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Distribution pattern of *Caragana* species under the influence of climate gradient in the Inner Mongolia region, China

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Abstract: There is a strong climate gradient in the Inner Mongolia region, China, with solar radiation and air temperature increasing but precipitation decreasing gradually from the northeast to the southwest. Sixteen *Caragana* species exist in the Inner Mongolia region. These *Caragana* species exhibit a distribution pattern across moisture zones and form a geographical replacement series. In order to examine the mechanisms responsible for *Caragana* species distribution pattern, we selected 12 *Caragana* species that exhibit a distinct distribution pattern across multiple moisture zones in the Inner Mongolia region, and determined the relationships between the leaf ecological and physiological traits of these *Caragana* species and the aridity index and solar radiation. Along with the climatic drought gradient and the solar radiation intensification from the northeast to the southwest, leaf ecological characteristics of *Caragana* species changed drastically, i.e. the leaf shape gradually turned from flat into tegular or tubbish; the leaf hair became denser, longer and lighter in color; the leaf area, leaf biomass and specific leaf area (SLA) decreased significantly; the leaf thickness and the ratio of leaf thickness to leaf area increased significantly; and the leaf chlorophyll content decreased significantly. As the climatic drought stress increased, osmotic potentials of the main osmotic adjustment substances and the cytoplasmic ion concentration of *Caragana* species increased significantly. Meanwhile, the total and free water contents and water potential of leaves decreased significantly; the ratio of bound to free water increased significantly; the stomatal conductance and transpiration rate reduced significantly; and the water use efficiency (WUE) increased significantly. In addition, with the intensification of climatic drought stress, peroxidase (POD) and superoxide dismutase (SOD) activities in leaves increased significantly. As a result, the malondialdehyde (MDA) content increased while the oxygen free radical content decreased. Our results showed that most of the leaf ecological and physiological traits of the 12 *Caragana* species varied in accordance with the climatic drought gradient in the Inner Mongolia region, which reflected the adaptation of the *Caragana* species to the local climate conditions. With relatively more active metabolism and faster growth, the *Caragana* species in the northeast had strong competitive abilities; on the other hand, with stronger resistance to climatic drought stress, the *Caragana* species in the southwest could survive in harsh environments. Based on our results, we concluded that both the environmental gradients and the adaptive responses of *Caragana* species to their environments played important roles in the formation of the *Caragana* species distribution pattern across the Inner Mongolia region.

Keywords: adaptation; climatic gradient; morphological trait; osmotic potential; water state; water metabolism; antioxidative enzyme

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Distribution pattern of plant species can be shaped by environmental factors, such as precipitation gradient (Pyke et al., 2001; Zhu et al., 2007), drought (Eeley et al., 1999), moisture and temperature gradients (McKenzie et al., 2003; Zhu et al., 2007), length of dry period, cumulative water deficit (Bongers et al.,

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1999), nocturnal warming (D'Odorico et al., 2013), habitat-mediated differences (Palmiotto et al., 2004), regional climate (Schmitt et al., 2013), belowground resource availability (John et al., 2007), and soil properties (Sollins, 1998). Meanwhile, it can also be influenced by the characteristics of plant species, such as drought tolerance (Engelbrecht et al., 2007; Comita and Engelbrecht, 2009) and plant responses to climate gradient (Retuerto and Carballeira, 2004). In short, distribution pattern of plant species reflects the evolutionary response of the species to the long-term environmental change (Thompson et al., 2013).

The climate in the Inner Mongolia region is characterized by a gradual increase in solar radiation and air temperature, and a gradual decrease in precipitation from the northeast to the southwest (Yang et al., 1987). The decrease in precipitation together with an increase in evaporation results in a climatic drought gradient from the northeast to the southwest. In the Inner Mongolia region, there are seven moisture-based zones from the northeast to the southwest: humid, sub-humid, semi-arid, droughty, very droughty, intensively droughty and extremely droughty zones. In accordance with these moisture-based zones, there are seven vegetation types: forest, meadow steppe, typical steppe, desert steppe, steppe desert, typical desert and extremely arid desert, respectively. With the climatic drought gradient, the Inner Mongolia region provides an ideal area for studying plant species adaptation to variable moisture conditions.

There are 16 *Caragana* species (deciduous shrubs) distributed in the Inner Mongolia region (Zhao, 1991). They exhibit a distribution pattern across moisture zones and form a geographical replacement series (Zhao, 1991; Zhou, 1996; Ma et al., 2012). Zhou (1996) studied the *Caragana* species in the Inner Mongolia region and suspected that the distribution pattern of these species in this region might be shaped by drought gradient, but the author did not demonstrate evidences in the aspects of climate gradient and corresponding plant traits. Examining the relationships between the climate gradient and the traits of *Caragana* species is critical in understanding the formation of *Caragana* distribution pattern in the Inner Mongolia region. However, very few studies have explicitly addressed this topic. In this study, we selected 12 *Caragana* species that exhibit a distinct distribution pattern across the moisture zones in the Inner Mongolia region, and determined the relationships between the leaf ecological and physiological traits of these *Caragana* species and the aridity index and solar radiation. The aim of this study is to illustrate the mechanisms, in the aspects of climate gradient and plant traits, responsible for *Caragana* species distribution pattern across the Inner Mongolia region.

