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Shoot growth and phosphorus–nitrogen relationship of grassland swards in response to mineral phosphorus fertilization

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The sustainable P management of grassland ecosystems require a better understanding of the effect of P fertilization on the growth of grassland swards and the relationship between shoot P and N concentrations with the overall goal of developing improved methods of predicting P requirements of grassland swards. Our objectives were to determine the importance of P supply during the primary growth in spring, to analyze the optimal relationship between shoot P and N concentrations for timothy (*Phleum pratense* L.) and multi-species swards under a wide range of P nutrition, and conditions of soil and climate, and to confirm for grassland swards the decrease in the N:P ratio during crop growth previously reported for annual crops. Experiments with varying rates of P fertilization were conducted for two to five consecutive years at sites with timothy swards in Canada (Lévis, Normandin, Charlottetown, and Quebec) and Finland (Maaninka), and at sites with multi-species swards from long-term P fertilization experiments in Switzerland (Les Verrières) and France (Ercé). Dry matter (DM) yield, and N and P concentrations were measured on four dates with one-week intervals from the vegetative to late heading stages of development during primary growth. Our results (i) indicate that a P deficiency mostly affects the early season growth of grassland swards corresponding to shoot biomass less than approximatively 1 Mg DM ha^{-1}, (ii) confirm the optimal relationship between shoot P and N concentrations for timothy and multi-species swards but with variations of the relationship with the level of N nutrition and between timothy and multi-species swards, and (iii) confirm for grassland swards the decrease in the N:P ratio during the primary growth and the close relationship of the N:P ratio to shoot biomass.

1. Introduction

Improving methods for predicting P requirements of grassland swards require a better understanding of the effect of P fertilization on their growth and the relationship between shoot P and N concentrations. Several studies, mostly with annual crops species, have shown that the early season P supply is critical for optimum crop yield (Grant et al., 2001). Few studies, however, have established the importance of the P supply for the early season growth of grassland swards.

The relationship between shoot P and N concentrations in above-ground shoot biomass during crop growth has been studied in several crop species (Ziadi et al., 2007, 2008; Bélanger et al., 2015a,b), including forage grasses (Duru and Ducrocq, 1997; Bélanger and Richards, 1999; Bélanger and Ziadi, 2008), mostly for the purpose of developing diagnostic tools of P and N deficiencies. Plant-based methods for identifying P and N deficiencies depend on the definition of (1) critical concentrations, that is, the minimum concentration of a given nutrient required to achieve maximum shoot growth and yield or (2) optimal ratios of N to P concentrations.

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Table 1
Site characteristics and soil (0–15 cm) information.

Soil/crop information	Lévis (CA)	Normandin (CA)	Charlottetown (CA)	Maaninka (FI)	Les Verrières (CH)	Ercé (FR)
Latitude	46°48′N	48°51′N	46°17′N	63°08′N	46°91′N	42°85′N
Longitude	71°23′W	72°32′W	63°07′W	27°19′E	06°47′E	01°29′E
Altitude, m.a.s.l.	65	114	53	90	1150	660
Annual rainfall, mm	899	612	887	612	1100	1200
Annual temperature, °C	4.2	0.9	5.7	3.2	10.2	12.7
Soil texture	Clay	Clay loam	Sandy loam	Silt loam	Silty clay	Silt loam
pH (water)	6.1	5.3	6.0	5.7	5.3–5.1[a]	4.9–5.2[b]
Organic matter, g kg^{-1}	65.6	45.8	36.0	153.8	71.4–70.3[a]	91.6–86.6[b]
P-M3, mg kg^{-1}	34.1	48.2	153.0	136.5	17.2–44.8[a]	12.9–94.8[b]
P-Olsen, mg kg^{-1}	25.4	22.5	–	84.0	14.1–22.8[a]	11.3–90.5[b]
P-AA-EDTA, mg kg^{-1}	17.7	18.0	–	131.8	12.2–61.8[a]	5.5–44.6[b]
N applied[c], kg N ha^{-1} yr^{-1}	70	70	75	97	25	60
Main species	Timothy	Timothy	Timothy	Timothy	Multi-species	Multi-species
Year of seeding	2007	2008	2012	2009	1991	–
Plot size, m^2	6.0	12.0	27.0	6.3	6.0	12.0

[a] Values following several years with no P applied and with 17 kg P ha^{-1}.
[b] Values following several years with no P applied and with 50 kg P ha^{-1}.
[c] N applied in spring before the beginning of the primary growth.

