Homeostatic responses and growth of *Leymus chinensis* under incrementally increasing saline-alkali stress

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Despite considerable tolerance to salt and alkali stress, *Leymus chinensis* populations on the southwestern Songnen Plain in northern China are threatened by increasing soil salinity and alkalinity. To explore the species' responses to saline-alkali stress, we grew it in substrates with varying concentrations of nitrogen (N) and phosphorus (P) while applying varying levels of saline-alkali stress (increasing in 14-, 17- or 23 -day intervals). We measured the plants' contents of N and P, and the N:P ratio, and calculated their homeostasis indices (H_N, H_P and $H_{N:P}$) under each nutrient and saline-alkali stress treatment.

The N content was found to be more sensitive to saline-alkali stress than the P content. The N and P contents were highest and the N:P ratio was stable at pH 8.4. At both pH 8.1 and 8.4, $H_{N:P} > H_N > H_P$, but the indices and their relations differed at other pH values.

Exposure to saline-alkali stress for the 14-day incremental interval had weaker effects on the plants. Rapid changes in salinity-alkalinity weakened both the positive effects of the weakly alkaline conditions (pH 7.5-8.4) and the negative effects of more strongly alkaline conditions (pH 8.7 or 9.3) on *L. chinensis*. When *L. chinensis* plants lack N, applying N fertilizer will be extremely efficient. The optimal concentrations of N and P appeared to be 16 and 1.2 mmol/L, respectively. When the *L. chinensis* plants were N- and P-limited, the specific growth rate correlated positively with N:P, when limited by N it correlated positively with the environmental N concentration, and when limited by P it was weakly positively correlated with the environmental P concentration.

INTRODUCTION

Ecological stoichiometry (ES) is the study of the balances of elements and energy in ecosystems, which have profound effects on living organisms, their interactions, and associated ecological processes (Cambardella and Elliott, 1993; Elser *et al.*, 2000; Güsewell, 2004; Bradshaw *et al.*, 2012). ES theory focuses primarily on elements required by all living organisms, so it can be readily generalized across taxa and systems (Sanders and Taylor, 2018). A key concept is homeostasis: a system's capacity to maintain constant conditions internally when external conditions vary, a fundamental property of organisms (Kooijman, 1995; Cooper, 2008; Halvorson *et al.*, 2019). Characterizing both the degrees and consequences of homeostasis is important for understanding responses to environmental changes (Meunier *et al.*, 2014; Halvorson *et al.*, 2019). In ES-based analyses, the homeostasis concept is used to infer organisms' responses to changes in ratios of elemental resources, and predict their feedback effects on resources' availability through changes in the acquisition, storage, and release of nutrients, particularly limiting nutrients such as nitrogen (N) and phosphorus (P) (Halvorson *et al.*, 2019). A general assumption is that stoichiometric homeostasis is stronger in heterotrophs than autotrophs (Sterner and Elser, 2002), so environmental stoichiometry is believed to be reflected more closely by plant and algal stoichiometry than by animals' stoichiometry. However, Yu *et al.* (2011) found that homeostatic regulation of N and P varies widely in vascular plants, so the patterns and processes involved are complex and far from fully understood.

Another core concept of ES is the growth rate hypothesis (GRH), which postulates that organisms' specific growth rates correlate positively with their phosphorus (P) contents because P is essential for the ribosomal RNA production needed to sustain growth (Moody *et al.*, 2017) and for many other processes including photosynthesis, respiration, enzyme production, and

24   generation of ATP (Li *et al.*, 2019). Thus, organisms with high specific growth rates have high

25   nutrient demands and (*inter alia*) low tissue C:nutrient ratios, low N:P ratios, and potentially

26   competitive advantages in high-P environments, but disadvantages in low-P environments.

27   Hence, plants' abilities to compete for nutrients depend on both their tissue nutrient contents and

28   life history traits (Mulder and Elser, 2009; González *et al.*, 2010; Sanders and Taylor, 2018).

29   ES has been widely applied in various areas of plant science, including in studies on the

30   growth of individual plants, population dynamics, limiting elements, succession, and stability of

31   vegetative communities (Güsewell, 2004). Soil is a key component of terrestrial ecosystems

32   because it supports numerous ecological processes (Normand *et al.*, 2017; Wang *et al.*, 2018),

33   plays crucial roles in plant growth, and directly affects plant communities' composition, stability,

34   and succession (Wardle *et al.*, 2004). N and P are key elements with profound effects on plants

35   growth because they play major roles in microbial dynamics, litter decomposition, food webs,

36   and the accumulation and cycling of nutrients in soil (Elser et al., 2003). Changes in plants' N:P

37   ratios (*inter alia*) may reflect adaptations to environmental conditions (Tang *et al.*, 2018),

38   including (of particular interest here) salinity and alkalinity.

