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RESEARCH ARTICLE

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Long-term straw incorporation benefits the elevation of soil phosphorus availability and use efficiency in the agroecosystem

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Abstract

Soil pH and organic matter are important factors influencing phosphorus (P) fertilizer use efficiency. Long-term crop straw incorporation alters soil pH and soil organic matter. To explore the influence of crop straw incorporation on P fertilizer use efficiency, this research was conducted in a long-term field experiment (30 years) with a wheat-soybean cropping system and selected four treatments: no fertilization, mineral fertilization (NPK), mineral fertilization + 3750 kg/ha wheat straw (WS/2-NPK) and mineral fertilization + 7500 kg/ha wheat straw (WS-NPK). Results show that long-term straw incorporation not only accentuates soil acidification, but also elevates crop yields and soil P availability. Consequently, compared with the NPK treatment, straw incorporation contributed to higher P fertilizer use efficiency, which increased from 43% in 1983 to 72% in 2012 for WS/2-NPK, from 46% to 69% for WS-NPK, and from 34% to 60% for NPK treatments, respectively. Moreover, the P fertilizer use efficiency in all fertilization treatments could be categorized as follows: slowly increasing stage in 1982-2002, stable stage in 2003-2006, and rapidly increasing stage in 2007-2012. Correspondingly, the annual P balances of the WS/2-NPK and WS-NPK treatments ranged from positive to negative in the 1982-2003 and 2004-2012. Therefore, compared with mineral fertilization alone, long-term wheat straw incorporation has the associated benefit of elevating the P fertilizer use efficiency. However, to maintain sustainable high crop productivity, it is necessary to elevate the dose of P fertilizer input and reduce the soil acidification under wheat straw incorporation.

Additional keywords: crop management; Fertilization; phosphorus fertilizer use efficiency; wheat straw.

Abbreviations used: APB (Annual phosphorus balance); AVP (Soil available phosphorus); FUE (Fertilizer use efficiency); NF (No fertilization); NPK (Mineral fertilization); Pi (Inorganic phosphorus); Po (Organic phosphorus); SBD (Soil bulk density); STP (Soil total phosphorus); TN (Soil total nitrogen); WS-NPK (Mineral fertilization + 7500 kg/ha wheat straw); WS/2-NPK (Mineral fertilization + 3750 kg/ha wheat straw).

Authors' contributions: Conceived and designed the experiments: ZG, DW and CH. Performed the experiment: ZG and KH. Analyzed the data: ZG & HL. Wrote the paper: ZG. All authors read and approved the final manuscript.

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Introduction

Phosphorus (P) is an essential and limiting element for plant growth. At the global scale, about 30% of total croplands suffer from P deficiency (Lynch, 2007; MacDonald *et al.*, 2011). To maintain or increase crop yields, a large amount of P fertilizers are annually input into croplands (Roy *et al.*, 2017). However, only 10-30% of the P fertilizers are taken up by plants in the year of application because of the low mobility, high fixation of P in soils, and the inefficient P fertilizer management (Zhang *et al.*, 2008a, b). Consequently, a substantial component of the added P fertilizers in croplands accumulates in the soil as residual P and lowers the P use efficiency in the year of application (Wironen *et al.*, 2018). In the context of finite rock phosphate reserves in the world, it is vital to increase the P use efficiency in the year of application to develop sustainable agriculture and maintain or increase current agricultural productivity (George *et al.*, 2016).

Soil P forms include inorganic P (Pi) and organic P (Po). Of these, the Pi could be fractionated into Ca-P, Fe-P, Al-P and O-P, and its bioavailability are closely related to soil pH and the anions that compete with P ions for ligand exchange reactions (Hinsinger, 2001). The Po, which is less mobile than Pi, and accounts for 20%-80% of soil total P (Hansen *et al.*, 2004; Fransson & Jones, 2007), represents a major reserve of potentially available P and is available to plants after mineralization by soil organisms or by phosphatase (Soltangheisi *et al.*, 2018). The Po in soils is originally derived from microbial, plant, or animal residues, and its mineralization processes are also closely related to soil pH (Zhou *et al.*, 2016; Hou *et al.*, 2018). Therefore, the concentration of soil available P (mainly $H_2PO_4^-$ and HPO_4^{2-}) is strongly affected by soil pH.

Besides pH, soil organic matter is another important factor influencing soil available P content (Zhou *et al.*, 2016). For example, organic acids released during organic matter decomposition are important mechanisms to reactivate insoluble inorganic phosphorus to plants under organic fertilizer application alone or combined with mineral fertilizer (Ayaga *et al.*, 2006; Martins *et al.*, 2008). However, soil organic matter is relatively stable and needs a long time to accumulate in the agroecosystem (O'Brien & Jastrow, 2013; Guo *et al.*, 2014). Thus, previous innovative fertilization regimes and bio-technology for P fertilizer use efficiency improvement have mainly focused on enhancing soil P availability by decreasing soil pH over a short-term period (Jing *et al.*, 2012; Liu *et al.*, 2013; Mahanta *et al.*, 2014).

Agriculture in China produces the most crop straw in the world, approx. 0.8 billion tonnes/yr. To develop sustainable agriculture, crop straws are widely used for soil quality improvement (Guo *et al.*, 2014; Boitt *et al.*, 2017; Soltangheisi *et al.*, 2018). Studies have shown a higher level of soil organic matter and lower soil pH under long-term crops straw incorporation than under mineral fertilization alone (Guo *et al.*, 2014). Therefore, compared to conventional mineral fertilization, less P fertilizer input may be possible because of the elevated soil organic matter and decreased pH with long-term wheat straw incorporation. Under the above hypothesis, we implemented this research based on a long-term field experiment to explore the relationship between long-term wheat straw incorporation and P fertilizer use efficiency and guide the P fertilization management in a P-limited calcareous soil in the Huang-Huai-Hai plain of China.

