

# Critical plant and soil phosphorus for wheat, maize, and rapeseed after 44 years of P fertilization

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**Abstract** Phosphorus (P) crop fertilization requires optimal management to avoid the waste of a non-renewable resource and water pollution, but current methods for assessing soil phyto-available P and plant P requirements are not sufficiently precise to meet this goal. The objectives of the present study were to (1) evaluate the effect of long-term P fertilization on the grain yield of winter wheat, maize, and rapeseed, (2) validate or establish models of critical shoot P concentration ( $P_C$ ) based on relationships of shoot P concentration with either shoot biomass or shoot nitrogen (N) concentration, and (3) assess both plant-based and soil-based diagnostic tools for managing P fertilization. A long-term field experiment with

contrasted P fertilizer treatments, established in 1971 by Agroscope in Changins (Switzerland), was used to measure the shoot biomass and P concentration of winter wheat in 2011, maize in 2012, and rapeseed in 2014 weekly during the growing period and the grain yield at harvest. Soil available P in the 0–0.20 m soil layer was assessed by three chemical extractions: ammonium acetate EDTA (P-AAE), sodium bicarbonate (P-NaHCO<sub>3</sub>), and CO<sub>2</sub>-saturated water (P-CO<sub>2</sub>). Long-term P fertilization increased soil available P extracted by P-CO<sub>2</sub> (+ 24%), P-AAE (+ 200%), and P-NaHCO<sub>3</sub> (+ 155%), shoot growth and grain yield by 8.4% and 26.2% for winter wheat and rapeseed respectively but had no effect on maize. The relationships between  $P_C$  and shoot biomass or N concentration were described respectively by allometric and linear models ( $R^2 > 0.85$ ,  $n = 21, 28$  and  $32$  for winter wheat, maize and rapeseed respectively; slope  $P$  values for linear models  $< 0.05$ ). The  $P_C$ –shoot N concentration model (slope: 0.083, intercept: 0.88) for winter wheat confirmed results from previous studies and can be used for calculating the P nutrition index. For the three soil available P indicators, threshold values needed to achieve 95% of the maximum yield for the three crops were less than those currently used in the official fertilization guidelines in Switzerland. Our results obtained after 44 years of contrasted P fertilization confirm the relationship between  $P_C$  and shoot N concentration for grain crops and the need to revise P fertilizer recommendations based on currently used soil P tests.

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## Introduction

Phosphorus (P) is an essential crop nutrient (Marschner 1995) and its application to agricultural soils is crucial to achieve optimum crop production. Phosphorus fertilizer applied in excess to crop requirements, however, is an important environmental concern because soil P accumulation increases the risk of P losses to surface and ground waters, with detrimental effects on aquatic ecosystems through eutrophication (Haygarth and Jarvis 1999; Carpenter 2005). In addition, reserves of economically exploitable phosphate rocks are decreasing and some scenarios, even if controversial, predict its depletion in the next 100 years (Gilbert 2009). Therefore, for economic and environmental reasons, appropriate diagnostic systems and management practices are required to improve soil and fertilizer P use efficiency by crops.

Fertilization recommendations are generally based on soil P analyses that approximate the amount of available P to plants using chemical extractions (Demaria et al. 2005). The degree of success of soil chemical extractions in assessing plant-available P and predicting the amount of P fertilizer required to reach optimal crop yields is often limited. The absence of a yield response to P fertilization in fields with an expected positive response based on soil P analyses was reported for perennial forage grasses (Bélanger and Ziadi 2008; Messiga et al. 2015), wheat (Valkama et al. 2011; Bélanger et al. 2015b), maize (Ziadi et al. 2014) and rapeseed (McKenzie et al. 2003; Grant et al. 2009; Bélanger et al. 2015a). Those studies, however, were conducted over only a few years and the effects of long-term P fertilization on soil available P and grain yield were not investigated.

Plant-based diagnostic methods can be used as an alternative to or alongside soil analyses to evaluate crop P requirements. The critical P concentration ( $P_C$ ), defined as the minimal plant P concentration for maximal yield, is necessary for determining the crop P status and managing P fertilization for optimal plant nutrition. Most plant nutrient concentrations, such as N, P and K, decrease with increasing shoot biomass

and plant development. Consequently, the P concentration in itself has no diagnostic value unless it is interpreted for a given level of shoot biomass. To account for this dilution effect, models of critical concentrations were established based on the relationship between the nutrient concentration and shoot dry matter (DM) yield under non-limiting conditions. This approach was first developed for N in grasslands (Salette and Lemaire 1981), and then tested for winter wheat (Greenwood et al. 1990; Justes et al. 1994), spring wheat (Ziadi et al. 2010), and maize (Ziadi et al. 2008b); it was then also used for P in various crops, including spring wheat (Bélanger et al. 2015a) and forage grasses (Bélanger and Ziadi 2008).

As a result of the dilution of both nutrients with increasing shoot biomass, P and N concentrations are positively related. This relationship has been used to develop models of  $P_C$  expressed as a function of N concentration for spring wheat (Ziadi et al. 2008a; Bélanger et al. 2015b), maize (Ziadi et al. 2007), rapeseed (Bélanger et al. 2015a), and forage grasses (Bélanger and Ziadi 2008; Bélanger et al. 2017). The  $P_C$  models can then be used to evaluate the plant P nutrition status via the P nutrition index (PNI), calculated as the ratio between measured P concentration and  $P_C$  (Duru and Thélier-Huché 1997). Values of PNI equal to or greater than 100% indicate that P is not limiting, while values smaller than 100% indicate P deficiency. The PNI is a plant-based diagnostic tool independent from soil types and climate conditions. Along with providing a measure of the level of P nutrition, it also provides information on soil P availability.

Obtaining P limiting conditions in field experiments is often difficult as soils are often rich in available P. For spring wheat and maize (Ziadi et al. 2007, 2008a), the models of  $P_C$  were first established from experiments with only one non-limiting P fertilization rate. In subsequent studies with contrasted P fertilization rates, there was no response of spring wheat to P fertilization on eight different sites (Bélanger et al. 2015b), while rapeseed responded to P fertilization at only one out of five sites (Bélanger et al. 2015a). Models of  $P_C$  for wheat, maize, and rapeseed, discussed above, need to be validated under conditions of P deficiencies.

This study focuses on three crops: winter wheat (*Triticum hybernum* L.), maize (*Zea mays* L.), and rapeseed (*Brassica napus* L.) grown in a long-term

field experiment with contrasted P mineral fertilization rates conducted in Changins (Switzerland). The objectives were to (1) evaluate the effect of long-term (> 40 years) P fertilization on the grain yield of winter wheat, maize, and rapeseed, (2) validate or establish models of  $P_C$  based on relationships of shoot P concentration with either shoot biomass DM yield or shoot N concentration under contrasted P conditions including P deficiency, and (3) assess both plant-based and soil-based diagnostic tools for determining the need for P fertilization. This unique context of large variations in soil P contents due to very long-term contrasted P fertilization will contribute to the development of better predictors of P requirements.

## Materials and methods

### Site description and experimental design

A field experiment was conducted at Agroscope in Changins (VD, Switzerland; 46°23'55.72''N, 06°14'24.72''E; altitude 432 m) on a Gleyic Cambisol (FAO classification system) with 525 g clay  $\text{kg}^{-1}$  and 163 g sand  $\text{kg}^{-1}$  in the top 0.20-m soil layer. A 4-year rotation of rapeseed, winter wheat, maize, and winter wheat was initiated in 1971. Winter wheat was replaced by sugar beet (*Beta vulgaris* L.) in 1982, and by spring wheat in 1983, 1993, and 2001. The soil was plowed to a depth of 0.20–0.25 m from 1971 to 1985 and only disked to 0.12–0.15 m with a harrow after 1985. Crop residues were left on the field after harvest.