39   According to incomplete UNESCO and FAO statistics, the area of saline-alkali land is

40   growing globally (Kerr, 2017) and amounts to 9.5438×10^8 hm^2, including 0.9913×10^8 hm^2 in

41   China (Li *et al.*, 2017a). This is a severe problem because salinization impairs seed germination,

42   root extension, and plant development, resulting in land degradation, reductions in cultivated

43   land quality, and limitation of agricultural development (Wu *et al.*, 2016). Most saline-alkali soil

44   in northeast China is on the Songnen Plain, and its area is increasing through grassland

45   salinization (Li, 2000). The dominant species in saline-alkali grassland on the Songnen Plain is

46   *Leymus chinensis (L. chinensis)*, which can provide high yields of high quality gramineous

47   forage if the salinity and alkalinity are not too severe. It can play an important role in restoring

48   saline-alkali land (Liu *et al.*, 2014), but there is a need to reduce the salinity and alkalinity of

49   affected grassland and improve the species' yield and stress tolerance. It is therefore important to

50   study the ecological stoichiometric homeostasis of N and P, associated ecological processes, and

51   the effects of saline-alkali stress on the growth rate and survival strategy of *L. chinensis*.

52   Previous research on saline-alkali tolerance in *L. chinensis* has mainly focused on

53   physiological processes (such as the osmoregulatory roles of ions, organic acids, soluble sugars

54   and other substances) and morphological traits (such as tiller buds, root distribution, and

55   rhizomes' internode length and branching angles) (Zheng *et al.*, 2017). C, N, and P dynamics in

56   soil-*L. chinensis* systems have also been addressed (Li *et al.*, 2018). The secret of *L. chinensis*

57   adapting to saline-alkali stress has been discussed deeply from many perspectives. However, soil

58   salinization is a long-term dynamic process, and there is relatively little information on relations

59   between the effects of saline-alkaline stress (and of variation in its duration) and ecological

60   stoichiometric characteristics. And few studies point out a certain range in which *L. chinensis*

61   can adapt to lasting saline-alkali stress. We make the hypothesis that the adaptability of *L.*

62   *chinensis* under different saline-alkali stress conditions is different (positive effects? or negative

63   effects?) and there is a certain range. Here, we demonstrate that the ecological stoichiometric

64   characteristics of *L. chinensis* are influenced by the duration of saline-alkali stress and the

65   available concentrations of both N and P.

66   ## MATERIALS AND METHODS

67   **The experimental site**

68   We performed experiments in an outdoor terrace on the campus of the College of Earth Sciences,

69   Jilin University, Changchun, Jilin Province, northeast China. This is in the north temperate

70   continental climatic zone, which has four distinct seasons. The mean annual temperature and

71   precipitation are 4.8°C and 567 mm, respectively, and mean winter and summer temperatures are

72   -14 and 24 °C, respectively (Zhang *et al.*, 2017).

73   **Experimental design**

74   **Experimental preparation**

75   Experiments were performed from August 7 to November 6, 2017. To minimize interference

76   from impurities in the soil, the substrate used was fine nutrient-poor sand, sieved with 10 and 75

77   mesh sieves and thoroughly washed with distilled water to minimize its nutrient content. To

78   avoid effects of different soil loadings on growth of experimental plants, the sand was air-dried

79   and equal portions were placed in plastic flowerpots (diameter 30 cm, height 23 cm). The

80   portions were roughly equivalent to two-thirds of the pots' volume. *L. chinensis* seeds were

81   immersed in water for 48 hours and air-dried in the dark to avoid dormancy and increase the

82   germination rate, then about 100 seeds were sown evenly in each flowerpot. About seventy

83   percent of the seeds survived, and all pots were similar with respect to the number, size, and

84   vitality of surviving seedlings.

85   **N and P addition**

86   After the seeds germinated, a 200 ml portion of Hoagland's nutrient solution with adjusted N and

87   P concentrations was added to each pot once every two days. The elemental composition of

88   Hoagland's solution is described in detail elsewhere (Li et al., 2018). In five sets of nine pots, the

89   N concentration was adjusted using ammonium nitrate to establish five N concentration

90   treatments (2, 4, 8, 16, and 24 mmol/L) in which the concentration of P was fixed at 1 mmol/L.

91   In another five sets of nine pots, the P concentration was adjusted using potassium dihydrogen

92   phosphate to establish five P concentration treatments (0.3, 0.6, 1.2, 2.4, and 4.8 mmol/L) in

93   which the concentration of N was fixed at 14 mmol/L. The experiment thus involved 10 nutrient

94   treatments and a total of 90 pots (10 treatments×3 replicates×3 salinity stress intervals) in a

95   randomized complete block design. The N and P concentration gradients were set on the basis of

96   previously reported results (Yu et al., 2011).