Material and methods

Description of the experimental site

This study was conducted based on a long-term field experiment located in MengChen County of AnHui province, China (32°13'N, 116°37'E). The long-term experiment started in 1982 and details of the experiment are given in our previous research (Guo *et al.*, 2014). During the experiment, the annual precipitation from 1986 to 2011 is 792 mm (see Fig. 1 of Guo *et al.*, 2014), and the average temperature in the period of 1987-2012 (except for 1991 and 1992) is 16.5°C. According to the USDA Soil Taxonomy, the soil is a vertisol with pH ranging from 6.0 to 8.6, and consists of 25–26% fine clay (<0.001 mm), 43–45% fine silt (0.001–0.01 mm) and 33–50% coarse silt (0.01–0.05 mm) (Soil Survey Staff, 2010; Li *et al.*, 2011). At the beginning of the experiment in 1982, the soil in the 0-20 cm layer contained 10.40 g/kg of soil organic matter, 0.96 g/kg of total nitrogen (TN), 84.5 mg/kg of alkali-hydrolysis nitrogen, 0.28 g/kg of total P, 9.8 mg/kg of available P and 111 mg/kg of available potassium (K). In this study, four treatments were selected as follows: no fertilization (NF), mineral fertilization (NPK), mineral fertilization plus 3750 kg/ha wheat straw (WS/2-NPK) and 7500 kg/ha wheat straw (WS-NPK). There were four replicates for each treatment, and all plots were arranged randomly. The area of each plot was 70 m^2 (14.9 × 4.7 m). Across the experiment, the crops were wheat (var. Yannong19)-soybean (var. Zhonghuang13) except from 1993 to 1997 during which a wheat (var. Yannong19)-corn rotation (var. Yedan13) system was

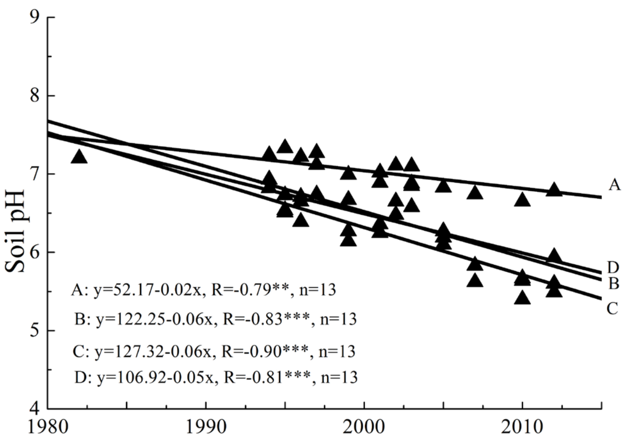

Figure 1. Changes of soil pH under no fertilization (NF) (A), NPK plus 3750 kg/ha wheat straw (WS/2-NPK) (B), NPK plus 7500 kg/ha wheat straw (WS-NPK) (C) and NPK (D) treatments across the experimental period (1982-2012). In order to make the graphs more clear, the error bars in Figures 3, 4 and 5 are not shown (the same as below).

used. The application rates of the mineral fertilizers were 180 kg/ha N, 90 kg/ha P, and 135 kg/ha K. The wheat straw incorporated in the cropland with WS/2-NPK contained 1807.5 kg C/ha, 20.6 kg N/ha, 4.5 kg P/ha and 43.1 kg K/ha, and 3615.0 kg C/ha, 41.2 kg N/ha, 9.0 kg P/ha and 86.2 kg K/ha for WS-NPK treatment. Every autumn, prior to winter wheat seeding, the wheat straw (chopped into small pieces) and mineral fertilizers were both applied as basal fertilizers with conventional tillage. For the soybean growth season, no tillage practice was implemented. In our research, the conventional tillage consisted mainly of using mouldboard ploughing (to a 15 cm depth). After harvest, wheat and soybean/corn straws were removed from the cropland.

Soil and plant sampling and measurements

Soil samples at depth of 0-20 cm were taken annually each October during the experimental period. An auger with an 8 cm internal diameter was used to collect soil samples three times in each plot. The collected soil samples were then air dried, sieved with a 2.0 mm sieve, and stored for analysis of soil physicochemical properties. Additional soil samples from the 0-20 cm layer were taken using ring kits (V=100 cm^3) in October of 1995, 2005 and 2012 for soil bulk density (SBD) analysis. After oven drying at 105 °C for 24 h, SBD was measured.

Wheat and soybean straw and grain yields were measured from 1982 to 1993 and 1998 to 2012, respectively. Wheat grain and straw with three replicates were taken in each plot in July 2012 for the P content analysis, and for soybean grain and straw in October 2012. The collected crop samples were dried at 70 ºC for more than 72 hours, and then fine ground to pass a 0.15-mm sieve.

Chemical analysis

Soil samples from all treatments were analyzed for soil pH, TN and total and available P, and soil samples taken in 2005 and 2011 were further analyzed for P fractions. Specifically, soil pH was determined in a 1:1 soil solution ratio (water/soil) (Peech, 1965). Soil TN was measured by the semimicro Kjeldahl digestion method (Kieltec Foss 2200, Denmark) (Bremner & Mulvaney, 1982). Soil available P using the molybdenum-blue colorimetric method after extracting soil with 0.5 M $NaHCO_3$ at pH 8.5 (Olsen & Sommers, 1982) and total P (STP) by colorimetric measurement after digestion using perchloric and sulfuric acid. The inorganic P fractions, including Ca_2-P, Ca_8-P, Al-P, Fe-P, occluded P and Ca_{10}-P, were determined according to a modified sequential fractionation scheme for calcareous soils (Jiang & Gu, 1989) modified from Chang & Jackson (1957) and Hedley *et al.* (1982). Briefly, 1.0 g soil sample was weigh into a 100-mL centrifuge tube, and then: (1) Ca_2-P was extracted by shaking with 0.25 mol L^{-1} $NaHCO_3$ solution at pH 7.5; (2) Ca_8-P was extracted with 0.5 mol/L NH_4Ac at pH 4.2; (3) Al-P was extracted with 0.5 mol/L NH_4F at pH 8.2; (4) Fe-P was extracted with 0.1 mol/L NaOH-0.1 mol/L Na_2CO_3; (5) occluded P was extracted with 0.3 mol/L Na_3(citrate)-$Na_2S_2O_4$-0.5 mol/L NaOH solution; and (6) Ca_{10}-P was extracted with 0.25 mol/L H_2SO_4. The organic P was determined in the $NaHCO_3$, NaOH and concentrated HCl fractions by persulfate oxidation, and its value was calculated as the difference between total P and inorganic P (Tissen & Moir, 1993).