Each year, five fertilization treatments were applied: (1) 0P0K: no P and K applied; (2) 0PK: no P applied and K applied in quantities equivalent to the theoretical crop uptake; (3) P0K: P applied in quantities equivalent to the theoretical crop uptake and no K applied; (4) PK: P and K applied in quantities equivalent to the theoretical crop uptake; (5) PK > exp.: P and K applied in quantities equivalent to double the theoretical nutrient uptake. The theoretical uptake is calculated from averaged local grain yield and nutrient concentration data for most Swiss crops. It is regularly updated and used as reference value in the Swiss official fertilization guidelines (Sinaj et al. 2017). The P and K fertilizers were respectively applied as triple superphosphate [ $\text{Ca}(\text{H}_2\text{PO}_4)_2$ ] and salt of potash (KCl) prior to plowing or disking for all

three crops. The five treatments were set in a randomized complete block design with four replications. Plot size was 15 × 8 m with a 1-m separation between plots. Nitrogen was applied as ammonium nitrate ( $\text{NH}_4\text{NO}_3$ ) at the same rate in all fertilization treatments according to the Swiss fertilization guidelines for each crop species (Sinaj et al. 2017). Herbicides were applied depending on weed infestation, and standard phytosanitary protection was applied according to integrated crop protection principles (Häni et al. 1990). Potassium in 0K treatments was considered non-limiting in this study, given the initially very high soil K content (data not shown).

The current study was undertaken in 2010. Winter wheat (cv. *Arina*) was sown (180 kg seeds  $\text{ha}^{-1}$ ) on 8 October 2010 and harvested on 12 July 2011, maize (cv. *Ricardinio*) was sown (94,000 seeds  $\text{ha}^{-1}$ ) on 15 May 2012 and harvested on 1 October 2012, while rapeseed (cv. *Visby*) was sown (500,000 seeds  $\text{ha}^{-1}$ ) on 30 August 2013 and harvested on 6 July 2014.

Mean annual temperature and precipitation were, respectively, 9.2 °C and 999 mm for the period 1961–1990, and 10.2 °C and 954 mm for the period 1981–2010. The mean annual precipitation was below the 30-year averages (810 mm) in 2011 but above in 2012 (1199 mm) and 2014 (1287 mm). All 3 years were warmer than the 30-year averages, with mean annual temperatures of 11.8, 11.1, and 11.8 °C in 2011, 2012, and 2014, respectively.

### Soil sampling and analysis

Soil samples (0–0.20 m and 0.20–0.50 m) were collected for general characteristics analysis in August 2014 after the rapeseed harvest, and in 2011, 2012 and 2014 for available P analysis. At least eight cores with a diameter of 2.5 cm were taken randomly from each plot. Plant residues were removed from the soil and individual core samples were mixed to form one composite soil sample per plot. Soil samples were air-dried and sieved ( $\leq 2$  mm) prior to analysis. Soil total P (Total P) was obtained by digestion of 0.25 g of soil previously treated in 5 ml of hydrofluoric acid (40%) and 1.5 ml of  $\text{HClO}_4$  (65%) according to the AFNOR standard X31-147 (1996) followed by molybdate colorimetric measurement (Murphy and Riley 1962). Soil available P was evaluated by three methods used in routine analyses. The first method operates at an acidic pH (4.6) in the presence of ammonium acetate

and EDTA as a complexing agent (FAL et al. 2004, P-AAE), the second operates at an alkaline pH (8.5) with sodium bicarbonate (Olsen et al. 1954, P-NaHCO<sub>3</sub>), while the third operates at soil pH and at a very low ionic strength (FAL et al. 2004, P-CO<sub>2</sub>). The P-AAE and P-CO<sub>2</sub> are the two methods used in routine soil available P tests in Switzerland (Sinaj et al. 2009), whereas the P-NaHCO<sub>3</sub> method is the most widely used indicator for soil available P worldwide.

#### Plant sampling and analysis

Shoot biomass was collected weekly on a 1-m<sup>2</sup> area in each plot using pruning shears during 7 weeks between tillering and joint stage (CD23–31 to CD45 in BBCH scale, Meier 2001) for winter wheat in 2011, during 6 weeks between leaf development and senescence (CD14–16 to CD99) for maize in 2012, and during 8 weeks between inflorescence emergence and ripening (CD53–55 to CD85) for rapeseed in 2014. At harvest, grain yield was measured in each plot on an area of 31.5 m<sup>2</sup> for maize and rapeseed, and 29.5 m<sup>2</sup> for winter wheat.

Shoot biomass samples were weighed before and after shredding and oven-drying at 55 °C for 72 h. Thereafter, the samples were ground in a Retsch rotor mill. Residual humidity was evaluated at 103 °C. Dry ashes and organic matter were evaluated by calcination (480 °C). Total N was measured after combustion, using the Dumas method (Masson et al. 2010). Total P was determined by radial ICP-AES (Varian Vista RL Simultaneous or Varian 725ES Simultaneous) after incineration (480 °C for 5 h) and solubilization in hydrofluoric acid (Masson et al. 2010). Crop grain quality was analyzed by Near Infrared Spectroscopy (NIRS) for the determinations of grain protein concentration for winter wheat, maize and rapeseed, and grain oil concentration for rapeseed.

#### Statistical analysis, calculations, and modeling

Data were checked for normality and variance homoscedasticity. Shoot biomass, shoot P and N concentrations, grain yield, and grain P, protein and oil concentration were subjected to an analysis of variance (ANOVA) with fertilization treatments as a fixed effect and replicates as a random effect. Mean differences between pairs of fertilization treatments were evaluated by a Tukey HSD test. All statistics and

model calculations were performed with R software package (R Development Core Team 2011).

Shoot growth was analyzed with a linear parallel curve analysis with grouped data, in which any difference in the response curves of shoot biomass to the number of days from the first day of sampling among fertilization treatments was determined. Using the FIT directive of GENSTAT (VSN International 2011), the response curves were described by the following models:

$$SB = a + b \times \text{Days} \quad (1)$$

where SB is the response variable, Days is the explanatory variable, and *a* and *b* are the estimated intercept and slope parameters. The procedure initially calculated one equation for the five fertilization treatments, which described the average response to the explanatory variable. In the next step, separate *a* parameters for SB were estimated for each fertilization treatment to determine the vertical distance between parallel lines (i.e., response curves). The following step estimated separate linear parameters *b* for the slope (i.e., the interaction between the linear portion of the fertilization treatment and the explanatory variable). At each stage, statistical significance was calculated for the change in the mean square explained by the addition of another parameter (fertilization treatments and fertilization treatments by the explanatory variable interaction) to the model. Slopes of the linear regressions between shoot biomass and the number of days are estimates of shoot growth rates. This approach was used to separate the effect of fertilization rates between early season shoot growth up to the first sampling day and rates of shoot growth later in the season (Bélanger et al. 2015b). Statistical significance was assessed at  $P \leq 0.05$ .

Then, the relationship between shoot P concentration and shoot biomass was described by a power function of allometric type:

$$Psc = c \times SB^{-d} \quad (2)$$

where Psc is the shoot P concentration (g kg<sup>-1</sup> DM), SB is the shoot biomass (t DM ha<sup>-1</sup>), *c* is the estimated scaling factor, and *d* is the estimated power parameter. The relationship between shoot P and N concentrations was described by a linear regression:

$$Psc = e \times Nsc + f \quad (3)$$

where  $N_{sc}$  is the shoot N concentration,  $e$  is the estimated slope and  $f$  is the estimated intercept. The P dilution in shoot biomass and the P linear relationship with N were calculated for all five treatments in all three crops. Then, the treatment with the lowest P input within the treatments that resulted in maximum grain yield (maize and rapeseed) or maximum shoot growth (winter wheat) was considered closest to critical P concentration and was chosen as reference. Those reference treatments corresponded to PK for winter wheat and rapeseed, and OPK for maize.

An allometric function was also fitted on N concentration data and compared to  $N_C$  established in a previous study to evaluate if N nutrition could be considered optimal. Sampling dates when shoot biomass was below  $1 \text{ t DM ha}^{-1}$  were excluded because, at these early stages, the relationship between P and N concentrations is not linear (Lemaire and Gastal 1997). Consequently, five sampling dates for winter wheat and maize, and seven sampling dates for rapeseed were used.

