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ROOT MORPHOLOGICAL CHARACTERISTICS OF *LESPEDEZA DAVURICA* (L.) INTERCROPPED WITH *BOTHRIOCHLOA ISCHAEMUM* (L.) KENG UNDER WATER STRESS AND P APPLICATION CONDITIONS

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Abstract

Root morphological traits such as root surface area (RSA), root average diameter (RAD), total root length (TRL), specific root area (SRA) and specific root length (SRL) per plant of *Lespedeza davurica* L. (a C_3 perennial leguminous subshrub) were investigated, when intercropped with *Bothriochloa ischaemum* (Linn.) Keng (a C_4 perennial herbaceous grass) in a pot experiment using a replacement series design. The two species were planted in the same pot at density ratios of 12:0, 10:2, 8:4, 6:6, 4:8, 2:10, and 0:12 under three levels of soil moisture 80% ± 5% FC (field capacity), sufficient water level (HW); 60% ± 5% FC, moderate water level (MW) and 40% ± 5% FC, severe low water level (LW) and two levels of P fertilization (no P (-P) and 0.1 g P_2O_5 per kg dry soil (+P)). Phosphorous application increased root biomass (RB) of *L. davurica* under LW, while decreased the root:shoot ratio (RSR) values. P application strengthened the effect of water deficit on root surface area (RSA). Significant linear regressions were observed between RSA and RB at three water levels without P application. While under P application, significant regression equation was only observed at LW. Significant linear regressions were observed between RSA and total root length (TRL) under both P treatments, irrespective of water treatments. Water level statistically significantly affected the RAD values of *L. davurica*. P application significantly decreased SRA and SRL under severe water stress. All these suggest that root proliferation of *L. davurica* was increased due to P application, to compete for limited soil resources and increase nutrient acquisition efficiency under water stress and competitive environment.

Introduction

The identification of adaptive strategies and an understanding of the physiological and ecological characters associated with are essential in understanding plant adaptation to water and P-limited ecosystems, especially in arid and semiarid regions (van Auken & Bush, 2010; Shah *et al*., 2011). In such conditions, plants display an array of responses, including morphological and architectural responses of the root system (Raghothama, 1999; Ho *et al*., 2004). Root morphological characters play an important role in belowground resource acquisition, and root data has become more significant important in evaluating environmental impacts in arid and semi-arid regions (Campbell & de Jong, 2001). Contrasting to aboveground competition for the single resource (e.g. light), much of the competition among plants takes place underground, and belowground competition often reduces plant performance more than does aboveground competition (Casper & Jackson, 1997). Many studies have been carried out on root distribution and morphology for crops, shrubs, trees and some grasses under pure stand, while less attention has been paid on plants under competitive environment or in mixture (Lynch & Brown, 2008; Bano, 2010; Riaz *et al*., 2010). The traits response will be most evident in competitive environments, including those experienced by wild plants, plants in agro-ecosystems and in the high-density genetic monocultures (Li *et al*., 2007, Lynch & Brown 2008). Theoretical modeling showed that interplant competition could be important in determining an optimal balance of plastic and non-plastic root phenotypes under P-stress and P and water stress combined conditions (Lynch & Brown, 2008). To minimize resource competition between plants, while maximizing the use of available resources, is central to improving yield and overall productivity in multi-species cropping systems (Li *et al*., 2007, Zamora *et al*., 2007). Therefore, only the analysis based on the aboveground appearance is not sufficient to clarify the essence of species competition in mixtures or natural community.

Lespedeza davurica L., and *Bothriochloa ischaemum* (Linn.) Keng are two co-dominant species in the natural grassland communities in the semiarid Loess Plateau of China. *L. davurica* is a C_3 perennial leguminous subshrub and *B. ischaemum* is a C_4 perennial grass species, and both are mainly distributed in the temperate zones of the world. Beside their potential as excellent natural pasture species, many agronomic attributes make the two wild species ideal forage species due to high adaptability and quality. Our previous results showed that there were complementary effects to grow the two species together to maximize biomass production (Xu *et al*., 2011a; Xu *et al*., 2011b). Therefore, we further investigated the morphological parameters such as root length density (RLD), specific root area (SRA) and specific root length (SRL) of the two species after the experiment. In this paper, we only reported the results of *L. davurica*, and the purposes were: 1) to clarify the root morphological characters of leguminous species in the mixture under different water and phosphorus levels; 2) to provide further understanding of the mechanisms in competition of the two species to water and phosphorus supply.

Material and Methods

Plant materials: Seeds of 2 species were obtained in the autumn of 2008 from the experimental fields at Ansai Research Station (ARS) of the Chinese Academy of Sciences (CAS) (36˚51´30"N, 109˚19´23"E, altitude 1068-1309 m a.s.l.), located at the center of the semiarid hilly-gully region on the Loess Plateau. Seed germination rates were above 90% when germinated on moist filter paper in Petri dishes at 25^{o}C.

