Changes in photosynthetic and growth characteristics of *Leymus chinensis* community along the retrogression on the Songnen grassland in northeastern China

L.-X. SHI and J.-X. GUO*

Key Laboratory for Vegetation Ecology, Ministry of Education, Institute of Grassland Science, Northeast Normal University, Changchun 130024, China

Abstract

Net photosynthetic rate, stomatal conductance, ratio of sub-stomatal to atmospheric CO_2 concentration, transpiration rate, and water use efficiency changed significantly and assimilation capacity dropped continuously along the salinization and alkalinization process in the afternoon. Assimilation capacity of *L. chinensis* leaf correlated negatively with the degree of salinization and alkalinization. The photosynthetic characteristics of *L. chinensis* determined its community formation. By changing the ratio of chlorophyll *a/b* in leaves and accumulating soluble saccharides in rhizome, *L. chinensis* could adapt to the saline-alkali condition.

Additional key words: carotenoids; chlorophyll; CO_2 concentration; retrogressive succession; salinity and alkalinity gradients; stomatal conductance; transpiration rate; water use efficiency.

Introduction

Songnen grassland is one of the main districts in which Chinese saline-alkali soil concentrates. The topographical features and climatic conditions of the area result in a very unique soil alkalization and salinization process and abundance of minerals in ground water (Kulakov *et al*. 1997, Qiu *et al*. 2003). Extensive distribution of the saline-alkali soil has influenced the growth and distribution of the area vegetation type (Zheng and Li 1995). In the past 20 years, various factors, especially human over-utilization of the grasslands (such as overgrazing), resulted in desertification, and especially soil alkalization and salinization processes are more serious than the primary ones (Zhang 1994, Li and Zheng 1997, Zhu 2004). The salinized land of the Songnen grassland forms approximately 60–70 % of the total grassland and is seldom farmland being mainly utilized as the mowing pasture and grazing field (Zhu 2004). Different plant communities are formed along the retrogression on the Songnen grassland, such as *L. chinensis, L. chinensis+Puccinellia chinampoensis*, *L. chinensis+P. chinampoensis+Chloris virgata*, *etc*. (Li and Zheng 1997, Zheng and Li 1999).

The Songnen Plain that displays dramatically the eastern Eurasian steppe is the distributing centre of *L. chinensis* grassland (Zheng and Li 1993). As a climax community, *L. chinensis* grassland occupies 65 % of total grassland area in the Songnen Plain (Li and Zheng 1997). *L. chinensis* is a perennial species of the Gramineae family. The strong rhizomes of *L. chinensis* adapt to saline-alkaline and dune conditions (Clayton and Renvoize 1999). Due to its high productivity and high protein content, this species serves as major gramineous forage in Northern China and the Mongolian plateau. *L. chinensis* can be a candidate grass for the establishment or renewal of artificial grassland (Chen *et al*. 1988). The research of photosynthesis of *L. chinensis* has already made great progress, especially the research in the double concentration of CO_2 and the comparison between the two ecotypes of *L. chinensis* (Qi *et al.* 1990, 1997, Wang *et al.* 1997, 1998, 1999, 2003, Lin *et al.* 1998, Shi *et al.* 1998a,b, 2002, Gao and Zhou 2001).

We compared the discrimination in net photosynthetic

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*Corresponding author; fax: 086-0431-5695065, e-mail: gjixun@nenu.edu.cn
Abbreviations: C_a – atmospheric CO_2 concentration; C_i – sub-stomatal CO_2 concentration; Car – carotenoids; Chl – chlorophyll; E – transpiration rate; EC – electrical conductivity; g_s – stomatal conductance; P_N – net photosynthetic rate; WUE – water use efficiency.
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rate (P_N), stomatal conductance (g_s), ratio of sub-stomatal to atmospheric CO_2 concentrations (C_i/C_a), transpiration rate (E), water use efficiency (WUE), and contents of photosynthetic pigments and soluble saccharides of *L. chinensis*. We tried to find the regulation of changes of the photosynthetic characteristics of *L. chinensis* growing naturally along the grassland retrogressive process.

