



## More aboveground biomass, phosphorus accumulation and remobilization contributed to high productivity of intercropping wheat

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### Abstract

Intercropping often results in increasing production than sole per unit land area, but the underlying mechanisms are poorly understood. Plants showed different physiological characteristics in intercropping and sole. However, less information was shown the relationships between plant aboveground biomass (AB), phosphorus accumulation (PB) and remobilization and the yield advantage. Here, field experiments were designed as split plot and carried out in 2012 and 2013 with three P levels (0, 40 and 80 kg P ha<sup>-1</sup>) in wheat (*Triticum aestivum* L.) / maize (*Zea mays* L.) relay intercropping and sole. The study measured grain yield, AB and P accumulation and remobilization of wheat. Averaged grain yield of intercropping wheat increased 3.9 Mg ha<sup>-1</sup> in 2012 and 2.7 Mg ha<sup>-1</sup> in 2013 compared with that of the corresponding sole and the grain yield of intercropping wheat changed with the border row (BR) > the inner row (IR) > the sole wheat (SR), the grain yield in BR was contributed by 58.2% to intercropping wheat. The PA was consistent with AB accumulation, which in intercropping was higher than that in sole over the entire growing season. Close correlations between yield and AB remobilization and P remobilization were observed. The yield of BR was higher from 39.3% to 88.0% than that of SR wheat, as mainly attributed to more AB and P accumulation across the whole growing season and more remobilization from pre-anthesis to grain filling stage. More than 40 kg P ha<sup>-1</sup> did not result in any further increasing in yield and did not enhanced the physiological processes associated with AB and P remobilization, indicating that P fertilizer and agronomic management should be intensified synchronously in field to achieve high yield and sustainability.

**Keywords:** Wheat/maize relay strip intercropping; Border-row effect; Aboveground biomass; Phosphorus.

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### Introduction

China produces around 121 million tons of wheat annually (National Bureau of Statistics of China, 2013), to meet the projected demands of the population growth, while the increasing consumption of global food demand is likely to double in 2050 (Thierfelder et al., 2012). Achieving and sustaining optimal yield is a continuous challenge for worldwide, especially in intensive agricultural region as China. It is difficult to improve yield potential in the short term through genetic improvement (Tilman et al., 2011), so improving agronomic management practices would be the expedient solution to meet further demands of wheat.

Intercropping (or relay strip intercropping) as an ancient practice have been widely practiced in many parts of the world (Li et al., 2001; Tollenaar et al., 2002; Rusinamhodzi et al., 2012; Yang et al., 2014). In northwest China, there are about 864,000 ha of cereal/ cereal intercropping which includes 75,000 ha of wheat/maize (W/M) intercropping in Gansu province and about 43% of the total cereal yields in Ningxia municipality were produced by intercropping system (Lu, 1999). In southwest China, about 85% of wheat yield was produced by intercropping systems, especially W/M relay strip intercropping in Sichuan province (Fan et al., 2009).

Intercrops typically grow in close proximity to a different species and thus have intrinsically different architecture, light use efficiency, length of the growth period and root interaction. Such differences results in complementary strategies for resource capture which are regarded as key factors driving the advantage of primary production in species-diverse crops (Yachi and Loreau, 2007), such as intercrops. Many studies have demonstrated that intercrops capture much more resource than sole contribute to productivity advantages. In pearl millet-groundnut intercropping system, higher yield of intercrops was associated with a commensurately greater uptake of radiation and nutrients (Gregory and Reddy, 1982; Marshall and Willy, 1983). Zhang et al. (2008) and Zhu et al. (2016) also found that increased radiation interception by intercropped wheat (especially in the border-row) in wheat-cotton or maize intercropping systems fully explained the high productivity and land-use efficiency of the system. However, most studies pay close attention to the results caused by plants growth environmental changed instead analyses how plant physiological processes response to the environmental changed and contribute to higher yield.

Wheat grain yield has been shown to be positively correlated with aboveground biomass (AB) and nutrient accumulation in crops and studies were certified that AB and nutrient accumulation for wheat primarily occurred at pre-anthesis and grain yield greatly depended on the remobilization of pre-anthesis assimilates and nutrient accumulation (Cox et al., 1985; Papakosa and Gagianas, 1991). However, a significant correlation has also been found between grain yield and AB accumulation of post-anthesis in high wheat yield production systems in China (Ye et al., 2011). Wheat yield potentials is determined by AB and nutrient accumulation, remobilization which generally affected by agronomic management strategies and growth environment conditions, such as weather conditions (Koutroubas et al., 2012), sowing date (Ferrise et al., 2010), soil types (Masoni et al., 2007), soil water availability (Ercoli et al., 2008), radiation interception (Wang et al., 2015b) and nutrient management (Meng et al., 2013). The overyielding of intercropped wheat through changing the growing conditions associated with water, nutrient and solar energy have been proved (Xia et al., 2013; Wang et al., 2015a; Wang et al., 2016b). As AB accumulation is a result of the combination of environmental factors and is a process for yield formation, hence, we focus on wheat yield associated with AB accumulation and remobilization in intercropping system here.

Phosphorus also influences crop growth and production (Arduini et al., 2006). For example, overuse of P fertilizer results in higher plant P content due to so-called “luxury accumulation” by crops, while deficiency of P in the soil decreases plant P content and sacrifices some grain yield. However, due to limited knowledge of P accumulation and remobilization contribute to plant production and generally P fertilizer provides in present management practices does not meet crop demand. Phosphorus uptake occurs throughout the life of the plant until physiological maturity, but farmers regularly apply all of P fertilizer for wheat before sowing, which results in a amount of P accumulation in soil because P can be dominantly adsorbed to soil mineral surfaces and occur as

sparingly available precipitates (Arai and Sparks, 2007). Intercropping through exploitation of the biological potential for efficient acquisition of P by interspecific interactions, which occur as probable to meet crop P demand (Xia et al., 2013). P management strategies for reduce P loss is vital in the Southwest part of China, as this area is located at the upstream of the Yangtze River and the arable field on the hilly landscape is easily eroded by rainfall (Zhang et al., 2004).