Growth conditions: The experiment was conducted under a rainout shelter in Yangling, Shaanxi Province, China (34˚12´N, 108˚7´E, 530 m a.s.l.). The mean annual temperature is 13.0˚C, with a maximum mean monthly temperature is 26.7˚C in July and a minimum temperature is -1~-2˚C in January. Mean annual precipitation is 650 mm. The loess soil for the study was collected from the upper 20 cm of an arable field in ARS. The soil was sandy loam with 55% porosity, and its bulk density was 1.2 g cm^{-3}. Soil gravimetric moisture content at field capacity (FC) and wilting point (WP) were 20.0% and 4.0%, respectively. The soil organic matter content was 0.33%, and the total N, P and K contents were 0.019%, 0.061% and 2.01%, respectively. The available N, P and K contents were 27.72, 3.99 and 83.40 mg kg^{-1}, respectively.

Seeds of each species were sown in plastic pots sized 17.6 cm × 19 cm × 14.7 cm (height × upper inner diameter × lower inner diameter) (3.67 L). Each pot contained 3.0 kg dry soil. A plastic pipe was inserted adjacent to the inner wall to supply water from the bottom of the pot. During the seedling establishment period, the soil moisture content was maintained above 80% FC.

Species combination design: A replacement series design was used with plants grown in pots at a density of twelve plants per pot. Seven planting ratios of the two species (12:0, 10:2, 8:4, 6:6, 4:8, 2:10 and 0:12) were used and each ratio was replicated four times.

Water and phosphorous treatments: The experiment began on 28 March, 2009. The water treatment started on 30 June, 2009 when the seedling biomass of each species was about 0.2 g per seedling dry weight (DW). Just before the water treatments, 40 g perlite was used to cover the soil surface of each pot (about 2.0 cm) to reduce the compaction caused by watering and evaporation from soil surface. During that time, *L. davurica* had no new branching and was about 3-4cm in height. From then, the pots were watered regularly after weighing to maintain three watering regimes: 80% ± 5% FC (HW, sufficient water supply), 60% ± 5% FC (MW, moderate water stress) and 40% ± 5% FC (LW, severe water stress). Two phosphorous treatments include one with phosphorous addition (0.1 g P_2O_5 as KH_2PO_4 per kg dry soil) during packing pot and the other with field soil.

Shoot and root samplings: Plant biomass was sampled before and after imposing the water treatments on 30 June and 5 October 2009, respectively. At each sampling time, 12 seedlings in 4 pots were harvested, and shoots and roots were collected and washed the roots carefully. Three randomly selected pots per treatment were collected for destructive sampling at the end of growth period in November, and they were transferred to the laboratory for biomass measurements. From the selected pots, the root samples were used for the root morphology estimations prior to biomass measurements. The root was separated from the soil, under a gentle water jet, using a sieve to collect any root fragments detached from the system. Due to the large size of the root, about 1/3 of total root was selected for morphological measurements. The selected root sample was dyed with 0.5% methylene blue solution for 5 minutes, then gently dried with absorbent paper, and was put in two transparent plastic sheets to keep it in a fixed position and improve the contrast of the root image. The sheet was placed on a scanner (BENQ colour scanner 5560) connected to a computer, and an image analysis system (DT-Scan, Delta T-Devices) was used to determine the total root length (TRL), the root surface area (RSA), root diameter (RD) and the total root length (TRL) (Tsakaldimi *et al*., 2005). Shoot biomass was further separated into leaf and stem. Dry weight (DW) of each part was determined after drying at 75°C for 48 h in an oven. Root:shoot ratio (RSR) was calculated as the ratio of root dry biomass to shoot dry biomass. Specific root length (SRL, cm g^{-1}) was calculated as the total root length divided by the root biomass for each individual plant (Trubat *et al*., 2006, Cortina *et al*., 2008), and specific root surface area (SRA) was surface area per unit dry weight of the root biomass per plant (Hill *et al*., 2006).

Statistical analysis: Our treatments were a factorial combination of seven mixture ratios, two phosphorous treatments and three water levels, arranged into a completely randomized design inside the rainfall shelter. Treatments were in four replications. Data were processed using the Microsoft office Excel 2003 for windows. An analysis of variance about the main effects of mixture ratios, phosphorous treatments, water level and their interactions was conducted with SPSS 17.0 statistical software (SPSS, Chicago, IL, USA). Treatment means were compared using Tukey's HSD test at the 0.05 probability level. Root biomass (RB), TRL and RSA were then regressed against one another to explore treatment constraints on root characteristics.

Results

Root biomass (RB) and root:shoot ratio (RSR): Under HW and MW treatments, P application increased the root biomass (RB) except when the proportions of *B. ischaemum* to *L. davurica* were 2:10, 4:8 and 10:2, while under LW, P application increased RB of *L. davurica* in each proportion (Table 1). In the same mixture ratio, water stress disproportionately decreased the RB of *L. davurica*. There were no consistent changes in RB with the proportion changes of *L. davurica* in the mixtures (Table 1). Water, phosphorous, mixture ratio and their interactive effects statistically significant affected the RB of *L. davurica*, and the interactive effect of mixture ratio and phosphorous was also statistically significant (Table 2).

