MAPPING OF GEOGRAPHIC DISTRIBUTION OF C₃ AND C₄ SPECIES OF THE FAMILY CHENOPODIACEAE IN IRAN

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The patterns of geographic distribution of C_3 and C_4 species of the family *Chenopodiaceae* have been analyzed in Iran, using a database of 11151 georeferenced records. The members of *Chenopodiaceae* family with 45 genera and 204 species are widely distributed in Iran. There they show a ratio of about 2:1 between C_4 and C_3 species. All available data were organized in MS access database, and analyzed in point-to-grid analysis tool of DIVA-GIS software, using 10×10 km grid cells and the circular neighborhood option. The highest species numbers, with a peak at 83 per grid unit, occur in Tehran, Semnan, and East Azerbaijan provinces but the density of records is extremely uneven. Consequently, the same areas also are the hot spots of both C_4 and C_3 species. The comparison between the distribution patterns of carbon isotope composition ($\delta^{13}C$) values of species and the annual precipitation gradient map represents that C_4 Chenopods are well adapted to regions with high degree of aridity and low precipitation.

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Key words. Chenopodiaceae, C₃ and C₄ species, Iran, geographic distribution mapping, GIS analysis.

الگوی پراکندگی گونه های C₃ و C₄ از تیره اسفناج در ایران

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پراکندگی جغرافیایی گونه های C_3 از تیره اسفناج در ایران با استفاده از ۱۱۱۵۱ رکورد ژئورفرنس شده واکاوی شد. تیره اسفناج با داشتن که جنس و ۲۰۶ گونه، به طور وسیعی در ایران انتشار یافته است. تعداد گونه های C_4 در این تیره تقریباً دو برابر گونههای C_5 می باشد. اطلاعات سازمان دهی شده در برنامه MS access به کمک نرم افزار DIVA-GIS و با استفاده از خانه گریدهای C_5 کیلومتر و محدوده دایرهای همسایه نقشه برداری شد. بالاترین تعداد گونههای این تیره با حداکثر C_5 گونه در هر خانه گرید، در استانهای تهران، سمنان و آذربایجان شرقی مشاهده شد. اما میزان جمعآوری از مناطق مختلف به طور مساوی صورت نگرفته است. همچنین این استانها به عنوان نقاط داغ گونههای C_5 گونههای C_5 شناخته شدند. مقایسه بین پراکندگی جغرافیایی مقدار C_5 گونهها و میزان بارندگی منطقهای حاکی از این است که گونههای به خوبی با مناطق خشک و نیمه خشک و میزان بارندگی یایین سازگاری یافتهاند.

INTRODUCTION

The photosynthetic pathway composition (C₃/C₄ fraction) of vegetation is a fundamental physiological and ecological distinction in many land cover classes (Tieszen et al. 1997). The distribution patterns of C₃ and C₄ plant species in relation to climate variables can provide direct evidences for the environmental conditions that favor a particular photosynthetic pathway. It is known that plants of two photosynthetic pathways, in addition to biochemical and physiological differences, exhibit distinct ecological characteristics

(Pyankov et al. 2010). The basic distinction between the two patterns of carbon dioxide fixation is in the first product of CO_2 fixation which is a C-3 molecule (phosphoglyceric acid) in case of C_3 plants and a C-4 molecule (oxaloacetic acid) in case of C_4 plants (Akhani et al. 1997). Moreover, the leaves of C_4 plants have specialized anatomical features that differ from those of C_3 plants. C_3 plants have a single type chloroplasts in their leaf mesophyll cells in which both light and dark reactions of photosynthesis take place, while in C_4 plants (except for *Bienertia* and

Borszczowia), photosynthetic processes are partitioned in two types of photosynthetic cells: the mesophyll cells and the Kranz or Bundle sheath cells (Brown 1975, Muhaidat et al. 2007, Kadereit et al. 2003). As C₄ photosynthesis is a CO₂ concentrating mechanism, it allows plants to perform well even when stomata need to be partially closed to reduce loss of water by transpiration. Therefore, the abundance of C₄ species is highly correlated with climate factors such as temperature, precipitation, and degree of aridity (Pyankov et al. 2010).