Wheat/maize relay strip intercropping system occupies a dominant position for wheat yield produce in southwest China, where is low light intensity and soil P availability. In Wheat/Maize relay intercropping system, strips of winter wheat are sown first, with a strip of bare soil between the wheat strips. Maize is intersown in these bare strips when the wheat plants are approximately at the flowering stage. After wheat harvest, maize continues to grow as a sole crop until its harvest. Every wheat strip contains four rows and maize strip contains two rows. Wheat experiences a favorable light and nutrient environment at early stage of development because of the absence of maize. Thus, more exertions should be made to understand AB, P accumulation and remobilization in intercropping as an efficiently agronomy management with special crop growing condition to improve management efficiency and at the same time gain higher grain yield.

To assess the contribution of physiological processes responses of wheat to yield advantage in the relay strip intercropping with maize, we analyzed the wheat AB, P accumulation, remobilization and yield in three situations: (a) wheat grown as a sole crop, (b) wheat plants in border rows of the wheat strips in a wheat-maize intercropping system with alternating sets of four rows of wheat and two rows of maize and (c) wheat plants from the inner rows of the wheat strips in the intercropping system.

## Materials and Methods

### *Site description*

The experiments were carried out in 2011-2012 and 2012-2013 at the department of Sichuan Agricultural University Ya'an Experimental Station (29°58' N, 102°58' E) in Sichuan Province, China with an altitude of 600 m above sea level. Annual mean temperature of 15.4 °C with a maximum and minimum temperature of 25.4 °C and 6.1 °C, respectively. Annual precipitation of 1500 mm and potential evaporation of 838 mm. Annual sunshine of about 1019 hours and total solar radiation averages around 3,750 MJ m<sup>-2</sup> yr<sup>-1</sup>. The monthly average temperature and rainfall values during the current experimental period showed that generally the temperature and rainfall were very low in January and very high in August (Figure 1). The experimental soil was classified as Purple soil (Luvic Xerosols) and Olsen-P content is low.

### *Experiment design and crop management*

The field experiments were designed as a split plot with four replicates over two years in the same location. The main plot treatments comprised a zero P and two application rates of P and the sub-plot treatments consisted of wheat/maize relay strip intercropping system and sole wheat. The area of each individual plot was 4×9 m<sup>2</sup> for intercrops and sole (S<sub>1</sub> Figure). Each intercropping plot consisted of four strips with 1.0 m in width. Each strip contained four rows of wheat and two rows of maize (Figure 2), while each sole plot contained 16 rows of wheat. Wheat was sown in rows at spacing of 25 cm between rows in intercropping and sole system. Two maize seedlings were transplanted per hole at a spacing of 40 cm between holes and 50 cm between rows. The space between wheat and maize was 25 cm (Figure 2). Density of

intercropped wheat was about 240 plants m<sup>-2</sup>, maize was 5 plants m<sup>-2</sup> and sole wheat was about 480 plants m<sup>-2</sup>. Wheat was sowed on 11<sup>th</sup> November in 2011, 10<sup>th</sup> November in 2012 and harvested on 30<sup>th</sup> May in 2012, 11<sup>th</sup> May in 2013, respectively. Maize was transplanted on 15<sup>th</sup> April in 2012, 7<sup>th</sup> April in 2013 and harvested on 6<sup>th</sup> August in 2012, 6<sup>th</sup> August in 2013, respectively (S<sub>2</sub> Figure). The coexisting periods of wheat with maize were approximately 45 days (Figure 1).

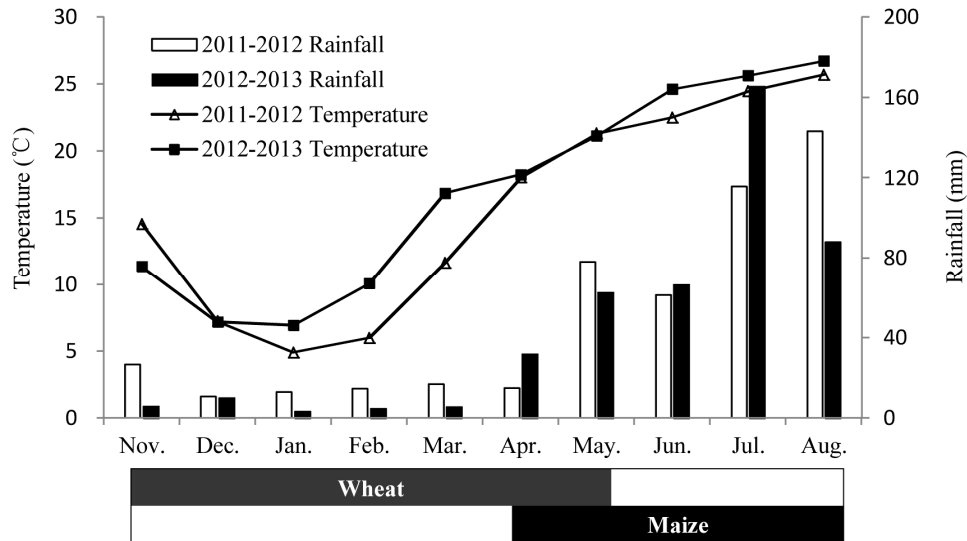


Figure 1. Monthly average rainfall (mm) and temperature (°C) at the experimental spot in 2012 and 2013 and planting/harvest times of the two crops during growth period. (1) Shading indicates that the crops are growing in the field; (2) Wheat intercropped with maize has a co-growth period of almost 45 days.