Table 1. Root biomass (RB) and root:shoot ratio (RSR) per plant of *Lespedeza davurica* at three levels of water and two levels of phosphorous treatments in the mixtures.

Water treatments	Planting scheme (B/L)	RB(g)		RSR	
		-P	+P	-P	+P
HW	0/12	0.51 ± 0.05	0.47 ± 0.03	1.01 ± 0.08	0.62 ± 0.04
	2/10	0.47 ± 0.02	0.47 ± 0.03	1.05 ± 0.07	0.76 ± 0.04
	4/8	0.48 ± 0.10	0.41 ± 0.07	1.20 ± 0.09	0.80 ± 0.05
	6/6	0.39 ± 0.08	0.48 ± 0.04	1.15 ± 0.10	0.79 ± 0.06
	8/4	0.30 ± 0.02	0.53 ± 0.07	0.90 ± 0.04	0.76 ± 0.03
	10/2	0.46 ± 0.10	0.46 ± 0.02	1.06 ± 0.10	0.89 ± 0.18
MW	0/12	0.49 ± 0.03	0.45 ± 0.08	1.12 ± 0.09	0.75 ± 0.09
	2/10	0.44 ± 0.06	0.41 ± 0.05	1.08 ± 0.10	0.82 ± 0.12
	4/8	0.44 ± 0.02	0.40 ± 0.04	1.22 ± 0.09	0.84 ± 0.06
	6/6	0.35 ± 0.04	0.46 ± 0.06	1.18 ± 0.09	0.77 ± 0.04
	8/4	0.31 ± 0.03	0.44 ± 0.08	1.01 ± 0.15	0.80 ± 0.13
	10/2	0.45 ± 0.04	0.45 ± 0.05	1.14 ± 0.21	1.13 ± 0.21
LW	0/12	0.29 ± 0.01	0.33 ± 0.05	1.25 ± 0.16	0.94 ± 0.08
	2/10	0.29 ± 0.01	0.32 ± 0.05	1.23 ± 0.10	1.03 ± 0.10
	4/8	0.31 ± 0.01	0.32 ± 0.05	1.27 ± 0.10	0.99 ± 0.12
	6/6	0.24 ± 0.05	0.31 ± 0.03	1.17 ± 0.04	0.95 ± 0.07
	8/4	0.23 ± 0.02	0.33 ± 0.07	1.16 ± 0.02	0.96 ± 0.19
	10/2	0.19 ± 0.04	0.37 ± 0.02	1.10 ± 0.12	1.14 ± 0.03

The replacement series B/L represents the density of *Bothriochloa ischaemum* as the first species versus *Lespedeza davurica* in each pot (HW: sufficient water supply, 80%±5% FC; MW: moderate water stress, 60%±5% FC; LW: severe water stress, 40%±5% FC). Values within a column followed by different letters are significantly different (P<0.05), while within a row for each index followed by different letters in brackets are also significantly different (P<0.05).

Table 2. Summary (F and P values) of analysis of variance for the effects of mixture ratio, water and phosphorous treatments on root biomass (RB) and root:shoot ratio (RSR) per plant of *Lespedeza davurica* in mixtures.

Effect	df	RB(g)		RSR	
		***F*-value**	***P*-value**	***F*-value**	***P*-value**
Phosphorous level (PL)	1	24.257	**<0.001**	152.640	**<0.001**
Water level (WL)	2	128.302	**<0.001**	27.422	**<0.001**
Mixture ratio (MR)	5	4.642	**0.001**	5.257	**<0.001**
WL × PL	2	3.067	0.051	1.939	0.149
MR × PL	5	11.160	**<0.001**	5.992	**<0.001**
MR × WL	10	0.730	0.695	1.208	0.294
MR × WL × PL	10	2.443	**0.011**	0.439	0.924

Probabilities considered statistically significant (P<0.05) are indicated in bold typeface

P application decreased the RSR values of *L. davurica* under each mixture ratio and water treatment, except in the ratio 10:2 of *B. ischaemum* to *L. davurica* under LW (Table 1). There was also no obvious trend in RSR change of *L. davurica* with mixture ratio under three water levels and two P applications (Table 1). Water, phosphorous, mixture ratio and their interactive effects statistically significantly affected the RSR of *L. davurica*, and the interactive effect of mixture ratio and phosphorous was also statistically significant (Table 2).