Since the discovery of C₄ photosynthetic pathway in the mid-sixties, numerous researches have been continuously performed on the quest for species with this carbon metabolism, and ecological comparisons between C₃ and C₄ plants. Many studies on the geographic distribution of C₃ and C₄ plants and its relation to climate variables have been performed on Central Asia (Pyankov et al. 1992a, 1997, Pyankov & Molotkovskii 1992, Pyankov & Vakhrusheva 1989), the Middle East (Shomer-Ilan et al. 1981, Akhani et al. 1997), Japan (Okuda & Furukawa 1990, Ueno & Takeda 1992), Europe (Collins & Jones 1985, Mateu1993, Pyankov et al. 2010), Africa (Ellis et al. 1980, Batanouny et al. 1988, Schulze et al. 1996), Australia (Hattersley 1983), North America (Teeri & Stowe 1976, Teeri et al. 1980), and South America (Powel & Still 2009).

Indeed, results of recent investigations about the distribution of C₃ and C₄ plants along latitudinal and altitudinal gradients suggest that climate factors alone or in combination with other ecological factors, are correlated with their abundances.

However, the correlation between the geographic distribution of plants and their photosynthetic pathways as well as the mapping of plant species according to photosynthetic pathways data have not been completely done yet in Iran. The present study provides an overview of the geographic distribution analysis of photosynthetic pathways among species Chenopodiaceae from Iran.

Ecologically and economically, Chenopodiaceae is one of the most important families in such inhospitable places as deserts, semi-deserts, and salt marshes. Chenopods are often important sources of forage for grazing livestock. Some of them also provide a useful source of fuel, while others have been used as a commercial source of potash or alkali. Many members of this family are succulent and late flowering and fruiting, which has historically made collections difficult to identify, with many specimens lacking the necessary characters for species identification (Hedge et al. 1997, Akhani et al. 2007). Here, we consider Chenopodiaceae as a separate family and do not integrate it in Amaranthaceae as proposed recently (APG II 2003, APG III 2009).

One of the important characteristics of this family, which correlates with the morphology, ecology, and even with taxonomy, is the types of photosynthetic pathways of the species. C₃ and C₄ photosynthetic pathways are frequently found in Chenopodiaceae (Carolin et al. 1975, Winter 1981, Pyankov et al. 1992a, Akhani et al. 1997, Kadereit et al. 2003). The family, which has the highest number of C₄ species among any of the dicot families, provides a valuable source of species to explore the variation and evolution of C₄ Kranz-anatomy and biochemical mechanisms in plants. Five types of Kranz-anatomy (Atriplicoid, Kranz-Suaedoid, Kochioid, Salsoloid, Conospermoid) and two C₄ biochemical subtypes (NAD-ME and NADP-ME) were described in this family (Carolin et al. 1975, Freitag and Stichler 2002). Furthermore, plants performing the C₃ and C₄ photosynthesis differ in their carbon isotopes composition (δ^{13} C) values. Because discrimination made by C₃ and C₄ plants in the initial fixation of carbon between the two carbon isotopes (12C and ¹³C), analysis of the photosynthetic products will help to identify the photosynthetic pathways by which these products have been made. (Nelson et al. 2006, Hobbie & Werner 2004).

Despite various studies on photosynthetic pathways of Chenopodiaceae species (e.g. Carolin et al. 1978, Winter 1981, Shomer-Ilan et al. 1981, Pyankov & Vakhrusheva (1989), Pyankov (1991), Akhani et al. 1997, Akhani & Ghasemkhani 2007, and Kadereit et al. 2003, 2012), there has been no comprehensive study on the geographic distribution pattern of C₃ and C₄ species of this important family in Iran which we try to provide in this contribution.