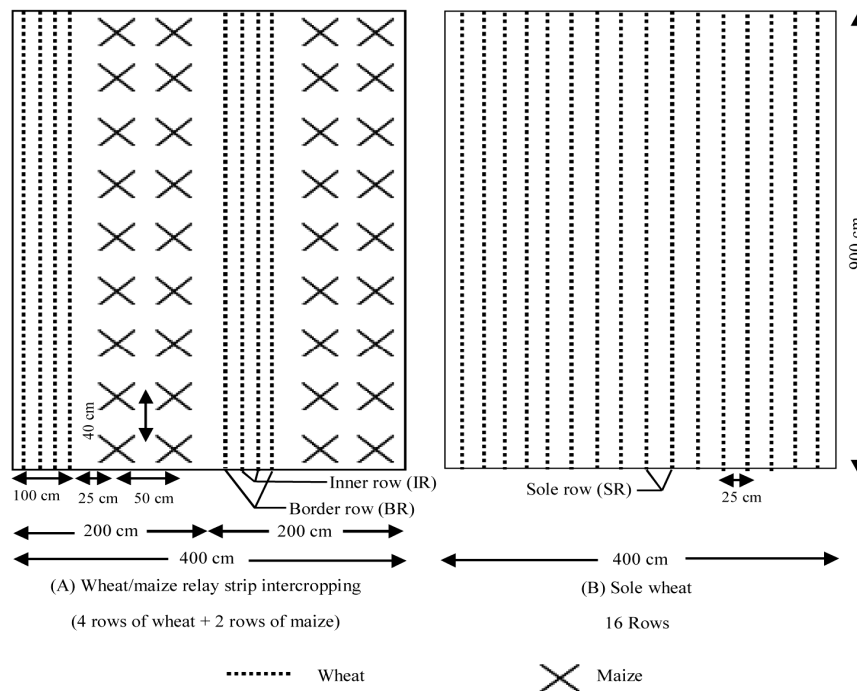
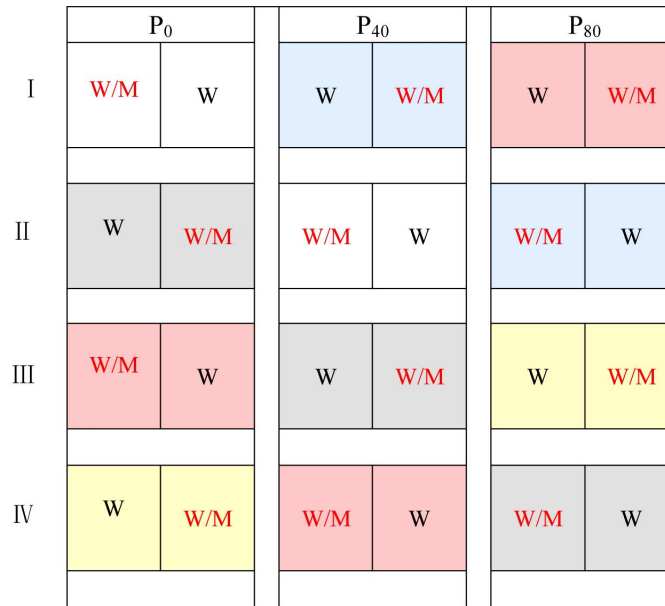


Figure 2. Diagram showing the arrangement of wheat intercropped with maize (A) and sole wheat (B) in the field plot.



S<sub>1</sub> Figure. Diagram of the integrated field experiment with three P levels and four replications. W/M, wheat/maize relay strip intercropping; W, sloe wheat.



S<sub>2</sub> Figure. Wheat/maize relay strip intercropping system. The pattern of the W/M/S system is shown as pictures (A, B, C). Wheat is first sown in November but maize strip is blank (A). Maize is transplanted in April of the following year when wheat is at flowering stage (B). After wheat was harvested in May in the following year, maize was growth as a sole crop (C).

The P rates for wheat were 0, 40 and 80 kg P ha<sup>-1</sup> marked as P<sub>0</sub>, P<sub>40</sub> and P<sub>80</sub>, respectively, the rates for maize were 0, 32 and 64 kg P ha<sup>-1</sup>, both of two crops were applied as triple superphosphate. The total N application rates for wheat and maize were 120 kg N ha<sup>-1</sup> and 225 kg N ha<sup>-1</sup> as urea, respectively. The K application rates for wheat and maize were 75 and 87 kg K ha<sup>-1</sup>, respectively as potassium chloride. All of the P and K fertilizers were applied as basal fertilizer by hand before wheat or maize sowing, while N fertilizer was split to a basal fertilization and two topdressings which were applied by hand at the tillering stage and the stem elongation stage of wheat and applied at the stem elongation and the tasseling stage of maize. The proportions of basal fertilizer and two topdressings were 40-30-30 in the percentage of the total N fertilizer applied at wheat and 30-30-40 applied at maize. No organic manure was applied. During the growth period, all the plots were well irrigated and weeded manually or by chemical control.

*Sample collection and measurement*

Before the start of the study, 0~20 cm depth of soil was sampled randomly in the field by a soil auger. The characteristics (Blake and Hartge, 1986) of soil sample were

pH (water) 6.2, organic matter content 32.1 g kg<sup>-1</sup>, total N 2.10 g kg<sup>-1</sup>, available N 112 mg kg<sup>-1</sup>, Olsen-P 13.2 mg kg<sup>-1</sup>, exchangeable K 71 mg kg<sup>-1</sup> and Cation Exchange Capacity 21.5 cmol kg<sup>-1</sup>, the soil N and K content is medium but the P is low. Measurements of wheat growth were done at anthesis (on 12<sup>th</sup> Apr. 2012 and 2013, respectively) and maturity. Intercropping wheat with 4 rows, the first and fourth named as border row (BR), the second and third row named as inner row (IR) (Figure 2). The sampling areas for each occasion were 0.4×1.0 m<sup>2</sup> for BR, IR and sole row (SR) wheat.

All plants were cut at ground level and subdivided into stover (content leaves, stems, culms, chaff) and grain. All samples were oven dried at 70 °C to constant weight for shoot biomass determination at anthesis and maturity. Plant samples were wet-digested with concentrated H<sub>2</sub>SO<sub>4</sub> and H<sub>2</sub>O<sub>2</sub> (30%) for P determination by the vanadomolybdate method (Page, 1982) and P uptake were calculated by multiplying the P concentration with dry weight. Harvest index (HI) (the ratio of yield to aboveground biomass), phosphorus harvest index (PHI) (the ratio of seed P content to shoot P content) and ratio of HI/PHI were measured.

As wheat/maize intercropping comprises two crops, wheat only occupied 50% area of the whole system. In this study, the weighted means of sole wheat grain yield were calculated as follow:

$$\text{Weighted mean of sole wheat grain yield} = Y_{\text{sole}} \times P_{\text{wheat}}$$

where  $Y_{\text{sole}}$  is the grain yield of sole wheat,  $P_{\text{wheat}}$  is the proportions of the area occupied by wheat in the intercropping.  $P_{\text{wheat}}$  is determined as  $P_{\text{wheat}} = \frac{1}{2} = W_{\text{wheat}} / (W_{\text{wheat}} + W_{\text{maize}})$  where  $W_{\text{wheat}}$  and  $W_{\text{maize}}$  are width of crops in the strip. This formula was also used to calculate the weighted means of shoot P uptake and by sole.

