#### **RESEARCH ARTICLE**

# Effects of Phosphorus Application in Different Soil Layers on Root Growth, Yield, and Water-Use Efficiency of Winter Wheat Grown Under Semi-Arid **Conditions**

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## Abstract

Deep phosphorus application can be a usefull measure to improve crops' performance in semi-arid regions, but more knowledge of both its general effects and effects on specific crops is required to optimize treatments. Thus, the aims of this study were to evaluate the effects of phosphorus (P) application at different soil layers on root growth, grain yield, and water-use efficiency (WUE) of winter wheat grown on the semi-arid Loess Plateau of China and to explore the relationship between root distribution and grain yield. The experiment consisted of four P treatments in a randomized complete block design with three replicates and two cultivars: one drought-sensitive (Xiaoyan 22, XY22) and one drought-tolerant (Changhan 58, CH58). The four P treatments were no P (control, CK), surface P (SP), deep P (DP), and deep-band P application (DBP). CH58 produced larger and deeper root systems, and had higher grain yields and WUE, under the deep P treatments (DP and DBP) than under SP, clearly showing that deep P placement had beneficial effects on the drought-tolerant cultivar. In contrast, the grain yield and root growth of XY22 did not differ between DP or DBP and SP treatments. Further, root dry weight (RW) and root length (RL) in deep soil layer (30-100 cm) were closely positively correlated with grain yield and WUE of CH58 (but not XY22), highlighting the connections between a well-developed subsoil root system and both high grain yield and WUE for the drought-tolerant cultivar. WUE correlated strongly with grain yield for both cultivars ( $r=0.94$ ,  $P<0.001$ ). In conclusion, deep application of P fertilizer is a practical and feasible means of increasing grain yield and WUE of rainfed winter wheat in semi-arid regions, by promoting deep root development of drought-tolerant cultivars. **Examplication can be a usefull measure to improve crops' perform an eg in semi-arid regional all effects and effects on specific crops is required to optimize treatments. Thus, the air est est of phosphorus (P) applicatio** 

Key words: water stress, phosphorus application, soil layers, grain yield, root growth, water-use efficiency

## INTRODUCTION

Available phosphorus (P) typically becomes increasingly concentrated in cultivated and fertilized topsoils due to shallow application of phosphate and the poor mobility of P in soil. Thus only a small proportion of the applied P reaches the subsurface soil (Yin and Vyn 2002; Mallarino and Borges 2006). In semi-arid environments, the mobility of P is further reduced by water deficits in the soil restricting diffusion. This is a common problem on the Loess Plateau in China, where the topsoil frequently dries out during the growing

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season (Wang *et al*. 2009). Hence, crops grown on the Plateau and in other semi-arid environments are prone to P deficiency, which may adversely affect their growth, yield, and water-use efficiency (WUE) (He *et al*. 2002; Fernández *et al*. 2009; Waraich *et al*. 2011).

It is generally believed that increasing use of water stored in deep layers is essential for improving crop productivity in semi-arid regions (Walter and Morio 2005). However, deep placement of P fertilizer in the subsoil may alleviate the low availability of P in deep layers of soil in water-limited environments, where plants may have continued access to substantial amounts of water, even if the topsoil is dry. Plants can redistribute water and nutrients from moist subsoil layers to a drier topsoil through hydraulic lift by the roots (Huang 1999; Bauerle *et al*. 2008), but this process alone is not sufficient to overcome the reduced availability of nutrients in the topsoil unless the subsoil layers are periodically fertilized. Deep fertilization can therefore solve the problem of a spatial mismatch of nutrients and water. Fertilizing the subsoil encourages roots to grow deeper by creating patches with favourable physical and chemical conditions throughout the root zone (Grewal and Graham 1999). Because the subsoil retains its moisture better than the topsoil, this can increase the availability of nutrients in applied fertilizers to crops for longer periods during the growing season, thereby enhancing the uptake and utilisation of the nutrients by the crops (Ma *et al*. 2009), as illustrated by the following finding. Kuhlmann (1990) reported a positive correlation between P concentrations in spring-wheat shoots and available P in the subsoil (>30 cm) of a loess-parabrown soil in northern Germany. The absorption of water and nutrients from deep soil layers has been shown to improve both yield and WUE of soybean (Borges and Mallarino 2000). Alston (1976) found that under dry topsoil and moist subsoil conditions, placement of nitrogen (N) and P at 25 cm depth resulted in wheat crops with longer roots, as well as higher WUE, grain yields, and P concentrations than placement at 5 cm. In a study spanning 15 site-years, Borges and Mallarino (2001) observed minor differences in corn yield between broadcasting and deep-band (15-20 cm) P applications although the latter resulted in better yields at one site. However, *H* and the low availability of nurfacture that and the same of soil in water-limited environments, the shown that crop cenely and y and the constrained access to substantial efficiency of acquisition and util ther, even i