Total root length (TRL): P application significantly increased total root length (TRL) of *L. davurica* in each ratio (Fig. 1). Under no P application treatment, TRL decreased gradually as the proportion of *L. davurica* increased in the mixtures, except when *B. ischaemum* to *L. davurica* was at 10:2 under HW and MW, while under P application treatment no obvious was change observed (Fig. 1). The averaged TRL values for all mixtures under HW and MW were significantly higher than under LW, and there were significantly differences between the former two (Fig. 1). Under no P application treatment, the TRL values under monoculture decreased significantly as water stress increased, while under P application treatment, TRL values in MW were slightly higher than in HW, and both were significantly higher than in LW (Fig. 1). Water, phosphorous, mixture ratio and their interactive effects significantly affected the RB of *L. davurica*, and the interactive effects of mixture ratio with phosphorous and water with phosphorous were also statistically significant (Table 2). Under no P application, significant linear regressions were only observed between total root length (TRL, y) and root biomass (RB, x) in MW, and the regression equations were y= 11077x +57.10 (R^2=0.29, P=0.0173) (Fig. 2). While under P application, no regression equations for the three water levels could be obtained (Fig. 2). When results were combined under three water treatments, significant linear regressions were observed between total root length (TRL) and root biomass (RB), and the equations were y = 1713.5x - 148.08 (R^2 = 0.50, P<0.0001) and y = 1370.9x + 6.84 (R^2 = 0.87, P=0.0034) for no versus P application, respectively (Fig. 2).

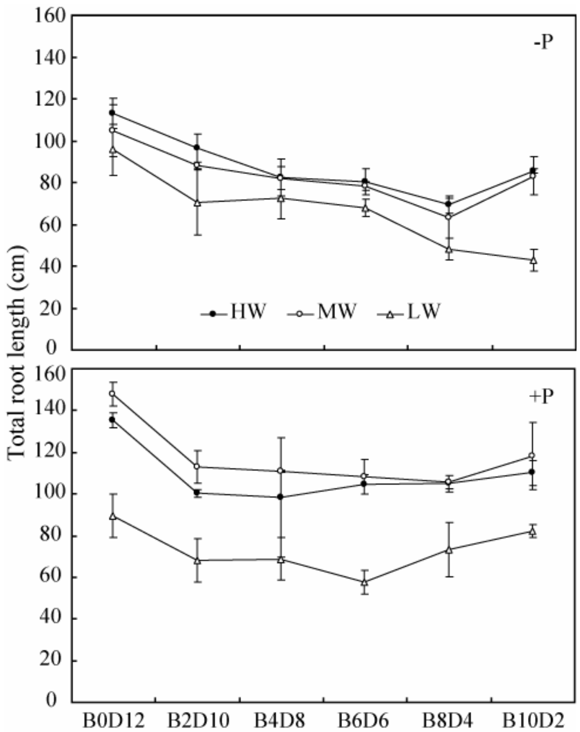

Fig. 1. Total root length (TRL) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for each water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). Bars are standard errors of means (n = 3).

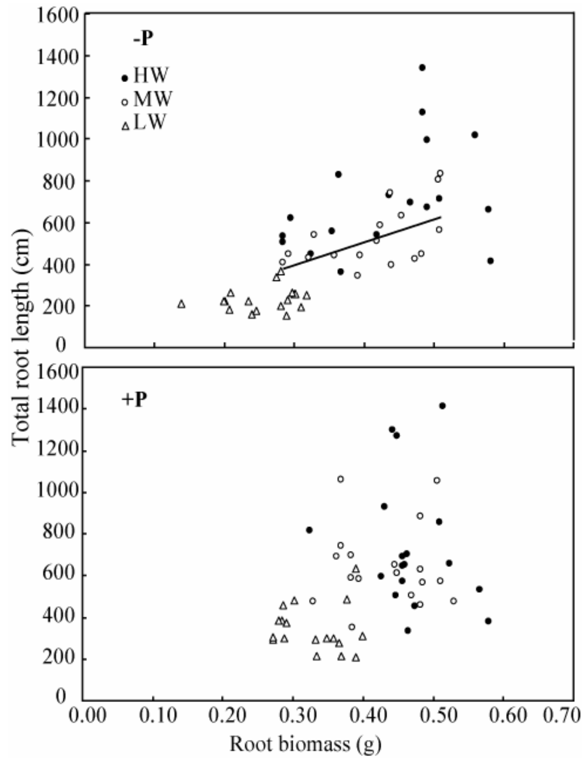

Fig. 2. Scatter plots of root biomass (RB) and total root length (TRL) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for all three water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). *Trend lines* fitted to linear regression