Materials and methods

Study site and plot selection: This research was conducted in the grassland ecosystem experimental station of Northeast Normal University, Chang Ling Horse Breeding Farm in Jilin Province (44°30'–44°45'N, 123°31'–123°56'E). This area is of typical mesothermal monsoon climate with plain topography in the south of the Songnen plain (altitude: 137.8–144.8 m). The main characteristics of the climate of the region are: cold, dry, frequently windy spring; warm, wet summer with frequent droughts; early autumn frosts; and long cold winter with relatively little snowfall. The mean annual temperature is 4.6–6.4 °C. The annual accumulated temperature is 2 545–3 374 °C, frost-free period is 136–146 d. The mean annual rainfall is about 400–500 mm which is mainly concentrated from June to August and accounts for more than 60 % of the rainfall of the whole year. The annual evaporation capacity is 2–3 times more than the rainfall. The salinized meadow soil is the main soil type on the Songnen grassland.

Five plots were selected for sampling, according to the different degree of retrogressive succession. Each plot area was 3×3 m, which could be distinguished according to the different density of *L. chinensis* community. The distance between the regions was less than 100 cm, so they shared nearly similar climatic conditions (*e.g.* precipitation, air temperature, and irradiation) and the same soil type. Our study was conducted from 2002 to 2003. In the middle ten days of July, the full expanded leaves of each plot were collected to measure their photosynthetic characteristics and each plot was used 3 times. At the same time, the soil samples were analyzed for pH and electrical conductivity (EC). Using routine methods, the density, height, and biomass of *L. chinensis* community were determined and each measurement was repeated 5 times.

Soil pH and EC: The magnetic force agitator mixed every 10 min and was 30 min quiet (Bao 2005). The pH was measured using a *HI98129* acidity meter and the survey of EC used *DDS-307* conductivity meter (*Shanghai Thunder Magnetism Instrument Plant*). Along the salinity and alkalinity gradient, the soil pH significantly increased from 8.23±0.01 to 10.29±0.04. EC reflected the ion concentration in soil, *i.e.* its salinity. It changed from 85±3 up to 612±8 μs cm^{-1}. The positive relationship between the soil alkalization and the soil alkalization was significant (r = 0.679, p<0.05) (Table 1).

Table 1. Changes in soil pH and conductivity along the retrogression on the Songnen grassland [μS cm^{-1}].

	1	2	3	4	5
pH	8.23±0.01	9.71±0.00	10.18±0.03	10.15±0.01	10.29±0.04
Electrical conductivity	85±3	169±5	226±5	318±7	612±8

P_N*, *g_s*, *C_i/C_a*, and *E of leaves were determined using portable open flow gas exchange system *LI-6400* (*LI-COR*, USA) in 1-h intervals from 08:00 through 16:00 h. WUE was calculated as the ratio of P_N/E. The photosynthetically active radiation (PAR) was 1 000±12 μmol·m^{-2}·s^{-1}, CO_2 concentration 350±2 cm^3 m^{-3}, leaf temperature 26.0±0.8 °C. Gas exchange was measured in full expanded leaves from the same adult plants. Five replications were made for each measurement, 3 replications in each plot, and measurements were done in 3 d in series in the middle ten days of July.

Chlorophyll (Chl) and carotenoid (Car) contents: 0.1 g leaf sample was dipped into 10 cm^3 of 80 % acetone/anhydrous ethanol mixture (1 : 1) to extract the photosynthetic pigments till the leaf became white. Spectrophotometric (*SpectrUV-754, Shanghai Accurate Scientific Instrument Co.*) determination at 440, 645, and 663 nm of each sample was done 3 times. The calculation used the formulae of Holm (1954).