We used Geographic Information System (GIS) to analyze a large georeferenced database of locations where the species were observed. The advent of special geographical tools such as DIVA-GIS has made possible large scale studies in which variables such as species richness are mapped on a grid, and hotspots or centers are usually identified by eye. Species richness is used, because it is a simple, widely used, well understood, and is a useful measure of taxonomic diversity (Gaston 1996) and because it is less sensitive than diversity indices to the problems of unsystematic sampling intensities and procedures (Hijman et al. 2000). This type of study often face the risk of a bias related to uneven collection intensity, particularly oversampling in areas of high species richness and in easily accessible areas (Hijman et al. 2000, Reddy & Davalos 2003). So, it is worth noting that, the bias created from collecting in regions where the botanists

consider a large number of species, has to be considered when interpreting maps of species richness.

METHOD AND MATERIAL Data collection

We prepared a checklist of *Chenopodiaceae* species from different zones of Iran as complete as possible based on available data. The plants taxonomic and distribution data were gathered from four main sources: i) Flora Iranica (Hedge et al. 1997) which is the major taxonomic and nomenclatural reference for this study. ii) Flora of Iran published by Research Institute of Forests and Rangelands (Assadi 2001). iii) Data of recent articles published relevant to this family (Ghobadnejhad & al. 2003, Rahiminejad & al. 2004, Rahiminejad & Ghaemmaghami 2005, Akhani et al. 2005, Akhani 2008, Fuentes-Bazan et al. 2012). iv) Information obtained directly from Shiraz University Herbarium specimen labels. Data, which supply scientific name, locality, collector, collected number, date, distributional points and related coordinates for each Chenopodiaceae species, were organized in MS access database. The accepted name of species used for the analysis, followed Flora Iranica and Flora of Iran nomenclature. But in cases where there was taxonomical disagreement between the two sources, we refer to Tropicos (www.Tropicos.org) and IPNI (www.IPNI.org) nomenclature to select the best scientific name for species with regard to the synonymization of Chenopodiaceae taxa, proposed in the recent classification (Akhani et al. 2007, Fuentes-Bazan et al. 2012).

Most of the data sets didn't include coordinates. Therefore, we used Google Earth (http://earth.google.com/) to georeference the locations. At the putative collection site, site coordinates were added to the data set. Some 671 specimens from the original database which lacked geographic coordinates were excluded from the database. All geographic coordinates were then subjected to an error checking exercise using Map info. The final dataset used in this analysis contained 11151 accurately geo-referenced entries.

The classification of species as C_3 or C_4 plants was based on the results of previously published literature, especially the $\delta^{13}C$ analyses, biochemical studies and anatomical traits, such as Kranz anatomy. We mainly used the results of studies reported by Akhani and Ghorbanli (1993), Akhani et al. (1997), Pyankov et al. (2000b), Akan & Ghasemkhani (2007), and Kadereit et al. (2012). The photosynthetic types of some species that were not available from experimental studies were identified based on closely related species and plant taxonomy for Chenopods. However, the

photosynthetic pathways of few species are not known yet. Finally, the carbon isotope composition (δ^{13} C) values of 95% of the species were added to the database according to available published works (Akhani et al. 1997, Akhani & Ghasemkhani 2007).

Geographic distribution

The data were then imported into DIVA-GIS using its data import tools which requires latitude and longitude expressed in decimal degree format. The general software DIVA-GIS version 5.2 was used to produce potential maps. The distributions Chenopodiaceae species in Iran were projected and hotspots were identified. To eliminate border effects caused by assigning the grid origin, and to have a smoother surface and obtaining results less sensitive to small changes in the coordinate data (Hijman & Spooner 2001), we used circular neighborhood method. When the circular neighborhood option is chosen for analysis/point-to-grid/main, calculations are made based not on the observations within each grid cells, but rather on the observation found within a circle with its center in the middle of each grid cells and on the specified radius (Bonham-Carter 1994, Cressie 1991). We compared the number of observations and species using 10×10 km grid cells, and the circular neighborhood option with a radius of 25 km was applied to eliminate border effects due to grid origin assignment.