To determine which of the two relationships ( $P_C-SB$  or  $P_C-N_{sc}$ ) should be used to calculate the PNI, the linear regression between measured and simulated values was estimated for all experimental plots, similar to the approach used by Pineiro et al. (2008). The coefficient of determination ( $R^2$ ) and the root mean square deviation (RMSD) were used to determine the relationship with the best predicting power for calculating the PNI. The PNI for winter wheat and rapeseed was calculated as follows (Lemaire and Salette 1984):

$$\text{PNI}(\%) = (P_{\text{measured}}/P_C) \times 100 \quad (4)$$

where  $P_{\text{measured}}$  is the measured shoot P concentration ( $\text{g kg}^{-1} \text{ DM}$ ) and  $P_C$  is the critical P concentration ( $\text{g kg}^{-1} \text{ DM}$ ).

Finally, crop relative grain yields were calculated by dividing the grain yield from a given fertilization treatment by the grain yield from the treatment PK and expressed as percent. Maximal grain yield was considered to be achieved in the PK treatment for all three crops. The relationship between relative grain yield (RGY) and P-NaHCO<sub>3</sub>, P-AAE and P-CO<sub>2</sub> for winter wheat, maize and rapeseed was described by the following hyperbolic function that takes in account yield reduction at very high fertilization rate:

$$\text{RGY} = gP - \text{NaHCO}_3 / (P - \text{NaHCO}_3 + h) - iP - \text{NaHCO}_3 \quad (5)$$

where  $g$ ,  $h$  and  $i$  are estimated parameters. The intersection between 95% of calculated maximal grain yield and the function was used to determine critical threshold values for P-NaHCO<sub>3</sub>, P-AAE and P-CO<sub>2</sub>.

The relationship between RGY and PNI was described by the following quadratic function:

$$\text{RGY} = j\text{PNI}^2 + k\text{PNI} + l \quad (6)$$

where  $j$ ,  $k$  and  $l$  are estimated parameters. The intersection between 95% of maximal grain yield and the function was used to determine a critical threshold value for PNI.

## Results and discussion

### Soil phosphorus and other soil characteristics after 44 years of fertilization treatments

Phosphorus fertilization significantly affected soil total P concentration and the three indicators of soil available P in the topsoil (0–0.20 m) after 44 years of cultivation and fertilization treatments (Table 1). Soil total P concentration ranged from 0.72 to  $1.14 \text{ g P kg}^{-1}$ , while soil available P concentration ranged from 0.62 to  $7.54 \text{ mg P kg}^{-1}$  for the P-CO<sub>2</sub> extraction, from 7.1 to  $72.5 \text{ mg P kg}^{-1}$  for the P-AAE extraction, and from 9.7 to  $60.7 \text{ mg P kg}^{-1}$  for the P-NaHCO<sub>3</sub> extraction. Soil total P and soil available P, evaluated by the three extraction methods, followed similar response patterns to the fertilization treatments (Table 1). Soil pH and CEC as well as soil organic C and total N concentrations were not significantly affected by the fertilization treatments (Table 1). For the subsoil layer (0.20–0.50 m), P fertilization did not significantly affect concentrations of total P (mean value of  $0.40 \text{ g kg}^{-1}$ ), P-CO<sub>2</sub> ( $0.09 \text{ mg kg}^{-1}$ ), organic C ( $11.4 \text{ g kg}^{-1}$ ), and pH (7.10). However, the PK > exp treatment with annual applications of  $52.4 \text{ kg P ha}^{-1}$  significantly differed from all other fertilization treatments for soil available P extracted by NaHCO<sub>3</sub> ( $7.52$  vs.  $3.54 \text{ mg kg}^{-1}$  across all other treatments) and AAE ( $2.96$  vs.  $1.41 \text{ mg kg}^{-1}$ ), indicating a downward movement to the solid phase of the

**Table 1** Selected soil characteristics, soil total and available P in 2014 in the topsoil (0–0.20 m) and the subsoil (0.20–0.50 m), and cumulative P budget (BcumP)

Treatments	Depth (m)	pH <sup>a</sup> H <sub>2</sub> O	CEC <sup>ca</sup> (cmol kg <sup>-1</sup> )	Organic C <sup>a</sup> (g kg <sup>-1</sup> )	Total N <sup>a</sup> (g kg <sup>-1</sup> )	Total K (g kg <sup>-1</sup> )	Total P (g kg <sup>-1</sup> )	P-CO <sub>2</sub> (mg kg <sup>-1</sup> )	P-AAE (mg kg <sup>-1</sup> )	P-NaHCO <sub>3</sub> (mg kg <sup>-1</sup> )	BcumP (kg P ha <sup>-1</sup> )
OP0K <sup>b</sup>	0–0.20	6.55a	27.9a	29.2a	2.90a	19.7ab	0.72a	0.62a	7.1a	9.7a	- 567
OPK		6.60a	26.9a	28.2a	2.98a	19.3a	0.72a	0.72a	8.7a	10.5a	- 209
P0K		6.58a	28.9a	29.7a	3.07a	19.3a	0.91b	2.37b	27.9b	24.4b	+ 543
PK		6.63a	28.1a	29.9a	3.03a	19.5ab	0.89b	1.98b	26.1b	26.8b	+ 667
PK > exp.		6.70a	27.3a	28.5a	3.08a	21.4b	1.14c	7.54c	72.5c	60.7c	+ 1554
P value		0.82	0.71	0.77	0.87	< 0.001	< 0.001		< 0.001	< 0.001	
OP0K <sup>b</sup>	0.20–0.50	7.12b		12.1b	1.32b	17.8a	0.40d	0.09d	1.32d	3.49d	
OPK		7.20b		10.9b	1.15b	17.6a	0.37d	0.09d	1.20d	2.91d	
P0K		7.10b		11.2b	1.22b	18.4a	0.42d	0.09d	1.56d	4.14d	
PK		7.05b		11.3b	1.17b	17.8a	0.39d	0.08d	1.55d	3.60d	
PK > exp.		7.00b		11.6b	1.14b	17.5a	0.41d	0.10d	2.96e	7.52e	
P value		0.77		0.73	0.49		0.70	0.69	< 0.01	< 0.01	

Results are the mean of four replicates. Means with different letters are significantly different at  $P < 0.05$ . Reported  $P$  values are from ANOVA

<sup>a</sup>pH, CEC (cation exchange capacity), organic C, and total N were determined according to FAL et al. (2004)

<sup>b</sup>OP0K: no P and K applied; OPK: no P applied, K applied in quantities equivalent to the crop uptake; P0K: P applied in quantities equivalent to the crop uptake and no K applied; PK: P and K applied in quantities equivalent to the crop uptake; PK > exp.: P and K applied in quantities equivalent to the crop uptake with an additional fertilization of 26.2 kg P ha<sup>-1</sup> and 166 kg K ha<sup>-1</sup>

subsoil. The cumulative P budget over the 44 years of the experiment was positive with the treatments having received P fertilization equivalent to theoretical crop P uptake (PK and POK) (Table 1). The P input was therefore greater than P exported in the grain crops. Potassium fertilization did not affect any of the soil characteristics, except for a slight increase in soil available K extracted by AAE and CO<sub>2</sub> (data not shown), and total K (Table 1) in the 0–0.20 m soil layer with the PK > exp treatment.