they later reported that deep-band P could reduce P losses due to surface runoff by reducing the accumulation of P near the soil surface (Borges and Mallarino 2003). In contrast, Singh *et al*. (2005) reported that deep-band of P (10-15 cm) increased wheat yields by 30-43% compared with conventional P application (5-10 cm). Thus, deep placement of nutrients can be highly advantageous, but the benefits may vary with agrotype (Adjetey *et al*. 1999), level of soil fertility (Alston 1980), and moisture content of the topsoil (Valizadeh *et al*. 2003).

It is known that crop genotypes can differ in the efficiency of acquisition and utilisation of water and nutrients (Ma *et al*. 2007; Chen *et al*. 2012), and the application of fertilizers is very important for increasing yield under water stress (Prasertsak and Fukai 1997; Vaemerali *et al*. 2009). Furthermore, the efficiency of water and nutrient acquisition is correlated with the uptake per unit root length and weight, especially in deeper roots (Ball-Coelho *et al*. 1998; Henry *et al*. 2009). Hence, deep rooting, high root-length density in deep soil and high proportions of deep roots have been identified as drought-adaptive traits that can increase crop yields in water-limited environments (Ludlow and Muchow 1990). In addition, under water stress, drought-tolerant genotypes have significantly more root dry matter and higher root length densities in deep soil than drought-sensitive genotypes (Matsui and Singh 2003). However, there is little knowledge of the differences in responses (if any) of drought-tolerant and drought-sensitive crop cultivars to deep placement of P fertilizer.

A better understanding of the effects of subsoil P availability on grain yields and WUE of winter wheat under semi-arid conditions could lead to better predictions of crops' responses to P fertilizer. Therefore, the ames of this study were: 1) to determine if grain yield of winter wheat can be improved by manipulating the redistribution of P; 2) to improve our understanding of optimal fertilization regimes for winter wheat in dryland agriculture; and 3) to assess the variation in root traits and the relationship between deeper roots and yield in a drought-tolerant and a drought-sensitive cultivar of winter wheat in response to water stress. A site on the Loess Plateau of China was chosen for the study, as it is a semi-arid environment with dry topsoil and moist subsoil.

# RESULTS

#### Grain yield and yield components

As shown in Table 1, the grain yield and yield components varied considerably between cultivars and P treatments. The drought-tolerant cultivar, Changhan 58 (CH58), produced significantly more (3.0-35.5%, *P*<0.05) spike numbers than the drought-sensitive cultivar, Xiaoyan 22 (XY22) under the condition of the lower rainfall during the early growth stage of winter wheat, which was only 29.2 mm precipitation from October to January. Consequently, CH58 had 10.8-46.0% higher grain yields than XY22. As expected, the addition of P significantly affected grain yield and spike number of both cultivars (*P*<0.05). For XY22, grain yields were 1.66-, 1.46-, and 1.46-fold higher under the surface P (SP), deep P (DP), and deep-band P (DBP) treatments, respectively, than under the control treatment (CK). However, there were no significant differences in yield between SP, DP, and DBP treatments. For CH58, grain yields were 1.48- and 1.49-fold higher in DP and DBP plots, respectively, than in CK plots, but there was no significant difference in this respect between DP and DBP plots. Both DP and DBP resulted in 1.16-fold higher grain yields than SP. The grain yield was 27.8% higher under the SP treatment than under CK. For 122 Junctic to contain of the lower of the control of the early growth stage of winter wheat, stages (Table 2). The RW of CF  $29.2$  mm precipitation from October to higher than XY22 under all treat equently, CH58 had

Correlation analysis showed that grain yield was closely correlated with spike number (*r*=0.85, *P*<0.001), panicle length (*r*=0.6863, *P*<0.001), and 1 000-kernel weight  $(r=0.74, P<0.001)$  in both cultivars (Fig. 1), indicating that not only P concentrations, but also the vertical distribution of P in the soil affected grain yield.