1 Study area and methods

1.1 Study area and plant species

Inner Mongolia region is located in the northern China (97°12′–126°04′E, 37°24′–53°23′N; 1,000 m asl), with a distance of 2,400 km from the east to the west and 1,700 km from the north to the south within the region. The total area of Inner Mongolia region is $1,183\times10^3$ km^2, accounting for 12.3% of the total land area of China. Inner Mongolia region is characterized by a temperate continental monsoon climate, with relatively less and unevenly distributed precipitation, strong wind and dramatic seasonal changes. The mean annual precipitation in the whole region is 50–450 mm, which decreases from the northeast to the southwest gradually. The average annual sunshine duration in the region is more than 2,700 h.

In this study, we selected 12 *Caragana* species that exhibit a distinct distribution pattern across the moisture zones as the target species. The distribution and morphological characteristics of these *Caragana* species are shown in Table 1. Field surveys on the 12 *Caragana* species were conducted at 14 study sites in the Inner Mongolia region in July of 2003, 2007 and 2012 (Fig. 1 and Table 2).

1.2 Measurements of leaf ecological and physiological traits

At each site, we set up three sampling plots (circle plots with a diameter of 50 m and distances of 20–40 km between them) for each *Caragana* species. In each plot, we took the following measurements on mature leaves from 30 *Caragana* plants (15–25 years old) to measure their leaf ecological and physiological traits.

Table 1 Distribution and morphological characteristics of the 12 *Caragana* species in the Inner Mongolia region

Species name	Distribution	Moisture type of distribution zone	Leaf shape	Leaf hair	Leaf hair color
C. sibirica	Forest and forest-steppe zones close to the northeast border	Humid to sub-humid	Flat	Few short	Bottle green
C. opulens	Shrubbery on the slope of Daqing Mountain in the mid-south	Semi-arid	Flat	Few short	Bottle green
C. microphylla	Meadow steppe and typical steppe in the northeast	Sub-humid to semi-arid	Flat	Few short	Bottle green
C. davazamcii	Desert steppe and steppe desert from the middle to the mid-west	Droughty to very droughty	Flat	Medium	Light green
C. brachypoda	Desert steppe, steppe desert and typical desert from the mid-west to the southwest	Droughty to very droughty to intensively droughty	Tegular	Medium	Bottle green
C. pygmaea	Desert steppe in the middle and mid-west	Droughty	Tegular	Medium	Light green
C. tibetica	Steppe desert and typical desert in the southwest	Very droughty to intensively droughty	Tubbish	Densely silky	Light green
C. stenophylla	Typical steppe, steppe desert and typical desert form the middle to the southwest	Semi-arid to very droughty to intensively droughty	Tegular	Medium	Bottle green
C. korshinskii	Steppe desert and typical desert in the southwest	Very droughty to intensively droughty	Flat	Densely fallen silky	Bright gray
C. roborovskyi	Typical desert and extremely arid desert in the southwest	Intensively droughty to extremely droughty	Flat	Densely straight silky hair	Bright gray
C. jubata	Shrubbery on the north slope of Helan Mountain in the southwest	Intensively droughty	Flat	Densely fallen silky	Bright gray
C. leucophloea	Typical desert and extremely arid desert in the southwest	Intensively droughty to extremely droughty	Tegular	Silky	Light green

The area of 100 leaves from 30 *Caragana* plants was measured using the CI-203 Portable Laser Area Meter (CID Co., USA), and the thickness of 100 leaf blades was measured with a vernier caliper. The biomass of 100 leaves was measured after drying, and then the specific leaf area (SLA) was calculated. Fresh leaves (2 g) were collected at 12:00–14:00 to determine the chlorophyll content (for methods and references, see Table 3).

In each plot, leaf samples (15 g) were collected at 12:00–14:00 and stored in liquid nitrogen in the laboratory to measure the osmotic potentials of the main osmotic adjustment substances (for methods and references, see Table 3). Osmotic potential was calculated according to the formula: $\psi s=RT\times ns/Vw$. Where ψs is solute potential, R is gas constant, T is temperature in degrees K, ns is the number of moles of solutes, and Vw is the total water volume of the cell (i.e. leaf water content; Peltier et al., 1997).

Fresh leaves (2 g) were collected at 12:00–14:00 in each plot to determine the cytoplasmic ion concentration. These fresh leave samples were cut into small pieces and mixed well, then 0.2 g subsample from the mixture was put into a tube with 10 ml of distilled water, and the tube was boiled for 15 min to enable adequate diffusion of ions in cell into the water. We measured the electrical conductivity of the solution with a conductivity meter. The total water in leaves was treated as solvent to calculate the cytoplasmic ion concentration (expressed as electrical conductivity).

In each plot, we also collected fresh leaves (2 g) at 12:00–14:00 to determine the total, free and bound water contents and water potential (for methods and references, see Table 3).

The net photosynthesis, transpiration and stomatal conductance were measured using the LI-6400 Portable Photosynthesis System (LI-COR Co., USA) every 2 h from 07:00 to 19:00. We measured each plant for a total of 30 measurements per cycle (2 h), and then calculated the mean values of net photosynthesis, transpiration and stomatal conductance, respectively. Water use efficiency (WUE) was