### Crop growth, and grain yield and quality

The shoot biomass of winter wheat in 2011 was significantly affected by the P fertilization treatments on all seven sampling weeks (Table 2) with generally greater shoot biomass when P was applied (POK, PK, and PK > exp) than with no applied P (OPOK and OPK). The fertilization treatments significantly affected the shoot biomass of maize in 2012 only in the first two sampling weeks (stage CD14–16; Meier

2001) and the shoot biomass of rapeseed in 2014 only in the fifth and sixth sampling weeks (CD53–63). Shoot growth was analyzed with a linear parallel curve analysis that indicated different crop responses to the P fertilization treatments. For rapeseed, the additional mean square contributed by Treatments was significant, while that contributed by Days × Treatments was not significant (Table 3). Hence, the regression lines of shoot biomass as a function of time for the different P rates were parallel with similar slopes and growth rates. Most of the effect of the P fertilization treatments on the shoot growth of rapeseed therefore occurred before the first sampling week (Table 3). Similar results were reported from a study with rapeseed conducted at two sites in Canada (Bélanger et al. 2015a). The results presented here and those of Bélanger et al. (2015a) confirm that an adequate P supply during early rapeseed growth is important (Grant et al. 2001, 2009). For winter wheat, the additional mean square contributed by both Treatments and Days × Treatments was significant. Hence,

**Table 2** Plant shoot biomass for winter wheat in 2011, maize in 2012, and rapeseed in 2014 as affected by fertilization treatments during a 6- to 8-week sampling period

Crops	Treatments	Sampling week							
		1 (t ha <sup>-1</sup> )	2 (t ha <sup>-1</sup> )	3 (t ha <sup>-1</sup> )	4 (t ha <sup>-1</sup> )	5 (t ha <sup>-1</sup> )	6 (t ha <sup>-1</sup> )	7 (t ha <sup>-1</sup> )	8 (t ha <sup>-1</sup> )
Winter wheat 2011	OPOK	0.26a	0.50a	1.08a	1.77a	2.67a	3.38a	4.36a	
	OPK	0.25a	0.56a	1.16a	2.04ab	2.92ab	4.09ab	4.96a	
	POK	0.48b	0.91b	1.66b	2.76c	3.81c	5.08bc	6.24b	
	PK	0.45b	0.9b	1.71b	2.60bc	3.65bc	5.11bc	6.37b	
	PK > exp	0.54b	1.00b	1.83b	2.94c	4.07c	5.81c	6.98b	
	<i>P</i> value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001
Maize 2012	OPOK	0.29a	0.70a	2.14a	3.44a	4.97a	6.75a		
	OPK	0.29a	0.83ab	2.04a	3.11a	4.26a	6.25a		
	POK	0.45b	1.17b	2.45a	4.30a	4.79a	6.61a		
	PK	0.40b	1.23b	2.61a	3.98a	5.37a	7.07a		
	PK > exp	0.43b	1.14b	2.39a	3.76a	5.64a	7.03a		
	<i>P</i> value	< 0.05	< 0.01	0.48	0.41	0.26	0.89		
Rapeseed 2014	OPOK	1.31a	1.80a	2.25a	3.01a	3.30a	4.24ab	5.84a	6.14a
	OPK	1.13a	1.64a	1.98a	2.93a	3.16a	4.21a	5.62a	6.82a
	POK	1.31a	1.43a	2.45a	3.37a	3.92ab	4.86ab	6.81a	7.90a
	PK	1.35a	1.75a	2.37a	3.15a	3.61ab	4.78ab	6.55a	6.93a
	PK > exp	1.40a	2.07a	2.44a	3.60a	4.61b	5.72b	6.74a	8.61a
	<i>P</i> value	0.60	0.50	0.44	0.54	< 0.05	< 0.05	0.49	0.86

Results are the mean of four replicates. Means with different letters are significantly different at  $P < 0.05$ . Reported  $P$  value are from ANOVA

**Table 3** Linear parallel curve analysis ( $P$  values)<sup>a</sup> for shoot biomass (SB) as a function of the number of days from the first sampling day for different fertilization treatments

Estimated parameters	<i>df</i>	Wheat 2011	Maize 2012	Rapeseed 2014
Days	1	< 0.001	< 0.001	< 0.001
Treatments	4 (5)	< 0.001	0.55	< 0.001
Days × Treatments	4 (5)	< 0.001	0.48	0.058
$R^2$		0.96	0.86	0.85

<sup>a</sup>Statistical significance of the additional mean square contributed to the model  $SB = a + b \times \text{Days}$  as individual parameters  $a$  and  $b$  for each treatment are estimated in steps

both shoot growth before the first sampling week and shoot growth during the seven weeks of sampling were affected by the P fertilization treatments. Other studies also showed that P nutrition is important throughout the growing season of winter wheat (Miller et al. 1994). For maize, the additional mean square contributed by both Treatments and Days × Treatments was not significant.

Grain yields with the PK treatment were similar to or slightly greater than the average yield since 1995 for winter wheat (5.54 vs. 4.68 t ha<sup>-1</sup>) and maize (8.37 vs. 8.03 t ha<sup>-1</sup>). The rapeseed grain yield in 2014, however, was lower than the average yield since 1995 (2.60 vs. 3.11 t ha<sup>-1</sup>), perhaps due to late planting or seasonally high precipitation in the winter and spring of 2014 (288 mm between January and March) that prevented an herbicide application, causing weed competition. It might also have caused nutrient leaching, creating limiting N conditions.

The fertilization treatments significantly affected the grain yield of winter wheat and rapeseed, but they did not affect the grain yield of maize (Table 4). For winter wheat, the OP0K treatment resulted in significantly lower grain yields than the treatments with the recommended P rate (PK and P0K). This result contrasts with several short-term studies reporting no wheat grain yield response to P fertilization (McKenzie et al. 2003, 2008; Valkama et al. 2009, 2011; Grant et al. 2009). More recently, in an eight site-year study conducted with contrasting P rates, Bélanger et al. (2015b) reported no spring wheat grain yield response to increasing P fertilization. The rapeseed grain yield for the treatments with no P (OP0K and OPK) was less than that with applied P (P0K and PK). Positive grain yield responses to P fertilization were reported by Brennan and Bolland (2007) in 21 out of 22 experimental sites and by Bélanger et al. (2015b) in two out

of five experimental sites, thus demonstrating the sensitivity of rapeseed grain yield to P deficiency. The absence of a grain yield response of maize after 44 years of P fertilization is probably due to its low crop P requirements or its deep rooting and P uptake from deeper layers (Boniface and Trocmé 1988; Gill et al. 2005). Numerous long-term experiments have reported no or little response of maize to P fertilization (Gallet et al. 2003; Magyar et al. 2006; Messiga et al. 2010).

Although not statistically significant, the treatment with the highest P rate (PK > exp) resulted in slightly but systematically lower grain yield compared to the treatments with the recommended P rate (PK, P0K) for all three crops (Table 4). This trend may be partly explained by antagonism or competition among nutrients. High soil P has been reported to decrease zinc (Zn) uptake in several crops including maize (Marschner 1995; Izsaki 2014). A Zn deficiency was observed in maize in 2012 at the beginning of the leaf development stage (CD11). Even though Zn was applied [foliar application (700 g ha<sup>-1</sup>)] in the form of zinc oxide (ZnO) at stage CD16 (6 leaves) to limit yield variation due to Zn deficiency, plants at harvest still had lower Zn concentrations for the PK > exp treatment (data not shown) than for the treatments with the recommended P rate.

The grain P concentration of all three crops was significantly positively affected by the fertilization treatments (Table 4). Plants from over-fertilized treatments have achieved a “luxury consumption”, which is defined by the fact that not all P taken up by the plants was used for biomass production. The grain protein concentration of winter wheat and rapeseed was significantly negatively affected by fertilization treatments, while that of maize was not affected. The grain protein concentration tended to be lower with



**Table 4** Grain yield and concentrations of P, protein, and oil of winter wheat in 2011, maize in 2012, and rapeseed in 2014 as affected by fertilization treatments

Crops	Treatments	Grain yield (t ha <sup>-1</sup> )	P (g kg <sup>-1</sup> DM)	Protein (g kg <sup>-1</sup> DM)	Oil (g kg <sup>-1</sup> DM)
Winter wheat 2011	OP0K <sup>a</sup>	4.67a	3.84a	146.4a	
	OPK	5.11ab	4.15ab	142.6ab	
	P0K	5.66b	4.57b	138.2ab	
	PK	5.54b	4.34b	136.0b	
	PK > exp	5.34ab	4.53b	135.0b	
	<i>P</i> value	< 0.05	< 0.01	< 0.05	
Maize 2012	OP0K	7.67a	1.89a	86.3a	
	OPK	7.59a	1.97ab	84.0a	
	P0K	7.51a	2.27ab	85.4a	
	PK	8.37a	2.04ab	80.4a	
	PK > exp	6.90a	2.35b	84.8a	
	<i>P</i> value	0.53	< 0.05	0.71	
Rapeseed 2014	OP0K	2.06a	5.38a	164.2a	522.4a
	OPK	2.06a	5.35a	162.4a	522.3a
	P0K	2.63b	6.23b	158.3ab	526.7a
	PK	2.60b	6.33b	156.5ab	526.7a
	PK > exp	2.44ab	6.52b	151.8b	527.5a
	<i>P</i> value	< 0.001	< 0.001	< 0.01	0.57

