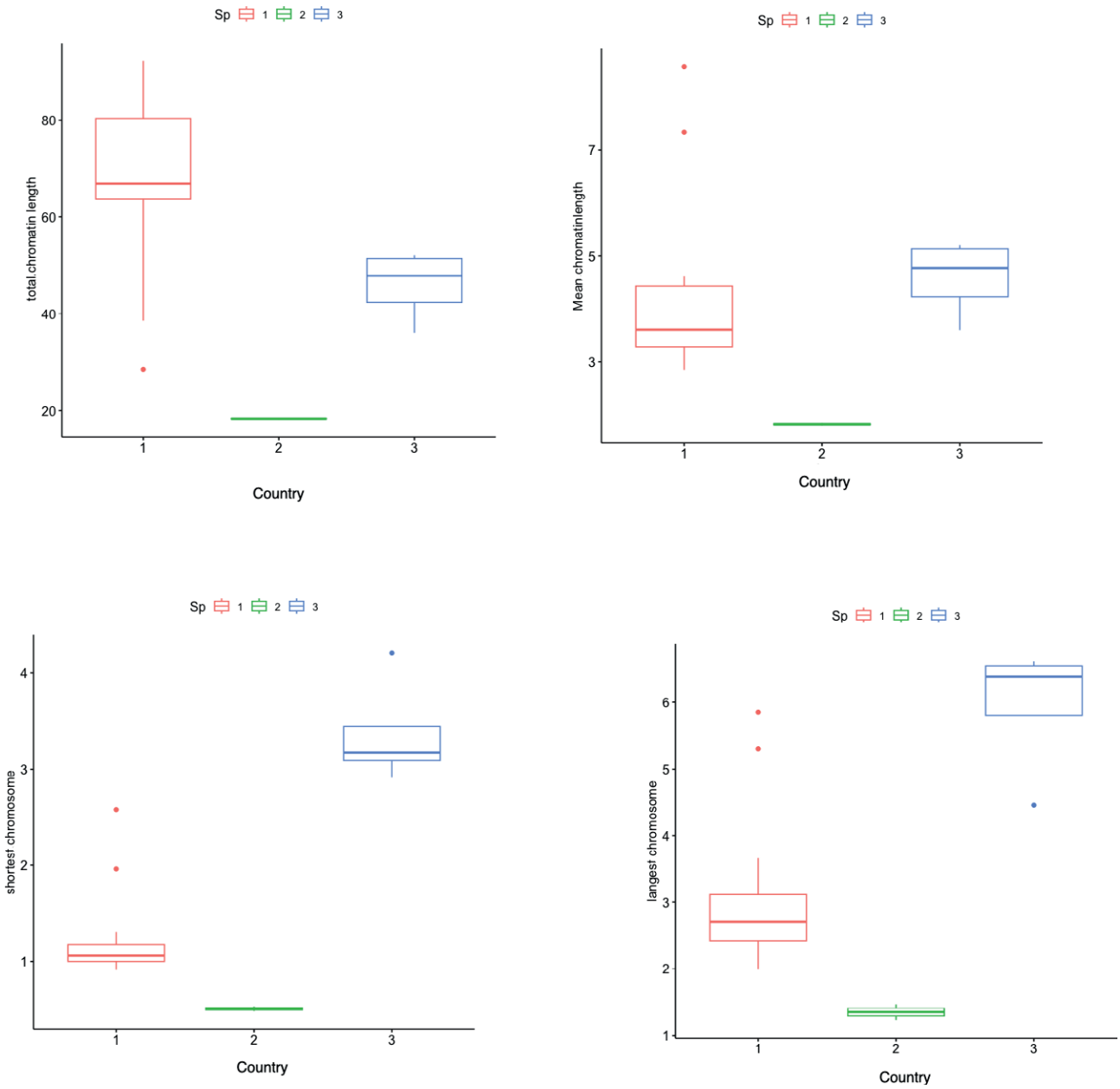


Figure 1. Representative box plots of Karyotypic data ANOVA between *Asparagus* species based on the countries. (Abbreviations for the country are 1= India, 2=Bangladesh, and 3 = Algeria).

Heterozygote translocations

Some of the studied *Asparagus* species showed the occurrence of heterozygote translocations in the pachytene stage of meiosis (Fig. 5, A). These translocations would result in multivalent formation in the metaphase stage (Fig. 5, B and C), (Sheidai 1985).

Genome size (C-value) analysis

The 1C value of the *Asparagus* species obtained from the published freely available work (Plath et al.2022) and their country of origin with spatial variables (longitude and latitude) are presented in Table 3. ANOVA performed on the amount of C-value produced significant results among the *Asparagus* species studied ($p< 0.01$, Fig. 6). The RDA analysis (Fig. 7) also showed a signifi-