97   **Saline-alkali stress**

98   On 7 Aug 2017, after the *L. chinensis* plants had grown to an average height of about 15 cm,

99   plants in triplicate pots were subjected to the saline-alkali stress treatments shown in Table 1, by

100   adding $NaHCO_3$ to the previously described treatment solution to simulate various degrees of

101   land salinization encountered in the western regions of Jilin Province. To avoid excessively

102   stressing the plants, the pH was increased at intervals of 14, 17, or 23 days. The pH was raised

103   from 7.5 to 8.1, and then to 8.4, 8.7, and finally 9.3. Aboveground parts of *L. chinensis* plants

104   subjected to these treatments were harvested (cutting from the ground level) according to the

105   schedule shown in the table 1.

106   **Sample collection and testing methods**

107   Aboveground parts of *L. chinensis* plants were cut according to the harvesting schedule shown in

108   Table 1. The samples were rinsed with distilled water, dried with absorbent paper, deactivated

109   for 15 min in a dry air oven (105 °C) and dried for 12 h at 65 °C to eliminate water completely.

110   After cooling the samples to room temperature, they were weighed, crushed, screened with a 100

111   mesh sieve, and finally quartered. The resulting sub-samples were then packed in sealed bags for

112   analysis.

113   Total N and total P in the *L. chinensis* samples were respectively determined by Chinese

114   standard methods LY/T 1269-1999 (using a SAN++ Continuous Flow Analyzer; Skalar,

115   Netherlands) and LY/T 1270-1999 (using an ICPS-7500 inductively coupled plasma atomic

116   emission spectrometer; Shimadzu, Kyoto, Japan).

117   **Data analysis**

118   The acquired data were statistically analyzed using SPSS 24 (SPSS Inc., USA). Homeostasis

119   indices were calculated using the stoichiometric homeostasis model $y = cx^{\frac{1}{H}}$. Here, x is the

120   measured content of an element in the soil, y is the measured content of the same element in L.

121   *chinensis*, c is a constant coefficient, and H is the homeostasis index. The results were visualized

122   using Sigmaplot 12.5 (Systat Software, Inc.). We calculated specific growth rates based on

123   recorded changes in dry mass and instantaneous growth rates using the equation $u = \ln(M_t / M_0)/t$,

124   where u is the specific growth rate (day^{-1}), M_t and M_0, are the final and initial dry mass,

125   respectively, and t is experiment duration in days. Details of several statistical analyses are

126   included in Appendix 1 and Appendix 2.

127   ## RESULTS

128   **Effects of incremental increases in saline-alkali stress on N, P contents and N:P ratios**

129   The 14-day stress increment yielded the highest N and P contents in the plants when the N

130   concentration in the substrate was low (2-8 mmol/L), but not when the N concentration in the

131   substrate was higher (16 or 24 mmol/L). The variation in the N and P contents, and the N:P ratio,

132   was similar under all three stress intervals: the N content and N:P ratio were highest with 17-day

133   intervals, while the P content was highest with 14-day intervals (Fig. 1).

134   **Homeostasis characteristics of *L. chinensis***

135   The homeostasis index of N (H_N) in *L. chinensis* ranged from 2.35 to 7.25, and first rose then fell

136   as salinity-alkalinity increased, independently of interval length (Fig. 2). Under the 14-, 17- and

137   23-day intervals, H_N ranges were 3.10-7.25, 2.35-5.36 and 3.19-3.87, respectively. It was

138   consistently highest at pH 8.4, and higher at pH 8.1 and 8.7 than at pH 7.5 and 9.3 (Table 2).

139   Under the 14-day stress intervals, the changes in H_N were moderate, and H_N was high at all pH

140   values. The index was lower at both pH 8.1 and 8.7 under the 17- and 23-day intervals.

141   Under our treatments, H_P of *L. chinensis* ranged from 2.60 to 5.33. Under all three saline-

142   alkali stress intervals, it first rose and then declined (Fig. 2). Under the 14-, 17-, and 23-day

143   saline-alkali stress intervals, the ranges of H_P were 2.84-5.33, 2.65-4.90 and 2.60-4.10,

144   respectively, and the highest values of H_P occurred at pH 8.1, 8.7, and 8.1, respectively (Table 2).

145   At pH 8.1, H_P was significantly higher under 14-day intervals than under longer intervals. At pH

146   8.4, it was highest under 17-day intervals, and at pH 8.7 it declined in the order 17-day > 14-day >

147   23-day intervals.

148   Plants have strong self-regulating mechanisms, and generally keep their N:P ratios within a

149   narrow range by adjusting their N and/or P contents in response to environmental changes. For

150   example, the $H_{N:P}$ of *L. chinensis* varied from 3.06 to 9.23 in plants exposed to different P

151   concentrations, while its ranges under the 14-, 17- and 23-day stress interval regimes were 3.15-

152   9.23, 3.11-6.83 and 3.06-6.44, respectively, and its peak values occurred at pH 8.4, 8.4, and 8.1,

153   respectively (Table 2). *L. chinensis* plants are thus able to effectively regulate their *N:P* ratio in

154   the pH range 8.1 to 8.7, but less so at pH 7.5 and 9.3.