Table 2 Environmental data for the study sites

Study site	Longitude (E)	Latitude (N)	Altitude (m)	Annual precipitation (mm)	Annual average temperature (°C)	Sunshine duration (h)	Soil water content (1–100 cm; %)	Aridity index (I_{dm})	Vegetation type
Zhalantun	122°48′02″	48°00′00″	482	480	2.40	2,800	9.05	38.71	Forest
Hailar	119°23′04″	49°07′02″	650	350	–2.20	2,800	2.73	44.87	Forest-steppe zone
Xiwu	117°36′00″	44°34′08″	996	340	1.47	2,870	4.09	29.64	Typical steppe
Xilinhot	116°18′00″	43°34′02″	990	281	2.35	2,932	3.70	22.75	Typical steppe
Abaga	114°58′02″	44°04′00″	1,126	245	1.20	3,047	3.09	21.88	Desert steppe
Suniteyou	112°57′00″	42°28′02″	1,151	211	4.93	3,067	2.40	14.13	Desert steppe
Erenhot	111°54′08″	43°22′08″	966	142	3.40	3,055	2.68	10.60	Desert steppe
Horinger	111°52′08″	40°19′08″	1,063	420	5.60	2,942	9.21	26.92	Mountain slope shrubbery
Damao	110°25′08″	41°42′00″	1,377	256	4.01	3,061	2.16	18.27	Desert steppe
Wulatezhong	108°31′02″	41°34′02″	1,288	199	5.07	3,168	2.18	13.21	Steppe desert
Otog	107°58′02″	39°07′02″	1,500	265	6.40	3,050	2.56	16.16	Steppe desert
Hangjinhou	107°30′00″	41°15′00″	1,285	150	5.82	3,179	2.10	9.48	Steppe desert
Alashanzuo	105°40′08″	38°51′00″	1,561	110	7.80	3,200	1.73	6.18	Typical desert
Alashanyou	101°40′08″	39°12′00″	1,400	72	8.40	3,105	1.68	3.91	Extremely arid desert

Note: Climate data are the mean values from 1961 to 2000; and soil water content data are the mean values from 3–6 sampling plots. Aridity index (I_{dm}) was calculated using de Martonne method (Meng et al., 2004).

calculated according to the formula: WUE=daily average value of net photosynthesis/daily average value of transpiration.

For each plot, leaf samples (2 g) were collected at 12:00–14:00 and stored in liquid nitrogen in the laboratory to measure the antioxidative enzymes (peroxidase (POD), catalase (CAT) and superoxide dismutase (SOD)) activities and cell injury indices (malondialdehyde (MDA) content and oxygen free radical content). The measurement methods and references of these variables are shown in Table 3.

Soil gravimetric water contents at depths of 30, 60 and 100 cm in each plot were measured respectively.

1.3 Statistical analysis

The mean value of each variable for each plot over 2003, 2007 and 2012 was calculated, and then these mean values were used in the statistical analyses. Each variable was analyzed using one-way ANOVA with species as factor and plot as replicate. Correlation analyses were performed between each variable (mean value for each species) and aridity index. The mean aridity index of the 14 study sites was 3.91–41.79. In addition, correlation analyses were also conducted between chlorophyll content and local annual sunshine duration. The mean annual sunshine duration in the 14 study sites ranged from 2,800 to 3,200 h. All the statistic analyses were performed using SPSS 16.0.

2 Results

2.1 Leaf ecological traits

Along with the climatic drought gradient and the intensification of solar radiation from the northeast to

