



White clover living mulch reduces the need for phosphorus fertilizer application to corn



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ABSTRACT

White clover living mulch (LM) increases the uptake of phosphorus (P) and the yield of the main crop by promoting the colonization of arbuscular mycorrhiza (AM). However, the extent to which the P fertilizer application rate can be reduced by using LM is not yet known. This study aimed to address this question. Two field experiments were conducted from 2008 to 2009 (Experiment 1) and from 2009 to 2010 (Experiment 2) at the fields where the available P of soil fluctuated near the lower limit of the optimum P level (43.6 mg kg⁻¹; Truog method). Experiment 1 had a randomized block design, and Experiment 2 had a split-plot design with a factorial arrangement of two cropping systems (LM and no LM) with three P application treatments (0 kg ha⁻¹, 43.6 kg ha⁻¹, and 87.3 kg ha⁻¹). LM increased P concentrations in the early stages of growth and the yield of corn. This can be attributed to the increased AM colonization rate in the early stages of growth. The yield and total digestible nutrient yield of corn in LM with no P application was comparable to the maximum yield in no LM with or without P application. Therefore, LM could make unnecessary P fertilization in soils where P fertilization is required for silage corn.

1. Introduction

Phosphorus (P) is an essential mineral nutrient required by plants in large amounts (Marschner, 1995). MacDonald et al. (2011) reported that agronomic inputs of P exceed its removal by harvested crops on a global scale. However, the current global reserves of P have been estimated to become depleted in 50–100 years (Cordell et al., 2009). Therefore, reducing P fertilizer application is necessary for ensuring more sustainable use of P resources in agriculture.

Living mulch (LM) is a cultivation technique in which a cover crop is planted before the main crop to cover the ground throughout the growing season (Hartwig and Ammon, 2002). LM provides several benefits for crop production, including soil erosion prevention (Hall et al., 1984; Wall et al., 1991), soil organic matter (Kumwenda et al., 1993) and soil organism increase (Nakamoto and Tsukamoto, 2006), insect damage reduction (Brandsøter et al., 1998), nitrogen (N) loss by leaching decrease (Liedgens et al., 2004), and weed growth suppression (Ilnicki and Enache, 1992; Uozumi et al., 2004; Deguchi et al., 2015). White clover (*Trifolium repens* L.) LM has been reported to increase

arbuscular mycorrhiza (AM) colonization and the P concentration of corn (*Zea mays* L.) in the early stage of growth (Deguchi et al., 2005); this increases the yield of corn (Deguchi et al., 2007) because white clover acts as a host plant for AM in winter. AM increases the P uptake and growth of host plants (Smith and Read, 2008), and its colonization is influenced by cultivation management practices (Harrier and Watson, 2003). P uptake and the growth and yield of crops can be increased by promoting AM colonization by modifying cultivation management practices, such as tillage techniques (Kabir et al., 1997), crop rotation (Gavito and Miller, 1998; Arihara and Karasawa, 2000; Karasawa et al., 2001), and winter cover crops (Boswell et al., 1998; Kabir and Koide, 2000, 2002; Njeru et al., 2013). However, few studies on cultivation management have suggested that the P fertilizer application rate could be reduced while producing an adequate yield by using indigenous AM fungi under field conditions (Oka et al., 2010).

LM increases the P uptake in the main crop by promoting AM colonization by indigenous fungi (Deguchi et al., 2012). Thus, LM might reduce the P application rate needed for the main crop via the action of indigenous AM fungi. However, evidences for the extent to

Abbreviations: LM, living mulch; P, phosphorus; AM, arbuscular mycorrhizal; TDN, total digestible nutrient; CEC, cation exchange capacity

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which P fertilizer application rate can be reduced by using LM under field condition are scarce. This can be of particular interest in soils with high P adsorption capacity, such as volcanic soils, where P efficiency is usually very low, and thus high fertilizer rates are required to avoid P deficiency. This study aimed to estimate the extent to which the P fertilizer application rate can be lowered by using white clover LM without tillage while ensuring that the yield of silage corn is comparable to that from conventional cultivation.

2. Materials and methods

2.1. Cultural practices

Two experiments were conducted in different fields from 2008 to 2009 (Experiment 1) and from 2009 to 2010 (Experiment 2) on a volcanic ash soil (Pachic Melanudands) (Soil Survey Staff, 2003) at NARO Tohoku Agricultural Research Center, Morioka, Japan (39°42'N, 141°09'E). Morioka is located in the northern area of Japan, and its average temperature and precipitation are 9.3 °C and 1182.8 mm, respectively. The soil properties were as follows: (Experiment 1) pH, 6.9; total carbon, 63.8 g kg⁻¹; total N, 4.8 g kg⁻¹; cation exchange capacity (CEC), 38.2 cmol kg⁻¹; exchangeable Mg content, 3.0 cmol kg⁻¹; total P content, 1567 mg kg⁻¹; and available P content, 20.5 mg kg⁻¹ (Truog method; Committee of Soil and Environment Analysis, 1997) in August 2008; and (Experiment 2) pH, 6.3; total carbon, 100.2 g kg⁻¹; total N, 7.4 g kg⁻¹; CEC, 46.4 cmol kg⁻¹; exchangeable Mg content, 2.9 cmol kg⁻¹; total P content, 2125 mg kg⁻¹; and available P content, 17.0 mg kg⁻¹ in August 2009.