The following parameters, related to AB and P accumulation and remobilization within the wheat plant, were calculated with the method of Masoni et al., 2007.

- (1) AB remobilization (ABR) = AB of the whole plant at anthesis – stover at maturity.
- (2) Phosphorus remobilization (PR) = P uptake of the whole plant at anthesis – P uptake of stover at maturity.
- (3) AB remobilization efficiency (ABRE) = (ABR/ AB of the whole plant at anthesis) × 100%.
- (4) Phosphorus remobilization efficiency (PRE) = (PR/P uptake of the whole plant at anthesis) × 100%.
- (5) Contribution of AB remobilized assimilates to grain (ABRC) = (ABR/ AB of grains at maturity) × 100%.
- (6) Contribution of phosphorus remobilized assimilates to grain (PRC) = (PR/ P uptake of grains at maturity) × 100%.

For the estimation of ABR it was assumed that all the AB lost from vegetative plant parts was remobilized to developing grain, since losses of AB due to plant respiration during grain filling was not determined.

### Statistical analysis

Data from the split-plot design experiments were subjected to analysis of variance (ANOVA) using SAS software (SAS Institute, USA) and mean values (n=4) were compared by least significance difference (LSD) at the 5% level. The relationships of

grain yield and aboveground biomass remobilization, grain P uptake and P remobilization were analyzed also using SAS software.

## Results

### *Yield and AB accumulation*

Intercropping had a considerable impact on wheat yield, averaged values in intercropping over three P application rates ranged from 2.9 to 4.7 Mg ha<sup>-1</sup> across the two years and were significantly higher than in sole (Table 1). There was significant difference ( $P < 0.000$ ) in grain yield between P application rates and no significant difference between the two years ( $P > 0.215$ ). Irrespective of intercropping and sole, the average yield of wheat increased significantly from 2.7 to 3.8 Mg ha<sup>-1</sup> and from 2.6 to 3.9 Mg ha<sup>-1</sup> from 0 to the 40 kg ha<sup>-1</sup> P application rate in 2012 and 2013 respectively, and 80 kg ha<sup>-1</sup> P application rate did not result in any further increase in yield compared with 40 kg ha<sup>-1</sup> P application rate (Table 1).

The yields of wheat in BR significantly increased compared with in both IR and SR ( $P < 0.000$ ). Wheat yields of BR were from 39.3% to 88.0% higher than that of SR wheat. The wheat yields in IR had no advantage compared with wheat in the SR, except the treatment of no P application rate in 2013 (Table 1).

Table 1. Grain yield affected by cropping systems and P application rates.

Year	P rate (P kg ha <sup>-1</sup> )	Grain yield (Mg ha <sup>-1</sup> )		Grain yield in rows (g m <sup>-1</sup> row <sup>-1</sup> )		
		Inter.	Sole	BR	IR	SR
2012	0	3.2 <sup>a</sup>	2.2 <sup>b</sup>	14.5 <sup>a</sup>	10.4 <sup>b</sup>	8.9 <sup>b</sup>
	40	4.7 <sup>a</sup>	2.9 <sup>b</sup>	22.0 <sup>a</sup>	15.2 <sup>b</sup>	11.7 <sup>b</sup>
	80	3.6 <sup>a</sup>	2.5 <sup>b</sup>	16.3 <sup>a</sup>	12.2 <sup>b</sup>	10.1 <sup>b</sup>
2013	0	2.9 <sup>a</sup>	2.2 <sup>b</sup>	13.5 <sup>a</sup>	10.6 <sup>b</sup>	8.8 <sup>c</sup>
	40	4.6 <sup>a</sup>	3.2 <sup>b</sup>	22.0 <sup>a</sup>	15.2 <sup>b</sup>	12.7 <sup>b</sup>
	80	3.6 <sup>a</sup>	3.0 <sup>a</sup>	17.0 <sup>a</sup>	11.4 <sup>b</sup>	12.2 <sup>b</sup>
ANOVA	Year	0.215		0.533		
	P rate	0.000		0.000		
	Cropping system	0.000		0.000		
	P rate × Cropping system	0.001		0.002		

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different between Inter. (intercropping) and sole, or among BR, IR SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities ( $P$  values) of the source of variation. BR, wheat in border row; IR, wheat in inner row; SR wheat in sole row.

The average AB accumulation of wheat over all P application rate were not influenced by the intercropping at anthesis and maturity in 2012, but in 2013, the AB accumulation of intercropping wheat increased only with 80 kg ha<sup>-1</sup> P application rate at anthesis and maturity compared with the sole (Table 2). Regardless of cropping system, P fertilizer have an significantly effect on AB accumulation compared in 2012 and 2013 at anthesis and maturity ( $P < 0.000$ ), but much more P not lead further increased of AB accumulation. The stover of wheat in BR significantly increased compared to in IR and SR at anthesis and which at maturity in 2013 showed the same tendency as in anthesis.

The harvest index (HI) of wheat differed greatly between intercropping and sole and were also affected by P application rates ( $P < 0.000$ ) (Table 3). The average HI of BR wheat over 2012 and 2013 was 55.2%, the contrast HI of sole wheat was 43.7% at 40 kg ha<sup>-1</sup> P application rate.

Table 2. Aboveground biomass of wheat at anthesis and maturity under different cropping systems and P application rates.