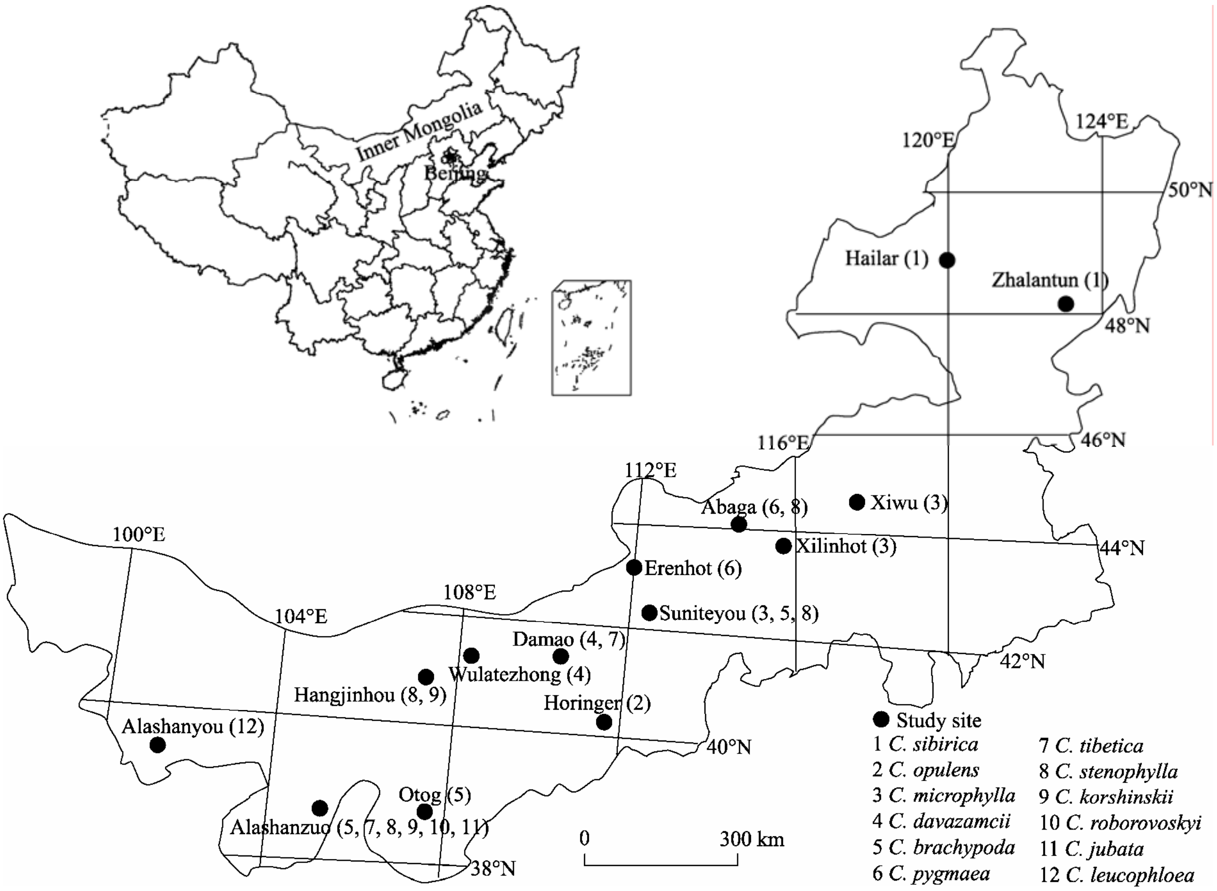

Fig. 1 Study sites and 12 *Caragana* species in the Inner Mongolia region

Table 3 The variables measured and methods (or references) used in the extermination of leaf physiological traits

Variable	Method or reference
Chlorophyll content	Lichtenthaler et al., 1996
Soluble protein content	Bradford, 1976
Free amino acid content	Lee and Takahashi, 1966
Organic acid content	Sinclair et al., 1945
Soluble sugar content	Buysse and Merckx, 1993
K^+, Na^+, Ca^{2+}, Mg^{2+}, Cu^{2+}, Zn^{2+} and Mn^{2+} contents	Atomic Absorption Spectr-photometer
Cl^- content	Schales and Schales, 1941
NO_3^- and NO_2^- contents	Marzinzig et al., 1997
Total, free and bound water contents	Pissis et al., 1987
Water potential	HR-33T dew point instrument
Peroxidase (POD) activity	Naveh et al., 1981
Catalase (CAT) activity	Goldblith and Proctor, 1950
Superoxide dismutase (SOD) activity	Beyer Jr and Fridovich, 1987
Malondialdehyde (MDA) content	Agarwal and Chase, 2002
Oxygen free radical content	Bors et al., 1978

the southwest, the leaf shape of different *Caragana* species in the study sites gradually turned from flat into tegular or tubbish (Table 1). With the intensification of drought and solar radiation, the leaf area decreased (F=419.719, P<0.01), while the leaf thickness increased significantly (F=15.307, P<0.01), and the ratio of leaf thickness to leaf area also increased significantly (F=93.405, P<0.01). Additionally, all these three variables were significantly correlated with aridity index (Figs. 2a–c). The leaf biomass (F=139.473, P<0.01) and SLA (F=55.169, P<0.01) decreased significantly with the drought intensification. Moreover, the two variables were also significantly correlated with aridity index (Figs. 2d and e). Thus, the light absorbent area and transpiration area of leaf reduced gradually with the intensification of climatic drought stress and solar radiation.

As the climatic drought stress and solar radiation increased, the foliar hair of *Caragana* species became denser, longer and lighter in color from the northeast