#### Root weight and its spatial distribution

Total root dry weight (RW, 0-100 cm) was affected by cultivar, P treatments, and the interaction between cultivar and P treatments  $(P<0.01)$  in all four growth stages (Table 2). The RW of CH58 was significantly higher than XY22 under all treatments (*P*<0.05). RW was the highest under the deep P treatments (DP and DBP), followed by the surface treatment (SP) and the lowest under CK. There was little or no difference in RW between the DP and DBP treatments. This pattern was found for both cultivars and at all growth stages. Analyses based on the whole soil profile and samples from discrete depths gave similar results. Phosphorus enrichment (SP, DP, and DBP) significantly increased RW, by 18.1-41.2%, compared with CK at the jointing stage, but there were no differences in this respect between the other treatments (SP, DP, and DBP). Overall, the RW values were the highest at the booting stage and lower at the milking stage. This decrease was more pronounced under the CK and SP treatments than under DP and DBP. At maturity, RW was significantly higher (15.7-24.4%) under the deep P treatments (DP and DBP) than under the surface treatment (SP).





<sup>1)</sup> CK, no P application; SP, surface application; DP, deep application; DBP, deep band application.

Different letters within each column and cultivar indicate significant differences at *P* ≤0.05. \* and \*\* represent probabilities of ≤0.05 and ≤0.01, respectively. ns, not significantly different.

The same as below.



**Fig. 1** Relationships between grain yield and spike number (A), panicle length (B), and 1 000-kernel weight (C) for Xiaoyan 22 (XY22) and

**Table 2** Changes of root dry weight in the 0-100 cm soil profile under different treatments (g m<sup>-2</sup>)

Cultivar	Treatments	Jointing	Booting	Milking	Maturity
Xiaoyan 22 (XY22)	CK	65.7 b	119.1 <sub>b</sub>	97.6c	79.4 c
	SP	75.4 a	147.4 a	126.5 <sub>b</sub>	106.2 b
	D₽	77.2 a	147.9 a	133.2 a	113.8 a
	<b>DBP</b>	80.3a	150.0a	132.8 a	113.4 a
Changhan 58 (CH58)	CK	82.0 <sub>b</sub>	136.5 <sub>b</sub>	121.7c	101.2c
	<b>SP</b>	113.0 a	176.8 a	161.0 <sub>b</sub>	139.8 b
	DP	115.6 a	177.0 a	171.2a	147.6 a
	<b>DBP</b>	118.8 a	178.1 a	172.1a	150.2a
Source of variation					
Cultivar		$\pm\pm$	$\pm$ s	$8.8\,$	88
P		$\pm\pm$	$* *$	88	88
Cultivar×P		$\pm\pm$	**	**	**

Analysis of the vertical distribution of RW showed that it was consistently higher in the top soil layer (0- 20 cm) of SP plots than in plots subjected to any other treatment, for both cultivars at the four growth stages (Fig. 2). The proportion of RW found in the deep soil layers (30-100 cm) was higher for the drought-tolerant cultivar CH58 than for the drought-sensitive cultivar XY22 and the difference was most pronounced under the deep treatments.

Analyses of temporal changes revealed that the proportion of RW in deep soil layers (30-100 cm) increased notably after the jointing stage under the DP and DBP



**Fig. 2** The effects of different treatments on root weight density at jointing, booting, milking, and maturity stages for Xiaoyan 22 (XY22) and Changhan 58 (CH58). Data in boxes mean the proportions of root weight in deeper soil (30-100 cm) to the whole soil (0-100 cm). The same as below.

treatments, whereas changes were minor under the SP and CK treatments. At the jointing and booting stages, RWD in deep layers was significantly higher under the DP and DBP treatments than under both the SP and CK treatments. At the milking stage, RWD in deep layers

also increased more rapidly under the DP and DBP treatments than in earlier growth stages. In contrast, RWD decreased more under SP than under CK in all layers during this stage. At maturity, RWD was lower under all treatments and in all soil layers than in the milking stage, but the decrease was smaller under DP and DBP treatments than under CK and SP.