155   **The specific growth rate of *L. chinensis***

156   The specific growth rate of the plants rapidly increased then decreased as the N concentration in

157   the substrate increased, peaking at 16 mmol/L (Fig. 3). This indicates that when *L. chinensis*

158   plants lack N, initial N addition will be extremely efficient, but saturating or excessive amounts

159   will not promote (and may even hinder) further growth. Conversely, the plants' specific growth

160   rate first increased, then decreased, and then increased again with increases in environmental P

161   concentration. Specifically, it was lowest at a P concentration of 2.4 mmol/L, and higher at both

162   1.2 and 4.8 mmol/L. Thus, high addition of P is beneficial for plant growth but may lead to

163   wastage of resources. Therefore, the optimum P concentration under our experimental conditions

164   was 1.2 mmol/L.

165   In summary, the threshold concentrations of N and P in the substrate solution at which the

166   biomass of *L. chinensis* began to decrease (or stopped increasing) were 16 and 1.2 mmol/L,

167   respectively. Moreover, plants exposed to 1.2-4.8 mmol/L P were mainly restricted by N, while

168   those exposed to 16 and 24 mmol/L N were mainly restricted by P, and those exposed to lower

169   concentrations of P and N were restricted by P and N, respectively (Fig. 4). According to the

170   results of the correlation analysis, the specific growth rate of *L. chinensis* and the plant N:P ratio

171   is positively correlated in both N-limited and P-limited environments, and the correlation is

172   slightly stronger under P-restricted conditions, but neither of them passed the significance test. In

173   the N and P co-limited environment, the specific growth rate of *L. chinensis* has a positive

174   correlation with the plant N:P ratio, and the correlation coefficient is 0.46, which is significant at

175   the level of 0.01.

176   **DISCUSSION**

177   **N and P contents and the N:P ratio in *L. chinensis***

178   N and P are key nutrients that play major roles in myriad processes in plants and have important

179   effects on one-another's uptake and metabolism (Han *et al.*, 2009). N:P ratios in plants fluctuate

180   within relatively narrow ranges, and variations in their N and P contents are restricted by

181   homeostatic mechanisms. Although P and N contents can influence each other, N affects P

182   contents more strongly than vice versa, putatively due to the overall higher abundance of N in

183   plants (Yu, 2005). We found that *L. chinensis* plants absorbed limited amounts of P from soils

184   with a low N content, and changes in the soil's N contents between 2 and 8 mmol/L had minor

185   effects on the P content of their aboveground parts. However, their P contents were substantially

186   increased by higher N concentrations in the soil. Similarly, we found that N contents of

187   aboveground parts of *L. chinensis* are influenced by the environmental P concentration: they

188   changed greatly when the soil's P content was low but remained very similar at high P

189   concentrations. In summary, P content is the main constraint on plant growth when the soil's P

190   content is low, but N content becomes the main constraint of plant growth when the P content is

191   high.

192   Our experiment examined saline-alkaline environments with different pH values. The N and

193   P contents in aboveground parts of *L. chinensis* were both high at pH 8.4, indicating that the

194   plants maintained strong control over their N:P ratios at this pH and confirming the previously

195   reported finding that this pH promotes growth of *L. chinensis* (Li *et al.*, 2018). *L. chinensis* can

196   resist mild pH stress but it is inhibited by strong saline-alkali stress (Yan *et al.*, 2006). At high

197   pH (8.7 and 9.3), we found that its P content was relatively high when there were high soil

198   concentrations of either N (16 or 24 mmol/L) or P (2.4 or 4.8 mmol/L). However, plants need

199   more P to cope with pH stress. Plants' nutrient adsorption is related to pH, together with

200   indications that luxury uptake of N and P under moderate pH stress enables some plants (e.g.

201   *Rhizoma acori graminei* and *Lythrum salicaria*) to resist strong pH stress (Cheng *et al.*, 2017).

202   The N and P contents in aboveground parts of *L. chinensis* we observed at various pH values

203   indicate that N contents in plants are more sensitive to saline conditions than their P contents.