Root surface area (RSA): P application significantly increased average root surface area (RSA) of *L. davurica* in mixtures, while only significantly improved the values at HW and MW in monoculture (Fig. 3). Under no P application treatment, RSA decreased gradually as the proportion of *L. davurica* decreased in the mixtures, except when *B. ischaemum* to *L. davurica* was at the 10:2 ratio under HW and MW treatments (Fig. 3). Severe water stress (LW) significantly decreased the single plant root surface area (RSA) values of *L. davurica*, and P application strengthened the effect of water deficit (Fig. 3). Under no P application, MW and LW treatments averagely brought about 1.3% and 24.6% decrease of RSA of *L. davurica* in mixtures, while those were 11.2% and 19.5% in monoculture respectively, comparing with HW. Compared with MW treatment, LW decreased about 23.7% and 19.3% in mixtures and monoculture, respectively (Fig. 3). Under P treatment, MW and LW treatments averagely brought 11.1% and 38.3% decrease of RSA of *L. davurica* in mixtures, while those were 11.4% and 38.9% for monoculture, respectively, comparing with HW treatment. As compared with MW, LW treatments decreased about 30.6% and 31.0% of RSA of *L. davurica* in mixtures and monoculture, respectively (Fig. 3). The effects of phosphorous, water level, mixture ratio or the interactive effect of water with phosphorous were statistically significant on RSA of *L. davurica* (Table 3). Significant linear regressions were observed between root surface area (RSA, y) and root biomass (RB, x) at three water levels without P application, and the regression equations for HW, MW and LW were $y = 54.603x + 17.049$ (R^2=0.47, P=0.002), $y = 55.864x + 16.221$ (R^2=0.50, P=0.0012) and $y = 89.91x + 7.9816$ (R^2=0.34, P=0.0167), respectively (Fig. 4). While under P application treatment, significant regression equation was only observed at LW and which was $y = 105.32x - 2.06$ (R^2=0.46, P=0.0017) (Fig. 4). Significant linear regressions were also observed between root surface area (RSA) and total root length (TRL) (Fig. 5). Under no P application treatment, these were only observed at HW and MW, and which were $y = 0.0182x + 27.846$ (R^2=0.37, P=0.0078) and $y = 0.023x + 27.475$ (R^2=0.35, P=0.0092), respectively. Under P application treatment, significant linear regressions were observed for each water level, and they were $y = 0.0205x + 38.175$ (R^2=0.73, P<0.0001), $y = 0.0213x + 33.63$ (R^2=0.38, P=0.0061) and $y = 0.0301x + 22.51$ (R^2=0.23, P=0.0464) at HW, MW and LW, respectively (Fig. 5). When the water levels not considered, significant linear regressions were also observed between root surface area (RSA, y) and total root length (TRL, x) under both P treatments, and which were $y = 0.0204x + 27.175$ (R^2=0.43, P<0.0001) and $y = 0.0327x + 25.717$ (R^2=0.66, P<0.0001) respectively.

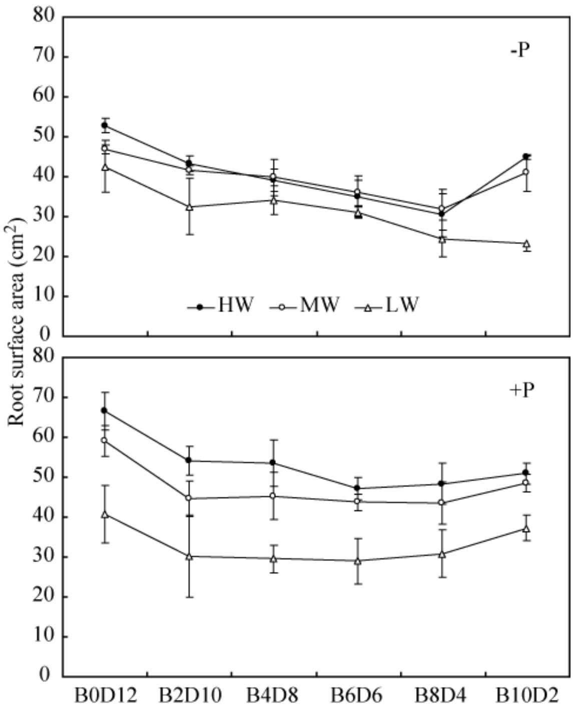

Fig. 3. Root surface area (RSA) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for each water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). Bars are standard errors of means (n = 3).

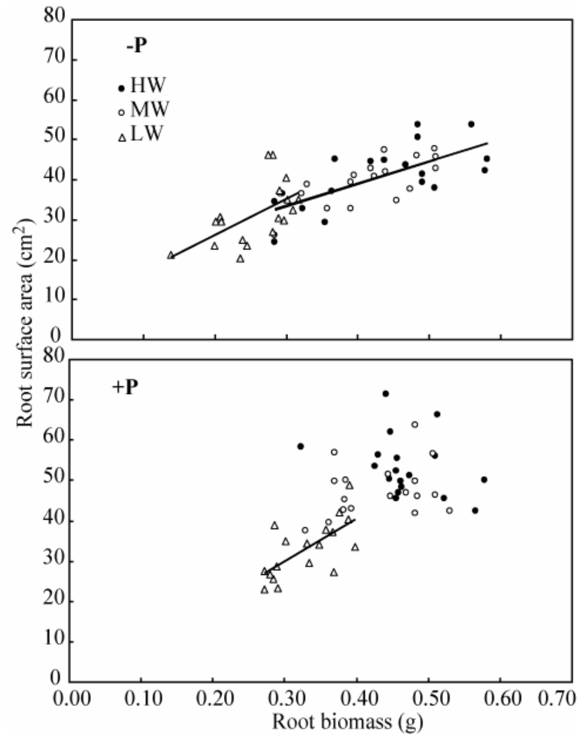

Fig. 4. Scatter plots of root biomass (RB) and root surface area (RSA) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for all three water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). *Trend lines* fitted to linear regression.