For the P content of the crop samples, the molybdenum-blue colorimetric method was used after digestion of the samples with H_2SO_4-H_2O_2 (Page *et al.*, 1982).

Calculations

The crop P uptake was calculated based on the grain and crop straw yields and their corresponding P concentrations. Based on the cumulative amount of P fertilizer input and crop uptake from 1982 to 2012, the accumulated fertilizer use efficiency (FUE, %) was calculated as follows:

$$FUE = (U_{Pn}-U_{0n}) \times 100/P_{rn},$$

where U_{Pn} (kg P/ha) is the total accumulated P uptake starting in 1982 by crops with P fertilization treatment in n years, and U_{0n} (kg P/ha) represents the total accumulated P uptake from 1982 by crops including grain and straw from the unfertilized treatment in n years, and P_{rn} (kg P/ha) is the total accumulated input of P fertilizer starting from 1982 in each fertilization treatment in n years (Hossain *et al.*, 2005).

The annual P balance (APB) (kg/ha) was calculated as follows:

$$APB = P_{input} - P_{output}$$

where P_{input} (kg P/ha) is the annual P input into cropland with P fertilization; and P_{output} (kg P/ha) represents P removed by the aboveground biomass of wheat and soybean.

Statistical analysis

In this research, the differences in wheat and soybean yields and straws and soil phosphorus under various treatments were analyzed using the Kruskal-Wallis post hoc Kruskal-Nemenyi test by R software (Vers. 3.4.1). The one-way analysis of variance procedure in SAS 9.1.3 was used to analyze the differences of soil P fractions among various treatments. When ANOVAs were significant, the least significant difference (LSD) test at the 0.05 confidence level was used to compare the means. The correlation analysis procedure in SAS 9.1.3 (SAS Institute, Cary, NC, USA) was used to determine the relationships between soil available P and soil P fractions.

Moreover, SigmaPlot 10.0 (Systat Software Inc., San Jose, USA) was used for linear regression analysis and to generate figures.

Results

Soil physicochemical properties

Figure 1 shows that soil acidification was accentuated under long-term wheat straw incorporation combined with mineral fertilizer application in comparison with the NPK and NF treatments. Specifically, soil pH under the NPK, WS/2-NPK and WS-NPK treatments in 2012 was lower by 0.8, 1.2 and 1.3, respectively, compared with the NF treatment at pH of 6.8.

Besides soil pH, crop straw incorporation significantly improves soil bulk density (SBD) (Table 1). When compared with NPK, the SBD decreased by 9% under the WS/2-NPK and 7% under the WS-NPK in 1995, by 9% under WS/2-NPK and 12% under WS-NPK in 2005, and by 3% under WS/2-NPK and 5% under WS-NPK in 2012, respectively.

Table 1. Soil bulk density (g/cm^3) of the 0-20 cm layer in 1995, 2005 and 2012 under the NF, WS/2-NPK, WS-NPK and NPK treatments.

Treatments	1995	2005	2012
NF	1.38±0.02a	1.36±0.01a	1.37±0.01a
WS/2-NPK	1.24±0.02b	1.24±0.01b	1.29±0.02ab
WS-NPK	1.26±0.01b	1.20±0.01b	1.27±0.02b
NPK	1.36±0.01a	1.36±0.01a	1.33±0.02a

Different letters in a column indicate significant treatment effect at the $p < 0.05$ level.

Total phosphorus and nitrogen, available P and soil P fractions

Long-term crop straw incorporation elevates soil total phosphorus and nitrogen and available P in the 0-20 cm layer (Figs. 2A-B). After regression analysis, we found that soil total P content with WS/2-NPK (R=0.83, p<0.001) and WS-NPK (R=0.86, p<0.001) treatments was significantly positively related to the number of years of fertilization, and there was a positive linear relationship between soil available P and total P in the WS/2-NPK (R=0.65, p<0.05) and WS-NPK treatments (R=0.59, p<0.05). Consequently, soil total P in 2012 was higher by 52% under NPK, 100% under WS/2-NPK and 124% under WS-NPK than under NF, and higher by 429% under NPK, 812% under WS/2-NPK and 824% under WS-NPK for soil available P. For soil TN, its concentration also increased under the WS/2-NPK and WS-NPK treatments (Fig. 2C). Specifically, compared to the average value of TN in the period of 1983-1987, the average TN in 2009-2012 was respectively higher by 10% with WS/2-NPK and 24% with WS-NPK treatments. In contrast, the average TN decreased by 23% under NF treatment.