Soluble saccharides: The rhizome and leaf of *L. chinensis* were treated with 120 °C for 10 min. Then dried mass was determined at 80 °C. A 50 mg sample was dipped into 3 cm^3 of 80 % ethanol. After 40 min in water bath (80 °C) the sample was centrifuged at 3 000×*g* for 15 min and the supernatant was collected. This course was repeated twice. Unified supernatants were then extracted and the dehydrated sample reacted then with furfural. The polycondensation with anthrone resulted in a blue compound (Zhou *et al*. 2002). The spectrophotometric estimation used the *UV-754* spectrophotometer at 620 nm; each measurement was repeated 3 times.

Statistical analyses of variance and correlation were compared using the statistical procedures of *Microsoft Excel 2000*.

Results

Quantitative changes: Plant density, height, dry mass, and underground dry mass of *L. chinensis* community were determined in different retrogressive stages (Fig. 1). Along the retrogression, these values consistently decreased. The changes were significant and positively correlated with the soil pH and EC (R_{pH} = –0.828, –0.905, –0.649, p<0.05, R_{EC} = –0.939, –0.634, –0.908, p<0.05).

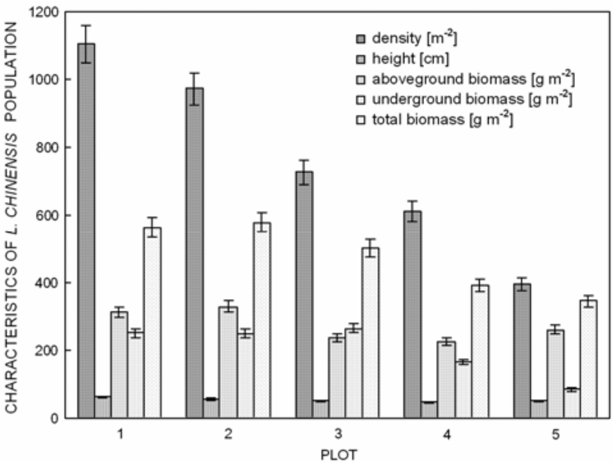

Fig. 1. Changes in quantitative characteristics of *L. chinensis* population along retrogression on the Songnen grassland.

Daily changes: P_N values of *L. chinensis* in different salty habitats were similar and could be expressed as two-hump curves, reflecting an obvious noon recess along the salinity and alkalinity gradient. The trend of P_N changes of *L. chinensis* differed greatly after 14:00 h. From 14:00 to 16:00, the P_N in plots 1 and 2 fell slowly or rose slightly and in plots 3 to 5 dropped rapidly (Fig. 2*A*). The daily average value of P_N in the different plots had a downward trend (y = –0.277 x + 9.044, r^2 = 0.446). The correlation coefficient of the daily average value of P_N was R_{pH} = –0.217, R_{EC} = –0.855. This indicated the daily change in P_N was mainly influenced by soil ions.

g_s did not exert significant differences in various plots before 14:00. From 14:00 to 16:00, the daily change of g_s in plots 1 to 3 went up significantly, but in plots 4 and 5 went down (Fig. 2*B*). The daily average value of g_s of *L. chinensis* leaf was positively correlated with the salinity and alkalinity of the soil (R_{pH} = –0.434, R_{EC} = –0.685, p<0.05). At 16:00, g_s of *L. chinensis* leaf in the different plots showed significant negative correlation with the salinization and alkalinization of the soil (R_{pH} = –0.786, R_{EC} = –0.898, p<0.05).

C_i/C_a did not differ significantly in the different plots, but it increased from plot 1 to plot 3 and in plots 4 and 5 decreased in the period of 15:00–16:00 (Fig. 2*C*). However, the daily average value of C_i/C_a differed significantly in different plots: from plot 1 to plot 2 went upward, from plot 2 to plot 5 downward. The mean value of daily change of C_i/C_a of *L. chinensis* was significantly related with the salinization and alkalinization of the soil (R_{pH} = –0.403, R_{EC} = –0.701, p<0.05).