Results are the mean of four replicates. Means with different letters and a crop are significantly different at  $P < 0.05$ . Reported  $P$  value are from ANOVA

increasing applied P most likely because of N dilution in the increasing grain biomass (Russell et al. 1958; Daccord et al. 2002). Although the fertilization treatments affected the grain protein concentration of winter wheat and rapeseed (Table 4), the total protein produced per hectare (data not shown) was not affected. These results are similar to those of Holford et al. (1992) who reported a decrease of protein concentration with repeated P applications. The oil concentration of rapeseed ranged from 522 to 528 g kg<sup>-1</sup> DM and was not significantly affected by P fertilization, which is in agreement with the results of Brennan and Bolland (2007).

Critical P concentration defined as a function of shoot biomass

Shoot P concentration decreased with increasing shoot biomass for the three crops and fertilization treatments (Fig. 1). Models of shoot  $P_C$  were determined using the data of shoot biomass and P concentration from treatments resulting in maximum shoot biomass growth with the lowest P fertilization rate: PK in

wheat, OPK in maize, and PK in rapeseed. The relationship between  $P_C$  and shoot biomass (SB) for the three crops were closely described by allometric functions:

$$P_{C_{\text{wheat}}} = 4.44 \times \text{SB}^{-0.41}, R^2 = 0.95, \quad (7)$$

$n = 21$  for winter wheat

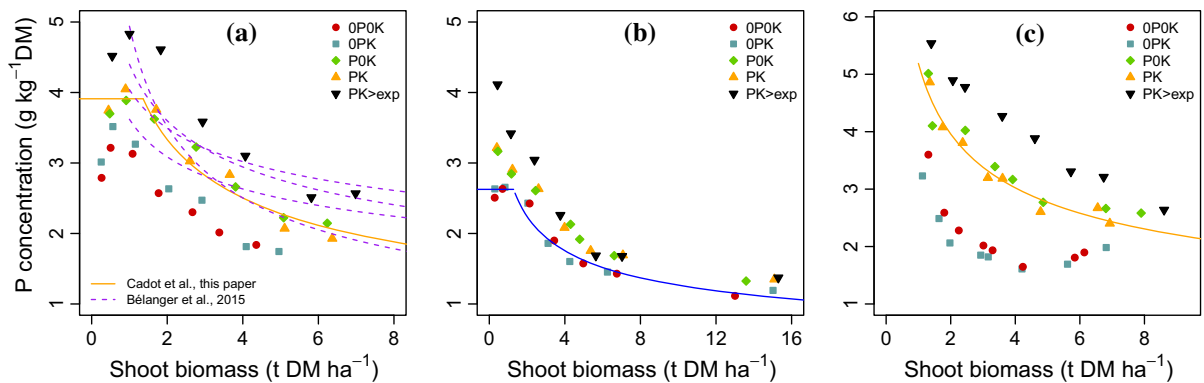
$$P_{C_{\text{maize}}} = 3.49 \times \text{SB}^{-0.19}, R^2 = 0.99, \quad (8)$$

$n = 28$  for maize

$$P_{C_{\text{rapeseed}}} = 5.18 \times \text{SB}^{-0.39}, R^2 = 0.89, \quad (9)$$

$n = 32$  for rapeseed

The model obtained in this study for winter wheat predicted  $P_C$  values close to the lowest values predicted by Bélanger et al. (2015b) (Fig. 1). Luxury consumption at some of the locations studied by Bélanger et al. (2015b) might be responsible for the variations observed in their study. The relationship between  $P_C$  and shoot biomass has never been tested in maize and rapeseed. Current results therefore provide a first estimation of their  $P_C$  as a function of shoot



**Fig. 1** Shoot P concentration as a function of shoot biomass (SB) for five fertilization treatments along with the estimated model of  $P_C$  for **a** winter wheat ( $P_{C_{\text{ww}}} = 4.44 \times SB^{-0.41}$ ,  $R^2 = 0.95$ ), **b** maize ( $P_{C_{\text{maize}}} = 3.49 \times SB^{-0.19}$ ,  $R^2 = 0.99$ ), and **c** rapeseed ( $P_{C_{\text{rapeseed}}} = 5.18 \times SB^{-0.39}$ ,  $R^2 = 0.89$ ) under non-limiting P conditions. For wheat, critical P concentrations expressed as a function of shoot biomass are presented for four

locations from Bélanger et al. (2015a, b). OPOK: no P and K applied; OPK: no P applied, K applied in quantities equivalent to crop uptake; POK: P applied in quantities equivalent to crop uptake and no K applied; PK: P and K applied in quantities equivalent to crop uptake; PK > exp: P and K applied in quantities equivalent to crop uptake with additional fertilization of 26.2 kg P ha<sup>-1</sup> and 166 kg K ha<sup>-1</sup>

biomass. Because of the lack of a grain yield response to P fertilization,  $P_C$  of maize may be over-estimated.

For shoot biomass less than 1 t DM ha<sup>-1</sup>, a constant  $P_C$  for winter wheat (3.9 g kg<sup>-1</sup> DM) and maize (2.6 g kg<sup>-1</sup> DM) was assumed and calculated as the mean P concentration from the reference treatments, PK for winter wheat and OPK for maize, as did Lemaire and Gastal (1997). To have a more precise biomass threshold value where the P dilution phase starts, we calculated the intersection between these constant initial values of  $P_C$  and the modeled dilution curve of  $P_C$ , as suggested by Justes et al. (1994) for N dilution in winter wheat. This analysis indicates that P dilution starts approximately at 1.36 t DM ha<sup>-1</sup> for winter wheat and 1.34 t DM ha<sup>-1</sup> for maize.

Although a strong relationship between  $P_C$  and shoot biomass was found, its use for assessing the crop P status might be hampered by other factors. For example, Bélanger et al. (2015b) concluded that the allometric relationship between  $P_C$  and shoot biomass in wheat differed among locations and, consequently, the wide use of the  $P_C$  model expressed as a function of shoot biomass is limited. Furthermore, previous research on perennial grasses (Bélanger and Ziadi 2008) has shown that the relationship between  $P_C$  and shoot biomass varied with the level of N nutrition. More research is needed to determine this variation of the  $P_C$ –SB relationship with a range of N nutrition in maize, winter wheat, and rapeseed.