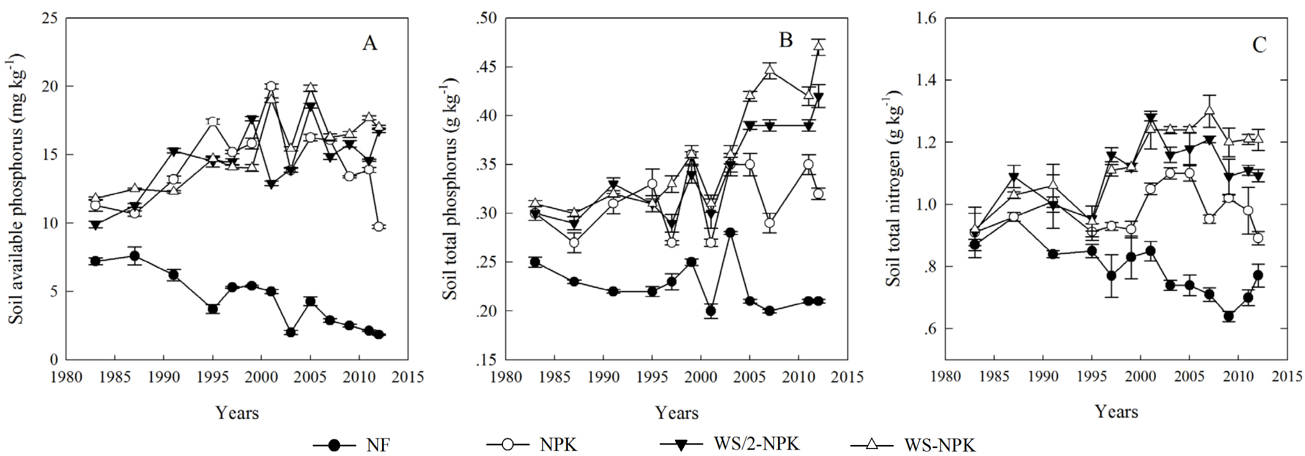

Figure 2. Dynamics of soil available P (A), soil total P (B) and total nitrogen (C) from 1983 to 2012 under the no fertilization (NF), NPK plus 3750 kg/ha wheat straw (WS/2-NPK), NPK plus 7500 kg/ha wheat straw (WS-NPK) and NPK treatments.

For inorganic P fractions, the concentrations of Ca_2-P, Ca_8-P, and Al-P under the WS/2-NPK and WS-NPK treatments were significantly higher than those under the NPK treatment in 2005 (Table 2). However, there were no significant differences in the levels of Ca_2-P, Ca_8-P, and Al-P in 2011 among all fertilization treatments. Moreover, crop straw incorporation significantly elevated soil organic P relative to mineral fertilization alone (Table 2). Specifically, soil organic P was increased by 21% under WS/2-NPK and 29% under WS-NPK relative to the NPK treatment in 2005, and by 37% and 43% in 2011, respectively. However, compared with inorganic P fractions such as Ca_2-P, Al-P and Fe-P, soil organic P played less important role in improving soil P availability in all fertilization treatments (Table 3).

Combined with the results on soil organic carbon of Guo *et al.* (2014), this research found that the ratio of soil organic carbon to total nitrogen (C/N) increased in all treatments after long-term fertilization (Fig. 3A). Specifically, in the periods of 1982-1991 and 2007-2012, the average C/N ratio ranged from 6.95 to 8.00 in NF, from 6.96 to 8.83 with NPK treatment, from 7.17 to 9.95 with WS/2-NPK treatment and from 6.78 to 9.98 under WS-NPK. Similarly, the ratios of soil organic carbon to available phosphorus (C/AVP) increased in all treatments (Fig. 3B). Between the periods of 1982-1991 and 2007-2012, the average C/AVP ratio ranged from 888.31 to 2471.57 with NF, from 573.80 to 632.48 with NPK, from 598.93 to 708.45 with WS/2-NPK and from 558.22 to 709.84 with WS-NPK treatments, respectively. Based on our analysis, the C/AVP ratio in NF was significantly higher than that of NPK, WS/2-NPK and WS-NPK treatments.

Crop productivities and P concentrations in crop grain and straw

Fertilization management elevates crop productivities (Figs. 4 and 5). In the 2007-2012 period, the average wheat grain under the NPK, WS/2-NPK, and WS-NPK treatments were 5263 kg/ha, 5400 kg/ha and 5607 kg/ha, respectively, and these values were higher than those in the period of 1982-1989 by 29%, 34% and 43%, respectively. For wheat straws, the increase was 21%, 21% and 31% above 1982-1989 levels, respectively. Similarly, the average soybean grain under the NPK, WS/2-NPK and WS-NPK treatments in 2007-2012 were 1879 kg/ha, 2013 kg/ha, and 2198

Table 2. Amounts of plant available phosphorus (AVP), inorganic phosphorus (IOP) including Al-P, Fe-P, Ca-bound (Ca_2-P, Ca_8-P and Ca_{10}-P) and occluded fractions (O-P: occluded P) and organic phosphorus (OP) in 2005 and 2011 in the 0-20 cm soil layer under NF, WS/2-NPK, WS-NPK and NPK treatments.

Year	Treatments	AVP (mg/kg)	IOP (mg/kg)						OP (mg/kg)
			Ca_2-P	Ca_8-P	Al-P	Fe-P	O-P	Ca_{10}-P	
2005	NF	8.14±0.80b	1.44±0.13c	2.21±0.20c	6.09±0.57c	11.91±1.05b	79.91±6.47b	34.21±3.17c	76.68±6.75c
	WS/2-NPK	18.59±1.11a	13.64±0.89a	17.99±1.59a	15.21±1.49ab	31.32±3.18a	105.19±9.79a	53.07±6.09b	155.08±10.32a
	WS-NPK	19.88±0.98a	15.33±1.32a	20.25±2.06a	19.09±2.11a	35.99±2.69a	105.80±11.23a	55.68±4.31b	164.96±12.19a
	NPK	16.25±1.00a	8.22±0.69b	10.98±0.89b	13.51±1.01b	28.38±3.17a	92.72±9.17a	69.77±7.80a	128.00±11.76b
2011	NF	3.60±0.27b	2.34±0.51b	2.17±0.38a	18.47±2.17b	24.87±2.35b	50.54±2.83b	64.09±5.65b	84.03±3.42c
	WS/2-NPK	14.60±2.21a	20.91±1.89a	2.30±0.22a	34.20±5.22a	49.48±3.19a	54.07±11.27b	81.43±3.56a	143.09±7.80a
	WS-NPK	17.70±1.99a	23.12±1.84a	2.67±0.35a	40.77±4.33a	50.69±3.94a	38.63±3.53c	73.87±3.11a	149.66±6.12a
	NPK	13.90±1.33a	22.86±3.36a	2.06±0.24a	43.08±7.55a	56.66±4.20a	97.70±8.36a	76.32±3.37a	104.37±8.24b

Different letters in a column indicate significant treatment effect at $p < 0.05$ level.