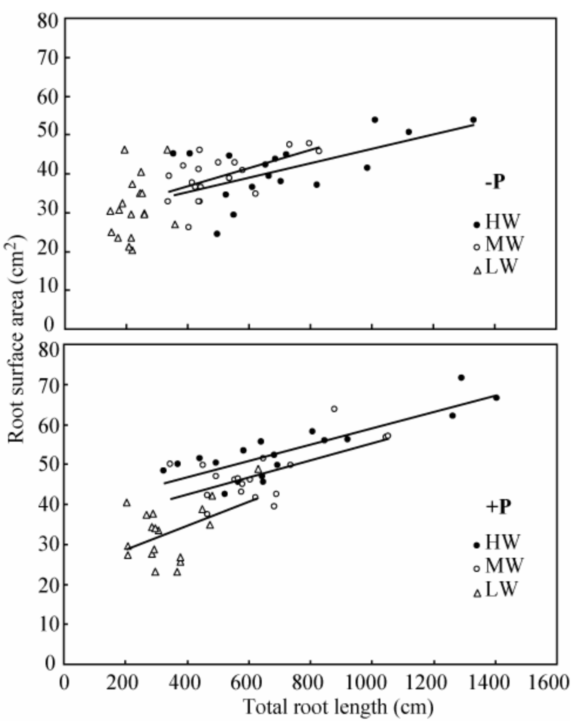

Fig. 5. Scatter plots of total root length (TRL) and root surface area (RSA) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for all three water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). *Trend lines* fitted to linear regression.

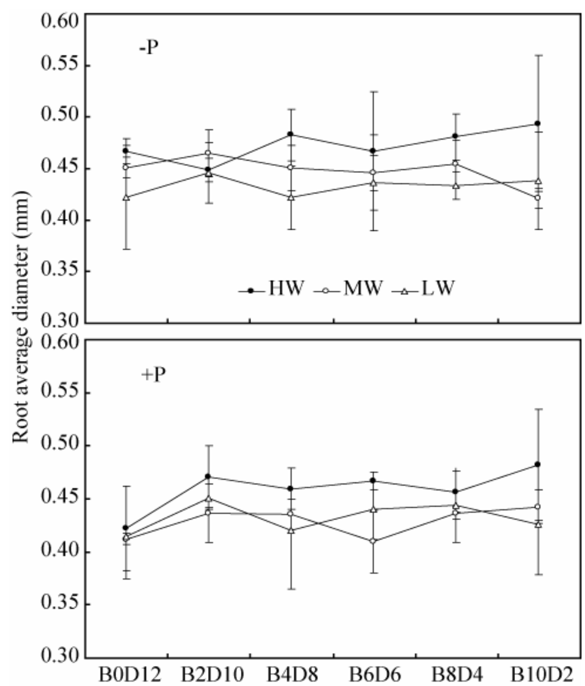

Fig. 6. Root average diameter (RAD) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for each water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). Bars are standard errors of means (n = 3).

Table 3. Summary (F and P values) of analysis of variance for the effects of mixture ratio, water treatment and phosphorous on root surface area (RSA), root average diameter (RAD), total root length (TRL) specific root area (SRA) and specific root length (SRL) of *Lespedeza davurca* in mixtures.

Effect	df	RSA(cm^2)		RAD(mm)		TRL(cm)		SRA($cm^2.g^{-1}$)		SRL($m.g^{-1}$)	
		***F*-value**	***P*-value**	***F*-value**	***P*-value**	***F*-value**	***P*-value**	***F*-value**	***P*-value**	***F*-value**	***P*-value**
Phosphorous level (PL)	1	71.030	**<0.001**	2.179	0.144	136.435	**<0.001**	1.017	0.317	8.661	**0.004**
Water level (WL)	2	106.055	**<0.001**	8.470	**<0.001**	124.027	**<0.001**	2.523	0.087	2.839	0.065
Mixture ratio (MR)	5	28.225	**<0.001**	0.985	0.433	35.194	**<0.001**	4.067	**0.003**	6.404	**<0.001**
WL × PL	2	12.927	**<0.001**	0.723	0.489	19.389	**<0.001**	15.390	**<0.001**	15.492	**<0.001**
MR × PL	5	1.939	0.098	0.514	0.765	6.032	**<0.001**	2.161	0.068	2.519	**0.037**
MR × WL	10	0.820	0.611	0.454	0.914	0.745	0.680	0.892	0.545	0.481	0.897
MR × WL × PL	10	1.880	0.062	0.446	0.918	2.080	**0.037**	0.864	0.570	1.042	0.418

Probabilities considered statistically significant (P<0.05) are indicated in bold typeface