In addition, the grain yield of CH58, but not XY22, correlated with RW in deep soil layers (30-100 cm) across all growth periods (Table 1 and Fig. 2), indicating that during times of water stress the grain yield of the drought-tolerant cultivar is dependent on the extent of its roots in the deep soil layers.

### Root length and its spatial distribution

The root length (RL) and its spatial distribution were presented in Table 3 and Fig. 3. The total RL varied significantly between cultivars and P treatments both across the whole soil profile (0-100 cm) and in individual layers. RL varied with cultivar, P treatments, and cultivar×P treatments interaction (*P*<0.01) at each of the four growth stages. The RL of CH58 was significantly longer than that of XY22, while the response of RL to the different P treatments was similarly for the two varieties. The roots of both cultivars were the longest (i.e., RL was the highest) under the deep P treatments (DP and DBP) at all four growth stages. The SP treatment resulted in lower RL and roots were the shortest (i.e., RL was the lowest) in control (CK) plots. The difference in RL of both cultivars between DP and DBP was negligible at all growth stages. Similarly, the root length density (RLD) in deep soil layers (30-100 cm) was higher for CH58 than for XY22 throughout the experiment and the difference was most pronounced under the deep P treatments. At the jointing stage, P enrichment significantly increased RL by 41.1-93.7%, relative to CK. In addition, RL was significantly higher under DP and DBP treatments than under SP at this stage. After the jointing stage, there **h** (RL) and its spatial distribution were the prediments occurred at the pilkinhole 3 and Fig. 3. The total RL varied three for RWD it occurred at the miliking the soil profile (0-100 cm) and in individent and the case of

was a significant overall increase in RL in DP and DBP plots. In the deep soil layers (30-100 cm), this led to an increased difference in RL between the deep P and shallow P/control treatments. Under the deep P treatments, the accelerated increase in deep soil RLD continued through to the milking stage. In contrast, there was an overall decrease in RLD under the SP treatment during this period, and at the milking stage it decreased more in the control. The key difference in the responses of RL and RW to the type of P enrichment was their timing. For RLD, a significant difference between shallow and deep P treatments occurred at the jointing stage, whereas for RWD it occurred at the milking stage. This difference in timing indicates that the deep applications of P accelerated the development of deeper roots and did so earlier than shallow application of P.

The changes of grain yield in response to changes in deep soil RL were similar to those observed for RW (Table 1 and Fig. 3). The relationship between grain yield and deep soil RL was cultivar-dependent (*P*<0.05). For CH58, but not XY22, changes in grain yield were consistent with changes in deep soil RL at all growth stages. Thus, during times of water stress, grain yield in the drought-tolerant cultivar was improved by a well-developed root system in deeper soil layers.

### Water-use efficiency

Water-use efficiency (WUE) was affected by cultivar, P treatments, and cultivar×P treatments interaction (*P*<0.01) (Table 1). It was significantly higher for CH58 than XY22 across all treatments. For CH58, WUE was significantly higher under DP and DBP treatments than under SP, but there was no significant difference in this

	<b>Table 3</b> Changes of foot length in the 0-TOO cm soft profile under different treatments (cm cm -)				
Cultivar	Treatments	Jointing	Booting	Milking	Maturity
Xiaoyan 22 (XY22)	CK	46.7 <sub>b</sub>	89.5 c	79.1c	66.8 c
	<b>SP</b>	72.8 a	117.8 <sub>b</sub>	105.5 b	95.1 b
	DP	78.7 a	154.0 a	153.5 a	140.1a
	<b>DBP</b>	78.3 a	157.5 a	154.3 a	144.6 a
Changhan 58 (CH58)	CK	72.0c	101.1c	97.2c	82.8 c
	<b>SP</b>	101.6 <sub>b</sub>	139.5 <sub>b</sub>	135.1 <sub>b</sub>	115.3 <sub>b</sub>
	DP	137.3 a	193.3 a	202.6 a	183.3 a
	<b>DBP</b>	139.5 a	195.0a	208.0a	185.3 a
Source of variation					
Cultivar		**	**	88	**
P		88	$* *$	88	**
Cultivar $\times$ P		$\pm\pm$	$* *$	$8*$	**

**Table 3** Changes of root length in the 0-100 cm soil profile under different treatments (cm cm<sup>-2</sup>)



**Fig. 3** The effects of different treatments on root length density at jointing, booting, milking, and maturity stages for Xiaoyan 22 (XY22) and Changhan 58 (CH58).

respect between the DP and DBP treatments. The WUE values of XY22 were the highest under SP, followed by DP and DBP. However, no differences in WUE between DP, DBP and SP treatments were significant for this cultivar. These results suggested that deep applications of P may increase WUE of drought-tolerant cultivars, but not drought-sensitive cultivars, under dry soil conditions.