Table 3. Relationships among the soil P fractions in the 0-20 cm soil layer in NPK, WS/2-NPK and WS-NPK treatments (n=24).

	Ca_2-P	Ca_8-P	Ca-Al	Ca-Fe	O-P	Ca_{10}-P	OP
AVP	0.836***	-0.097	0.645***	0.665***	0.193	0.093	0.106
Ca_2-P		-0.168	0.675***	0.651***	0.208	0.065	0.142
Ca_8-P			-0.082	0.119	0.044	0.190	0.223
Ca-Al				0.592***	-0.128	0.105	0.153
Ca-Fe					0.367*	0.191	-0.001
O-P						0.179	-0.500**
Ca_{10}-P							0.127

AVP: soil available phosphorus. OP: organic phosphorus. Significant level: * p <0.05, ** p <0.01, *** p <0.001.

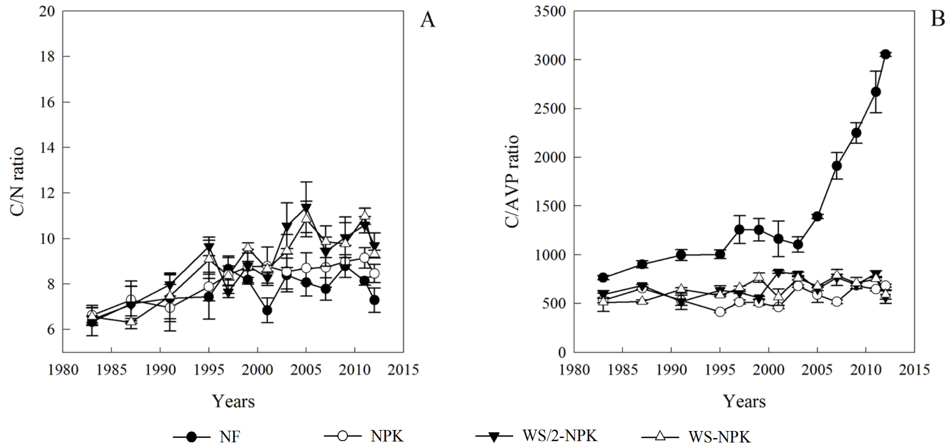

Figure 3. The ratios of C/N (soil organic carbon/total nitrogen) (A) and C/AVP (soil organic carbon/available phosphorus) (B) in the period of 1983-2012 under the no fertilization (NF), NPK plus 3750 kg/ha wheat straw (WS/2-NPK), NPK plus 7500 kg/ha wheat straw (WS-NPK) and NPK treatments.

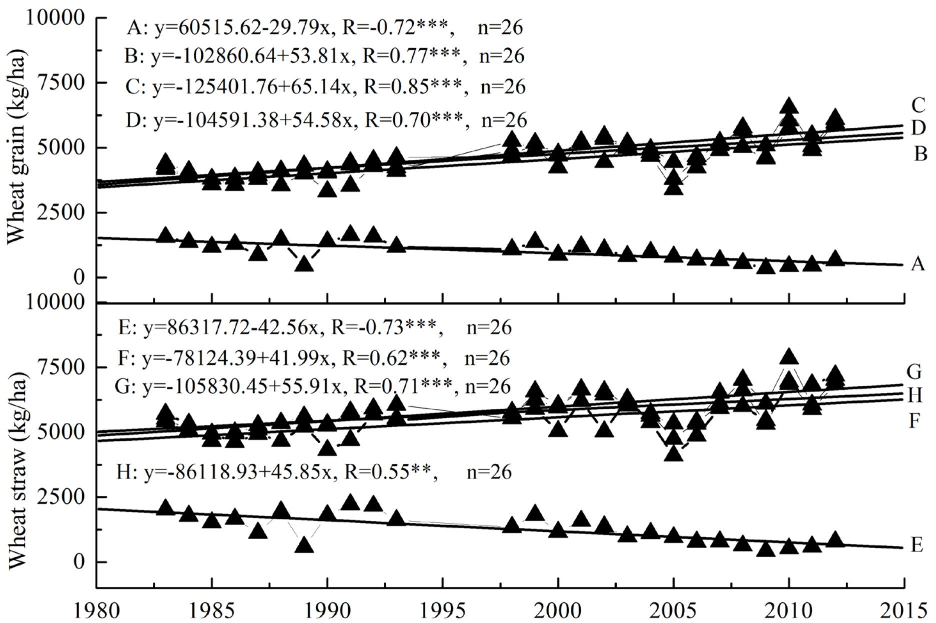

Figure 4. Wheat grain yield and wheat straw under no fertilization (NF) (A and E), NPK plus 3750 kg/ha wheat straw (WS/2-NPK) (B and F), NPK plus 7500 kg/ha wheat straw (WS-NPK) (C and G), and NPK (D and H) treatments in the experimental period (from 1982 to 2012).

kg/ha, respectively, which was higher by 40%, 28%, and 36% higher than the equivalent values in 1982-1989. The soybean straws increased by 40%, 28% and 36%, respectively, relative to the 1982-1989 period. In the 2004-2006 period, the wheat grain under the NPK, WS/2-NPK, and WS-NPK treatments averaged 4100, 4385, and 4683 kg/ha, significantly lower than the values in 2001-2003 and 2007-2012. Similarly, wheat straw, soybean grain and straws in 2004-2006 were all significantly lower than those in 2001-2003 and 2007-2012.

The concentrations of P in crop grain and straw were also closely related to fertilization regime. In 2012, the P concentrations of wheat grain in NF, NPK, WS/2-NPK, and WS-NPK treatments were respectively 2.80, 2.62, 3.17, and 3.50 g/kg, and 0.33, 0.15, 0.42, and 0.35 g/kg, respectively, for wheat straw. The soybean grain P concentrations in the NF, NPK, WS/2-NPK, and WS-NPK treatments were respectively 9.85, 8.94, 9.26, and 9.69 g/kg, and 4.97, 4.19, 4.24, and 5.02 g/kg, respectively, for soybean straw.