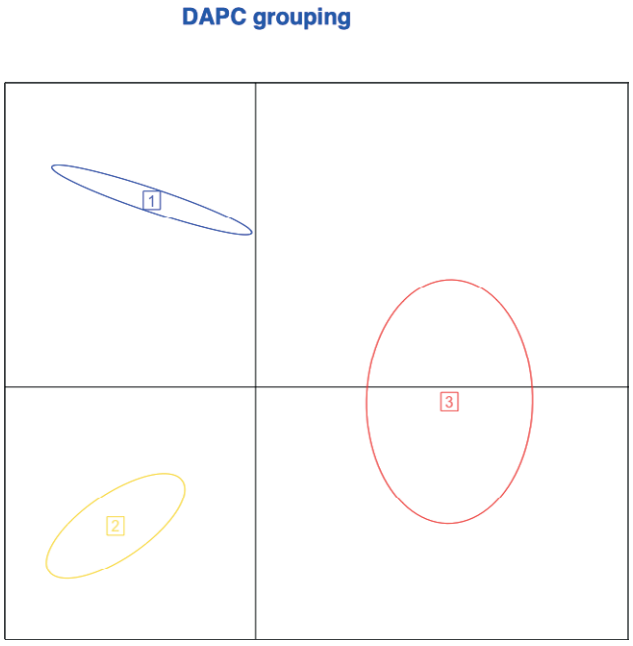


Figure 2. DAPC grouping of *Asparagus* species based on karyotype data. Abbreviations: 1= India, 2= Bangladesh, and 3 = Algeria.

cant association (p= 0.01), and spatial variables. Therefore, the longitudinal as well as latitudinal distribution of *Asparagus* species analyzed affect the ploidy level and their 1C-value genomic content. The longitude and latitude data are in degrees, and minutes, respectively.

Spatial principal components analysis (sPCA) of 1C-value data

The results of sPCA are presented in Fig. 8, A-D. The preliminary analysis of sPCA Eigenvalues showed the presence of strong positive and global spatial variables over the ploidy level, and the genomic 1C-value content of the studied *Asparagus* species (Fig. 8, A). This was supported by a significant global test obtained (p =0.10). Similarly, the isolation by distance test (IBD), did not produce a significant result (p= 0.01), indicating that the 1C-value content difference among *Asparagus* species is not increased with increasing geographic distance.

The connection network (Fig. 8, B), showed similarities (common shared cytogenetic features) between the species studied in different countries, and the genome size clines plot (Fig. 8, C), showed that the species studied in all these countries formed ploidy and 1C-value content clines due to their spatial adaptation. Moreover, Moran's I test was not significant (p> 0.1), indicating that the similar spatial and geographical regions have similar effects on the studied cytogenetic features.

The contribution plot (Fig. 8, D), revealed that the ploidy level plays a more pronounced role compared to that of 1C-value content in the analyzed *Asparagus* species in response to spatial variables.

Species distribution modeling (SDM) results

SDM analysis of selected *Asparagus* species provides insight into the climatic variables affecting the growth and occurrence of these important species worldwide and can predict their response to climate change in the future. These findings help conservation programs.

The results of present-time predicted distribution versus *Asparagus* species distribution by the year 2050 are presented in Figs. 9 and 10. The probable distribution of these species under the influence of climate change obtained from both BIOCLIM and Maxent models was almost the same. These results revealed that the area under cultivation for the studied species would be much reduced in extent by the year 2050. This statement holds true, particularly for *Asparagus verticillatus*.

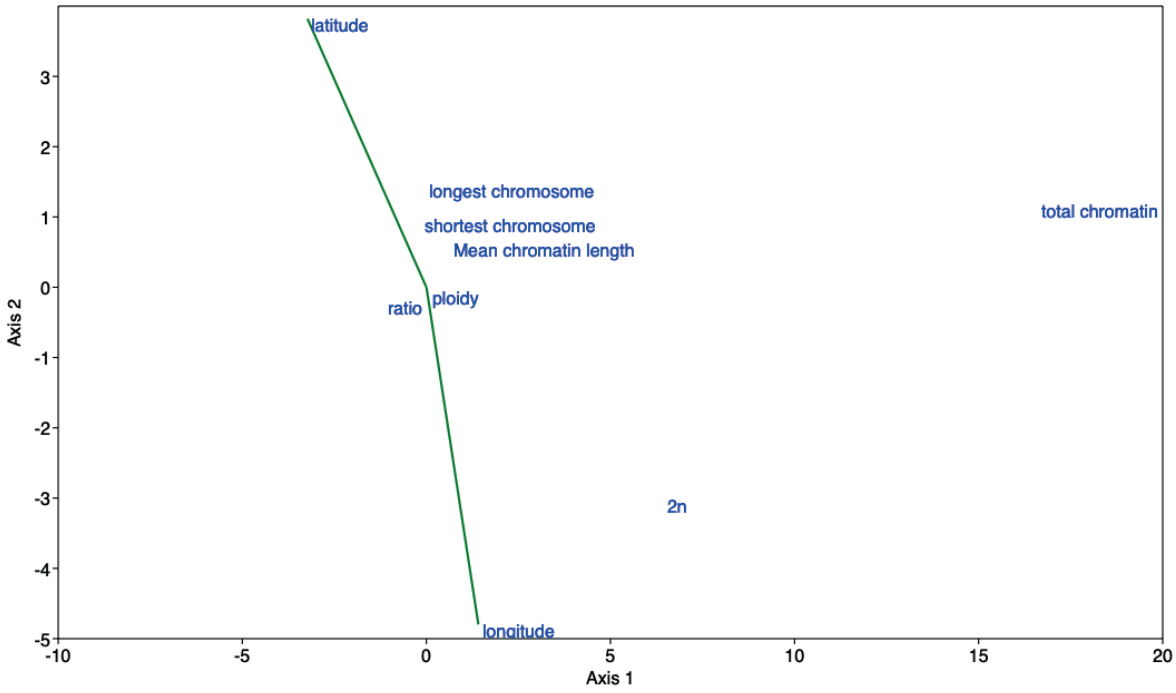
The importance of climatic variables based on the Jackknife method and ROC curve (AUC value) are presented in Fig. 10. The AUC values obtained for both present time prediction and by the year 2050, were all above 0.90 which supports the modeling results.