Year	P rate (P kg ha <sup>-1</sup> )	Aboveground biomass accumulation (Mg ha <sup>-1</sup> )						stover biomass accumulation (g m <sup>-1</sup> row <sup>-1</sup> )					
		Anthesis			Maturity			Anthesis			Maturity		
		Inter.	Sole		Inter.	Sole		BR	IR	SR	BR	IR	SR
2012	0	3.9 <sup>a</sup>	3.8 <sup>a</sup>	3.4 <sup>a</sup>	3.1 <sup>a</sup>	3.4 <sup>a</sup>	16.9 <sup>a</sup>	14.7 <sup>b</sup>	15.3 <sup>b</sup>	12.8 <sup>b</sup>	12.0 <sup>c</sup>	13.7 <sup>a</sup>	
	40	5.7 <sup>a</sup>	5.6 <sup>a</sup>	4.2 <sup>a</sup>	4.1 <sup>a</sup>	4.2 <sup>a</sup>	24.7 <sup>a</sup>	20.6 <sup>c</sup>	22.3 <sup>b</sup>	17.0 <sup>a</sup>	15.7 <sup>b</sup>	16.8 <sup>a</sup>	
	80	5.4 <sup>a</sup>	5.9 <sup>a</sup>	4.5 <sup>a</sup>	4.0 <sup>a</sup>	4.5 <sup>a</sup>	23.2 <sup>a</sup>	20.5 <sup>b</sup>	23.8 <sup>a</sup>	16.5 <sup>b</sup>	15.5 <sup>c</sup>	18.1 <sup>a</sup>	
2013	0	4.6 <sup>a</sup>	4.4 <sup>a</sup>	3.1 <sup>a</sup>	3.2 <sup>a</sup>	3.1 <sup>a</sup>	20.0 <sup>a</sup>	16.7 <sup>b</sup>	17.7 <sup>b</sup>	13.9 <sup>a</sup>	11.5 <sup>b</sup>	12.4 <sup>b</sup>	
	40	6.0 <sup>a</sup>	4.6 <sup>b</sup>	3.7 <sup>a</sup>	4.2 <sup>a</sup>	3.7 <sup>a</sup>	26.4 <sup>a</sup>	21.6 <sup>b</sup>	18.4 <sup>c</sup>	18.6 <sup>a</sup>	15.1 <sup>b</sup>	14.7 <sup>b</sup>	
	80	5.4 <sup>a</sup>	4.5 <sup>b</sup>	3.6 <sup>b</sup>	4.0 <sup>a</sup>	3.6 <sup>b</sup>	23.9 <sup>a</sup>	19.5 <sup>b</sup>	18.1 <sup>b</sup>	17.0 <sup>a</sup>	15.0 <sup>b</sup>	14.2 <sup>c</sup>	
ANOVA	Year	0.052						0.870					
	P rate	0.000						0.000					
	Cropping system	0.000						0.000					
	P rate × Cropping system	0.002						0.003					

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different between inter. (intercropping) and sole, or among BR, IR, SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities (*P* values) of the source of variation. BR, wheat in border row; IR, wheat in inner row; SR wheat in sole row.



Table 3. Harvest index of dry matter and Phosphorus.

Year	P rate (P kg ha <sup>-1</sup> )	Harvest index (%)					
		Yield			Phosphorus		
		BR	IR	SR	BR	IR	SR
2012	0	53.0 <sup>a</sup>	46.3 <sup>b</sup>	39.5 <sup>c</sup>	79.9 <sup>a</sup>	75.5 <sup>a</sup>	65.8 <sup>b</sup>
	40	56.2 <sup>a</sup>	49.1 <sup>b</sup>	41.1 <sup>c</sup>	76.3 <sup>a</sup>	72.9 <sup>a</sup>	64.6 <sup>b</sup>
	80	49.7 <sup>a</sup>	43.9 <sup>a</sup>	35.8 <sup>b</sup>	73.7 <sup>a</sup>	72.2 <sup>bc</sup>	60.3 <sup>c</sup>
2013	0	49.4 <sup>a</sup>	47.7 <sup>a</sup>	41.5 <sup>b</sup>	72.0 <sup>a</sup>	68.5 <sup>a</sup>	65.5 <sup>b</sup>
	40	54.2 <sup>a</sup>	50.0 <sup>a</sup>	46.3 <sup>b</sup>	74.9 <sup>a</sup>	68.1 <sup>bc</sup>	66.0 <sup>c</sup>
	80	50.0 <sup>a</sup>	42.9 <sup>a</sup>	46.1 <sup>a</sup>	70.7 <sup>a</sup>	65.2 <sup>c</sup>	64.6 <sup>c</sup>
ANOVA	Year	0.157			0.000		
	P rate	0.000			0.000		
	Cropping system	0.000			0.000		
	P rate × Cropping system	0.642			0.998		

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different among BR, IR, SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities (*P* values) of the source of variation. BR, wheat in border row, IR, wheat in inner row; SR wheat in sole row.

#### Total P accumulation

Total P accumulation followed generally the variation in AB accumulation. Wheat P accumulation was affected by cropping system and P fertilization treatments at anthesis, while only P application rates had impact on wheat P accumulation at maturity in 2012 and 2013 ( $P < 0.000$ ). P accumulation of intercropping wheat averaged over all three P application rates and two years was increased by 12.7% compared with sole wheat at anthesis (Table 4). The averaged P accumulation of intercropping and sole wheat increased by 42.5% and 30.8% at 40 kg ha<sup>-1</sup> P, 42.9% and 45.1% at 80 kg ha<sup>-1</sup> P application rate compared with no P application, respectively (Table 4).

The P accumulation in BR significantly increased compared to IR and SR wheat at anthesis ( $P < 0.000$ ), however, there were no difference among BR, IR and SR wheat at maturity ( $P < 0.461$ ). The P harvest index (PHI) (Table 3) differed greatly between intercropping and sole wheat. Averaging the PHI over three P application rates, intercropping wheat acquired significantly higher PHI than sole and the average PHI of BR, IR and SR wheat were 74.6%, 70.4% and 64.5%, respectively.

Table 4. Phosphorus (P) accumulations of wheat at anthesis and maturity under different cropping system and P application rates.