First, the number of different classes (Richness) method (Hijman et al. 2005) was used to map the distribution of species richness in order to identify hotspot regions. Hotspots are important basic units of extreme diversity that allow analysis of spatial patterns of biodiversity and permit prioritization of conservation activities and financing (Murray-Smith et al. 2008). To address this issue, the number of observation method (Hijman et al. 2005) was also used in DIVA to map the density of germplasm collections Chenopodiaceae species. Subsequently, C₄ species was mapped using the circular neighborhood point-to-grid richness analysis tool based on 10×10 km grid cells with a radius of 25 km. The same procedure was also used for C₃ species richness analysis. Finally, in order to represent the geographic occurrence of both C₃ and C₄ Chonopods on a map, the distribution pattern of species carbon isotope composition (δ^{13} C) values was mapped using point-to-grid statistics of DIVA-GIS, based on 10 by 10 km grid cells and 10 km radius in circular neighborhood method. In this statistical analysis, the mode (most frequent observations) of δ^{13} C values of the species was determined for each grid cell. The radius of "circular neighborhood" is reduced from 25 to 10 km, because there is a deep correlation

between δ^{13} C values of plants and ecological factors in their habitats. This neighborhood size reflects estimated areas in which two definite ranges of $\delta^{13}C$ values of plants may occur.

RESULTS

A total of 11151georeferenced datasets were added to Herbarium of Shiraz University (HSU) database. According to this database, the Chenopodiaceaehas 45 genera and approximately 204 species in Iran. Some of the largest genera are: Salsola L. with 39 species, Atriplex L. with 21 species, and Suaeda Scop with 16 species. Genera with one species are: Camphorosma L., Ceratocarpus L., Hablitzia M. B., Halocnemum M. B., C. Halostachys A. Mey. ex Schrenk, Krascheninnikovia Gueldenst., Microcnemum Ungern-Sternb., and Piptoptera Bunge. The first two grid based maps show the number of observations (Fig. 1) and species richness (Fig. 2) of Chenopodiaceae. The mapping of the density of plant collections for all Chenopodiaceae species (Fig. 1) shows the most highly sampled areas with > 477 unique accessions collected, which includes Tehran, Semnan, and Fars Provinces. Plant collection sites are concentrated in Tehran, Ghom, Semnan, Fars, and East Azerbaijan provinces. Other provinces have lower observations. Fig. 2 depicts the species richness using 10×10 km grid cells which are highlighted with different colors. The hotspots (the highest richness values) could be found in an area which includes Tehran and Semnan provinces. Protected areas such as Kavir protected region (Southeast of Tehran), Kavir National Park (West of Semnan), Kavir-e Meyghan (Northeast of Markazi), and Turan protected region (East of Semnan) also belong to the high species rich areas. An overlap of species range hotspot and areas of high observations occur in Tehran and Semnan provinces. On the other hand, Zagros highlands which belong to the west mountainous region of Iran with a very few plant collections, have the lowest number of species, and this area is defined as a limit for growing most of Chenopods. In addition, comparison of these two maps (Fig. 1 and Fig. 2) shows that some parts of Kerman, Khorasan-e- Razavi, and Esfahan provinces which are identified as species rich areas do not correspond to areas of high collections, and further collections in these provinces are required.

The 204 species records available at HSU database, were divided into two distinct groups based on their photosynthetic pathways: The first group includes 127 C₄ species with 8956 records and the second is 2784 records of 64 C₃ species. Photosynthetic types of 13 species are not known yet. According to our study, the

ratio of C₄ to C₃ Chenopodiaceae species in Iran is about 2:1. Our results are in agreement with the previous studies indicating that C₄ photosynthesis species in this family were found in the subfamily Salsoloideae, tribes Salsoleae and Suaedeae, and in tribes Atripliceae and Camphorosmeae of the subfamily Chenopodioideae (Osmond et al. 1980, Gamely 1985, Pyankov et al. 1992a, 1997, Akhani et al. 1997).