Grain yield was strongly correlated with WUE for both cultivars (*r*=0.94, *P*<0.0001), indicating a possible effect of WUE on grain yield (Fig. 4). For CH58, the changes in WUE mirrored changes in both RL and RW in deep soil layers throughout the experiment. However,



**Fig. 4** Relationships between grain yield and WUE for Xiaoyan 22 (XY22) and Changhan 58 (CH58). The regression line was fitted for

there was no such pattern for XY22 (Table 1; Figs. 2 and 3). Thus, a well-developed deep root system seemingly promoted higher WUE in the drought-tolerant cultivar under water stress.

### **DISCUSSION**

The present study reports quantitative changes in root weight and root length of a drought-tolerant and a drought-sensitive cultivar of winter wheat resulting from applications of P fertilizer at different soil placements, under field conditions. The differences in P placement significantly influenced the growth, abundance and vertical distribution of roots. However, responses of the drought-tolerant cultivar (CH58) and drought-sensitive cultivar (XY22) to the placement of P application differed, indicating a difference in root sensitivity between the two cultivars. This is consistent with previously reported variations between cultivars of wheat and cotton in root sensitivity to the depth of fertilizer application under controlled experimental conditions (Wang *et al*. 2009). Although CH58 had consistently higher root weights and lengths than XY22, the cultivars exhibited similar trends in root weight and length throughout the growth season (Tables 2 and 3; Figs. 2 and 3). For both cultivars, the application of P (SP, DP and DBP) resulted in significantly higher RW and RL, relative to CK (*P*<0.05) and the increases were particularly pronounced under the DP and DBP treatments. RW and RL were **Example 19** 10 12<br> **Example 18** 18 10 1825) were two the transition of the state of the st

significantly higher under the DP and DBP than under the SP and CK treatments (*P*<0.05) at both the milking and maturity stages, indicating that deep P application enhances the growth of root systems and delays root senescence. In addition, RWD and RLD differed between the P treatments, reflecting the degree of root branching. RWD and RLD in the deep soil layers (30- 100 cm) were higher under the DP and DBP than under the SP and CK treatments, at each of the four observed growth stages. The findings are consistent with various previous findings. Nearly 30 years ago, Gajri and Prihar (1985) suggested that increases in RWD and RLD may be associated with increases in wheat development and growth. Roots in the subsoil also play an important role in grain yield (Sharma and Chaudhary 1983), and increase rates of uptake of water and nutrients, thereby improving yields (Asseng *et al*. 1998). Furthermore, in a pot experiment presented by Lotfollahi *et al*. (1997), application of N to the subsoil (60 cm) two weeks after anthesis greatly increased grain yield, root growth, and WUE of wheat. They concluded that the increased RLD in the subsoil appeared to be a major factor promoting the efficient use of subsoil N.

In the study presented here, the grain yield and WUE of both cultivars were affected by the placement of P fertilizer. However, deep P placement (DP and DBP) had contrasting effects on grain yield and WUE of the drought-sensitive and drought-tolerant cultivar (Table 1). It increased the grain yield of CH58 by 15.8% (DP) and 16.3% (DBP), compared to surface application (SP). In contrast, the DP and DBP treatments reduced grain yields of XY22 by 12.1 and 11.7%, respectively, compared to SP. These results suggest that drought-tolerant cultivars respond much more vigorously than drought-sensitive cultivars to P applied to the subsoil. In a previous study we found that grain yield of CH58 increased by 11.2% under deep P application (30-60 cm), compared with surface application (0-30 cm). We also found positive correlations between grain yield and both panicle length and 1 000-kernel weight in pot experiments (Kang and Li 2012). In the present study grain yield was strongly correlated with spike number, panicle length and 1 000-kernel weight (Fig. 1). There was also a strong correlation between grain yield and WUE of both cultivars (Fig. 4). However, effects of WUE responses to P application on grain yield of the two cultivars at different soil placements differed, as P applications at all depths were closely correlated with grain yield and WUE for CH58, but not XY22 (Table 1, Figs. 2 and 3). Thus, for the drought-tolerant cultivar, more developed root systems in the subsoil led to higher grain yields. Our results are consistent with those of other studies. For instance, Fang *et al*. (2000) found that the application of P approximately 20 cm below the soil surface significantly increased panicle length and grain yield of drought-tolerant winter wheat compared with a surface P application in the rainfed Weibei Highlands of China. Similarly, Liu *et al*. (2002) reported that deep applications of fertilizer (20-40 cm) in dryland farming increased the spike number and grain yield of drought-sensitive winter wheat by 17.8 and 19.9%, respectively, compared with surface application (0-20 cm), although it had no effect on 1 000-kernel weight.

