Differences in root morphological features and shoot P accumulation at different soil P levels

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## Abstract

The morphology and architecture of roots affect plant nutrient uptake. The objective of this study was to investigate how soil initial phosphorus (P) fertility level (Fair P 6,2 mg  $\Gamma^1$  soil, Good P 17,5 mg  $\Gamma^1$  soil) and annual P application (20 kg ha<sup>-1</sup>) affected root morphology, P content of silage grass and concomitantly its nutritive value. Soil P levels were created in a 40-year fertilization experiment (1973-2013). In a field trial (Kotkaniemi experimental farm) shoot and root samples were taken at the beginning of the growing season 2018, at the time of the 1<sup>st</sup> and 3<sup>rd</sup> harvests and twice after 1<sup>st</sup> and 3<sup>rd</sup> harvests. Roots were washed, photographed and root area, mean root length, and diameter were analysed with ImageJ program. Root and shoot nutrient content was analysed with ICP-OES. P application increased root area during the 1<sup>st</sup> harvest but decreased at 3<sup>rd</sup> harvest. Soil fertility P and P fertilization had a positive effect on root dry weight. Root area was negatively correlated with increasing shoot P content, which indicates that increase in root area compensated P uptake when less P was available. However, increased P foraging capacity was insufficient to elevate grass P content to an adequate level.

Keywords: Grass root, P uptake, root area, root diameter, soil P level, P fertilization

# Introduction

Over 60% of living grass biomass consists of roots (Gibson, 2009). In the roots of most grass species, 70–90% grow at a depth of 0–20 cm (Bolinder *et al.*, 2002). Nutrient deficiency affects root morphology by enhancing root growth and root hair cylinder volume (result of root diameter, length, and root hair length) when nutrients are less available (Haling *et al.*, 2016). When comparing plants with a fine extensive root system to plants with a thick and small root system, P deficiency was found to affect root morphology (Hill *et al.*, 2006). Changes in the root mass fraction and specific root length indicated the adaptation of roots to P stress. Although root diameter and the size of the root system had more important roles in describing plant P requirements. Although plants can adapt to different soil P levels, P availability has been shown to increase plant N and P uptake (Perez-Corona & Verhoeven, 1996; Xu *et al.*, 2016). Our aim was to investigate how P availability from soil and fertilizer affect grass root morphology and plant P uptake and to determine whether the change of the root morphological features correlate with root P uptake.

# Materials and methods

Root growth and nutrient uptake of the 3<sup>rd</sup> year silage grass ley (containing timothy (*Phleum pratense* L.), meadow fescue (*Schedonorus pratensis* Huds.), and tall fescue (*Schedonorus arundinaceus* Schreb.)) were investigated in a field trial located at Kotkaniemi experimental farm (Yara Suomi Ltd., Finland). Soil fertility P levels (Fair: 6,2 P mg l<sup>-1</sup> soil; Good: 17,5 P

mg  $1^{-1}$  soil) of the grass ley were created in a 40-year, long-term fertilization experiment with increasing NPK levels and the effect of soil fertility P levels and annual P fertilization (P0: 0 P kg ha<sup>-1</sup> and P20: 20 P kg ha<sup>-1</sup>) were evaluated. In 2018, ley was fertilized with N-P-K-S fertilizer (250/250–0/20–200/200–0/1,2) and harvested 3 times during the growing period. Due to a long period of water deficit, the ley was irrigated (30 mm) on June 11 and July 31. Growth stage during the  $1^{st}$  harvest was 31 and at the time of the  $3^{rd}$  harvest 21 (Simon & Park, 1983).