The daily course of *E* differed greatly in different plots, increasing in plots 1 and 2 and decreasing in plots 3 to 5 (Fig. 2*D*). The daily average value of *E* of *L. chinensis* was negatively related with salinization and alkalinization of the soil (R_{pH} = –0.493, R_{EC} = –0.893, p<0.05).

The daily changes of WUE were similar to those of P_N in *L. chinensis*, showing a two-hump curve. At 14:00, the difference in WUE was the greatest, WUE in plot 1 being 1.495 times that of plot 5 (Fig. 2*E*). The daily mean of WUE of *L. chinensis* decreased with the increasing salinization and alkalinization of the soil (R_{pH} = –0.508, R_{EC} = –0.970, p<0.05).

The contents of Chl *a* and *b* and Car in leaves were remarkably reduced by salinization and alkalinization (Table 2). The remarkable negative relation was found between the reducing trend and the salinization and alkalinization of the soil (R_{pH} = –0.681, –0.715, –0.563, R_{EC} = –0.638, –0.812, –0.922, p<0.05). However, Chl *a/b* showed increasing trend and remarkable positive correlation with the salinization and alkalinization of soil (R_{pH} = 0.634, R_{EC} = 0.957, p<0.05).

The content of soluble saccharides in leaf and rhizome of *L. chinensis* did not decrease along the salinity and

Table 2. Changes in contents of photosynthetic pigments (Chl – chlorophyll, Car – carotenoids) [g kg^{-1}(FM)] along the retrogression on the Songnen grassland.

Plot	Chl *a*	Chl *b*	Chl *a*+*b*	Chl *a/b*	Car	Total pigments
1	1.45±0.02	0.66±0.00	2.11±0.02	2.20±0.02	0.48±0.04	2.59±0.06
2	1.33±0.04	0.57±0.03	1.90±0.05	2.33±0.04	0.46±0.06	2.35±0.04
3	1.39±0.06	0.61±0.06	2.00±0.04	2.26±0.08	0.48±0.07	2.48±0.06
4	1.14±0.05	0.44±0.07	1.58±0.07	2.58±0.08	0.38±0.09	1.94±0.08
5	1.24±0.00	0.45±0.01	1.69±0.03	2.81±0.05	0.33±0.01	2.02±0.02