Year	P rate (P kg ha <sup>-1</sup> )	Aboveground P accumulation (kg ha <sup>-1</sup> )						Stover P accumulation (mg m <sup>-1</sup> row <sup>-1</sup> )					
		Anthesis			Maturity			Anthesis			Maturity		
		Inter.	Sole		Inter.	Sole		BR	IR	SR	BR	IR	SR
2012	0	10.0 <sup>a</sup>	9.0 <sup>a</sup>	3.8 <sup>a</sup>	2.9 <sup>b</sup>		44.9 <sup>a</sup>	35.0 <sup>b</sup>	36.0 <sup>b</sup>	12.8 <sup>b</sup>	10.6 <sup>c</sup>	15.0 <sup>a</sup>	
	40	14.7 <sup>a</sup>	14.3 <sup>a</sup>	6.1 <sup>a</sup>	5.6 <sup>a</sup>		66.0 <sup>a</sup>	51.4 <sup>b</sup>	57.4 <sup>b</sup>	25.1 <sup>a</sup>	19.6 <sup>b</sup>	24.6 <sup>a</sup>	
	80	13.6 <sup>a</sup>	14.3 <sup>a</sup>	6.0 <sup>a</sup>	4.8 <sup>b</sup>		62.0 <sup>a</sup>	46.8 <sup>b</sup>	57.1 <sup>a</sup>	21.5 <sup>ab</sup>	17.3 <sup>b</sup>	23.9 <sup>a</sup>	
2013	0	11.4 <sup>a</sup>	9.4 <sup>b</sup>	3.7 <sup>a</sup>	3.9 <sup>a</sup>		50.9 <sup>a</sup>	40.2 <sup>b</sup>	37.6 <sup>b</sup>	16.8 <sup>a</sup>	14.5 <sup>a</sup>	14.6 <sup>a</sup>	
	40	15.8 <sup>a</sup>	12.0 <sup>b</sup>	5.0 <sup>a</sup>	6.2 <sup>a</sup>		72.0 <sup>a</sup>	54.8 <sup>b</sup>	47.9 <sup>c</sup>	27.7 <sup>a</sup>	22.3 <sup>b</sup>	20.1 <sup>b</sup>	
	80	14.4 <sup>a</sup>	12.4 <sup>b</sup>	5.1 <sup>a</sup>	5.4 <sup>a</sup>		59.5 <sup>a</sup>	54.9 <sup>a</sup>	49.7 <sup>b</sup>	22.4 <sup>a</sup>	20.6 <sup>a</sup>	20.3 <sup>a</sup>	
ANOVA	Year	0.765			0.981			0.152			0.112		
	P rate	0.000			0.000			0.000			0.000		
	Cropping system	0.000			0.461			0.000			0.000		
	P rate × Cropping system	0.042			0.228			0.120			0.211		

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different between inter. (intercropping) and sole, or among BR, IR SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities (*P* values) of the source of variation. BR, wheat in border row; IR, wheat in inner row; SR, wheat in sole row.

### *AB and P remobilization*

In the both years of the study, ABR was affected by P and intercropping. P applied increased wheat ABR and ABRE ( $P < 0.000$ ) and resulted in higher contribution of ABRC (Table 5). Intercropping wheat showed the higher ABR and ABRE compared with sole wheat among P levels and years, especially in 2013 (Table 5). BR wheat obtained the highest ABRE than IR and SR wheat. In contrast, sole wheat showed higher ABRC than intercropping wheat (Table 5).

Phosphorus remobilization and remobilization efficiency ranged from 5.3 to 9.6 kg P ha<sup>-1</sup> and from 58.2% to 70.8%, respectively (Table 6). Intercropping enhanced wheat PR, but it had no impact effect on PRE and PRC to grain. In contrast, sole wheat showed higher PRC than intercropping. The highest PRC was in SR and then in IR, the lowest was in BR. PR was increased significantly with increasing P application, while the PRE and PRC decreased in intercropping and sole wheat in two years of this study (Table 6). The highest PRE and PRC showed at no P application in both years when compared to P application treatments, except for the PRC of sole wheat in 2012.

### *Relationship between ABR and grain yield, PR and grain P accumulation*

Aboveground biomass and PR from the vegetative parts to the grain during grain-filling period were significantly correlated with the grain yield and grain P uptake at maturity (Figure 3). The liner model described the relationship between ABR and yield well in BR ( $R^2=0.83$ ,  $P < 0.01$ ), IR ( $R^2=0.80$ ,  $P < 0.01$ ) and SR wheat ( $R^2=0.50$ ,  $P < 0.01$ ) (Figure 3). The  $b$  value representing the slope parameter for BR wheat was 1.06 and was 32.5% and 112.0% higher than that for IR and SR wheat, respectively. The liner model also described the relationship between PR and grain P accumulation well. The  $b$  value for BR wheat was 1.11 and was 19.3% and 40.5% higher than that of IR and SR wheat. As we know, AB and PR from the vegetative parts to grain during grain-filling period were independent, the ratio of HI to PHI could describe the proportion of grain carbon compared to the proportion of grain P. In Figure 4, the ratio of HI to PHI in intercropping wheat was higher than in sole, the highest was in BR wheat. Irrespective of cropping system, ratio of HI to PHI was increased with 40 kg ha<sup>-1</sup> P application compared to in no P. However, with 80 kg ha<sup>-1</sup> P application did not led to any further increase compared with 40 kg ha<sup>-1</sup> P.

Table 5. Aboveground biomass remobilization efficiency and contribution of pre-anthesis to seed as affected by cropping systems and different P application rates.

Year	P rate (P kg ha <sup>-1</sup> )	ABR (Mg ha <sup>-1</sup> )		ABRE (%)			ABRC (%)		
		Inter.	Sole	BR	IR	SR	BR	IR	SR
2012	0	0.8 <sup>a</sup>	0.4 <sup>b</sup>	24.0 <sup>a</sup>	18.3 <sup>a</sup>	10.5 <sup>b</sup>	31.4 <sup>b</sup>	32.3 <sup>b</sup>	39.6 <sup>a</sup>
	40	1.6 <sup>a</sup>	1.4 <sup>a</sup>	31.4 <sup>a</sup>	24.1 <sup>b</sup>	24.3 <sup>b</sup>	32.8 <sup>b</sup>	36.1 <sup>a</sup>	36.2 <sup>a</sup>
	80	1.5 <sup>a</sup>	1.4 <sup>a</sup>	28.9 <sup>a</sup>	24.1 <sup>a</sup>	23.6 <sup>a</sup>	41.2 <sup>b</sup>	41.3 <sup>b</sup>	45.5 <sup>a</sup>
2013	0	1.4 <sup>a</sup>	1.3 <sup>a</sup>	30.3 <sup>a</sup>	30.2 <sup>a</sup>	30.0 <sup>a</sup>	38.5 <sup>b</sup>	41.8 <sup>a</sup>	43.6 <sup>a</sup>
	40	1.8 <sup>a</sup>	0.9 <sup>b</sup>	29.6 <sup>a</sup>	30.0 <sup>a</sup>	19.8 <sup>b</sup>	37.8 <sup>b</sup>	40.7 <sup>a</sup>	41.7 <sup>a</sup>
	80	1.4 <sup>a</sup>	1.0 <sup>b</sup>	28.9 <sup>a</sup>	22.1 <sup>a</sup>	21.1 <sup>a</sup>	40.4 <sup>b</sup>	41.6 <sup>b</sup>	44.4 <sup>a</sup>
ANOVA									
	Year		0.127			0.026			0.582
	P rate		0.000			0.000			0.041
	Cropping system		0.000			0.000			0.021
	P rate × Cropping system		0.230			0.886			0.650

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different between inter. (intercropping) and sole, or among BR, IR, SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities (*P* values) of the source of variation. BR, wheat in border row; IR, wheat in inner row; SR, wheat in sole row.