204   **Homeostasis characteristics of N, P, and N:P in *L. chinensis***

205   Homeostasis theory postulates that plants can adjust their growth rate, and rates of both resource

206   allocation and utilization in mechanisms that maintain their internal homeostasis (Mendez and

207   Karlsson, 2005). Homeostasis is stronger in animals than in plants, and in higher plants than

208 lower plants (Zeng *et al.*, 2016), suggesting that homeostatic mechanisms have become more

209 powerful over the course of evolutionary history. We found that the homeostasis indices for N

210 content, P content and the N:P ratio in *L. chinensis* were within reported ranges for algae, fungi

211 (Sterner and Elser, 2002), and animals (Yu, 2005; Karimi and Folt, 2006). On the basis of

212 experiments on Inner Mongolian grassland and sand culture, Yu *et al.* (2009) concluded that

213 aboveground parts of *L. chinensis* have slightly higher H_N, H_P and $H_{N:P}$ values (5.88-8.80, 3.37-

214 6.67 and 4.49-9.46, respectively) than those observed in our study, possibly due to differences in

215 climatic conditions.

216   We also found that all three homeostasis indexes first increased then decreased with

217 increases in pH. At pH 8.1 and 8.4, they were all high, showing that *L. chinensis* can maintain

218 stable levels of nutrients in weakly alkaline environments. However, the indices were lower

219 under more strongly alkaline conditions (pH>8.7), indicating that such conditions have toxic

220 effects. This is consistent with previous observations that saline and alkaline stress has both

221 physiological and biochemical effects on roots and leaves of *L. chinensis* (Liu *et al.*, 2014).

222   Another finding is that $H_{N:P}>H_N>H_P$, indicating that the stronger homeostasis of N content

223 than of P content in plants is mainly due to their significantly higher N content. Accordingly,

224 homeostasis in zooplankton is reportedly highest for macronutrients, followed by micronutrients,

225 and lowest for non-essential elements (Karimi and Folt, 2006). Since high N contents in plant

226 tissues promote high P contents and plants' N:P ratios vary less than their N and P contents, the

227 N:P ratio may be subject to stronger homeostasis than either N or P individually (Sterner and

228 Elser, 2002). Plants maintain N:P stability by their adjusting resource allocation and utilization

229 of resources. The degree of stability may reflect their environmental adaptability, so the N:P ratio

230 appears to be more important than N and P contents in this respect.

231   The plants' homeostasis is related to the duration of the incremental increases in salinity-

232   alkalinity stress. With 14-day intervals (rapid changes in alkalinity), the positive effects of a

233   weakly alkaline environment (pH 7.5-8.4) on leaves of *L. chinensis* are stronger than those

234   observed with longer intervals: the homeostasis indices of the aboveground parts are relatively

235   high, and further increases in alkalinity (to pH 8.7 or 9.3) induce relatively little damage to

236   leaves. $H_{N:P}$ and H_P are strongly affected by changes in the duration of the incremental increases.

237   For example, the variation in $H_{N:P}$ of aboveground parts is higher with 23-day intervals than with

238   either 14- or 17-day intervals (rising to a peak and then quickly falling with increases in pH, as

239   shown in Fig. 2).

240   **Specific growth rate**

241   Specific growth rates are crucial indicators of organisms' adaptation to (and thus ability to

242   survive and reproduce within) an environment. Clearly, variations in growth capacity can be

243   linked to variations in elemental demand (Moody *et al.*, 2017). Our results confirm previous

244   findings (and intuitive expectations) that adding N can significantly enhance growth of N-

245   deficient plants. However, they also show that adding N can inhibit plants' growth if the

246   environmental content of N exceeds the plants' requirements. The optimal N and P contents in

247   the soil under our experimental conditions were 16 and 1.2 mmol/L, respectively. Adding excess

248   P will not have inhibitory effects, and may even further stimulate growth slightly, but would be a

249   waste of resources.

250   Plants can absorb elements in soils selectively. Moreover, The N:P ratio has a relatively

251   complex relationship with specific growth rates: they are positively correlated at low specific

252   growth rates, but negatively correlated once the specific growth rate exceeds a certain threshold

253   (Agren, 2004). Additionally, specific growth rates of belowground parts of plants were positively

254   correlated with N:P under N constraints, but negatively correlated with N:P under P constraints

255   (Yu, 2009). In our experiment, growth of *L. chinensis* was restricted by both N and P. The

256   specific growth rate of aboveground parts was positively related to N:P under N constraint or P

257   constraint, but the positive correlation was strengthened under the co-constraint of N and P. This

258   is consistent with the conclusions of Yu *et al.* with respect to the effects of N constraint, but not

259   those of P constraint. We speculate that this was due to the plant's generally low specific growth

260   rate in the experiment, and also possibly to effects of salinity-alkalinity on the relationships

261   between plant growth and other environmental variables. Although effects of plants' nutrient

262   storage on their specific growth rates have been discussed (Sterner and Elser, 2002; Yu, 2009),

263   the relationship between their specific growth rates and C:N:P ratios clearly requires further

264   study.