Important and influential bioclimate variables identified by the Jackknife method revealed that BIO3= Isothermality, BIO5= Max Temperature of Warmest Month, BIO13= Precipitation of Wettest Month, BIO14= Precipitation of Driest Month, BIO15= Precipitation Seasonality, BIO17= Precipitation of Driest Quarter and BIO18= Precipitation of Warmest Quarter, are among the most important bioclimate variables affecting the distribution of *Asparagus* species.

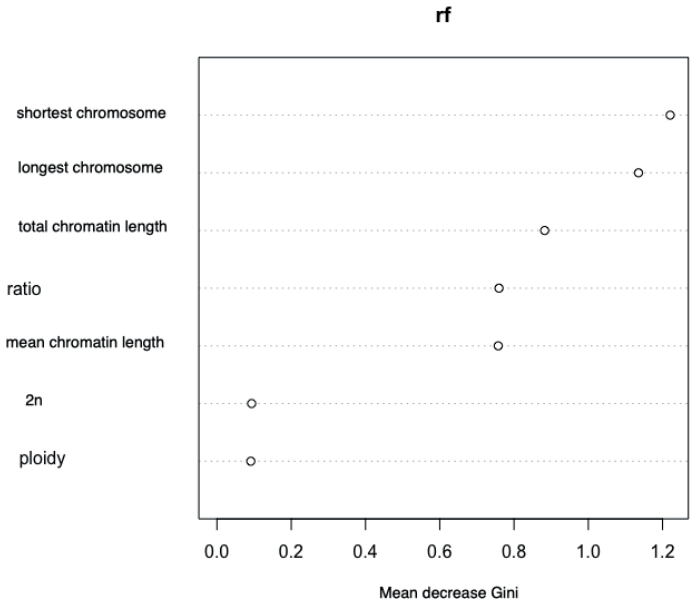
DISCUSSION

This study showed that global and local spatial patterns influence the genetic structure of *Asparagus* species through cytogenetic changes like polyploidy, structural changes of chromosomes (heterozygote translocations), chromosome size, and the plant genome size. Moreover, bioclimatic variables determine the geographical distribution of these plants worldwide.

Chromosomal evolution has played an important role in plant diversification and speciation, especially in the genus *Asparagus* (Plath et al. 2022). In the *Asparagus* genus, Plants with different ploidy levels within the same population are very common. For example, triploid, pentaploid, hexaploid, and octoploid plants were found in



A



B

Figure 3. A= RDA plot of karyotype data in the studied *Asparagus* species shows a significant association between ploidy level, longest to shortest chromosome ratio, and somatic chromosome number. B = Random Forest plot of the same data showing the importance of karyotype characters in differentiating the studied taxa.