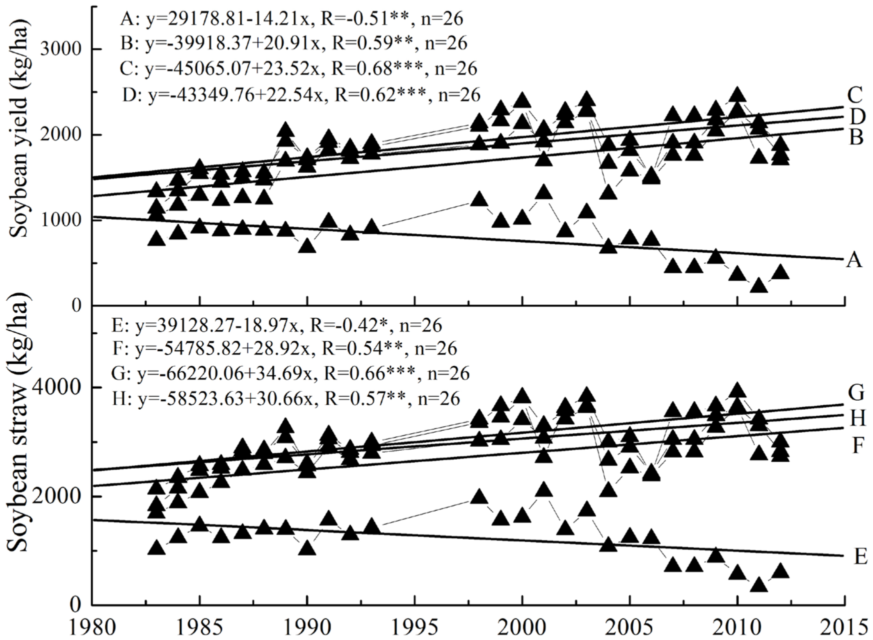

Figure 5. Soybean yield and straw under no fertilization (NF) (A and E), NPK plus 3750 kg/ha wheat straw (WS/2-NPK) (B and F), NPK plus 7500 kg/ha wheat straw (WS-NPK) (C and G), and NPK (D and H) treatments in the experimental period (ranging from 1982 to 2012).

Phosphorus use efficiency, soil P storage and annual P balance

Crop straw incorporation enhances P use efficiency (Figs. 6A, B and C). Specifically, the P use efficiency increased from 34% in 1983 to 60% in 2012 under the NPK treatment, from 43% in 1983 to 72% in 2012 under the WS/2-NPK treatment, and from 46% in 1983 to 69% in 2012 under the WS-NPK treatment. Regression analysis between P use efficiency and number of fertilization years showed that P use efficiency in all fertilization treatments could be categorized into three stages: slowly increasing stage in 1982-2002, stable stage in 2003-2006, and rapidly increasing stage in 2007-2012 (Fig. 6). In the slowly increasing stage (from 1982 to 2002), P use efficiency increased from 34% to 53% under the NPK, from 43% to 65% under the WS/2-NPK, and from 46% to 61% under the WS-NPK, respectively. In the stable stage (from 2003 to 2006), P use efficiency remained at 53%, 65% and 61% under the NPK, WS/2-NPK, and WS-NPK treatments, respectively. In the rapidly increasing stage (from 2007 to 2012), P use efficiency increased from 53% to 60% under the NPK treatment, from 65% to 72% under the WS/2-NPK treatment, and from 61% to 69% under the WS-NPK treatment, respectively.

The annual P balance was negatively related to the fertilization years and also categorized into two stages: 1982-2003 and 2004-2012. Specifically, the annual P balance in the period of 1982-2003 varied between 13.96 kg/ha in 1983 and -5.54 kg/ha in 2003 under the NPK treatment, between 12.85 and -6.27 kg/ha under the WS/2-NPK treatment, and between 13.78 and -4.17 kg/ha under the WS-NPK treatment, respectively. From 2004 to 2012, the annual P balance varied between 9.69 kg/ha in 2004 and 1.21 kg/ha in 2012 under the NPK treatment, between 3.53 and -1.42 kg/ha under the WS/2-NPK treatment, and between 3.68 and -0.27 kg/ha under the WS-NPK treatment, respectively.

Discussion

Soil pH and fertilization regime

Consistent with the results of Guo *et al.* (2010), who reported that long-term mineral fertilization could result in soil acidification, the results of the current study additionally show that soil acidification can be accentuated under long-term wheat straw incorporation combined with application of mineral fertilizers. In addition to the excessive N fertilizer application (Guo *et al.*, 2010), the significantly decreasing soil pH under the WS/2-NPK and WS-NPK treatments may also be due to the larger amount of organic materials mineralization annually in the WS/2-NPK and WS-NPK treatments, which maybe contribute formation of H_2CO_3 from H_2O and the released CO_2 leading to potential soil