Root average diameter (RAD): There were no obvious trends in root average diameter (RAD) of *L. davurica* in their various mixture proportions within the replacement series under each water level (Fig. 6). Under no P application treatment, the average RAD values were 0.47, 0.45 and 0.43 mm for HW, MW and LW for the mixtures, respectively. In monoculture, the average RAD values were 0.47 and 0.45 at HW and MW, respectively, and they were significantly higher than LW (0.42mm) (Fig. 6). Under P application treatment, the average RAD was 0.47, 0.43 and 0.44 mm for HW, MW and LW in mixtures, respectively, while those were 0.42, 0.41 and 0.41 mm, respectively in monoculture (Fig. 6). Only water treatment statistically significantly affected the RAD values of *L. davurica* (Table 3).

Specific root area (SRA) and specific root length (SRL): Under no P application, LW had significantly higher averaged SRA values in all mixture ratios than MW and LW treatments, and there was a significant difference between MW and LW (Fig. 7). While under P application, HW had significantly higher averaged SRA values than HW and LW, and there was no significant difference between the later two. P application significantly increased SRA at HW and MW, while decreased SRA at LW level (Fig. 7). The average SRL values in the mixtures under each treatment had the similar trend as SRA (Fig. 8). The mixture ratio and the interactive effect of water with phosphorous were statistically significant on SRA of *L. davurica* (Table 3). The phosphorous level, mixture ratio and the interactive effects of water with phosphorous and mixture ratio with phosphorous were all statistically significant on SRL of *L. davurica* (Table 3).

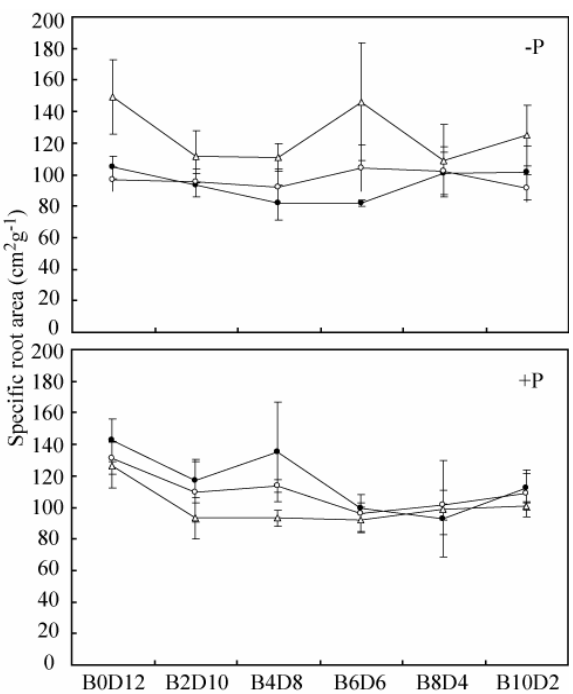

Fig. 7. Specific root area (SRA) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for each water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). Bars are standard errors of means (n = 3).

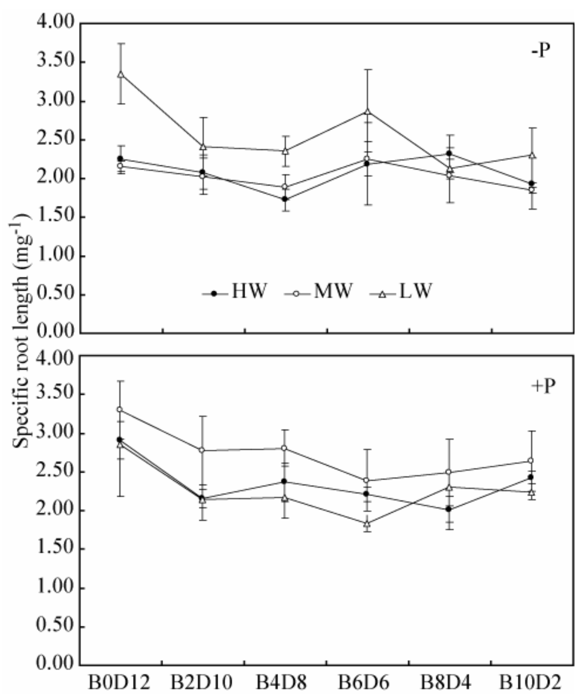

Fig. 8. Specific root length (SRL) of *Lespedeza davurica* in the replacement series under both pure stands and mixed stands for each water treatment (HW: sufficient water supply; MW: moderate water stress; LW: severe water stress). Bars are standard errors of means (n = 3).

Discussion

The importance of root morphological traits has long been recognized as crucial for plants to cope with drought and nutrient deficiency conditions, especially in arid and semiarid terrestrial ecosystems (Raghothama, 1999; Ho *et al*., 2005; Zia-ul-hassan & Arshad, 2011). Although numerous studies have focused on the interactions of water and phosphorus availability for plant growth, studies describing specific root traits that enable a plant co-optimise acquisition of both these resources under intercropping system were not well documented (Ho *et al*., 2005, Hinsinger *et al*., 2011).