The main difference between the two cultivars tested in the study was in their sensitivity to water stress. The drought-tolerant cultivar produced root systems with higher proportions in deep soil than the drought-sensitive cultivar (Figs. 2 and 3). Previous studies have shown that a larger root system and an increased proportion of roots in deeper soil are beneficial for water absorption and lead to higher grain yields (Fueki and Takeuchi 2010), and that growth of wheat roots in deeper soils is especially important when soils dry out (Asseng *et al*. 1998). Collectively, these results suggest that high WUE and yield formation mechanisms in winter wheat grown under semi-arid conditions are related to the production of an efficient, extensive root system in deep soils. Weibel Highlands of China. Similarly,<br>
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difference between the two cu

# **CONCLUSION**

Our results confirm that improvements to P fertilization practices can boost yields of winter wheat in semi-arid environments, provided that appropriate cultivars are used. We found that winter wheat growing in dryland soil is highly responsive to application of P in the subsoil. The deep P application (DP and DBP) treatments resulted in higher grain yields and WUE than surface P application for the drought-tolerant cultivar (CH58), but lower yields and WUE for the drought-sensitive cultivar (XY22). These conflicting responses resulted from differences in the vertical distribution of root

weight and root length, and differences in the cultivars' sensitivity to water stress. However, for both cultivars, deep P placement resulted in higher RW and RL in >30 cm deep soil (which is important for water absorption from dry soils) than surface P application. These results indicate that deep application of fertilizer is a feasible, practical means of increasing grain yields and WUE of drought-tolerant cultivars by promoting the growth of deep roots in semi-arid rainfed agricultural regions. Since the deep broadcast treatment (DP) offered no significant growth or yield advantages over the deep-band treatment (DBP), we recommend the incorporation of P fertilizers in the soil at a depth of 20 cm.

# MATERIALS AND METHODS

## Site description

The field experiment was performed from 2010 to 2011 at the Changwu Agri-Ecological Station on the Loess Plateau (35.2°N 107.8°E, 1 200 m a.s.l.) in Shaanxi Province, China. In this semi-arid region, the loess layer is more than 100 m thick. According to the Chinese Soil Taxonomy, the soils are Cumuli-Ustic Isohumosols, and comprise 37% clay, 59% silt, and 4% sand (Gong *et al*. 2007). The soils typically have a pH of 8.4 and bulk density of 1.3 g cm<sup>-3</sup>. Analyses of soil at the experimental field prior to planting in 2010 indicated that the top 20 cm contained 11.5 g kg<sup>-1</sup> organic matter, 10.6 mg kg-1 available phosphorus (Olsen-P), 114.5 mg kg-1 available potassium ( $NH<sub>4</sub>OAc-K$ ), and 0.78 g kg<sup>-1</sup> total nitrogen, of which 3.46 mg kg<sup>-1</sup> was inorganic N. The dominant crop production system in this region is based on conventional tilling and a maize or wheat cropping regime that gives one harvest per year. Rainfed agriculture is practised, i.e., crops are not irrigated. The annual average temperature at Changwu station is 9.2°C and annual average rainfall amounts to 578 mm, of which approximately 40% falls during the winterwheat growing season from October to June. During the field experiment (2010-2011), average rainfall during the growing season amounted to 161.8 mm and the average temperature during the wintering season (October-January) was 9.0°C (Fig. 5). Thus rainfall during the growing season of the study year was 20% lower, and the temperature during the wintering season was 14% higher, than the 5-yr averages. Rain-fed agriculture is the dominant production system.