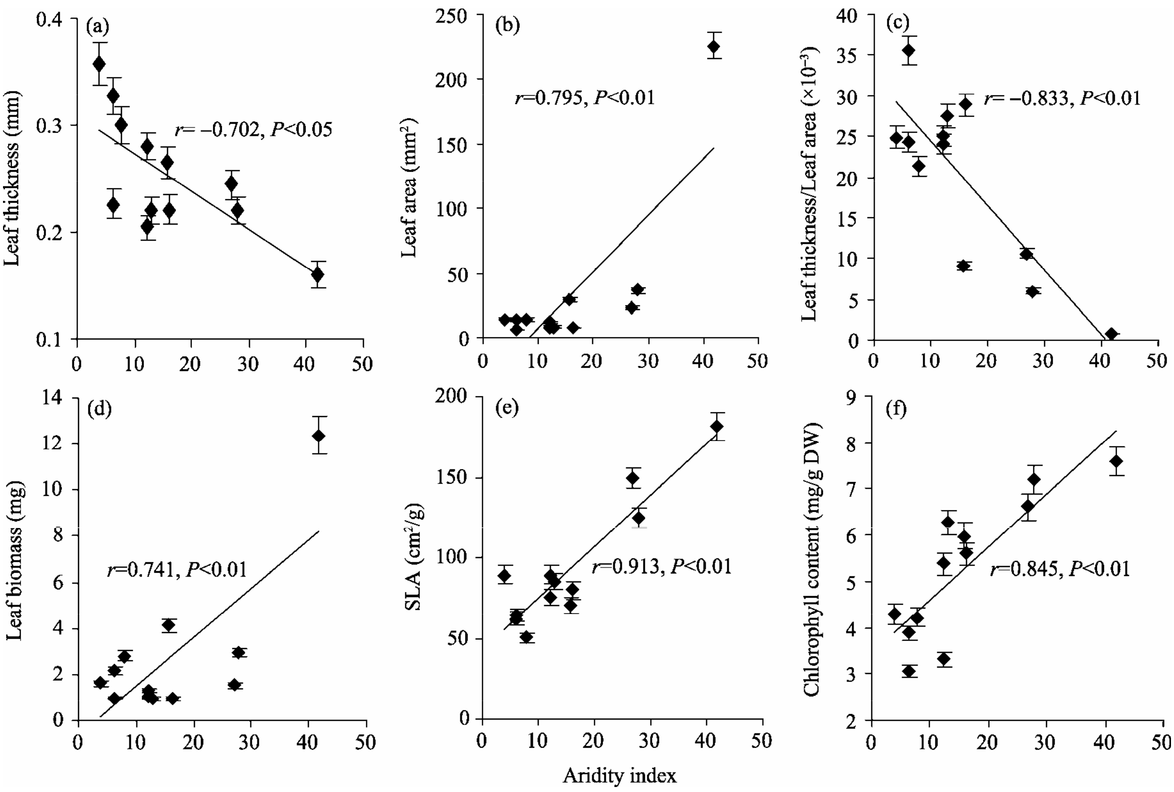

Fig. 2 Correlations between leaf ecological traits ((a), leaf thickness; (b), leaf area; (c), leaf thickness/leaf area; (d), leaf biomass; (e), SLA (specific leaf area); (f), chlorophyll content) of *Caragana* species and aridity index (mean±SE)

to the southwest (Tables 1 and 2). The chlorophyll content in leaves decreased gradually with the intensification of climatic drought stress and solar radiation (F=43.462, P<0.01), showing a significant positive correlation with aridity index (r=0.845, P<0.01; Fig. 2f) and a significant negative correlation with annual sunshine duration (r= –0.832, P<0.01). Thus, the light-reflecting ability of leaf in *Caragana* species gradually became stronger with the increases of climatic drought stress and solar radiation.

2.2 Leaf osmotic potentials of the main osmotic adjustment substances and cytoplasmic ion concentration

There were no clear relationships between the osmotic potentials of soluble protein, free amino acid and organic acid with the climatic drought gradient (Figs. 3a and b). While, as the climatic drought stress increased from the northeast to the southwest, the osmotic potentials of soluble sugar (F=12.593, P<0.01) and inorganic ions (K^+, F=15.550, P<0.01; Cl^-, F=21.727, P<0.01; Ca^{2+}, F=32.954, P<0.01; Mg^{2+}, F=27.755, P<0.01; Na^+, F=14.553, P<0.01; NO_3^- and NO_2^-, F=2.279, P=0.052; Cu^{2+}, F=2.199, P=0.059; Zn^{2+}, F=17.718, P<0.01; Mn^{2+}, F=4.476, P<0.01) in the leaves of *Caragana* species increased significantly. The osmotic potentials of soluble sugar and inorganic ions (except for Zn^{2+}, P>0.05) and the total osmotic potential showed significant negative correlation with aridity index (Figs. 3c–h). The cytoplasmic ion concentration in *Caragana* species leaves increased significantly (F=87.661, P<0.01) with the increase of climatic drought stress, and there was a significant negative correlation between cytoplasmic ion concentration and aridity index (Fig. 3i).

2.3 Leaf water states and water potential

With the increase of climatic drought stress from the northeast to the southwest, the total and free water contents of *Caragana* species leaves decreased significantly (F=85.835, P<0.01 and F=9.913, P<0.01, respectively), whereas the bound water content tended to increase (F=1.473, P=0.215). The total and free