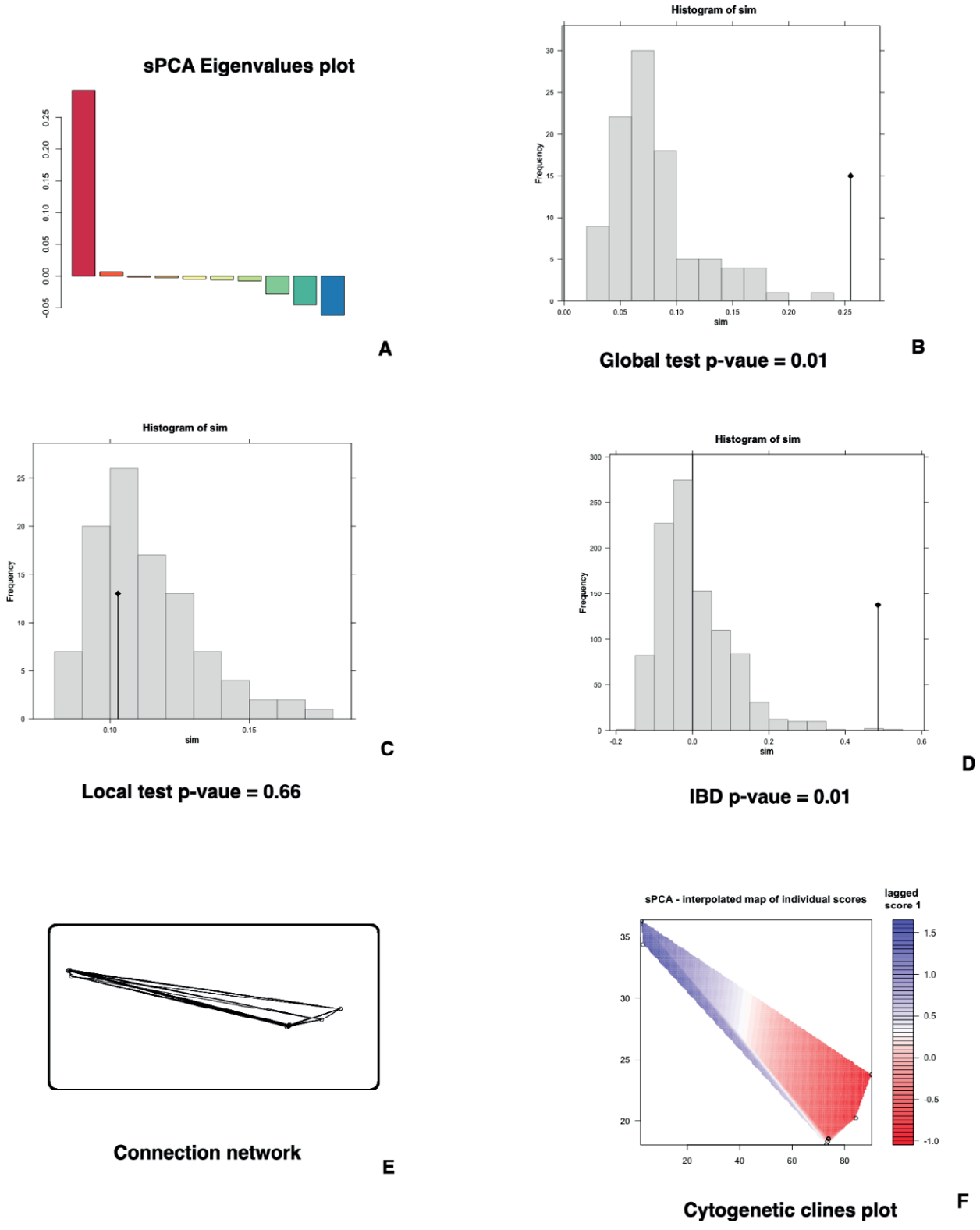
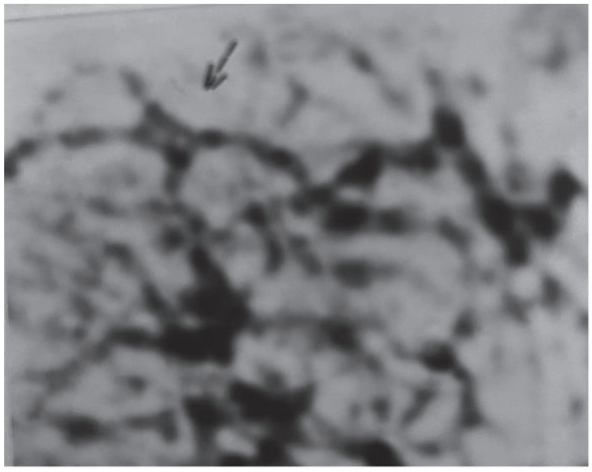


Figure 4. Representative sPCA plots of cytogenetic data in the studied *Asparagus* species. A = Plot of Eigenvalues revealed a strong effect of positive (global) spatial features for the studied taxa (the part shown in red), B-D = Global, local, and IBD test results showed significant p-values in the first two tests. E, and F = The connection and cytogenetic clines plots showed closer relationships between species from India and Bangladesh, with cytogenetic clines formed respectively in all three countries.



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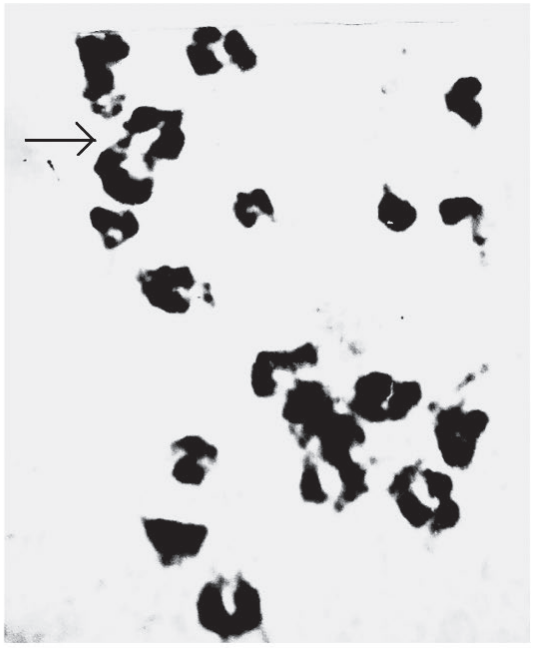