Table 6. Phosphorus remobilization efficiency and contribution of pre-anthesis to seed as affected by cropping systems and different P application rates.

Year	P rate (P kg ha <sup>-1</sup> )	PR (kg ha <sup>-1</sup> )		PRE (%)			PRC (%)		
		Inter.	Sole	BR	IR	SR	BR	IR	SR
2012	0	7.0 <sup>a</sup>	5.3 <sup>b</sup>	70.8 <sup>a</sup>	69.5 <sup>a</sup>	58.2 <sup>b</sup>	62.6 <sup>b</sup>	75.9 <sup>a</sup>	73.7 <sup>a</sup>
	40	9.1 <sup>a</sup>	8.2 <sup>b</sup>	62.1 <sup>a</sup>	61.3 <sup>a</sup>	57.1 <sup>a</sup>	50.9 <sup>b</sup>	59.9 <sup>b</sup>	74.4 <sup>a</sup>
	80	8.8 <sup>a</sup>	8.3 <sup>b</sup>	65.6 <sup>a</sup>	63.1 <sup>a</sup>	58.2 <sup>a</sup>	67.8 <sup>b</sup>	67.6 <sup>b</sup>	91.8 <sup>a</sup>
2013	0	7.5 <sup>a</sup>	5.8 <sup>b</sup>	66.9 <sup>a</sup>	63.4 <sup>a</sup>	61.2 <sup>a</sup>	81.5 <sup>a</sup>	80.9 <sup>a</sup>	83.5 <sup>a</sup>
	40	9.6 <sup>a</sup>	7.0 <sup>b</sup>	61.5 <sup>a</sup>	59.3 <sup>a</sup>	58.2 <sup>a</sup>	53.8 <sup>b</sup>	68.3 <sup>a</sup>	73.5 <sup>a</sup>
	80	9.1 <sup>a</sup>	7.4 <sup>b</sup>	62.4 <sup>a</sup>	62.3 <sup>a</sup>	59.2 <sup>a</sup>	70.3 <sup>b</sup>	79.6 <sup>a</sup>	89.7 <sup>a</sup>
ANOVA		Year		0.745			0.141		
		P rate		0.000			0.000		
		Cropping system		0.000			0.000		
		P rate × Cropping system		0.418			0.879		

Values are means of four replicates. Values followed by the same lowercase letters are not significantly different between inter. (intercropping) and sole, or among BR, IR, SR within the same P rate in one year at the 5% level by LSD. Values under ANOVA are the probabilities (*P* values) of the source of variation. BR, wheat in border row; IR, wheat in inner row; SR, wheat in sole row.

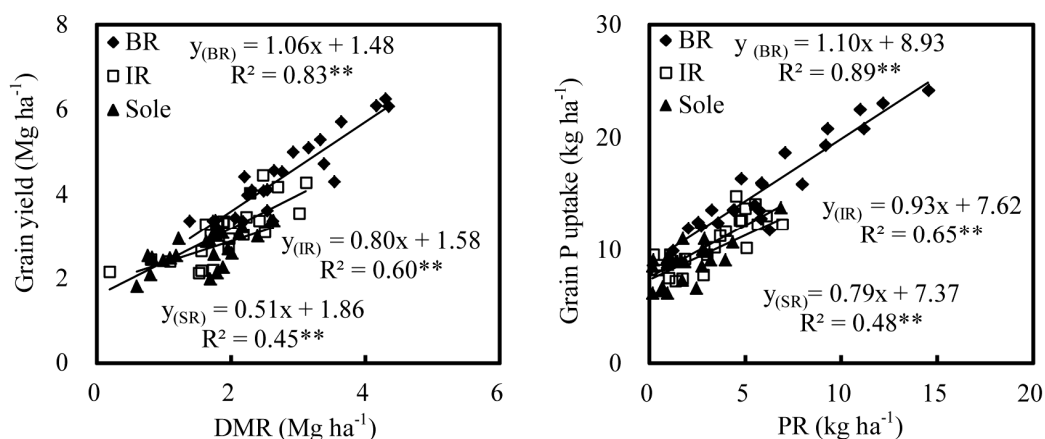


Figure 3. Relationships of grain yield (y axis) and DMR (x axis) (A), grain P accumulation (y axis) and PR (x axis) (B). BR, wheat in border row; IR, wheat in inner row; SR, wheat in sole row. Each data point was the mean of four replicates.

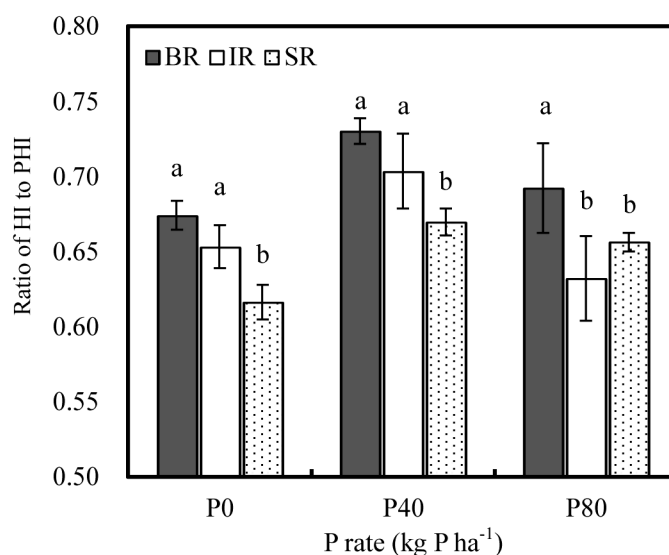


Figure 4. The ratio of HI to PHI. Different lowercase letters within the same P application rate indicate significant differences at  $P < 0.05$  by LSD among different rows of wheat; different capital letters indicate significant differences at  $P < 0.05$  by LSD among different P application rate, averaged over cropping system. BR, wheat in border row; IR, wheat in inner row; SR, wheat in sole row. Each data column was the average of 2012 and 2013 year over four replicates. Data bars show SE (n=4).