Critical P concentration defined as a function of shoot N concentration

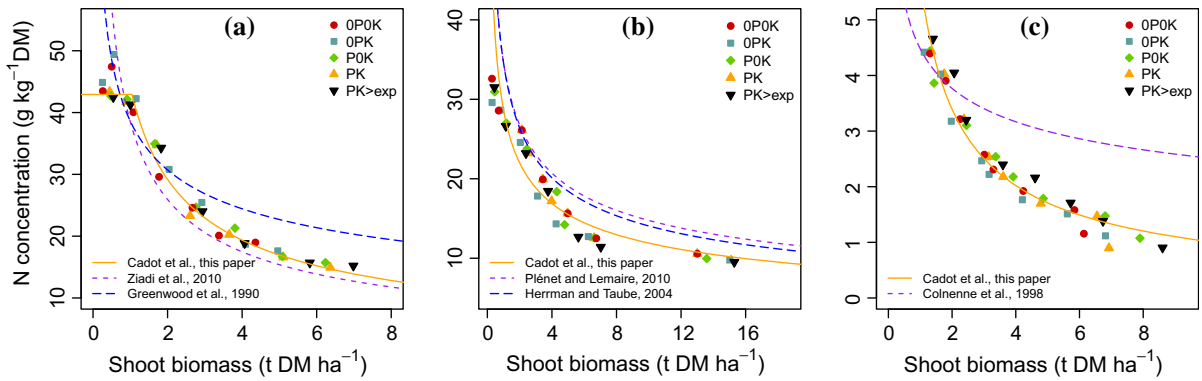
Because the relationship between critical P concentration and N concentration is affected by the level of N nutrition (Ziadi et al. 2007, 2008a), we first evaluated the level of N nutrition of the reference treatments (PK for winter wheat and rapeseed, and OPK for maize) by fitting allometric functions to our data, and comparing them with curves of shoot critical N concentration ( $N_C$ ) previously published on wheat by Ziadi et al. (2010;  $N_C = 38.5 \times SB^{-0.57}$ ), Greenwood et al. (1987;  $N_C = 38.6 \times SB^{-0.44}$ ) and Justes et al. (1994;  $N_C = 53.5 \times SB^{-0.44}$ ), on maize by Plénet and Lemaire (2000;  $N_C = 34.0 \times SB^{-0.37}$ ) and Herrman and Taube (2004;  $N_C = 34.0 \times SB^{-0.39}$ ), and on rapeseed by Colnenne et al. (1998;  $N_C = 44.8 \times SB^{-0.25}$ ) (Fig. 2). Our allometric functions were as follows:

$$N_{\text{ww}} = 44.0 \times SB^{-0.59}, R^2 = 0.93, n = 21 \quad (10)$$

$$N_{\text{maize}} = 30.6 \times SB^{-0.46}, R^2 = 0.84, n = 28 \quad (11)$$

$$N_{\text{rapeseed}} = 57.0 \times SB^{-0.75}, R^2 = 0.89, n = 32 \quad (12)$$

where  $N_{\text{ww}}$  and  $N_{\text{rapeseed}}$  are respectively the N concentrations of winter wheat and rapeseed for the PK treatment, and  $N_{\text{maize}}$  is the N concentration of maize for the OPK treatment. Values of  $N_{\text{ww}}$  in this experiment were comparable to  $N_C$  predicted by Ziadi



**Fig. 2** Shoot N concentration as a function of shoot biomass (SB) for **a** winter wheat ( $N_{ww} = 44.0 \times SB^{-0.59}$ ,  $R^2 = 0.93$ ), **b** maize ( $N_{maize} = 30.6 \times SB^{-0.46}$ ,  $R^2 = 0.84$ ) and **c** rapeseed ( $N_{rapeseed} = 57.0 \times SB^{-0.75}$ ,  $R^2 = 0.89$ ) for five fertilization treatments, as compared to critical N concentrations determined by Greenwood et al. (1990) and Ziadi et al. (2010) for winter wheat, Herrman and Taube (2004) and Plénet and Lemaire (2000)

for maize, and Colnenne et al. (1998) for rapeseed. OP0K: no P and K applied; OPK: no P applied, K applied in quantities equivalent to crop uptake; P0K: P applied in quantities equivalent to crop uptake and no K applied; PK: P and K applied in quantities equivalent to crop uptake; PK > exp: P and K applied in quantities equivalent to crop uptake with additional fertilization of 26.2 kg P ha<sup>-1</sup> and 166 kg K ha<sup>-1</sup>

et al. (2010), while values of  $N_{maize}$  were only slightly lower than  $N_C$  predicted by Plénet and Lemaire (2000) and Herrman and Taube (2004). The N nutrition of winter wheat and maize was thus considered optimum or near optimum, while the N nutrition of rapeseed was below the optimal N nutrition proposed by Colnenne et al. (1998).

Shoot P concentration increased with increasing shoot N concentration for all three crops and fertilization treatments (Fig. 3). As for the relationship between  $P_C$  and shoot biomass, models of  $P_C$  as a function of shoot N concentration were determined with data of shoot P and N concentrations with maximum shoot growth and lowest P fertilizer application rate: PK in wheat, OPK in maize, and PK in rapeseed. The  $P_C$  models were:

$$P_{Cww} = 0.083N + 0.88, R^2 = 0.85, n = 21 \text{ for wheat} \tag{13}$$

$$P_{Cww} = 0.291N - 1.557 - 0.004N^2, R^2 = 0.98, n = 21 \text{ for wheat} \tag{14}$$

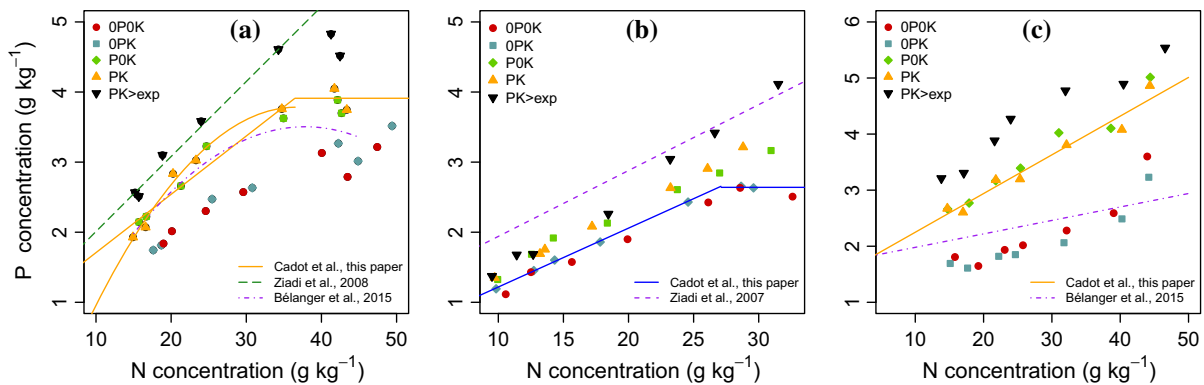
$$P_{Cmaize} = 0.083N + 0.39, R^2 = 0.94, n = 28 \text{ for maize} \tag{15}$$

$$P_{Crapeseed} = 0.657N + 1.67, R^2 = 0.85, n = 32 \text{ for rapeseed} \tag{16}$$

Ziadi et al. (2008a) also determined a linear model of  $P_C$  as a function of shoot N concentration ( $P_C = 0.18 N + 0.94$ ) for spring wheat. Their initial model probably over-estimated  $P_C$  because of plant luxury consumption, as no limiting P situations were identified in their experiment. Subsequently, Bélanger et al. (2015b) proposed a quadratic relationship between  $P_C$  and shoot N concentration ( $P_{C-} = 0.221 N - 0.677 - 0.003 N^2$ ,  $R^2 = 0.82$ ,  $P < 0.001$ ) in an experiment with several P rates over several locations. Even though grain yield in the latter study barely responded to P fertilization, results from our study were similar to those reported by Bélanger et al. (2015b).

The present model for maize predicts lower  $P_C$  values than those predicted by the model ( $P_{C-} = 0.094 N + 1.0$ ) proposed by Ziadi et al. (2007). Luxury P consumption probably also occurred in their study. This implies that the  $P_C$  model for maize developed in this study should be validated with a dataset of crops responding to P fertilization in order to ensure no bias due to luxury consumption.

The model obtained in this study for rapeseed predicted greater  $P_C$  values than those predicted by the model ( $P_C = 0.024 N + 1.74$ ) proposed by Bélanger et al. (2015a). Differences between the  $P_C$  models can be explained by different cultivars used or by the larger than average precipitation observed in our study which may have caused N leaching and reduced crop



**Fig. 3** Shoot P concentration as a function of shoot N concentration for five fertilization treatments along with the critical P concentrations expressed as a function of shoot N concentration for **a** winter wheat with a linear ( $P_{Cw} = 0.083 N + 0.88$ ,  $R^2 = 0.85$ ) and a quadratic model ( $P_{Cw} = 0.291 N - 1.557 - 0.004 N^2$ ,  $R^2 = 0.98$ ), **b** maize ( $P_{Cmaize} = 0.083 N + 0.39$ ,  $R^2 = 0.92$ ) and **c** rapeseed ( $P_{Crapeseed} = 0.657 N + 1.67$ ,  $R^2 = 0.85$ ). Previously published models for wheat [Ziadi et al. (2008a),  $P_{Cw} = 0.107 N + 0.94$ ; Bélanger et al. (2015a, b),  $P_{Cw} = 0.221 N$

$- 0.677 - 0.00292 N^2$ ], maize [(Ziadi et al. (2007),  $P_{Cmaize} = 0.094 N + 1.0$ )], and rapeseed [Bélanger et al. (2015a, b),  $P_{Crapeseed} = 0.094 N + 1.0$ ]] are also presented. OPOK: no P and K applied; OPK: no P applied, K applied in quantities equivalent to crop uptake; POK: P applied in quantities equivalent to crop uptake and no K applied; PK: P and K applied in quantities equivalent to crop uptake; PK > exp: P and K applied in quantities equivalent to crop uptake with additional fertilization of 26.2 kg P ha<sup>-1</sup> and 166 kg K ha<sup>-1</sup>

growth, in addition to increased weed competition for N because of the absence of an herbicide treatment.