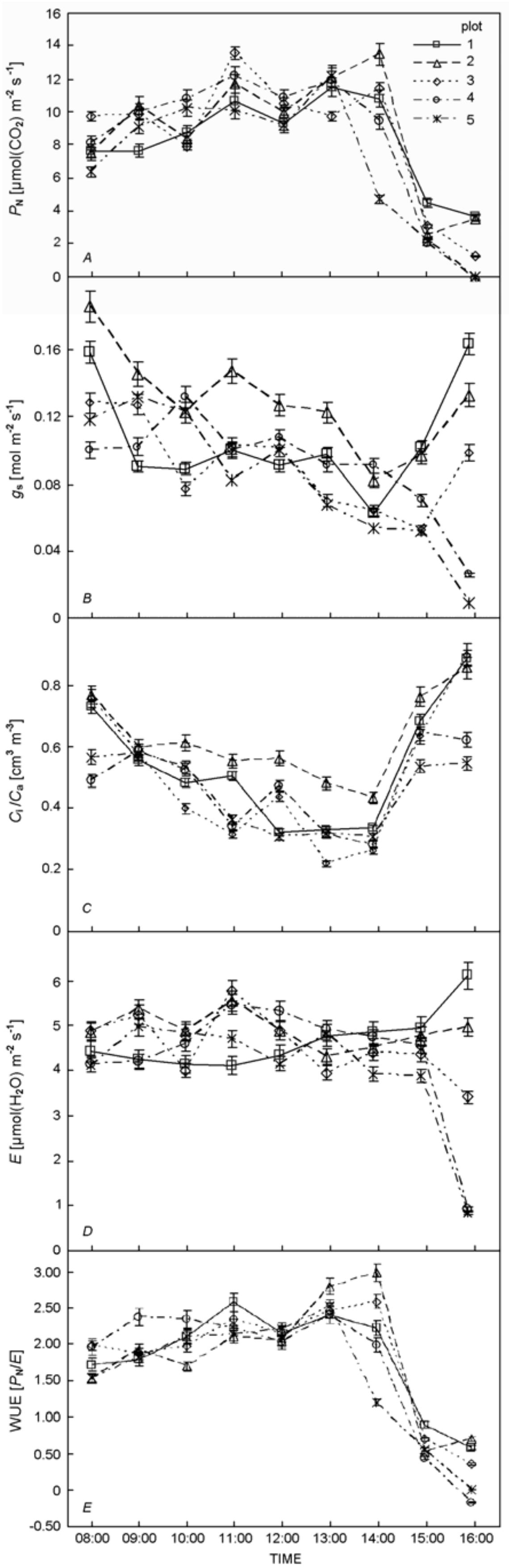

Fig. 2. Daily changes in (*A*) net photosynthetic rate, P_N, (*B*) stomatal conductance, g_s, (*C*) ratio of stomatal and sub-stomatal CO_2 concentration, C_i/C_a, (*D*) transpiration rate, *E*, and (*E*) water use efficiency, WUE of *L. chinensis* along the retrogression on the Songnen grassland.

alkalinity gradient, whereas it showed a specific increase (Fig. 3). The soluble saccharide content was related with the salinization and alkalinization of soil, and especially manifested the significant relation with the soil salt content (R_{leaf} = 0.500, $R_{rhizome}$ = 0.664, p<0.05). Maybe the soluble saccharides as the matter to regulate osmosis enabled the plant toleration of the salinity-alkali stress.

The above mentioned characteristics had close relations with the density, height, and biomass of *L. chinensis* community (Table 3).

Discussion

Most plants are sensitive to salinity and alkalinity, especially crops (Kingsbury *et al.* 1984, Zhang 1999). Because of the saline and alkaline stress, plant grew slowly, leaves became yellow, and subsequently died and fell. Even if a leaf did not fall down, the photosynthetic pigments engaged in photon absorption, transmission, and transformation would be degraded in the leaf; the fine structure of chloroplasts disintegrated and the components and structure of thylakoid membrane changed (Seemann and Critchley 1985). Thus the saline-alkali stress influenced seriously photosynthesis of *L. chinensis* leaves, depressed P_N, and sometimes induced plant death (Shi *et al*. 1998a,b, 2002, Zhang 1999).

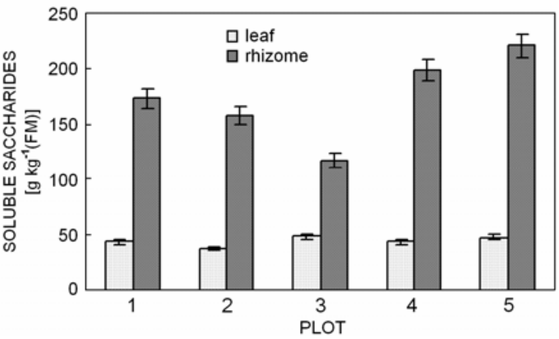

Fig. 3. Changes in contents of soluble saccharides of *L. chinensis* along the retrogression on the Songnen grassland.

The soil on the Songnen plain, which contains NaCl, Na_2SO_4, and a large amount of Na_2CO_3 and $NaHCO_3$, is the typical saline-alkali soil (Zheng *et al*. 1999). In the retrogressive process of the grassland, *L. chinensis* can grow well when EC is in the range of 85±3–612±8 μs cm^{-1} and the soil pH is 8.23±0.01–10.29±0.04. This shows that *L. chinensis* is a salt-tolerant plant.

Table 3. Correlation coefficients between photosynthetic characteristics and quantitative characters in the *L. chinensis* community. *,**Correlations at 0.05 and 0.01 levels of probability, respectively; ^{NS} = not significant. $r_{0.05}$ = 0.381, $r_{0.01}$= 0.487, n = 27.