#### Experimental design and treatments

Two winter wheat (*Triticum aestivum* L.) cultivars were



**Fig. 5** Monthly temperatures and rainfall during the growing period of winter wheat. Tmax, maximum temperature; Tmin, minimum

subjected to four phosphorus treatments, in triplicate, using a randomised complete block design with 24 plots in total, each covering  $12 \text{ m}^2 (3 \text{ m} \times 4 \text{ m})$ . The two cultivars were Xiaoyan 22 (drought-sensitive, XY22) and Changhan 58 (drought-tolerant, CH58). The four P treatments were surface P (SP), deep P (DP), and deep-band P (DBP) application and a control with no P added (CK). In each plot of the P treatments 120 kg  $P_2O_5$  ha<sup>-1</sup> was applied in the form of calcium superphosphate  $(Ca(H_2PO_4)_2)$ . In the SP treatment, the fertilizer was surface broadcast, and then ploughed into the soil. In the DP treatment, the topsoil (0-20 cm) was removed and the fertilizer was broadcast, and then ploughed into the soil (30 cm) before returning the topsoil. In the DBP treatment, P was furrowed in 30 cm below the soil surface, in 40-cm-wide rows (between wheat rows). Each plot also received a base fertilizer consisting of 210 kg N ha<sup>-1</sup> in the form of urea (N  $46\%$ ) and 75 kg K ha<sup>-1</sup> in the form of potassium sulphate  $(K_2O 45%)$ , which were broadcast over the soil. The fields were then ploughed, thus turning the soil and distributing the nutrients to below the surface. On 23 September 2010, winter wheat was planted in 20 cm-wide rows at a seeding rate of 150 kg ha $\,$ <sup>1</sup> (CH58) or 135 kg ha $^{-1}$  (XY22). Fig. 5 Monthly temperatures and minifill during the growing period<br>for winter wheat. Thmx, maximum temperature, Tmin, minimum<br>temperature.<br>
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#### Sampling procedures and measurements

Root samples were collected from soil samples taken from each plot at four stages of growth: jointing, booting, milking, and maturity. An auger with an internal diameter of 10 cm was used to obtain cores from two positions: in-row between plants and between-row 10 cm from the plants. Each core was divided into seven depth intervals: 0-10, 10-20, 20-30, 30-40, 40-60, 60-80, and 80-100 cm. The core sections and associated roots were soaked overnight in plastic buckets filled with tapwater. The next day, the soil in the bucket was gently disaggregated by hand to form a soil-water suspension. The root system was retrieved by pouring the soil-water

suspension onto a 0.25-mm mesh sieve and gently washing the roots with running water. The roots were thoroughly cleaned and straightened by repeated dipping and rinsing in buckets of clean water, then scanned using an EPSON Perfection 4870 scanner. Root images were analyzed to determine root length (RL) using WinRhizo Pro 2004a software (Regent Instruments Inc., Canada). The samples were then dried at 80°C to constant weight and weighed. At both planting time and physiological maturity, the water content of soil in each plot was gravimetrically determined by oven-drying (105°C for 24 h) samples taken at 20-cm intervals down the 200-cm long cores. Plants were harvested from each plot in 22-29 June 2011 for measurements of grain yield, spike number, panicle length per unit area, 1000-kernel weight, and individual seed weight.

# Calculations and statistical analyses

Root weight density  $(RWD, g m<sup>-3</sup>)$  and root length density (RLD, cm cm-3) were computed as follows:

$$
RWD = \frac{RW}{Vt}
$$

$$
RLD = \frac{RL}{Vt}
$$

Where, RW is root dry weight, RL is root length, and Vt is the total volume of the core.

In agronomy, water-use efficiency (WUE) is commonly defined as the ratio of crop yield (usually economic yield) to the amount of water needed to produce the yield, calculated as follows:

$$
WUE = \frac{GY}{ET} = \frac{GY}{P + I - \Delta SWS}
$$

Where, GY is grain yield, ET is actual evapotranspiration, P is precipitation (here: 161.8 mm), I is irrigation (here: 0 mm), and △SWS is the difference in soil water storage between the harvest and seeding stages. Means and standard errors were calculated for three replicates from each treatment.

The data were explored through analysis of variance, as well as comparisons of means using the least significant difference (LSD) test. All statistical analyses were performed using SPSS 13.0 and differences were deemed significant if *P*<0.05.

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