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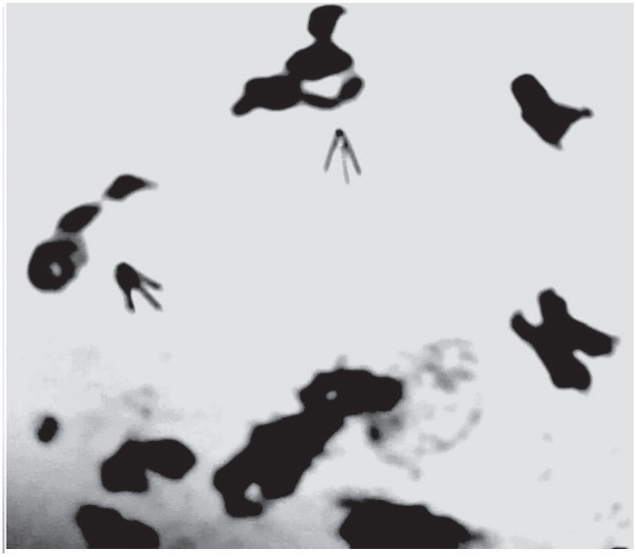

C

Figure 5. A = A heterozygote translocation (arrow) in *Asparagus racemosus* sub *acesora,* B, and C = Multivalent formation (arrows) in Asparagus gonoclados, and *A. officinalis*, respectively. (Figures are from one of the coauthors i.e. Sheidai 1985, Ph.D thesis)

the Spanish landrace 'Morado de Huetor' (Moreno et al. 2006). Additionally, Ozaki et al. (2014) discovered spontaneous triploid *Asparagus* plants from crosses with diploid parents.

Mousavizadeh et al. (2021), studied the influence of climate on the geographical distribution of diploid and polyploid *Asparagus* plants of *A. officinalis*, *A. persicus*, *A. verticillatus*, and *A. breslerianus* growing in Iran and reported changes in the ploidy levels of vegetation across different zones. These changes are related to humidity, average minimum and maximum temperatures, and soil salinity. They observed that the species with 8X and 10X species live at higher altitudes and are able to adapt to drier and more salinity lands than 2X and 4X plants.

The number and shape of plant chromosomes, the amount and composition of nuclear DNA, the frequen-

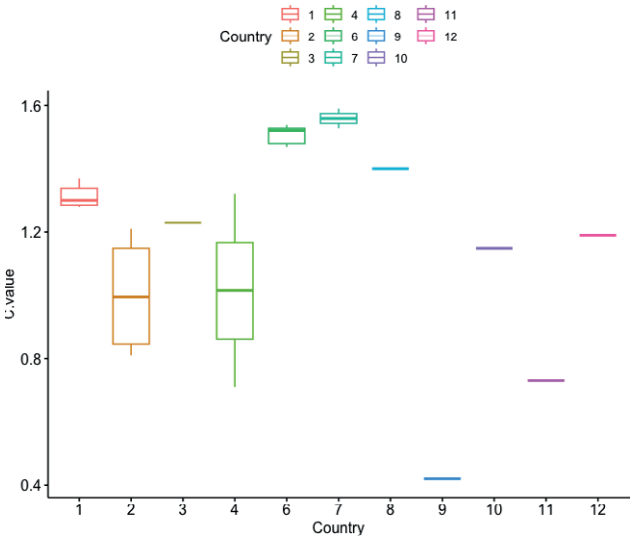

Figure 6. Box-plot of 1C-value quantity among the countries of origin of *Asparagus* species (The country code 1-12, are as in Table 3).

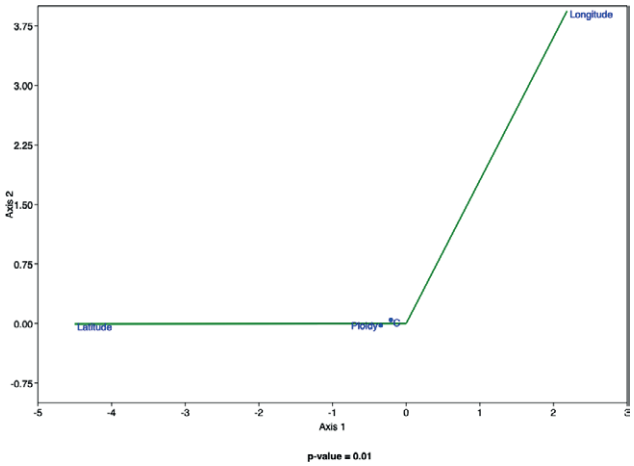

Figure 7. RDA plot shows a significant association (p=0.01), between the polyploidy level and 1C-value with spatial variables.

cy of chiasmata, and the chromosomal meiotic behavior of chromosome pairs vary greatly among plant species. In particular, meiotic behavior is genetically regulated, and changes in the frequency and location of crossovers within chromosome arms affect the genetic diversity of the offspring (Rees and Jones 1977). Therefore, the existence of significant differences in the chiasmata frequency and distribution, and ring and rod bivalents, among *Asparagus* species growing in different parts of the world may indicate their genomic differences (Sheidai et al. 2002) and may act as the genomic adaptation to environmental variables that have been reported in other plant crops (see for example, Sheidai et al. 2012).

Genetic variation can be exploited through local environmental and climatic selection to achieve ecological diversity even in the absence of physical barriers. Because new beneficial mutations or chromosomal rearrangements are unlikely to accumulate rapidly, it has been suggested that rapid adaptation may involve selection based on persistent genetic variation i. e. the genetic variation present in ancestral populations before divergence occurred (Ortiz-Barrientos et al. 2016).