The grid based map of C₄ species richness (Fig. 3) represents a main hotspot which occurs in Tehran, Ghom, and Semnan provinces. Arid zones of Esfahan, Fars, Kerman, and Khorasan-e-Razavi Provinces in Irano-Turanian region are also rich in terms of C₄ Chenopods. From fig. 4, it is clear that the highest number of C3 species concentrates into two distinct centers which include Tehran and East Azerbaijan provinces. Fars, Golestan, and West Azerbaijan are also defined as high C3 species rich provinces. Moreover, the overlap of C₃ and C₄ species richness hotspots occurs in central and west regions of Tehran.

Afterwards, the point-to-grid statistical analysis of δ^{13} C values of species were used to show the distribution pattern of C₃ and C₄ species in green and red cells (Fig. 5). This analysis represents where C₃ and C₄ Chenopods are found on the map, but not their richness or hotspots. Green cells with δ^{13} C ranging from a minimum negative of -20 ‰ to a maximum negative of -30 ‰ are related to C₃ species. They are sparsely distributed in different regions of Iran, particularly in northern regions along the Caspian Sea. A few number of C₃ species are also observed in arid zones of Irano-Turanian region. On the other hand, red cells in which their δ^{13} C values fall between -10 ‰ and -15 ‰ are the indicators of C₄ species. Chenopods with C₄ photosynthesis are widely distributed in Iran especially in arid and hyper-arid zones of Irano-Turanian lowlands and Saharo-Sindian region, and on floristic basis, comprise a higher percentage of all terrestrial flora of the country.

Results from the δ^{13} C values distribution analysis show a correlation between the geographic distribution pattern of C₃ and C₄ species of Chenopodiaceae and the regional precipitation gradients. It is obvious from the comparison between the data given in Fig. 6 and the regional precipitation map (Fig. 6) that the grid cells of C₃ species are associated with relatively high rates of precipitation, and C4 species are more distributed in arid regions with lower precipitation gradients.

DISCUSSION

This is the first report in which the C₃ and C₄ species of

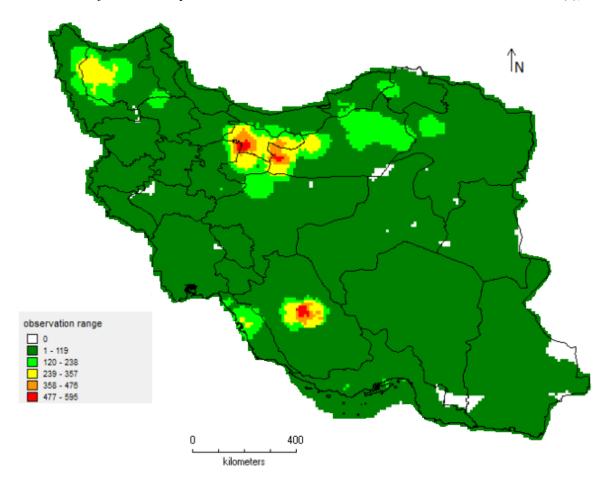


Fig. 1. Number of observations of *Chenopodiaceae* species per 10×10 km grid cell. A circular neighborhood with a radius of 25 km was used to assign observations to a grid cell.