The use of the relationship between shoot P and N concentrations to predict the critical P concentration in forage grasses (Bélanger and Richards, 1999; Bélanger and Ziadi, 2008) and other crops (Ziadi et al., 2007, 2008; Bélanger et al., 2015a,b) is based on the well-established strong dependence between P and N in crops. This dependence is evidenced by the simultaneous decrease in both P and N concentrations during crop growth and biomass accumulation, and the decrease in P concentration with a decrease in N concentration due to a N deficiency (Bélanger and Richards, 1999; Bélanger and Ziadi, 2008). Hence, the definition of critical P and N concentrations should take this decrease into account. The development of a model of critical P concentration as a function of shoot N concentration was first proposed for perennial grasses in Europe (Salette and Huché, 1991; Duru and Ducrocq, 1997). It was later investigated for timothy (*Phleum pratense* L.) in eastern Canada (Bélanger and Richards, 1999; Bélanger and Ziadi, 2008) and a model of critical P concentration (P_c) was developed under conditions where P was assumed not limiting for growth ($P_c = 1.07 + 0.063N$; Bélanger and Ziadi, 2008). Until now, this model has been validated only for timothy swards of eastern Canada in situations of P sufficiency. It has not been assessed in a wide range of conditions of P nutrition, soil and climate conditions, and types of grassland swards.

Even with this strong relationship and dependence between P and N concentrations in crops, there is strong evidence that the N:P ratio is not constant during crop growth. Greenwood et al. (2008), theoretically and experimentally, concluded that the N:P ratio of the shoot biomass of different annual crops decreases during crop growth because of changes in the relative proportion of growth-related tissues and storage/structure-related tissues. This decrease in the N:P ratio implies that the dilution of P and N in increasing shoot biomass operates at a greater rate for N than for P. During growth with near optimal levels of nutrients, Greenwood et al. (2008) proposed that the N:P ratio was primarily a function of the crop biomass. This variation of the N:P ratio during crop growth and the relationship of the N:P ratio to shoot biomass has never been studied in grassland swards.

Our objectives were to (i) determine the importance of P supply during the primary growth of grassland swards in spring, (ii) analyze the optimal relationship between shoot P and N concentrations for timothy and multi-species swards under wide range of conditions of P deficiency, and soil and climate conditions, and (iii) confirm for grassland swards the decrease in the N:P ratio during crop growth previously reported for annual crops.

2. Materials and methods

2.1. Field experiment, sampling, and laboratory analyses

The experiment was conducted at six sites (Table 1). Four rates of P fertilizer (0, 10, 20, and 40 kg P ha^{-1}) were applied at Lévis (CA), Normandin (CA), Charlottetown (CA), and Maaninka (FI) where timothy was the dominant grass species, while three rates (0, 8, and 17 kg P ha^{-1}) in Les Verrières (CH) and two rates (0 and 50 kg P ha^{-1}) in Ercé (FR) were used on multi-species-based swards from long-term P fertilization experiments. Site characteristics are presented in Table 1. Nitrogen and potassium were applied to ensure non-limiting conditions. All fertilizers were applied in early spring. A split-plot design was used with P rates as main plots and sampling dates as subplots with four replications at all sites. Sampling during the primary growth was done on four sampling dates, one week apart from vegetative to late heading stages of development (Tables 2–6). At each sampling date, dry matter (DM) yield was measured by cutting at a 5-cm height an area of at least 1 m^2. All plots were harvested again during the rest of the growing season but no measurements were taken. A fresh sample of around 300–500 g was taken in each plot, dried at 55 °C for 3 d, and ground to 1 mm. Plant samples were analyzed for P and N concentrations. Dried and ground forage samples of 100 mg were mineralized using a mixture of sulfuric and selenious acids, as described by Issac and Johnson (1976). The N and P concentrations in plant tissue were measured on a QuikChem 8000 Lachat autoanalyzer (Lachat Instruments) using the Lachat methods 13-107-06-2-D and 13-115-01-2-A, respectively.

At each site, soil samples (0–15 cm) were taken prior to applying P in the first year (Table 1). A composite sample per replicate was taken for a total of four samples per site. Soil samples were air-dried and sieved to 2 mm. Organic matter concentration was determined by wet oxidation (Tiessen and Moir, 1993). Soil pH was measured in distilled water with a 1:2 soil:solution ratio (Hendershot et al., 1993). Soil available P was determined with three methods: Mehlich 3 (Mehlich, 1984), Olsen (Olsen et al., 1954), and ammonium acetate EDTA (Demaria et al., 2005).

2.2. Data analysis

At each site-year, analyses of variance for shoot biomass, and P and N concentrations were done for each sampling day with the GENSTAT 14 statistical software package (VSN International, 2011) with P fertilizer rates as fixed effect and replications as a ran-