Van Belleghem et al. (2018) presented a scenario for the emergence of persistent genetic variation that describes the demographic history of a population or species. When alleles involved in adaptation arise from independent mutations or chromosomal changes, they occur either at different loci or randomly in lineages from the same locus. Therefore, lineages are not identical because new adaptive mutations may occur in different haplotypes in different regions. On the other hand, if ecological differentiation is based on alleles or genetic loci that exist as persistent genetic variation in an ancestral population, derived alleles have the same origin but differ greatly in their evolutionary history.

Plath et al. (2022) reported that 2C DNA content can vary not only across accessions within a species but also across Asparagus species growing in different geographical regions of the world. The causes of these changes are thought to be polyploidization and differences in chromosome size.

However, other cytogenetic abnormalities and mechanisms, such as aneuploidy (Sheidai and Inamdar 1992; Ozaki et al. 2004), the presence of B-chromosomes (Sheidai and Inamdar 1993), cytomixis (Sheidai et al. 1993), or desynapsis (Sheidai 1992), are the other potentially effective cytogenetic changes found in the genus *Asparagus*.

Landscape genetics and population-level cytogenetic studies can reveal habitat fragmentation and identify the genetic clines within the geographic range of a plant species (Anderson et al. 2011). Studying global climate change also tests the ecological and evolutionary responses of species to predicted conditions. Knowledge and understanding of how habitat fragmentation affects adaptive evolution under projected climate change is very limited (Anderson et al. 2011). The present study found that *Asparagus* species could see their geographic distribution significantly reduced in the future due to climate change.

It has been suggested that environmental stresses (e. g., climate change) may result in inbreeding depression. As a result, inbred, fragmented populations may have a lower ability to adapt to contemporary and changing conditions compared to large, unfragmented