265   According to the specific growth rate hypothesis, high amounts of ribosomal RNA (rRNA),

266   and thus P, are needed to synthesize the large quantities of proteins required to sustain high

267   specific growth rates (Sterner and Elser, 2002). Therefore, organisms with high specific growth

268   rates have relatively high P contents and low N:P ratios. This hypothesis is supported by both

269   theoretical considerations and empirical observations of zooplankton, arthropods, and bacteria

270   (Elser *et al.*, 2003; Watts *et al.*, 2006; Hessen *et al.*, 2007). However, the relationships may be

271   more complex in higher plants. Accordingly, we observed the positive correlation between the

272   specific growth rate and N:P ratio of *L. chinensis*, rather than the negative correlation predicted

273   by the hypothesis. This may have been because we monitored adult *L. chinensis* plants rather

274   than juveniles, and/or because the specific growth rate of the studied plants was strongly

275   influenced by the variation of several environmental factors (N content, P content, and pH) and

276   thus does not reflect their intrinsic potential specific growth rates.

CONCLUSION

The findings presented here demonstrate that *L. chinensis* has the homeostasis ability under a certain degree of salinity-alkalinity stress. And the N content of *L. chinensis* is more sensitive to the environmental pH than its P content. At substrate pH values of 8.4 and 8.7, *L. chinensis* possesses good environmental adaptability. In particular, at a substrate pH of 8.4, *L. chinensis* were well able to control their contents of N and P as well as the N:P ratio. Weak alkalinity (pH 7.5-8.4) is beneficial for growth and N accumulation in *L. chinensis*, but more strongly alkaline conditions (pH 8.7 or 9.3) inhibit its growth. At pH values above 8.7, the interval between stress increments clearly affected the plants' contents of N and P as well as the N:P ratio. The relationship between the specific growth rate and N:P ratio may become more complex (not simple linear) because of the salinity-alkalinity stress.

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Saline-alkali stress treatments, including durations of exposure to each pH, dates of exposure, and harvest dates.

Notes: There were 10 nutrient treatments and a total of 90 pots were used in the experiment. The salt-alkali stress experiment at incremental interval 23 days with the pH of 9.3 was not carried out due to the sudden drop of local temperature at the end of October.

N-P level	Duration (days)	pH	Dates of exposure	Harvest date
		7.5	2017/8/07-2017/8/17	2017/8/18
		8.1	2017/8/18-2017/8/31	2017/9/01
	Incremental interval 14 days	8.4	2017/9/01-2017/9/14	2017/9/15
3×N1-P		8.7	2017/9/15-2017/9/28	2017/9/29
3×N2-P		9.3	2017/9/29-2017/10/12	2017/10/13
3×N3-P		7.5	2017/8/07-2017/8/22	2017/8/23
3×N4-P		8.1	2017/8/23-2017/9/08	2017/9/09
3×N5-P	Incremental interval 17 days	8.4	2017/9/09-2017/9/25	2017/9/26
3×N-P1		8.7	2017/9/26-2017/10/12	2017/10/13
3×N-P2		9.3	2017/10/13-2017/11/05	2017/11/06
3×N-P3		7.5	2017/8/07-2017/8/27	2017/8/28
3×N-P4		8.1	2017/8/28-2017/9/19	2017/9/20
3×N-P5	Incremental interval 23 days	8.4	2017/9/20-2017/10/12	2017/10/13
		8.7	2017/10/13-2017/11/05	2017/11/06
		9.3	/	No Harvest

Homeostasis indices (H) of aboveground parts of *L. chinensis*, and their linear regression correlation coefficients (R^2), under the indicated saline-alkali stress treatments.

Notes: "—" indicates that no valid data were obtained, and "*" indicates an outlier.

pH	Incremental interval 14 days		Incremental interval 17 days		Incremental interval 23 days		Average H_N
	H_N	R^2	H_N	R^2	H_N	R^2	
7.5	3.23	0.93	3.20	0.92	3.81	0.93	3.41
8.1	5.53*	0.22*	3.70	0.98	3.87	0.95	4.37
8.4	7.25	0.95	5.36	0.83	15.58*	0.34*	6.31
8.7	5.57	0.64	2.35	0.96	3.19	0.93	3.70
9.3	3.10	0.89	—	—	—	—	3.10
	H_P	R^2	H_P	R^2	H_P	R^2	Average H_P
7.5	2.97	0.99	3.19	0.99	3.60	0.92	3.25
8.1	5.33	0.80	3.92	0.96	4.10	0.79	4.45
8.4	3.84	0.70	4.35	0.84	3.65	0.85	3.95
8.7	3.97	0.96	4.90	0.95	2.60	0.94	3.82
9.3	2.84	0.99	2.64	0.90	—	—	2.74
	$H_{N:P}$	R^2	$H_{N:P}$	R^2	$H_{N:P}$	R^2	Average $H_{N:P}$
7.5	3.15	0.96	5.47	0.82	3.06	0.95	3.89
8.1	4.84	0.82	3.68	0.93	6.44	0.79	4.98
8.4	9.22*	0.31*	6.83	0.86	4.76	0.89	6.94
8.7	5.27	0.84	5.89	0.91	3.94	0.84	5.03
9.3	3.46	0.86	3.11	0.96	—	—	3.29

Figure 1

Relationships between the N and P contents, and N:P ratios of *L. chinensis* plants and the intervals between saline-alkali stress increments. Solid circles, hollow circles and triangles refer to intervals of 14, 17, and 23 days, respectively.