## Discussion

In present study, yield of intercropping wheat had an advantage over corresponding sole (Table 1). We found that the yield of intercropping wheat increased dramatically by 41.2% compared with the weighted means of the corresponding sole averaged over the three P application rates and two years (Table 1). It was in agreement with previous studies in wheat/maize which showed that the yield of wheat had increased to 24-61% by intercropping regardless N, P application and plant density on a reclaimed desert soil in the arid zone of Gansu Province in northwest China (Li et al., 2001; Li et al., 2011). Wheat yield in border row (BR) were 39.3- 88.0% higher than in sole row (SR) and the

BR contributed 58.2% to intercropping wheat yield averaged over the three P application rates and two years (Table 1). The view that obtain a high yield in BR is important to achieve yield advantage in intercropping compare to sole, which was also consistent with some studies before (Zhang et al., 2007; Zhang et al., 2015a).

These results showed that the increase in wheat yield greatly depended on an increasing in AB and PB over the entire growing season. Our findings disagreed with some previous results, which showed a positive correlation between grain yield and pre-anthesis assimilate contribution to grain filling and greater AB translocation efficiency (Álvarez et al., 2008). However, some researches on modern wheat in China showed that, greater yield potential must come from increases in net primary productivity over the entire growing season (Ye et al., 2011; Meng et al., 2013). Intercropping wheat in pre-anthesis with a higher AB accumulation and a significant remobilization from vegetative parts to grain than sole occurred during grain filling (Table 5). Furthermore, the average ABRC of SR wheat over the three P application rates and two years was 41.8%, which was 7.2% and 13.0% higher than that of IR and BR (Table 5). Although, intercropping wheat showed a yield advantageous over sole, but the ABRC of intercropping wheat was lower than sole. These results demonstrated that intercropping wheat produced more AB than sole during grain filling, as the intercropping maize accumulated more AB and nutrient than sole in post-anthesis in previous studies (Li et al., 2001).

P accumulation of intercropping wheat is advantageous over corresponding sole wheat, and wheat PB increased with the application of P fertilizer (Table 4). P accumulation during grain filling was much less than P remobilization, so most of the grain P content originates from root absorbed during pre-anthesis (Papakosta, 1994; Dordas, 2009). However, the PRC of BR and IR wheat were lower than SR, although intercropping wheat had a higher PB than sole (Table 6). Intercropping wheat with a significant part of P was taken up in post-anthesis until maturity, especially in BR, which consistent with other report that found P uptake depending on plant demand (Batten, 1992). Intercropping wheat may absorb of additional P from maize strip to enhance P uptake during grain filling (S<sub>3</sub> Figure). Our present study demonstrated that with more P uptake after anthesis may lead to high grain yield.

Higher productivity of intercropping over sole was a result of increasing light capture, especially in border rows. Zhang et al., (2008) found radiation interception by relay strip intercropping systems were affected by the geometry of the canopies and systems with a higher fraction of BR usually captured more light on a reclaimed sandy loam soil in Henan province, China. In another wheat/maize intercropping system, six rows of wheat alternated with two rows of maize and twelve rows of wheat alternated with four rows of maize, which showed per row of inter-wheat intercepted 17% and 9% more radiation than sole in the Hetao irrigation district of China respectively (Wang et al., 2015a). Belowground interactions between intercrops also enhanced nutrient accumulation and grain yield of intercropping over sole (Liu et al., 2015; Zhang et al., 2015b). Root separation with solid plastic sheet between intercrops decreased grain yield compared with no root separation in wheat/maize intercropping conducted at Gansu province, China (Mu et al., 2013). If maize was not grown in the area between two wheat strips in the designed wheat/maize intercropping, the yield of BR less than the wheat/maize intercropping with maize (Li et al., 2001). Studies have been demonstrated that belowground nutrient exchange occurred during the common growth period of the two intercrops (Mu, 2011). In the present study, wheat was planted

130-140 days earlier than maize, allowing wheat plants to utilize additional nutrients from the maize strips (S<sub>3</sub> Figure), also wheat and maize had a co-growth period about 45 days (Figure 1). During the common growth period some exchanges of soil nutrients may occur between the rooting zones of the two intercrops, which perhaps one of the reasons for intercropping wheat with a higher P uptake than sole wheat during grain filling (Table 4). Overall, intercropping wheat especially in BR intercepted more radiation and utilized additional nutrient to affect the source-sink relations during the whole growing season, which enhanced the grain yield.

In the intensive agricultural region, such as southwest of China, pursuing high yield to satisfy the demand of the great population has been the top priority in policy and practice. In practice, the typical P application rate in wheat by farmers in the Southwest China is around 50 kg P ha<sup>-1</sup> (Mu, 2011). In present study our results showed more than 40 kg P ha<sup>-1</sup> did not result in higher yield (Table 1). Most of nutrient management studies focus on nitrogen fertilizer, especially overuse has the negative environmental effect of increasing N-related emissions from soil such as N<sub>2</sub>O, NH<sub>3</sub>, NO<sub>x</sub> and nitrate leaching (Erisman et al., 2008; Cui et al., 2013). However, P is commonly regarded as a key driving force of water body eutrophication, therefore reducing water pollution, especially at the hilly area, such as the Southwest of China is indispensable. In our previous study have demonstrated that intercropping winter wheat with 40 kg P ha<sup>-1</sup> application rate could met the P balance of inputs/ outputs and maintain the soil Olsen-P at an appropriate level (19.1 mg kg<sup>-1</sup>) (Chen et al., 2015). Thus, a rational P management could obtain high grain yield, mitigate the P efflux from the arable field, which is vital for protecting the ecological environment in the Southwest of China and the upstream of the Yangtze River.



S<sub>3</sub> Figure. Specific relationships between intercropping wheat and maize.

## Conclusions

Intercropping wheat overyielding correspondence sole is ascribed to special source-sink relationships, as higher AB and P accumulation of intercropping wheat across the whole growing season and higher ABRE and PRE than sole, especially of BR. Differ physiological processes of intercropping wheat to sole may ascribed to ameliorated light and nutrient environment. As in low light and soil P content agricultural region, for



increasing plant production ameliorate plant growth environment by agronomic practices like alter crop pattern, improve nutrient management is useful. Even though, much more knowledge need for exploring the underlying mechanisms of crop physiological characteristics response to growth environment.

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