Experiment 1 was a randomized block design of three replicates with two cropping systems × three P fertilizer application treatments. Experiment 2 was a split-plot design of four replicates with two cropping systems as main plots × three P fertilizer application treatments as subplots. The two cropping system treatments were (1) corn grown with white clover LM without tillage, and (2) corn grown with rotary tillage, without LM (no LM). The three P fertilizer application treatments were no P fertilizer application (P0), P fertilizer application at a rate of 43.6 kg ha⁻¹ (P1), and P fertilizer application at a rate of 87.3 kg ha⁻¹ (P2). Each experimental plot was 3 m × 4 m.

In June 2007, the field was planted with corn to establish uniform conditions throughout the field for Experiment 1. In September 2007, the corn was harvested. From October 2007 to July 2008, the field was kept bare by rotary tillage. The soil P availability of the field was lower than the optimum P level for Andisol uplands in Japan (43.6–436 mg kg⁻¹; Truog method; Ministry of Agriculture, Forestry and Fisheries 2008). Thus, in August 2008, dolomite and fused magnesium phosphate were applied for soil amendment to all the treatment plots at a rate of 2000 kg ha⁻¹ to improve the available P level. Next, soil was tilled using a rotary cultivator. On August 26, 2008, white clover (*T. repens* L. 'Fuia') was broadcasted by hand in the LM treatments at a rate of 20 kg ha⁻¹. No cover crops were sown in the no LM treatments. On May 26, 2009, the no LM treatments were tilled using the rotary cultivator. The LM treatments were not tilled. On May 27, 2009, the white clover shoots in the LM treatments were clipped and left in the field. On May 28, 2009, P was applied as indicated before, and nitrogen (N) fertilizer at a rate of 300 kg ha⁻¹ as ammonium sulfate and potassium (K) fertilizer at a rate of 249 kg ha⁻¹ as potassium chloride were applied as surface bands in all the treatment plots. The application rates of N and K were higher than the average of recommended fertilizer rates for silage corn in 9 prefectures of Japan (155 kg ha⁻¹ and 98.8 kg ha⁻¹; Kanazawa, 2009) to mask the effect of LM on the N and K concentration of corn. On the same day, corn (*Z. mays* L. '31N27') was sown using a strip tillage seeder at a rate of approximately 67,000 plants ha⁻¹ with 4 rows per plot with 0.75 m rows and 0.20 m interhill distances. Weeds were suppressed by applying herbicides (atrazine and metolachlor) in the no LM treatments. No herbicides were applied in the LM treatments.

For Experiment 2, from May to August 2009, corn was planted in a field different from that used for Experiment 1. After corn was harvested and soil was tilled, in August 2009, dolomite and fused magnesium phosphate were applied for soil amendment to all the treatments at the rates of 1000 and 4000 kg ha⁻¹, respectively. Next, white clover was sown in the LM treatments at a rate of 20 kg ha⁻¹. In May 2010, tillage in the no LM and white clover management in LM treatments were performed in the same manner as in Experiment 1. On June 1, 2010, corn was sown, and chemical fertilizers and herbicides were also applied in the same manner as in Experiment 1.

Monthly average air temperature, precipitation, and sunshine hours from August 2008 to September 2010 were obtained together with the 30-year averages (1971–2000; air temperature and precipitation) or the 15-year average (1986–2000; sunshine hours) from the meteorological observation system at the NARO Tohoku Agricultural Research Center.

2.2. Soil analysis

Surface soil samples (0–10 cm) were randomly obtained using a shovel from three sites in each plot just before fertilizer application on May 27, 2009 (Experiment 1) and May 28, 2010 (Experiment 2). The weight of soil obtained from each site was approximately 150 g. The available P content (Truog method) of the sieved air-dried soil (< 2 mm) was analyzed using the methods specified by the Committee of Soil and Environment Analysis (1997).

2.3. Plant analysis

On May 25, 2009 (Experiment 1) and May 28, 2010 (Experiment 2), the dry weight of white clover shoots in a 0.5 m × 0.5 m sampling quadrat was measured from each plot in the LM treatments as reported by Alley et al. (1998). The corn shoots of 10 plants were sampled from each plot on June 25, 2009 (28 DAS; Experiment 1) and June 29, 2010 (28 DAS; Experiment 2). For yield determination at final harvest, corn shoots in 3 m² plots were harvested on September 30, 2009 (Experiment 1) and September 24, 2010 (Experiment 2) from the two central rows of each plot, avoiding one row of each end and the plot edge by at least 0.5 m in each plot as border according to Japanese Society of Grassland Science (2004). After the ears were separated from the foliage, 1-kg subsamples were collected from each sample for analysis.

All shoot samples were oven-dried at 70 °C for 48 h, weighed for determining the dry matter yield, and ground. The N concentration of the samples was determined using the dry combustion method (Committee of Soil and Environment Analysis, 1997). The plant samples were digested with perchloric acid and nitric acid. The digests were used to determine the P concentration by using the colorimetric method (Harada, 2004). The P uptake of plant shoots was calculated as the dry weight multiplied by the P concentration.

The total digestible nutrient (TDN) yield of corn was estimated using the following equation: TDN yield = 0.582 × foliage yield + 0.850 × ear yield (Ishiguri, 1972).

2.4. Arbuscular mycorrhizal colonization

Roots of five corn plants were collected from all plots on June 25, 2009 (28 DAS; Experiment 1) and June 29, 2010 (28 DAS; Experiment 2). The roots were washed with water and cleaned with 100 g L⁻¹ of potassium hydroxide at room temperature for 3 days. The roots were rinsed with distilled water, acidified with diluted hydrochloric acid, and stained with 500 mg L⁻¹ aniline blue in lactoglycerol. The percentage of AM colonization was determined using the magnified intersection method (McGonigle et al., 1990).

2.5. Statistical analysis

Statistical analyses were performed using SAS Add-in for Microsoft

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