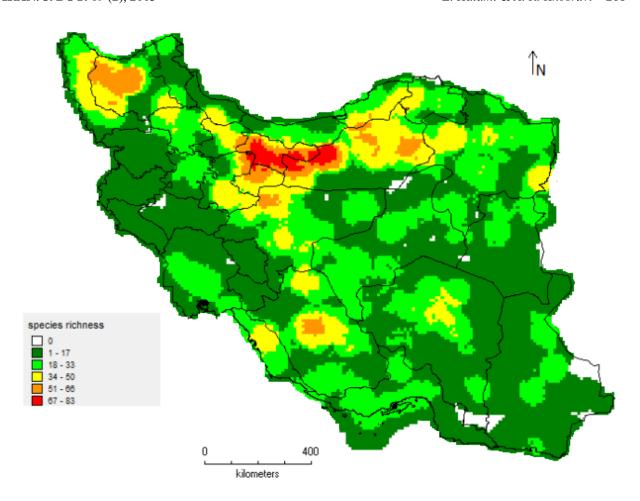


Fig. 2. Number of *Chenopodiaceae* species per 10 × 10 km grid cell. A circular neighborhood with a radius of 25 km was used to assign observations to a grid cell.

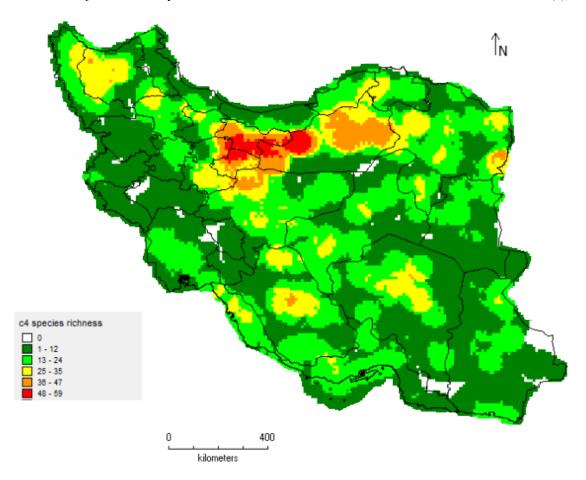


Fig. 3. Number of C_4 Chenopodiaceae species per 10×10 km grid cell. A circular neighborhood with a radius of 25km was used to assign observations to a grid cell.

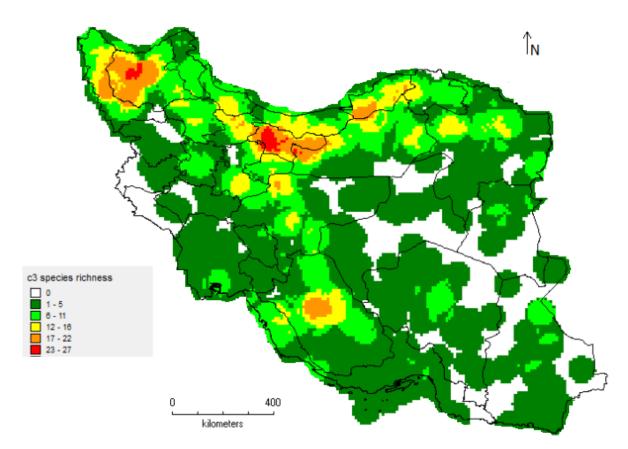


Fig. 4. Number of C_3 Chenopodiaceae species per 10×10 km grid cell. A circular neighborhood with a radius of 25 km was used to assign observations to a grid cell.

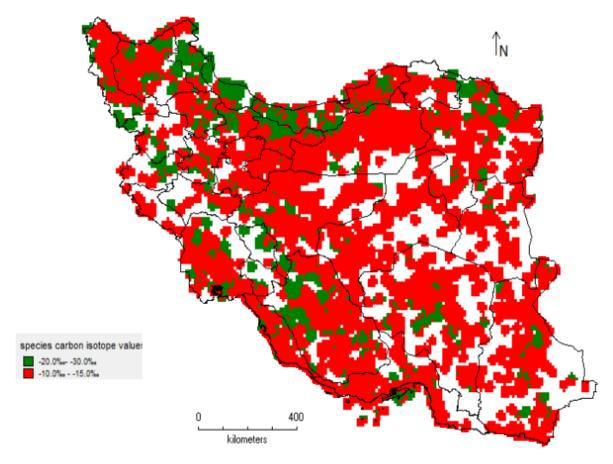


Fig. 5. Chenopodiaceae species $\delta^{13}C$ values mapped in 10×10 km grid cells. A circular neighborhood with a radius of 10 km was used to assign observations to a grid cell.