	Density	Height	Total biomass
P_N	0.694^{**}	0.240^{NS}	0.766^{**}
g_s	0.788^{**}	0.648^{**}	0.837^{**}
C_i/C_a	0.791^{**}	0.641^{**}	0.873^{**}
E	0.896^{**}	0.640^{**}	0.964^{**}
WUE	0.848^{**}	0.483^{*}	0.877^{**}
$P_{pigment}$	0.814^{**}	0.766^{**}	0.869^{**}

The soil is salinized and alkalinized distinctly along the retrogression on the grassland. *L. chinensis* is the main species on the Songnen grassland. It grows well, but the daily patterns of photosynthetic parameters were significantly different along the course of the retrogression on the grassland, especially in the afternoon, which immediately reduced leaf photosynthetic capacity. The accumulation of soluble saccharides in the rhizome has consumed a large amount of energy, which further influenced the change of the density, height, and biomass of *L. chinensis* community. This resulted in the retrogressive succession of *L. chinensis* community. We found that the photosynthetic capacity and the retrogressive succession of *L. chinensis* community were closely correlated.

In the retrogressive process, P_N, g_s, C_i/C_a, E, and WUE in plot 2 were relatively high along the salinity and alkalinity gradient. Thus the photosynthetic capacity was strengthened in the given saline-alkali condition. In addition, the contents of Chl *a/b* and soluble saccharides increased. This may be a specific physiological mechanism adapting *L. chinensis* to saline-alkali environment.

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Nátr, L.: **Země jako skleník. Proč se bát CO_2?** [Earth as a Greenhouse. Why to be Afraid of CO_2?] – Academia, Praha 2006. [In Czech.] ISBN 80-200-1362-8. 142 pp., 150 CZK.

The still increasing CO_2 concentration in the atmosphere and its effects on climate, vegetation, and human community, called 'global climate change', became one of the greatest problems of the recent world. The author of the textbook, professor of plant physiology on the Charles University in Prague, presents a unique analysis in 14 chapters. The first ones are devoted to carbon dioxide (carbon; oxygen; carbon dioxide; carbon monoxide; carbon dioxide as a part of carbon cycle on Earth; past and recent changes of atmospheric CO_2 concentration; carbon cycle on dry land and oceans; fuel consumption and CO_2 production; possible consequences of quantitative changes in carbon cycle on Earth, *etc.*). Chapters 4 and also 6 are fully devoted to discussion on CO_2 and greenhouse effect (greenhouse gases – water vapour, CO_2, methane, N_2O, ozone, chlorofluorocarbons, *etc.*). A short chapter 5 compares the greenhouse effects on Earth with those on the nearest neighbouring planets, Venus and Mars.

Further chapters (7−10) deal with the complexity of both atmospheric CO_2 and climate changes (limitations of mathematic modelling of climate, effects of aerosol particles in the atmosphere, the role of biosphere as carbon sink and source, mitigation costs of CO_2 sequestration, possible global climate change during the 21st century, *etc.*). The description of the scientific activities in the Czech Republic emphasizes experiments dealing with long-term effects of enhanced CO_2 concentration on forest ecosystems and review studies on the potential impact of global climate changes on both natural and agricultural ecosystems. Chapters 11−14 are devoted to the effects of CO_2 on plants, health of men, and general consequences for sustainability of human societies. The book is supplemented with extensive lists of both Czech and international literature sources (124 references of books, review articles, and original papers).

Generally, the book is an excellent source of information on the role of CO_2 in the global climate as well as reasons and consequences of the perturbations of the carbon cycle. The main text is accompanied by numerous boxes offering to the reader a comprehensive and reliable information of various terms, definitions, and processes. The book is well edited and printed. It also contains a short subject index (132 items). It can be recommended to readers understanding Czech (predominantly but not exclusively researchers and students) interested in environmental sciences and problems associated with global climate change. Further information on the book is available on www.academiaknihy.cz and www.academiabooks.cz.

J. ČATSKÝ (*Praha*)