Solid circles, hollow circles and triangles refer to intervals of 14, 17, and 23 days, respectively. Error bars are standard deviations.
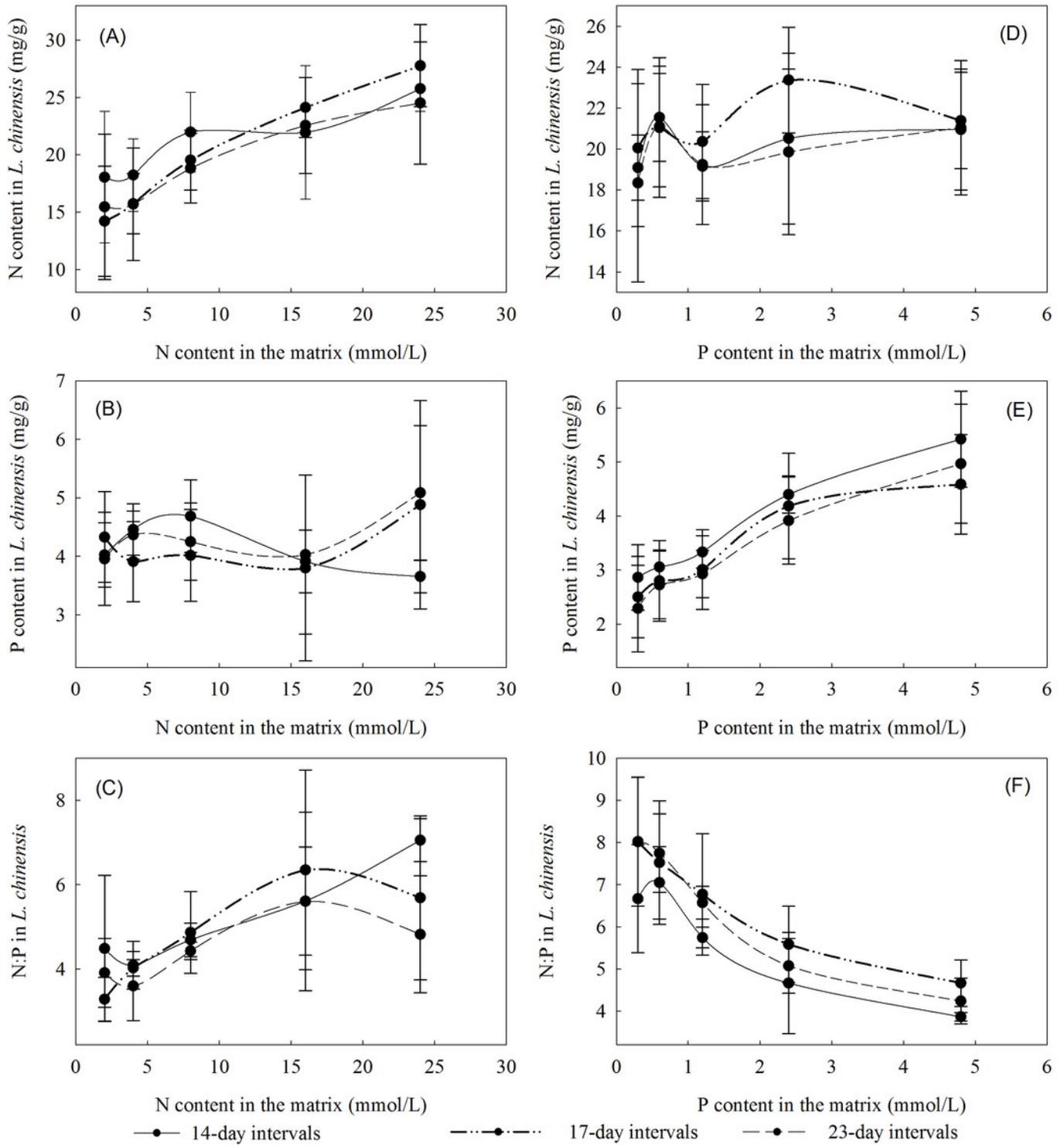
Figure 2