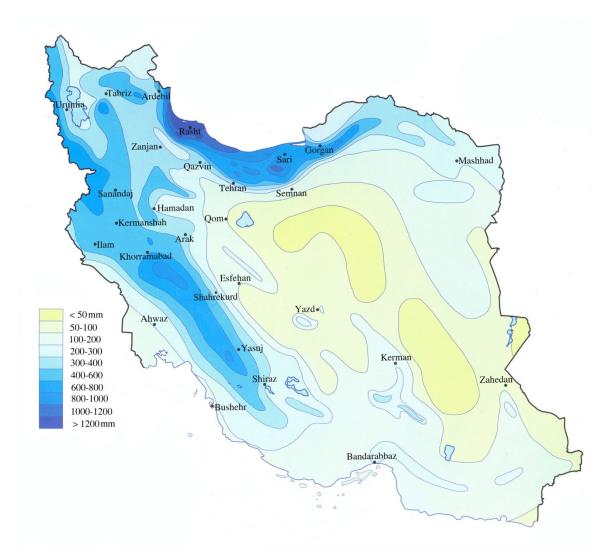


Fig. 6. Mean annual rainfall in Iran (adopted from best-forum.ir)

Chenopodiaeae family in Iran are systematically analyzed. Records of Chenopodiaceae species with their taxonomic and distribution data were organized in MS access database and the analyses were performed by personal computer using DIVA-GIS, whichprovides a visual representation of species distribution. Some of our records are recent, but many data, which are mentioned only in available Floras, date back for many years. Even in some cases, the habitat in which the species has occurred in the past has now been disappeared and the species may no longer be present there. But, these do not have much effect on the whole results. If the observations (specimens observed) would extend to the local herbaria in cities such as Esfahan, Kerman, and Tabriz, it would have impact on the results. Furthermore, data obtained from herbaria are of special values since permanently preserved specimens

can be physically examined and re-examined on many occasions, and any comments about the identification made can be noted (Hall 1994). Although specimen databases are extremely valuable, there are quality problems associated with collections data, that may limit the usefulness of products generated from these data. These include geographical bias (more accessible areas are sampled preferentially), taxonomic bias (greater representation of easy-to-study species), and temporal bias (records are based on collections during one season only).

Since, provinces are of different shapes and areas; we used equal-area grid cells to set up distribution of the species. Using small grid sizes have both high resolution and low geographic sampling bias (Mousavi & Khosravi 2010). The circular neighborhood approach produces a smoother grid that is less biased

by the origin of the grid and less sensitive to errors in coordinate data than standard grid cell approaches (www.diva-gis.org).

Results obtained in this study indicate that Chenopodiaceae species are distributed in almost all provinces of Iran mainly in such inhospitable places, such as deserts, semi-deserts, salty soils, and marshes. But regions with high altitude such as highlands of Zagros mountain ranges are defined as unsuitable habitats for most of Chenopodiaceae species. Tehran and Semnan provinces with the highest number of observations were also identified as the species range hotspots. Therefore, these areas are the best regions for collecting Chenopodiaceae. In addition, the low collections in provinces such as Esfahan, Kerman, and Khorasan-e Razavi which have high species richness should be given the top priority for future collections. It is possible that further explorations would result in discovering additional species, and solve the condition of nonrandom spatial distribution of species observation point. So, this type of study is valuable for identifying biodiversity hotspots, prioritization regions for conservation actions, and identifying gaps in collections. However, the main problem with such occurrence data is that the intent and methods of collecting are rarely known, so that, absences cannot be inferred with certainty. Due to the vast area of Iran, we know that the sample effort is not consistent throughout the range. Some regions could be richer than others, because they are more considerable and accessible for the botanists, and others could be considered poor in number of species because they are rarely or never sampled. Keeping this in mind, it may be assumed that the hotspot located in Tehran and Semnan is biased by high sampling effort in these provinces. But we do not think that high number of species follows casually from high number of observations.