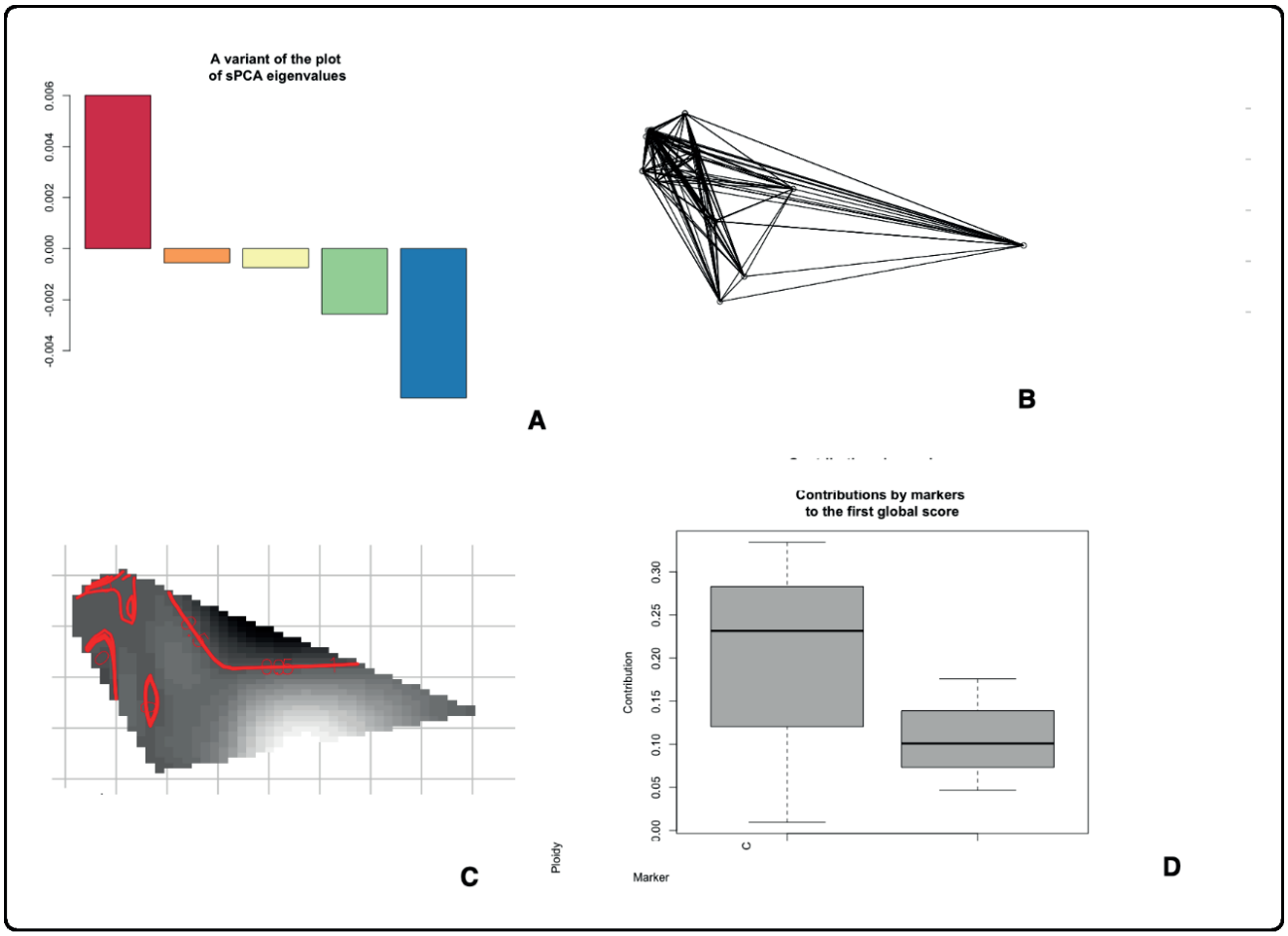

Figure 8. The sPCA analysis plots of the ploidy level and 1C-value content of *Asparagus* species. A the Eigenvalues plot, B the connection plot, C = Genetic clines formed due to both ploidy level and 1C-value data, and D = The contribution role of the studied variables viz. Ploidy and 1C-value.

populations. Fragmented populations with reduced genetic diversity may lack variation in key ecological traits such as drought tolerance. In such situations, assisted migration to suitable habitats along with the conservation of habitat corridors, may be necessary to prevent dramatic declines in species and genetic diversity (Anderson et al. 2011).

Polyploidy may play a role in adaptation to new habitats and environmental conditions. This appears to be positively related to latitude, altitude, and recent sea ice (Stebbins, 1984; Brochmann et al. 2004). Higher ploidy rates are generally observed at higher latitudes or altitudes than related diploids, especially in herbaceous perennial grasses (Zhang et al. 2019). Likewise, genome size is correlated with the environment and geographic distribution of species (Bottini et al. 2000; Bennett and Leitch 2011), and changes in DNA C values are correlated with many phenotypic traits of cells and organisms. This can affect important ecological traits of plant species in natural habitats, such as spring growth timing, cell size and leaf expansion rate early in the growing season, frost tolerance, and dry conditions (Zhang et al. 2019). Therefore, in conclusion, we report that both spatial and bioclimatic variables influence the genetic structure and geographical distribution of *Asparagus* species worldwide and that general conservation programs against climate change are needed.

AUTHOR CONTRIBUTION STATEMENT

Masood Sheidai and Fahimeh Koohdar: conceptualization of the project; Parisa Fouroutan; data collection and lab work