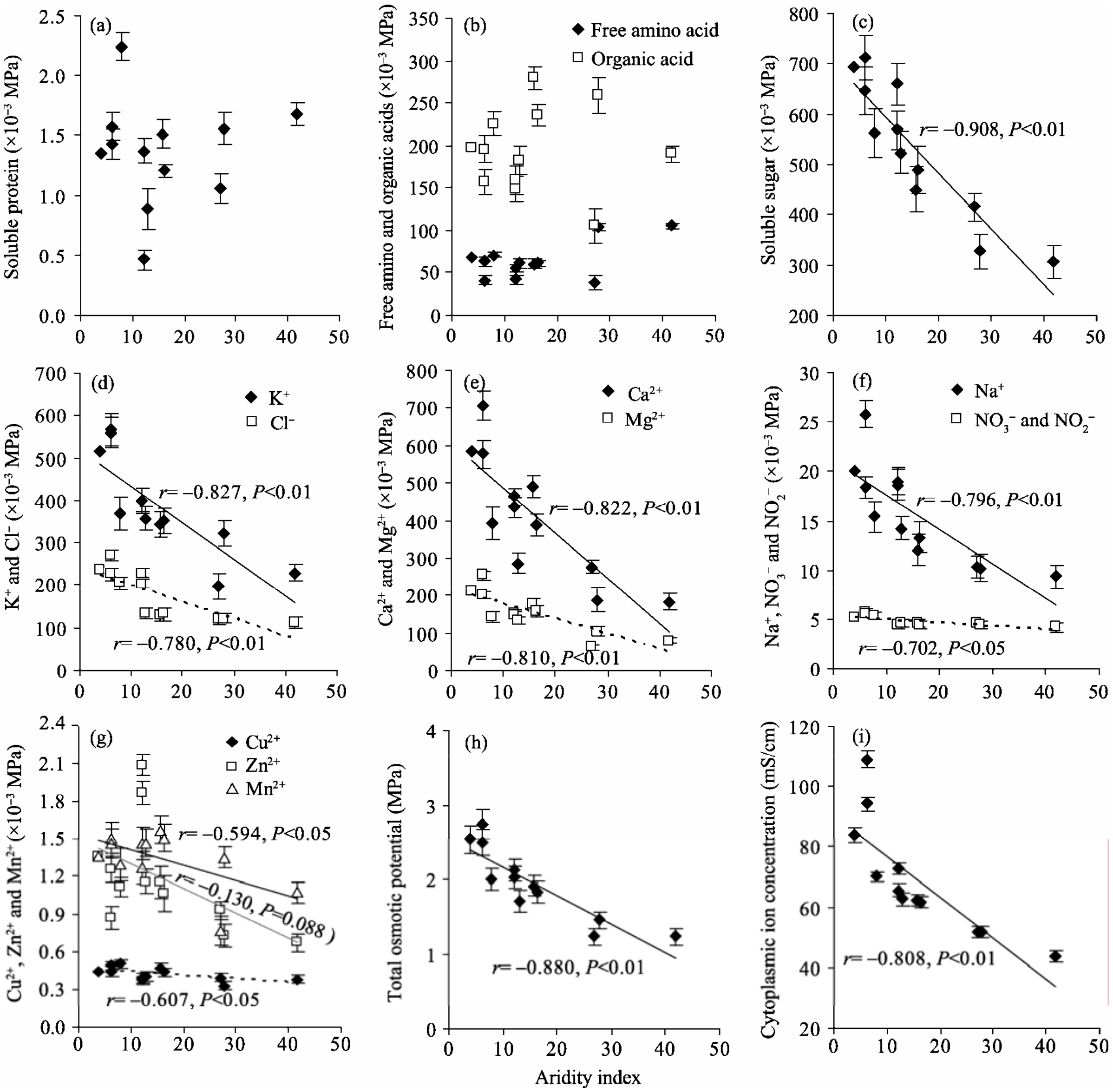

Fig. 3 Correlations between osmotic potentials of the main osmotic adjustment substances ((a), soluble protein; (b), free amino and organic acids; (c), soluble sugar; (d), K^+ and Cl^-; (e), Ca^{2+} and Mg^{2+}; (f), Na^+, NO_3^- and NO_2^-; (g), Cu^{2+}, Zn^{2+} and Mn^{2+}; (h), total osmotic potential) and cytoplasmic ion concentration (i) of *Caragana* species leaves and aridity index (mean±SE)

water contents were significantly positively correlated with aridity index, but the bound water content had a negative correlation with aridity index (Figs. 4a and b). The ratio of bound to free water content decreased significantly with the increase in aridity index (F=68.096, P<0.01; Fig. 4c), while the water potential increased significantly with the increase of aridity index (F=80.267, P<0.01; Fig. 4d).

2.4 Leaf water metabolisms

With the increase of climatic drought stress from the northeast to the southwest, the stomatal conductance (F=8.189, P<0.01) and transpiration rate (F=8.657, P<0.01) in leaves of *Caragana* species decreased significantly, and they both showed significant positive correlations with aridity index (Figs. 5a and b). It seemed that the ability of *Caragana* species to reduce water loss through stomatal adjustment increased gradually with the increase of climatic drought stress. The WUE in leaves of *Caragana* species increased significantly with the increase of climatic drought stress (F=13.345, P<0.01), presenting a significant negative correlation with aridity index (Fig. 5c).

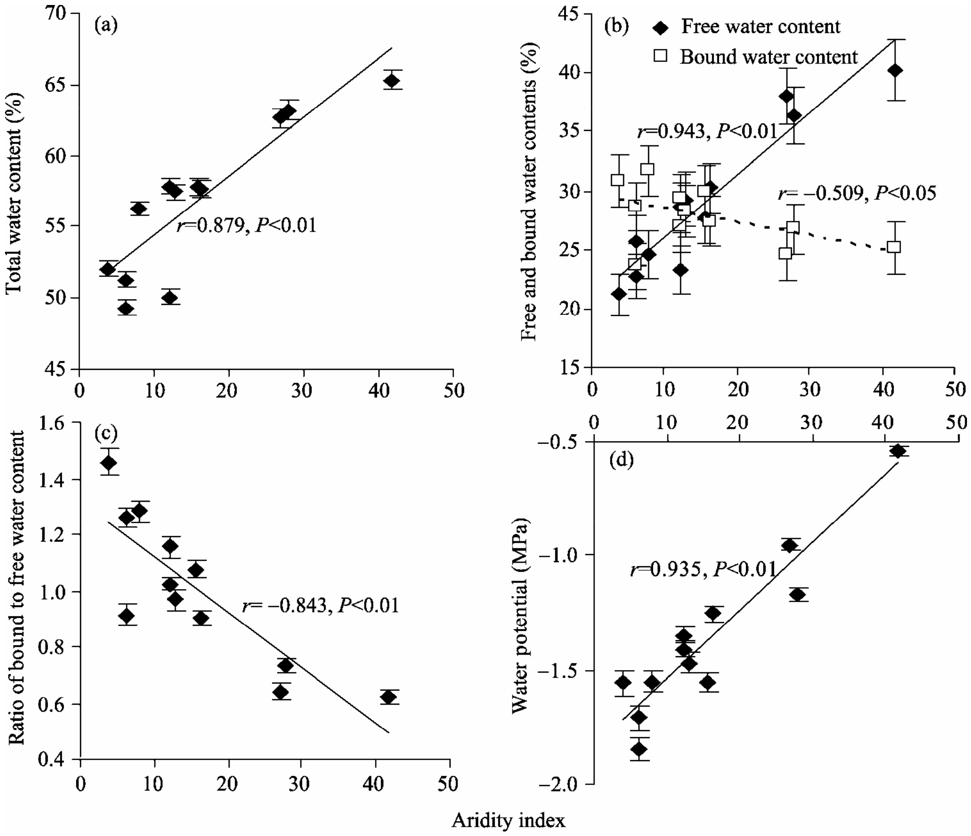

Fig. 4 Correlations between leaf water states ((a), total water content; (b), free and bound water contents; (c) ratio of bound to free water content) and water potential (d) of *Caragana* species and aridity index (mean±SE)