Our study emphasizes the relative cover abundances of C₃ and C₄ Chenopodiaceae species to interpret their geographic distribution. These two photosynthetic types have been segregated geographically with C4 plants occupying warmer and more arid areas. The findings of the present study are consistent with those reported by most other investigators: indices of C₄ species performance show a general pattern of increase toward warmer and drier environment, an apparent distributional reflection of physiological characteristics of photosynthetic pathway (Wentworth Chenopodiaceae is the leading family in abundance of C₄ species in Iran (Borchers et al. 1982). Most of diversification of C₄ Chenopods occurs in central deserts of Iran in a wide range of habitats from hypersaline to extremely arid conditions and they are

very important ecologically in saline and arid areas, like Irano-Turanian lowlands. The high proportion of C₄ Chenopods in Irano-Turanian lowlands is very interesting. Irano-Turanian region is characterized climatologically by continentally low precipitation, hot and dry summer, and a cold and harsh winter (Zohary 1973). Moreover, the less negative δ^{13} C values of C₄ species correspond to hot and arid conditions of their habitats. On the contrary, plants with the C₃ pathway cannot survive in such arid zones. They have higher rates of photosynthesis at low temperatures and irradiances, and the carbon isotope values in C₃ species are negatively correlated with water use efficiency of plants (Akhani 2008). According to our analyses (Fig. 4, Fig. 5), C₃ Chenopods are more abundant in humid areas along Caspian Sea from Golestan to West Azerbaijan provinces. But, sometimes the relative abundances of C₃ succulent desert Chenopods increase along the aridity gradient. A great number of C₃ species don't grow in natural or semi-natural ecosystems where they are directly exposed to the climatic parameters, but as weeds in irrigated fields or on ruderal places where climatic factors are more or less buffered. However, when both types occur in the same region, like in Tehran province in this study, they may be segregated along specific microenvironments because microclimatic, edaphic, or biotic Furthermore, ecological systems in areas characterized by marked seasonal differences in temperatures and precipitation will consist of mixtures of C3 and C4 species which will show a temporal separation in primary production. Comparison of Fig. 5 and Fig. 6 represents that regional precipitation gradients also impact the relative abundance of each photosynthetic pathway, and thus of vegetation δ^{13} C composition. It is also evident that the spatial patterns of the vegetation carbon isotope content (δ^{13} C), directly follow from the C₃/C₄patterns. As a result, the balance between C₃ and C₄ vegetation is believed to be potentially very responsive to climate changes, and the ecosystem functions potentially very responsive to changes in the C₃ and C₄ balance (Ehleringeret al. 1997). So, future studies should enhance the analytical tools and approaches to evaluate C₃ and C₄ plant species distribution and the related information, especially their habitats and physiological conditions.

As hot and arid conditions have favored C_4 plants evolution, increase in the abundance of C_4 plants might be the first signs of increasing aridity and desertification in a region. Unfortunately, desertification has a high rate in arid and semi-arid countries such as Iran (Zehtabian et al. 2005). We would like to propose reliable scientific and sociological means for remediating, restoring, and

balancing the management of deserts as stable productive economic ecosystems for Furthermore, mismanagement of some protected areas which occur in saline and arid regions has caused the destruction of some previously well conserved sites. The Turan and Kavir protected regions are the outstanding examples (Akhani & Ghorbanli 1993). Therefore, it is recommended to direct more attention toward these protected areas in the future.

As a final point, we suggest continuing researches, collections, identification of specimens, and further distributional studies for different groups of plants in Iran. An initial step would be to digitalize herbaria localities and to present them by their coordinates.

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