Results presented here from three crops grown under conditions of contrasted soil available P achieved after more than 40 years of fertilization treatments confirm the validity of the models used in previous studies to describe the relationships between shoot P concentration and either shoot biomass or shoot N concentrations. Furthermore, the model of  $P_C$  for wheat based on the  $P_C$ –shoot N concentration relationship was in agreement with the model developed by Bélanger et al. (2015b). For maize and rapeseed, however, more data are required to validate the respective  $P_C$  models.

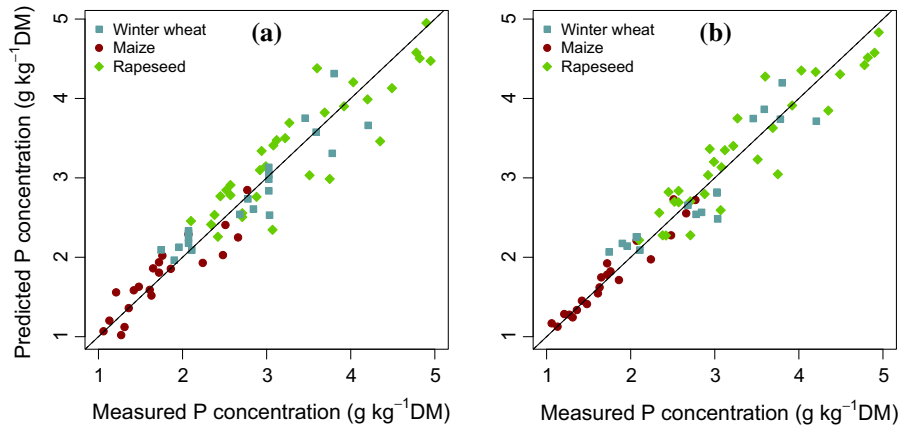
#### Agronomic use of $P_C$ and PNI

Two relationships based on the concomitant decrease of P and N concentrations with increasing shoot biomass were considered in our assessment of critical P concentration models for three important crops. Previous studies have shown that the  $P_C$ –shoot N concentration model was more universal than the  $P_C$ –SB model because the same model parameters could be used for several locations (Bélanger et al. 2015b; wheat) or for several levels of N nutrition (Bélanger and Ziadi 2008; perennial grasses). Furthermore, the

$P_C$ –shoot N concentration model does not require the determination of the shoot biomass, which makes it more practical for use in farmers' fields. The two models were also analyzed by comparing measured and predicted  $P_C$  values (Fig. 4). The  $P_C$ –shoot N concentration model appears, for all three crops, to predict  $P_C$  values with less dispersion around the 1:1 line ( $R^2 = 0.93$ ) as opposed to the  $P_C$ –SB model ( $R^2 = 0.90$ ). For all those reasons, the  $P_C$ –shoot N concentration model was chosen for calculating the PNI for winter wheat and rapeseed. Maize did not respond significantly to P fertilization and, therefore, the proposed model possibly overestimates  $P_C$  and potentially underestimates PNI.

The PNI of winter wheat ranged from 72 to 132%, while that of rapeseed ranged from 60 to 132% (Table 5). The relative grain yield increased with increasing PNI up to about 100% and then decreased, likely because of antagonism with zinc (Fig. 5). According to this relationship, 95% of maximum grain yield would be achieved with PNI of 82% for winter wheat and 81% for rapeseed. Because P fertilization cannot alleviate an early-season P deficiency (Barry and Miller 1989), producers would most likely use the PNI values to adjust fertilization for the following growing season. As suggested by Bélanger et al. (2015a), the PNI values could also be used for an

**Fig. 4** Predicted and measured  $P_C$  concentrations for maize, rapeseed, and winter wheat with values predicted by **a** the  $P_C$ -SB model ( $R^2 = 0.90$ ,  $n = 81$ ) and **b** the  $P_C$ -Nsc model ( $R^2 = 0.93$ ,  $n = 81$ ). The solid line represents the 1:1 line ( $y = x$ )



**Table 5** Phosphorus nutrition index (%) for winter wheat in 2011 and rapeseed in 2014 during a 7- to 8-week sampling period

Crop	Treatment	Sampling week					
		3	4 <sup>a</sup>	5	6 <sup>a</sup>	7	8
Winter wheat 2011	OP0K	85a	72	72a	75	74a	
	OPK	91a	72	76ab	73	75a	
	P0K	96ab	100	94bc	101	105b	
	PK	100b	98	105 cd	95	101ab	
	PK > exp	123c	114	124d	123	132c	
	<i>P</i> value	< 0.001		< 0.001		< 0.001	
Rapeseed 2014	OP0K	60a	60a	61a	56a	67a	78a
	OPK	55a	56a	58a	57a	63a	82a
	P0K	109b	101b	102b	97b	101b	109b
	PK	101b	96b	103b	93b	101b	106b
	PK > exp	127b	132c	126c	118c	124c	116b
	<i>P</i> value	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001

Means with different letters and a crop are significantly different at  $P < 0.05$ . Reported  $P$  value are from ANOVA

<sup>a</sup>Shoot P concentration of winter wheat was only analyzed for one replication in weeks 4 and 6. All other values are the mean of four replicates

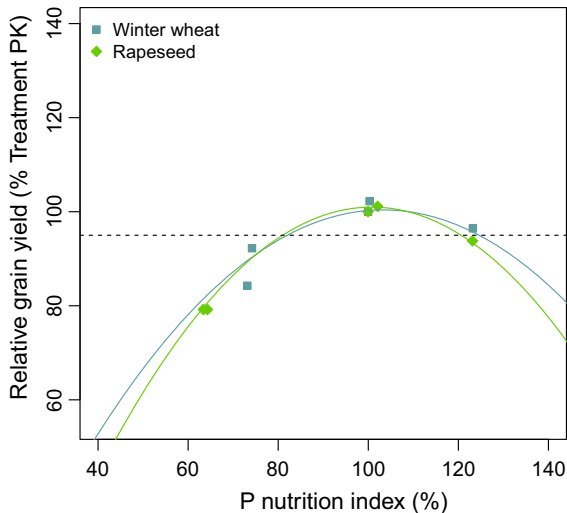
a posteriori diagnosis aimed at detecting limiting factors for crops in experimental trials or production fields.