One plant and root sample (cylinder height 20 cm,  $\emptyset$  10 cm) was taken from each plot (3 replicates) at the beginning of growing season (May 5), 1 week before the 1<sup>st</sup> (May 24) and 3<sup>rd</sup> (September 12) harvest and 2 times after the 1<sup>st</sup> (June 19 and 26) and the 3<sup>rd</sup> (September 24 and October 1) harvest. Soil samples were washed, and root samples were photographed and dried as reported by Knuutila *et al.* (2019). Samples were ground (1 mm sieve, Ultra Centrifugal Mill ZM 200, Retsch, GmbH, Haan, Germany) and P was measured with Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES, ISO 11885:2007(E)). Root area (m<sup>2</sup> m<sup>-2</sup>), mean root length (mm), and mean root diameter (mm) were analysed from the images using ImageJ program (version 1.52c, Wayne Rasband, National Institutes of Health, USA). Analysis of variance (ANOVA) was performed to compare differences between all treatments and sampling dates. Means were compared using Fisher's Least Significant Difference (LSD) when the F-value in the ANOVA was statistically significant (P < 0,05). Pearson's correlation was used to determine the correlations between variables.

## Results and discussion

Soil P fertility level and P fertilization affected the root area, mean root length, and mean root diameter. Root area was highest in the spring and decreased during the growing season. In the spring and around the 1<sup>st</sup> harvest root area (Figure 1a) increased when more P was available either from soil or fertilization. However, at the time of the 3<sup>rd</sup> harvest and afterwards root area decreased with increasing P fertilization. As an exception, the root area with a Good soil P fertility level increased 1 week and 2 weeks after the 3<sup>rd</sup> harvest.



Figure 1. a) Root area (m<sup>2</sup> m<sup>-2</sup>), b) mean root length (mm) and c) mean root diameter (mm) (soil layer 0–20 cm) means of four treatments (low soil P fertility 6,2 P mg l<sup>-1</sup> soil with 0 and 20 P kg ha<sup>-1</sup> fertilization and high soil P fertility 17,5 P mg l<sup>-1</sup> with 0 and 20 P kg ha<sup>-1</sup> fertilization) 1 week before 1<sup>st</sup> and 3<sup>rd</sup> harvest. The data shown are means  $\pm$  SE; n = 3.

After the 1<sup>st</sup> harvest, grass yield increased with increasing P fertilization and soil P fertility level as shown in Knuutila *et al.* (2019). Root area, mean root diameter and mean root length did not correlate with grass yield. However, shoot P content had a negative correlation with

root area ( $r^2 = 0,215$ , p < 0,000) whereas mean root length and mean root diameter did not correlate with shoot P content. In the 1<sup>st</sup> harvest shoot P content increased with P availability and was highest in treatment Good P20 (2,97 g kg<sup>-1</sup> DM) and lowest in Fair P0 (2,37 g kg<sup>-1</sup> DM). The treatments Fair P20 (2,67 g kg<sup>-1</sup> DM) and Good P0 (2,80 g kg<sup>-1</sup> DM) did not differ from each other or from Fair P0 and Good P20 at the 1<sup>st</sup> harvest. There was no difference in shoot P content between soil P fertility levels or P fertilizations in the 3<sup>rd</sup> harvest.

P fertilization decreased mean root length (Figure 1b) at the time of harvests but increased in the spring and after each harvest, which indicated improved root regrowth. Mean root diameter (Figure 1c) was lowest in the spring and increased during the summer. P fertilization increased root diameter at the Fair soil P fertility level. In addition, the root diameter was larger at higher soil P availability. Waddell *et al.* (2017) also found that increased P availability decreased the fine root (< 0,24 mm) portion in comparison to P application to several grass species. However, P application even decreased root diameter at the Good soil P level. Root diameter was unusually large in the summer, which may be due to a long period of water deficit during the spring and summer months. Water deficit has been shown to increase root diameter (Zhou *et al.*, 2014).

### Conclusions

P availability modified root morphological properties such as root area, mean root length and root diameter. Soil fertility P and P fertilization increased root biomass (Knuutila *et al.*, 2019), but available P decreased root area. Increased root area compensated for P uptake at lower P availability but was unable to compensate for adequate shoot P content or yield.

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