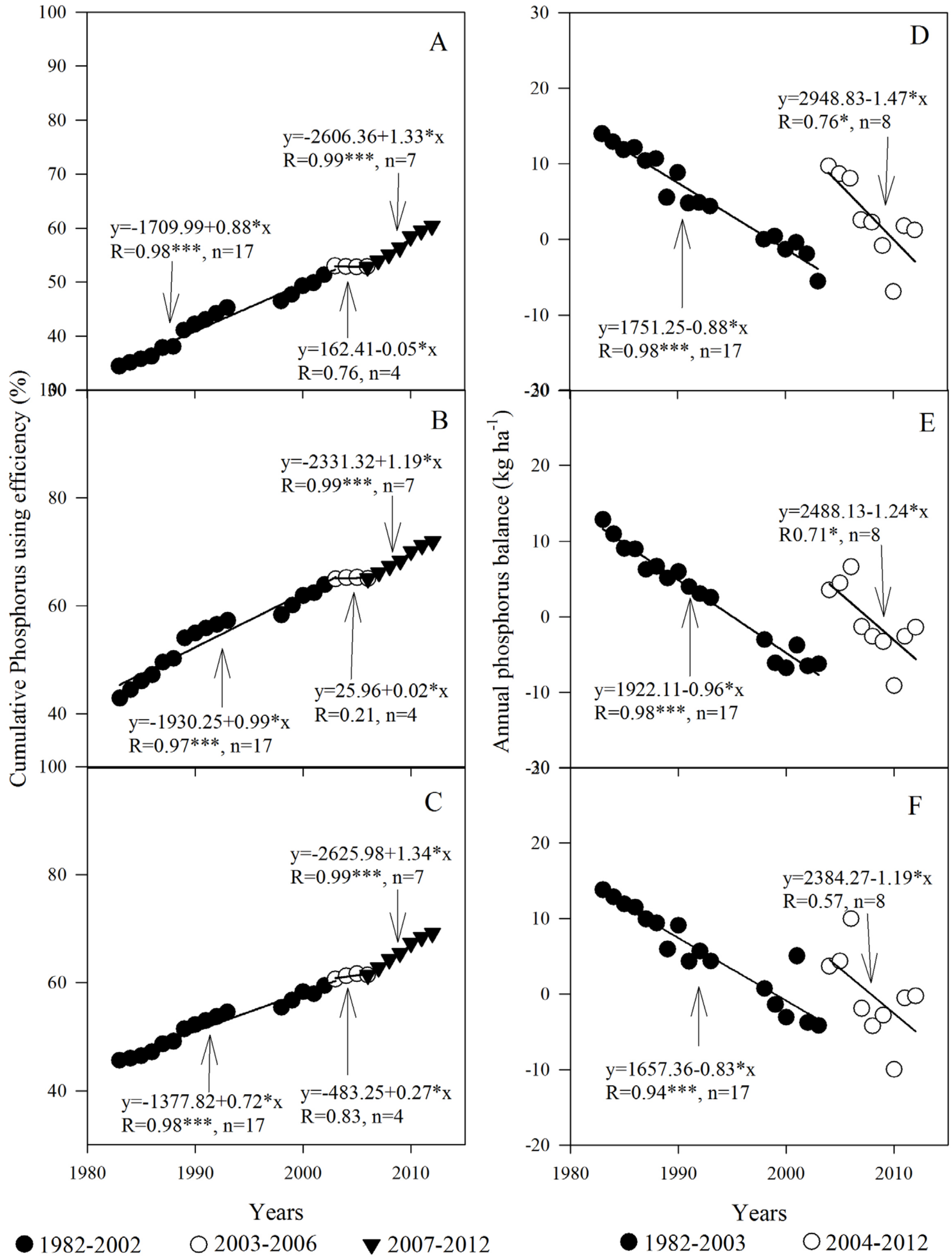

Figure 6. Cumulative P use efficiency (%) and annual P balance (kg/ha) under NPK (A and D), NPK plus 3750 kg/ha wheat straw (WS/2-NPK) (B and E), and NPK plus 7500 kg/ha wheat straw (WS-NPK) (C and F) treatments in the experimental period (ranging from 1982 to 2012). Arrows represent the regression equation in each experimental period (3 stages: 1982-2002, 2003-2006 and 2007-2012).

acidification. Combined with our previous results, the current study shows that the soil organic matter reaches its balanced point at the same time as the significant decrease of soil pH after long-term fertilization (Guo *et al.*, 2014). For example, the significant decreases of soil pH mainly occurred between 2004 and 2012, and the decrease rate/yr of pH was 0.10, 0.13 and 0.12 under the NPK, WS/2-NPK, and WS-NPK treatments, respectively. In contrast, the decrease rate/yr of pH between 1982 and 2003 was 0.02, 0.02 and 0.03, respectively.

Soil P, its availability and fertilization regime

Previous research suggested that the ratio of C:N:P was an important indicator for soil quality assessment (Tian *et al.*, 2010; Tipping *et al.*, 2016). In China, a general C:N:P ratio of 134:9:1 was found for the 0-10 cm organic-rich soil (Tian *et al.*, 2010). As a stable level of soil organic carbon existing in the NPK, WS/2-NPK and WS-NPK treatments after long-term fertilization (Guo *et al.*, 2014), compared to the study of Tian *et* *al.* (2010), the lower ratios of C/N in all fertilization treatments indicated that soil nitrogen in the C/N ratio was higher than its average level for the 0-10 cm organic-rich soil in a national scale. Therefore, soil N may be not the factor limiting crops productivity. Instead, soil AVP was the limited factor influencing crops growth. According to the results of Chen (1990) and Huang (2000) who reported that when C/AVP ratio were >300, soil microbial biomass carbon would increase rapidly, thereby immobilizing organic P and contributing to a competition between plant and microbes on soil AVP. In the present research, the C/AVP ratio was >300 in all treatments during the experimental period, suggesting that plant AVP was too low to meet the need of crops growth and a fierce competition existed between soil microbes and crops on soil AVP.

Soil acidification affects soil P availability (Zhou *et al.*, 2016; Hou *et al.*, 2018). As previous reported, plants assimilate P predominantly as $H_2PO_4^-$ and HPO_4^{2-} from the soil solution and take up more $H_2PO_4^-$ than HPO_4^{2-} (Prasad *et al.*, 2017a, 2017b). For the orthophosphate ions in soil solution, its dominant form is pH dependent. Generally, orthophosphate usually exists as $H_2PO_4^-$ ions at pH 4–5, while HPO_4^{2-} and PO_4^{3-} ions subsequently become more dominant as pH increases (Yadav & Verma, 2012). For soil Po, it could be solubilized and hydrolyzed by organic acids and phosphatase enzymes (Darch *et al.*, 2016). In our research, the low soil pH could partially inhibit soil microbial and the phosphatase enzymes activities (Eivazi & Tabatabai, 1977; Siles & Margesin, 2016). Consequently, the mineralizable organic P may be more accumulated. Therefore, the high P availability with fertilization treatments in 2007-2012 may be driven by soil acidification.