Soil analysis

The relationship between relative grain yield and  $P\text{-NaHCO}_3$ , the most widely used method for estimating soil available-P, and  $P\text{-AAE}$  and  $P\text{-CO}_2$ , the official methods used in Switzerland in routine soil

available-P tests, was described by a hyperbolic function for each crop (Fig. 6). This function differs from usual relationships that describe a relatively long plateau phase such as in the Mitscherlich model (Valkama et al. 2011). A deficiency in other nutrients such as Zn (Marschner 1995) due to an antagonism with P from over-fertilization may explain the decline in relative yield with high soil available  $P$  values. Minimum threshold values indicating optimal plant nutrition were calculated as the intersection between

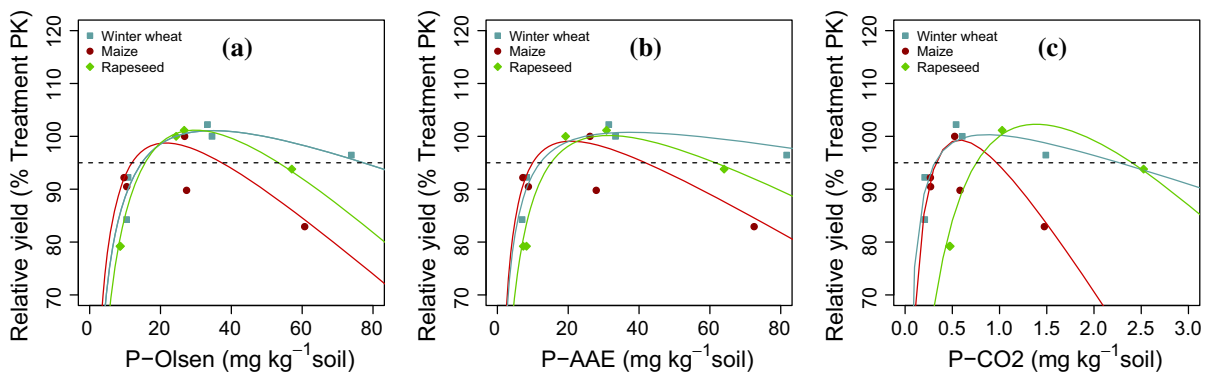
95% maximum yield and the hyperbolic curve fitting the relationship between crop yield and soil available P. Threshold values for winter wheat, maize, and



**Fig. 5** Relationship between relative grain yield (RGY) and the phosphorus nutrition index (PNI) (mean for all sampling dates) for winter wheat [ $RGY = -0.012PNI^2 + 2.46PNI - 26.4$ , Residual Standard Error (RSE) = 7.88,  $n = 5$ ] and rapeseed [ $RGY = -0.015PNI^2 + 3.08PNI - 54.1$ , RSE = 7.37,  $n = 5$ ]. Relative grain yields of winter wheat and rapeseed were calculated by dividing the grain yield from a given fertilization treatment by the grain yield from the PK treatment. The dashed horizontal line represents a relative grain yield of 95%

rapeseed were respectively 0.33, 0.31 and 0.70 mg P kg<sup>-1</sup> soil for P-CO<sub>2</sub>, 14.7, 11.9 and 15.6 mg P kg<sup>-1</sup> soil for P-NaHCO<sub>3</sub>, and 12.8, 10.0 and 15.2 mg P kg<sup>-1</sup> soil for P-AAE. These results are in accordance with that of Messiga et al. (2010) who found a critical value for maize of 8 mg P-NaHCO<sub>3</sub> on a slightly alkaline and sandy loamy soil. For winter wheat, Bollons and Barraclough (1999) reported a threshold of 9 mg P-NaHCO<sub>3</sub> kg<sup>-1</sup> soil on a silty-clay loam soil, whereas Morel et al. (1992), analyzing several French experimental sites, found a critical value that ranged from 6 to 12 mg P-NaHCO<sub>3</sub> kg<sup>-1</sup> soil depending on the soil type. Johnston et al. (1986) found a critical value of 11 and 18 mg P-NaHCO<sub>3</sub> kg<sup>-1</sup> soil on a sandy-clay loam for two different cultivars, and of 10, 12 and 8 mg kg<sup>-1</sup> on two silty clay loams and one sandy clay loam respectively, for 98% of maximum yield (Johnston et al. 2014). Brennan and Bolland (2007) assessed a critical P-NaHCO<sub>3</sub> value for rapeseed of 20 mg kg<sup>-1</sup> soil for the 0.01–0.10 m layer of various sandy soils of Australia.

To evaluate if the specific conditions of 2011–2014 data can be considered representative for this field, we analyzed the long-term relationship between yield and soil available-P, this time with the data collected throughout all 44 years of contrasted P fertilization. Calculated threshold for winter wheat, maize and rapeseed respectively are 0.42, 0.34 and



**Fig. 6** Relationship between relative grain yield (RGY) and a P-NaHCO<sub>3</sub> for winter wheat [ $RGY = 139.6x/(x + 2.81) - 0.66x$ , residual standard error (RSE) = 7.28,  $n = 20$ ], maize [ $RGY = 120.8x/(x + 3.25) - 0.27x$ , RSE = 11.39,  $n = 19$ ], and rapeseed [ $RGY = 144.1x/(x + 5.76) - 0.66x$ , RSE = 7.19,  $n = 20$ ], **b** P-AAE for winter wheat [ $RGY = 110.2x/(x + 1.72) - 0.40x$ , RSE = 6.91,  $n = 20$ ], maize [ $RGY = 116.7x/(x + 1.77) - 0.66x$ , RSE = 11.29,  $n = 19$ ], and rapeseed [ $RGY = 144.14x/$

$(x + 5.8) - 0.66x$ , RSE = 9.29,  $n = 20$ ], **c** P-CO<sub>2</sub> for winter wheat [ $RGY = 111.5x/(x + 0.05) - 6.27x$ , RSE = 7.83,  $n = 20$ ], maize [ $RGY = 136.4x/(x + 0.1x) - 29.69x$ , RSE = 11.09,  $n = 19$ ], and rapeseed [ $RGY = 165.8x/(x + 0.38) - 20.04x$ , RSE = 9.24,  $n = 20$ ]. Relative grain yields of winter wheat and rapeseed were calculated by dividing the grain yield from a given fertilization treatment by the grain yield from the PK treatment

0.45 mg P kg<sup>-1</sup> soil for P-CO<sub>2</sub>, and 22.3, 12.9 and 18.0 mg P kg<sup>-1</sup> soil for P-AAE (supplementary figure 1). This confirms the accuracy of the critical soil P thresholds for P-CO<sub>2</sub> and P-AAE calculated based on single-year data, despite the inevitable yearly variability.

Relationships between PNI, as an average of all sampling time points, and the three indicators of soil available P are shown on supplementary figure. The relationship is well described by a power function for all three crops. Winter wheat and maize start to reach a plateau at high soil available P values, showing that luxury consumption is also decreasing with each added unit of P-fertilizer.

Under our experimental conditions, the Swiss fertilizer recommendations (Sinaj et al. 2017) were found not to be adapted to crop specific requirements. Instead, the guidelines suggest no fertilization application for winter wheat and maize from 80 mg P-AAE kg<sup>-1</sup> soil only, while the fertilization response thresholds in this study for wheat, maize, and rapeseed were respectively 22, 13 and 18 mg P-AAE kg<sup>-1</sup> soil. Same conclusion with P-CO<sub>2</sub> for which official guidelines recommend no fertilization from 1.55 mg P-CO<sub>2</sub> kg<sup>-1</sup> soil, whereas our data indicate that 0.40, 0.30 and 0.50 mg P-CO<sub>2</sub> kg<sup>-1</sup> soil is enough for winter wheat, maize and rapeseed, respectively. These results suggest that similar adjustments might be needed for other crops and soil types.

## Conclusions

Long-term P fertilization applied over a period of 44 years affected soil available P, and the shoot growth and grain yield of winter wheat and rapeseed measured in the last 4 years but had not effect on maize shoot growth and grain yield. The relationships between P<sub>C</sub> and shoot biomass or N concentration were very good ( $R^2 > 0.85$ ) for all three crops. The P<sub>C</sub>–shoot N concentration model for winter wheat from this study confirmed results from previous studies and can be used for calculating PNI. For maize and rapeseed, however, further research is required to ensure situations with no P luxury consumption and N deficiency. Moreover, there is unavoidable variability from year to year and between local conditions and soil types, which makes further

studies including multiple sites and years necessary to confirm and ameliorate the precision of P<sub>C</sub> values.

Based on the relationship between grain yield and three indicators of soil availability, threshold values for 95% of the maximum yield for winter wheat, maize, and rapeseed were less than those currently used in the official fertilization guidelines in Switzerland. Our results indicate that the refinement of fertilization requirements with more sensitive diagnostic tools that take into account the long-term effects of continuous fertilization could result in substantial reductions in fertilizer application rates.

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