Water stress is the primary limiting factor for biomass production of *L. davurica*, regardless of mixture proportion or P application (Table 1). Under LW treatment, P application significantly improved root biomass production of *L. davurica* at each proportion, which meant that P was favorable for *L. davurica* growth under severe water deficit condition (Table 1). Increased relative allocation to root growth is obviously beneficial for phosphorus acquisition and drought tolerance, but may slow overall plant growth because of the increased respiratory burden of root tissue (Nielsen *et al*., 2001, Fan *et al*., 2003). Our results also confirmed that P application significantly decreased the RSR values of *L. davurica* as reputed by Lynch & Brown (2008) (Tables 1-2).

Root average diameter (RAD) has been taken as important root properties to evaluate plant adaptability (Gahoonia & Nielsen 2004). Decreased root diameter is one of the important mechanism for plants to adapt water deficit environment not only because it reduces the quantity of water that must be absorbed but also because it increases the surface to volume ratio, thus facilitating water uptake (van der Weele *et al*., 2000). Most species decreased root diameter in response to P deficiency (Hill *et al*., 2006), while some others especially *Arabidopsis thaliana*, its root diameters were larger in low phosphorus conditions (Ma *et al*., 2001). Plant root systems have inherent capability to adjust to prevailing environmental conditions through their morphological and physiological plasticity (Zamora *et al*., 2007). In this experiment, water stress had significantly effected on RAD, while it showed small fluctuations in all mixture ratios (Fig. 6), and which was also strongly supported by the highly significant positive relation between total root length (TRL) and root surface area (RSA) in all stands and treatments (Fig. 5).

Plants had greater rate of root surface area (RSA) production per unit of root length and root dry weight, and a greater rate of root length per unit root dry weight under P than without P application treatment, especially in LW conditions (Figs. 2, 4-5). The higher slopes of the linear regression of root surface area on root biomass under LW irrespective of P application, demonstrating a larger root surface area per unit of root biomass for *L. davurica* under sever water stress (Fig. 4). While the linear relationships between total root surface area and total root length had different slopes under all except under LW and without P application treatment (Fig. 5), and the straight-line slope was significantly higher, suggesting that *L. davurica* had a greater rate of root surface area production per unit of root length (Anderson *et al*., 2007).

Specific root area (SRA) and specific root length (SRL) are considered as good general characteristics of root structure, and has been applied to study variations in root morphology in relation to different nutrient levels, water content and soil types (Lohmus *et al*., 1989, Tsakaldimi *et al*., 2005, Zobel *et al*., 2006; Ostonen *et al*., 2007). Higher SRL implies thinner roots and a better exploitation efficiency (Nicotra *et al*., 2002, Trubat *et al*., 2006), and also indicates a lower constructive cost for the formation of root system under the condition of intensive competition environments (Eissenstat, 1992; Roumet *et al*., 2006). In the present study, significant higher SRL and SRA were only observed under LW without P application (Figs. 7-8). Changes in root SRL leading to increase SRA were important for most species adjusting to low P (Hill *et al*., 2006). Relatively larger SRL values under LW without P application were achieved by plant adjusting as a consequence of large decrease in RAD and root mass, while small decrease in RSA, resulting thinner roots under water and phosphorus deficit conditions (Figs. 7-8).

Across treatments, *L. davurica* converged on the same root surface area per unit of root length (Fig. 5), but it did so by employing different morphological strategies under different treatments. Assuming equal tissue mass densities between treatments, *L. davurica* arrived at its surface area by having greater root biomass and smaller root length under P application, and the negative relationship between root biomass and total root length implied that P application increased its root diameter, especially under severe water stress (e.g. LW treatment). Similar results were also reported by Xie *et al*., (2006), who presented that plant with long, fine roots should be more efficient in nutrient acquisition than a plant with short, thick ones suggesting that the formation of fine roots is favorable for plant to adapt to infertile or competitive environments.

In summary, P application significantly decreased the root:shoot ratios, while significantly increased total root length and root surface area of *L. davurica* in mixtures. Under severe water stress, P application could significantly decrease SRA, SRL and RAD, while increased RB of *L. davurica* within the mixtures. Because water is more mobile than phosphorus, root surface area and total length are not as critical for water acquisition as it is for phosphorus acquisition (Ho *et al*., 2005). Thus, the explanation for the morphological response of *L. davurica* was root proliferation under water stress due to P application, which was likely to aid in the competition for limited soil resources and increasing nutrient acquisition efficiency.

Our results suggest that, in the semiarid Loess Plateau of China, which has degraded loess soil with low P availability and limited rainfall, the improvement of root morphological traits by the application of a proper amount of P fertilizer might offset the adverse effects of water deficit on plant growth. Given the complexity of the underlying mechanisms in the intercropping system, the accurate amount of P application, the root development characteristics and intercropping ratio of the mixture need further research.

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