Homeostasis indices of *L. chinensis* at the indicated pH values and saline-alkali stress incremental intervals.
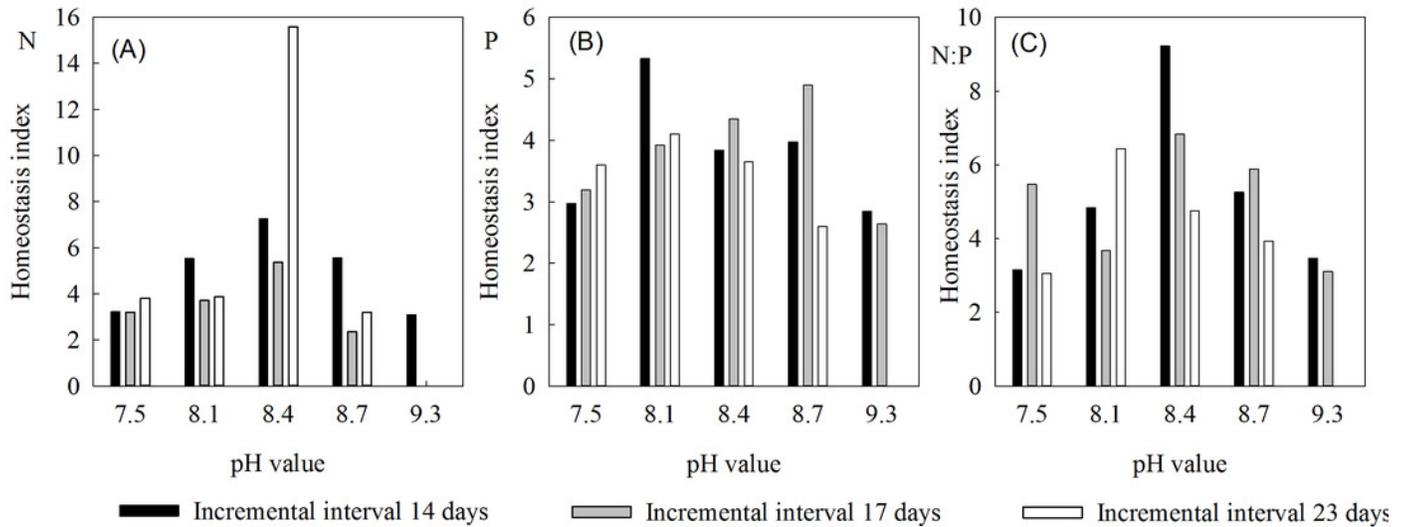
Figure 3

Specific growth rates of *L. chinensis* at the indicated environmental N and P concentrations, and pH.
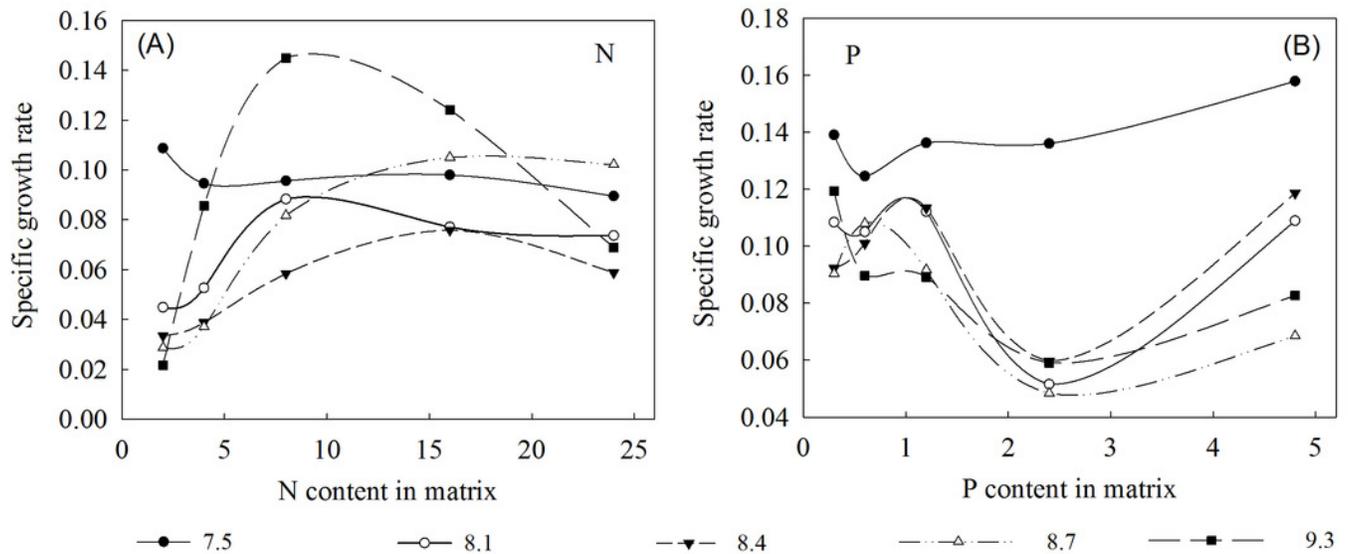
Figure 4

Relationship between the specific growth rate and the N:P ratio in the aboveground parts of *L. chinensis*.