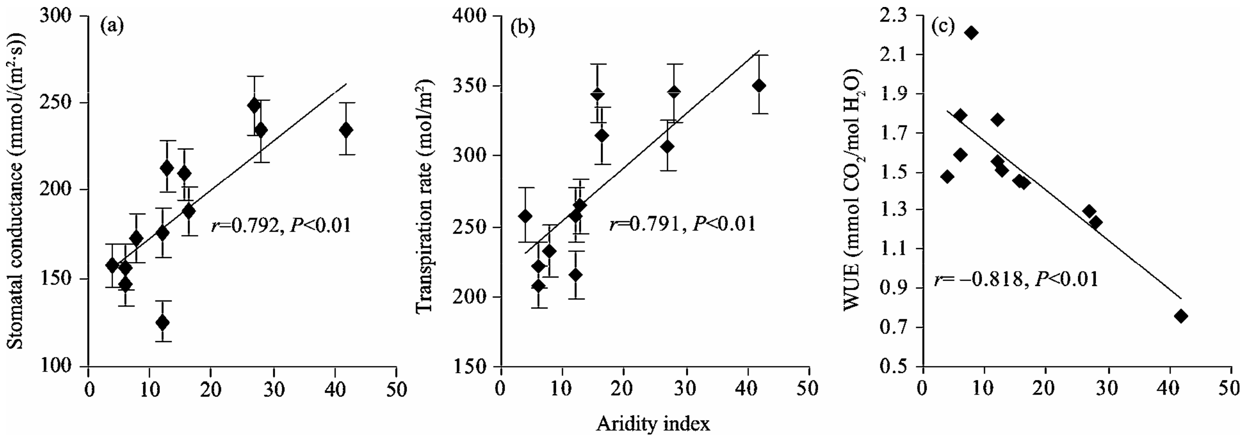

Fig. 5 Correlations between water metabolisms ((a), stomatal conductance; (b), transpiration rate; (c), WUE (water use efficiency)) in leaves of *Caragana* species and aridity index (mean±SE)

2.5 Leaf antioxidative enzyme activities and cell injury indices

As the climatic drought stress increased, the POD and SOD activities in leaves of *Caragana* species increased but the CAT activity did not show a significant increase. The POD and SOD activities were negatively correlated with aridity index (Table 4). Meanwhile, as the climatic drought stress increased, the MDA content increased while the oxygen free radical content decreased, and these two variables showed significant correlations with aridity index.

Table 4 Results of ANOVA and correlation analyses on the leaf antioxidative enzyme activities and cell injure indices of *Caragana* species across the climatic drought gradient

	POD activity	CAT activity	SOD activity	MDA content	Oxygen free radical content
ANOVA	F=134.972 P<0.01	F=31.530 P<0.01	F=5.550 P<0.01	F=1.510 P=0.201	F=9.682 P<0.01
Correlation with aridity index	r= –0.569 P<0.05	r=0.258 P=0.418	r= –0.607 P<0.05	r= –0.873 P<0.01	r=0.576 P<0.05

Note: POD, peroxidase; CAT, catalase; SOD, superoxide dismutase; MDA, malondialdehyde.

3 Discussion

3.1 Relationship between leaf ecological traits and climate gradient

Plants can adapt to environments with different moisture and radiation conditions through variable leaf ecological traits. The characteristics of reduced leaf area, increased leaf thickness, decreased leaf biomass, dense and bright leaf hair, and light leaf color may act to reduce light absorption and transpiration (Le Roux and Bariac, 1998; Quarrie et al., 1999; Jiang, 2000; Machado and Paulsen, 2001; Cabuslay et al., 2002). The plant species growing in environments with intensive sunlight, high temperature or serious drought stress usually have lower chlorophyll content (Pintado et al., 1997; Voronin et al., 2003; Zhang et al., 2003; Zhu et al., 2003). *Caragana* species (such as *C. sibirica* and *C. microphylla*) in the northeast Inner Mongolia region grow in humid forest or sub-humid to semi-arid steppes, where both solar radiation and air temperature are relatively lower than those in the southwest region. Larger light absorption area and stronger light absorption ability (few, short and green foliar hair, and high chlorophyll content) would favor photosynthesis, which enabled the *Caragana* species distributed in the northeast region to utilize solar energy more effectively, thereby maintaining high growth rate. In contrast, in the southwest region where the environment is extremely arid with strong solar radiation and high temperature, the smaller transpiration area and stronger leaf light-reflecting ability (dense, long and light-colored leaf hair, and low chlorophyll content) in *Caragana* species (such as *C. tibetica*, *C. roborovskyi* and *C. jubata*) would benefit the growth of these plants by reducing light absorption and decreasing transpiration (Fig. 5b). These ecological traits then generally would improve the water retention ability of these species, but simultaneously might reduce the growth and development rates of these plants.