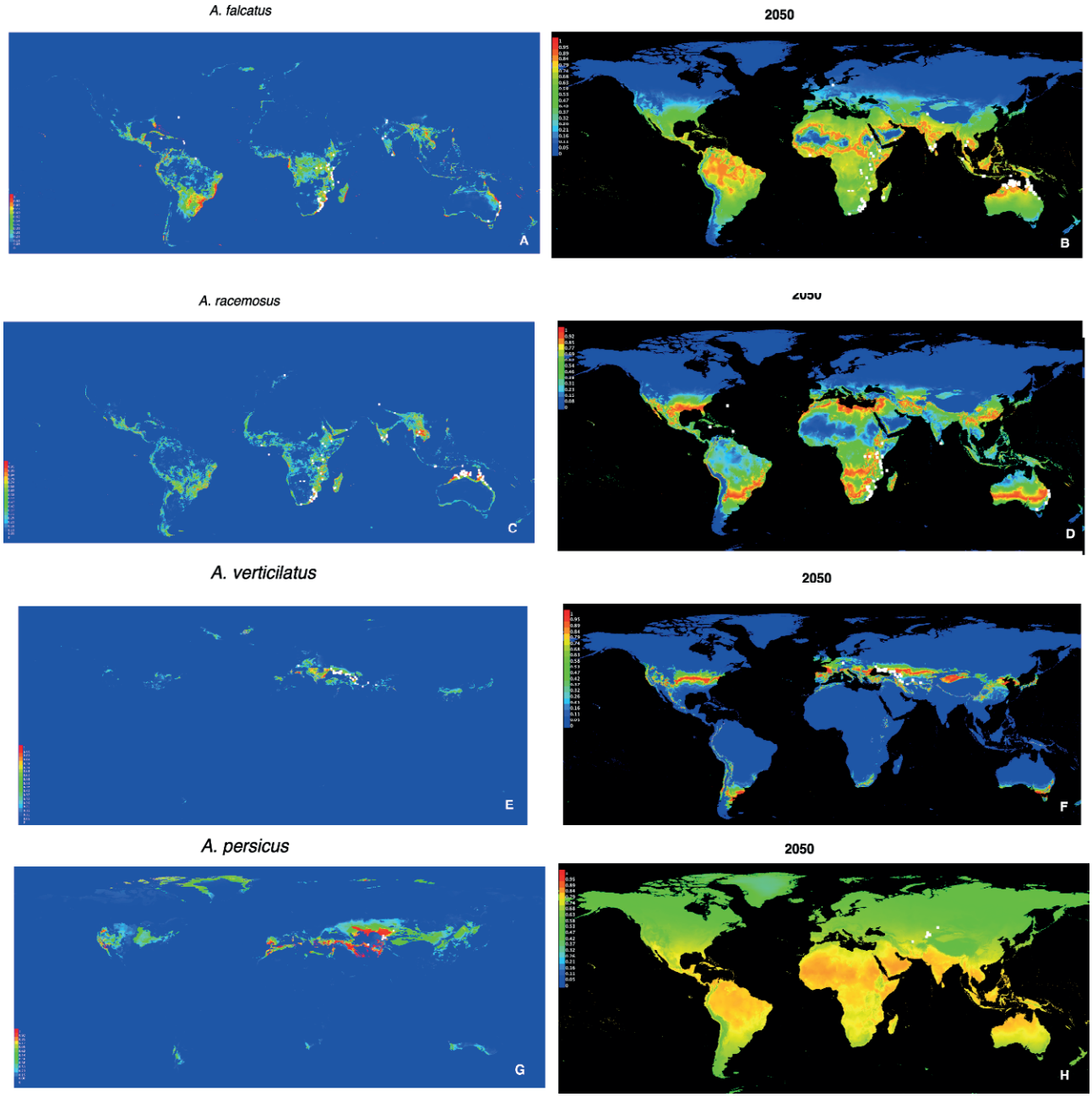

Figure 9. SDM results of the Maxent model show the predicted occurrence of *Asparagus* species at the present time versus the year 2050.

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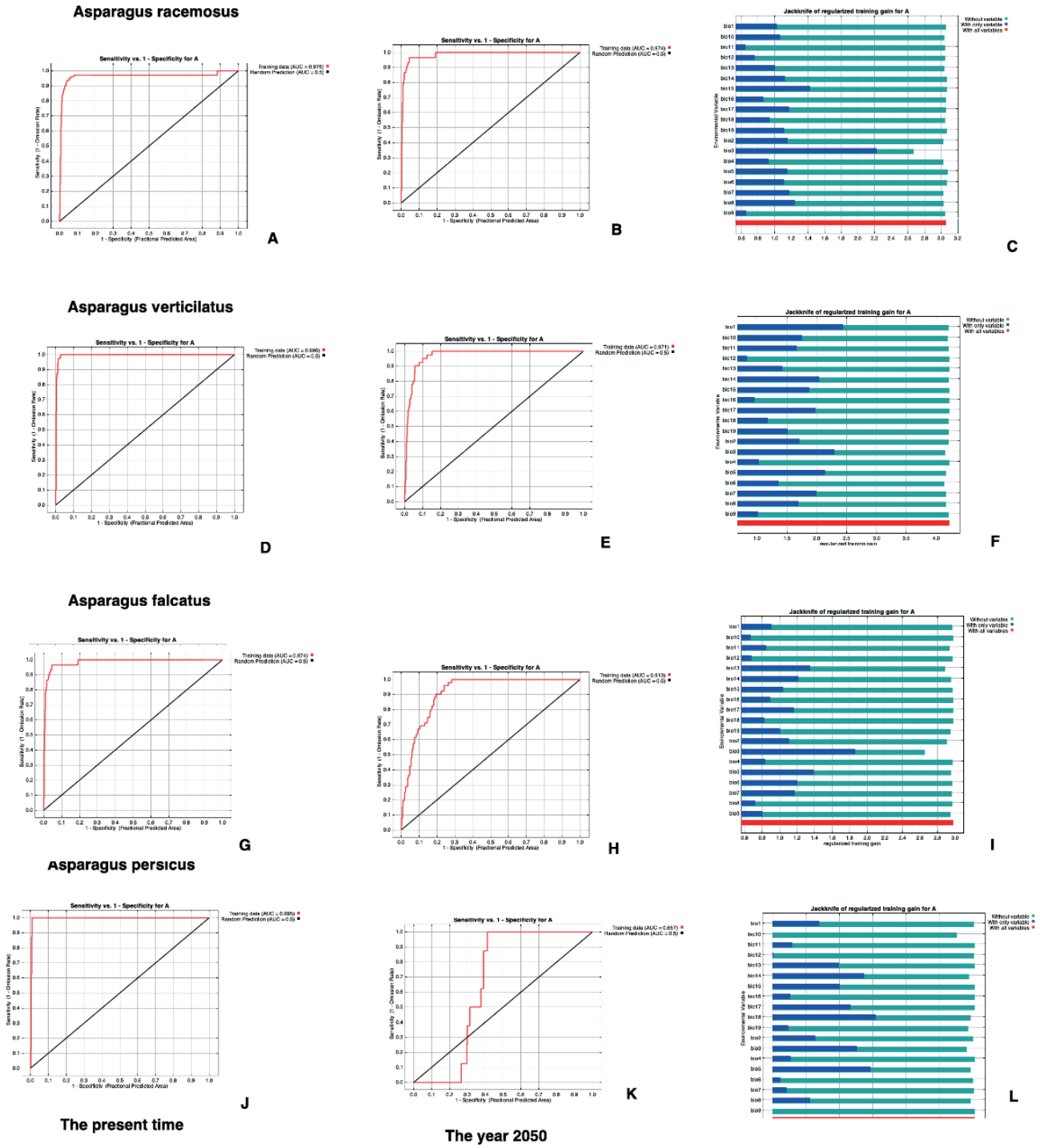

Figure 10. The representative plots of AUC values (ROC curve), and the importance of bioclimate variables affecting *Asparagus* species geographical distribution.

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