Furthermore, our research also suggests that mineral fertilization combined with wheat straw input is beneficial for enhancing soil P availability and P storage in the 0-20 cm layer compared with mineral fertilization alone. This may be attributed firstly to inorganic and organic forms of P contained in wheat straw, which could be released directly to soil as soluble orthophosphate or assimilated by microorganisms and subsequently released back into the soil (Boitt *et al.*, 2017, 2018). For example, the additional input of P contained in wheat straw is 4.5 kg/ha under the WS/2-NPK treatment and 9.0 kg/ha under the WS-NPK treatment. Secondly, the enhancement of P in soils amended with both mineral fertilizer and wheat straw may be associated with the higher soil organic P concentrations after wheat straw incorporation. Though soil organic P is less mobile than inorganic P, larger fractions of P available to crops were shown to be maintained because of a higher rate of cycling for organic P than inorganic P (Hansen *et al.*, 2004; Xavier *et al.*, 2009; Malik *et al.*, 2013). In this study, soil organic P was higher by 70% and 21% under the WS/2-NPK treatment, and by 78% and 29% under the WS-NPK treatment compared to NPK in 2005 and 2011, respectively.

Phosphorus use efficiency and fertilization regime

Based on several long-term field experiments, Tang *et al.* (2008) stated that the P fertilizer use efficiency in the wheat-corn cropping systems could be up to 29%. In contrast, we found that P fertilizer use efficiency was significantly higher than 29% and closely related to the fertilization regime in a wheat-soybean cropping system. The higher P use efficiencies under the NPK, WS/2-NPK, and WS-NPK treatments than that of Tang *et al.* (2008) may be attributed to the method of the P use efficiency calculation exploited by our research. Generally, only 15–20% of the applied P fertilizers are taken up by crops in the first year following application (Zhang *et al.*, 2008a, b). Wheat straw is an efficient activator for soil legacy P availability (Zhu *et al.*, 2018). Consequently, the remainder "(legacy P)" accumulates in the soil in forms of inorganic P and organic P, which can be used for crop nutrition during subsequent decades (Rowe *et al.*, 2016; Liu *et al.*, 2017; Withers *et al.*, 2018). In the present study, we took account of the legacy P reuse by crops in next growing season. In contrast, the method exploited by Tang *et al.* (2008) for P use efficiency calculation is apparent recovery efficiency, which is significantly lower than real P use efficiency (Wang & Zhou, 2014; Withers *et al.*, 2018). Another potential explanation for the high P use efficiencies in our study is the influence of the cropping system on the P loss from farmland (Soltangheisi *et al.*, 2018). In the current research, the amount of P loss under wheat-soybean cropping systems is much lower than that with wheat-corn cropping systems (Jiao *et al.*, 2011).

Moreover, we found that P fertilizer use efficiency in all fertilization treatments could be categorized into three stages: slowly increasing stage (1982-2002), stable stage (2003-2006) and rapidly increasing stage (2007-2012). In the slowly increasing stage in 1982-2002, both the increasing productivities of crops and the elevated soil organic matter affect the P fertilizer use efficiency (Maranguit *et al.*, 2017). According to our previous research, the positive effects of fertilization on soil organic matter lasted for about two decades, and then reached a saturated point (Guo *et al.*, 2014). For the stable organic matter, P is an essential nutrient and exists with carbon, N and sulfur in approximately constant ratios (Tipping *et al.*, 2016). Therefore, the applied P fertilizers may partly participate in the formation of stable soil organic matter and slow the increase rates of P use efficiency.

In 2003-2006, the P use efficiency stabilization could be accounted for by the significant decreases in crop productivities. The significantly lower productivities of crop and straw yields in 2004-2006 than that in 2001-2003 and 2007-2012 may be caused by the considerable change in soil pH occurring in all fertilization treatments (Lollato *et al.*, 2013; Schroder *et al.*, 2011). For example, we found that soil pH decreased by 1.05 under NPK, 1.02 under WS/2-NPK, and 0.96 under WS-NPK from 2003 to 2007. Previous research has already reported that significant soil acidification could result in a decrease of crop yield (Zhao *et al.*, 2010).

At the quickly increasing stage in 2007-2012, higher productivities of crop grain and straw were the major factor contributed to the quickly increases of P use efficiency in all fertilization treatments. The higher productivities of crop yields and straws in all fertilization treatments in 2007-2012 respect to those in 2004-2006 may be attributed to the increasing soil P availability. Past studies suggested that crop yields were often limited by low P availability in soils as the adsorption and precipitation reactions of both the indigenous soil P and the applied P with Fe, Al, or Ca components (Hou *et al.*, 2018; Zhu *et al.*, 2018). In our research, there was no significant difference in soil available P between 2005 and 2011 in all fertilization treatments under a constant annual P input and increasing crop productivities. This suggests that high soil P availability was present in all fertilization treatments in 2007-2012.

Combined with the results of Guo *et al.* (2014) and the ratios of C/N and C/AVP in this research, we found that soil nitrogen is not a limited factor affecting crops productivity. Instead, soil available phosphorus is too low to meet crops growth. In an agroecosystem, wheat straw incorporation practice combined with mineral fertilization could significantly increase crop grain yields and straw biomass relative to mineral fertilization alone, and thus improved P fertilizer use efficiency. Consequently, it is necessary to elevate the dose of mineral P fertilizer input in wheat straw incorporation treatment to meet the increases of crop productivities in the winter wheat-soybean rotation systems. However, compared with mineral fertilization alone, long-term wheat straw incorporation could accentuate soil acidification. Therefore, the positive effect of wheat straw incorporation on P fertilizer use efficiency may not be sustainable, and lessening soil acidification is urgent for developing sustainable agriculture with long-term wheat straw incorporation management in a P-limited calcareous soil in the Huang-Huai-Hai plain of China.

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