3.2 Relationship between leaf physiological traits and climate gradient

Plants can also adapt to environmental conditions through leaf physiological traits. It is well known that water regulation ability is vital for plants to adapt to different moisture environments (Sánchez et al., 1998). Osmotic adjustment is an important mechanism for plants to absorb water and maintain turgor (Morgan, 1984; Guicherd et al., 1997; Peltier et al., 1997; Patakas et al., 2002). As the climatic drought stress increased from the northeast to the southwest, the osmotic potential in leaves of *Caragana* species increased gradually, which enabled the *Caragana* species distributed in the southwest region to absorb sufficient water, and thereby survive in arid environments. The content of soluble sugars in the leaf tissue of *Caragana* species increased significantly with the intensification of drought stress, and these sugars made considerable contribution to the increase of cell osmotic potential (23% to 33%). This result suggests that, as climatic drought stress increases, *Caragana* species will allocate more photosynthetic products for maintaining cell osmotic potential and water balance. In the northeast region where the environmental conditions are relatively favorable for *Caragana* species, more photosynthetic products are devoted to the growth and development of these species. In contrast, in the southwest region with strong climatic drought stress, survivorship seems to be more important than growth and development for *Caragana* species.

It has been shown that the free and bound water levels can also play important roles in plant adaptation to different moisture environments. Plant species with more total leaf water and free water would have more active metabolism (Hanson and Hitz, 1982). On the other hand, species with more bound water would have stronger drought resistance (Al-Saadi and Wiebe, 1973; Jiang, 2000). Our results on the free and bound water in leaves of *Caragana* species indicated that, with the intensification of climatic drought stress from the northeast to the southwest, the drought resistance of *Caragana* species increased gradually. More total and free water in leaves of *Caragana* species distributed in the northeast region enabled them to have a more active metabolism. In contrast, higher content of bound water and higher ratio of bound to free water in leaves of *Caragana* species distributed in the southwest region reduced metabolic rate, while strengthened the drought resistance of these *Caragana* species, which enabled these species to survive in the harsh environments.

With the intensification of climatic drought stress, the leaf water potential of *Caragana* species decreased gradually. The lower leaf water potential in the *Caragana* species of the southwest region might enable them to extract more soil water to maintain water balance. This mechanism reflects the strong osmotic adjustment ability of *Caragana* species (Marigo et al., 2000; Ma et al., 2008).

Reduction of stomatal conductance and transpiration rate is critical for plants to maintain water balance in arid environments (Wang et al., 1996; Yang et al., 1997; Franca et al., 2000). High WUE is an important trait for drought-tolerant plants, and is a water-saving strategy for plants to survive in arid regions (Ares and Fownes, 1999; Ma et al., 2003a). The variation of stomatal adjustment ability among the 12 *Caragana* species was in accordance with the changes in the climatic conditions of their distribution areas. The greater stomatal conductance in the *Caragana* species of the northeast region was probably favorable to photosynthesis, although more water was consumed through transpiration (Fig. 5b). In the southwest region, although low stomatal conductance reduced water loss (Fig. 5b), it also restricted photosynthesis rate to some extent, so that *Caragana* species can survive in the southwest by sacrificing their growth rate. The leaf WUE of *Caragana* species increased significantly with the intensification of climatic drought stress, indicating that the *Caragana* species in the southwest region might take a water-saving strategy for adaptation to the arid environments.

Antioxidative enzyme system is an important physiological basis for plants to survive in harsh environments. SOD, POD and CAT are the major antioxidative enzymes that can remove oxygen free radicals from plants (Wang and Li, 2001; Lima et al., 2002). The activities of POD and SOD in *Caragana* species increased gradually from the northeast to the southwest. As a result, the oxygen free radical content might decrease with the increase of climatic drought stress. Having increasingly active antioxidative enzymes might be one of the important traits for the *Caragana* species in the southwest to adapt to the arid environments.

3.3 The formation of *Caragana* species distribution pattern across the Inner Mongolia region

By the middle stage of the Tertiary Period (approximately 20 million years ago (MYA)), the climate on the Eurasian Continent was temperate and moist, and the topography was flat and open. *Caragana* species originated from the same ancestor during the Miocene Period (16–14 MYA), and spread rapidly over the Asian Continent (Zhou, 1996; Ma et al., 2003b; Zhang and Fritsch, 2010). During the Miocene Period, with the Himalayan motion (21–17 MYA) and the rapid uplift of the Qinghai-Tibetan Plateau (approximately 8 MYA), the Central Asia region became progressively arid, but the climatic conditions had not changed substantially in Eastern Asia region (Guo et al., 2008; Zhang and Fritsch, 2010). As a result, a climatic drought gradient formed in the Inner Mongolia region. Shifts in climatic conditions have driven the evolution and diversification of *Caragana* species, which enabled these species to successfully adapt to the environments in the Inner Mongolia region (Zhang and Fritsch, 2010). Thus, the distribution pattern of *Caragana* species formed along with the establishment of the climatic gradients in the Inner Mongolia region. For the 12 *Caragana* species in this study, most of the leaf ecological and physiological traits varied in accordance with the climatic drought gradient. The leaf biological traits of *Caragana* species showed significant correlations with the climatic drought gradient, suggesting that *Caragana* species can well adapt to the local climatic conditions. With the relatively greater metabolism and faster growth rate, the *Caragana* species in the northeast have strong competitive ability; on the other hand, with stronger resistance to stress especially to climatic drought stress, the *Caragana* species in the southwest are able to survive in the harsh environments.

4 Conclusions

In this study, we examined the relationships between the leaf ecological and physiological traits of 12 *Caragana* species with aridity index and solar radiation in the Inner Mongolia region. Our results showed that most of the leaf ecological and physiological traits of the 12 *Caragana* species varied in accordance with the climatic drought gradient, which reflected the adaptation of the *Caragana* species to the local climate conditions. These results suggested that both the progressive development of the climate gradient pattern and the different adaptive strategies among the *Caragana* species to variable moisture environments played important roles in the formation of *Caragana* species distribution pattern across the Inner Mongolia region.

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