Circular Nutrients for Supplying Phosphorus and Closing Urban to Rural Nutrient Cycles in Organically Managed Cropping Systems

by

Jessica Nicksy

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ABSTRACT

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Phosphorus (P) nutrition is vital for crop growth and yield, but the global P cycle is broken. The current system is linear, with P entering the food system as fertilizer mined from rapidly depleting phosphate rock reserves, and leaving the food system via food and human wastes entering landfill or waterways. Excess P from urban wastes contributes to eutrophication and toxic algal blooms in freshwater bodies. Meanwhile, P deficiency can limit yields, particularly on organic farms which often have negative P balances. Improved recycling of P from urban areas onto farmland is essential for food system sustainability, reducing reliance on non-renewable mined P, diverting harmful P from waterbodies, and improving crop yields by eliminating P deficiency.

Urban recycled (circular) P fertilizers are understudied compared to manure and conventional P fertilizers. This thesis evaluated three circular nutrient sources for their capacity to supply P and improve yields on a P-depleted soil of the Canadian Prairies. Frass is the excreta of black soldier fly (*Hermetia illucens*) larvae fed a diet of urban pre-consumer food waste. Digestate is the product of anaerobic digestion of urban food processing waste. Struvite is a phosphate mineral precipitated from municipal wastewater streams. Frass, digestate, and struvite were compared with monoammonium phosphate (MAP), a soluble conventional P fertilizer, compost, a common organic amendment, and an unfertilized control. In a wheat (*Triticum aestivum*) experiment replicated in two years, frass, MAP, compost, and digestate increased P uptake and improved grain yields compared to the control. In an alfalfa (*Medicago sativa*) based

forage experiment monitored over two growing seasons, all nutrient sources improved P uptake and yield compared to the control in the second growing season. In a pot experiment using Italian ryegrass (*Lolium multiflorum*), where adequate nitrogen (N) was supplied to remove any N effect of the amendments, all nutrient sources improved P uptake and yields compared to the control.

All three circular nutrient sources showed the potential to supply P and improve yields in some organic cropping systems, with the degree of improvement varying depending on crop and nutrient source. These circular nutrient sources have the potential to play an important role in recycling P from urban areas to farmland.

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FOREWARD

This thesis has been prepared based on the guidelines of the Department of Soil Science at the University of Manitoba. Chapter 1 is a brief introduction to the context and literature relevant to this study. Chapter 2 is a manuscript for the ryegrass pot experiment, which will be submitted to the Canadian Journal of Soil Science as a short communication. I am the lead author on the manuscript, and the co-authors are my advisors Dr. Brian Amiro and Dr. Martin Entz. I was responsible for establishment, maintenance, data collection, analysis, and write-up of the pot experiment. Additional assistance in design, watering, and laboratory analysis were provided by Dr. Don Flaten, Joanne Thiessen Martens, and Anthony Buckley. Chapter 3 is a manuscript describing the wheat and alfalfa field experiments, which will be submitted to Renewable Agriculture and Food Systems. As with Chapter 2, I am the lead author on the manuscript and the co-authors are my advisors Dr. Brian Amiro and Dr. Martin Entz. I was responsible for managing both field experiments, performing statistical analysis, and writing the manuscript. Assistance with field work was provided by the Natural Systems Agriculture lab group and Joanne Thiessen Martens. Assistance with lab analysis was provided by Anthony Buckley. Chapter 4 is the overall synthesis of both manuscripts, and includes suggestions for future research directions.

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1 INTRODUCTION

1.1 Overview

Phosphorus (P) plays a pivotal role at the intersection of food, water, and energy security; it is a non-renewable resource that is an essential plant nutrient for food and biofuel crops, but a harmful environmental contaminant in waterways (Jarvie et al. 2015). Phosphate rock reserves, which are mined to produce P fertilizer, may be depleted in the next 70-140 years (Li et al. 2018). Simultaneously, P exported in crops from farms to cities can contribute to environmental problems like eutrophication when released to waterways (Schindler et al. 2012). P deficiency is common on organically managed farms of the Northern Great Plains (Entz et al. 2001) due to negative P balances in low-input cropping systems (Welsh et al. 2009). Circular nutrient sources, which divert food and human wastes back onto farmland, have the potential to ameliorate both the global scale issues of phosphate rock depletion and environmental contamination, and the farm-scale issue of P deficiency on organic farms. In order for circular nutrients to act as viable alternatives to conventional nutrient sources, their ability to supply P and improve yields must be demonstrated. This thesis evaluated the P-supplying capacity of three circular nutrient sources compared to typical conventional and organic sources.

1.2 Global phosphorus cycle

The challenges associated with the global P cycle are complex and multi-faceted, from geological resource limitation, to P deficiency on arable farmland, to environmental contamination via excess P fertilization. While no single solution can address all of the challenges associated with the P cycle, the principle of circularity provides a framework toward a more sustainable and food-secure P system.

Prior to the mid-late 19th century farms relied on local nutrient sources (e.g., manure and human excreta) for nutrient replenishment (Cordell et al. 2009). As P is a nutrient with no significant gaseous atmospheric source or sink (Liu et al. 2008), this made for fairly closed local P cycles. The use of mined phosphate rock as fertilizer, along with global food trade and reduced recycling of human excreta with the advent of flush toilets, shifted the system from one of local circularity to global linearity (Cordell et al. 2009). Phosphate rock deposits are formed on a geological time scale of millions of years, far slower than human activity is now depleting them (Filippelli 2011).

P enters the food system via fertilizer mined from non-renewable and often geographically distant phosphate rock reserves external to the food system, is exported to population dense urban centers via crops, and leaves the food system via food and human waste entering landfills and waterways (Figure 1.1). Phosphate rock reserves are a non-renewable resource, which will be depleted if changes aren't made to the global P cycle (Li et al. 2018). A model of global P flows estimates that only 17% of organic solid food waste P, and 10% of human waste P is recycled to arable soil (Cordell et al. 2009). Animal manure fares better, with over 50% of animal manure P being returned to arable soil (Cordell et al. 2009). However, animal manure is often abundant in regions with excess soil P, and scare in regions of soil P deficiency, due to concentration of animal production far from feed production (Jarvie et al. 2015).

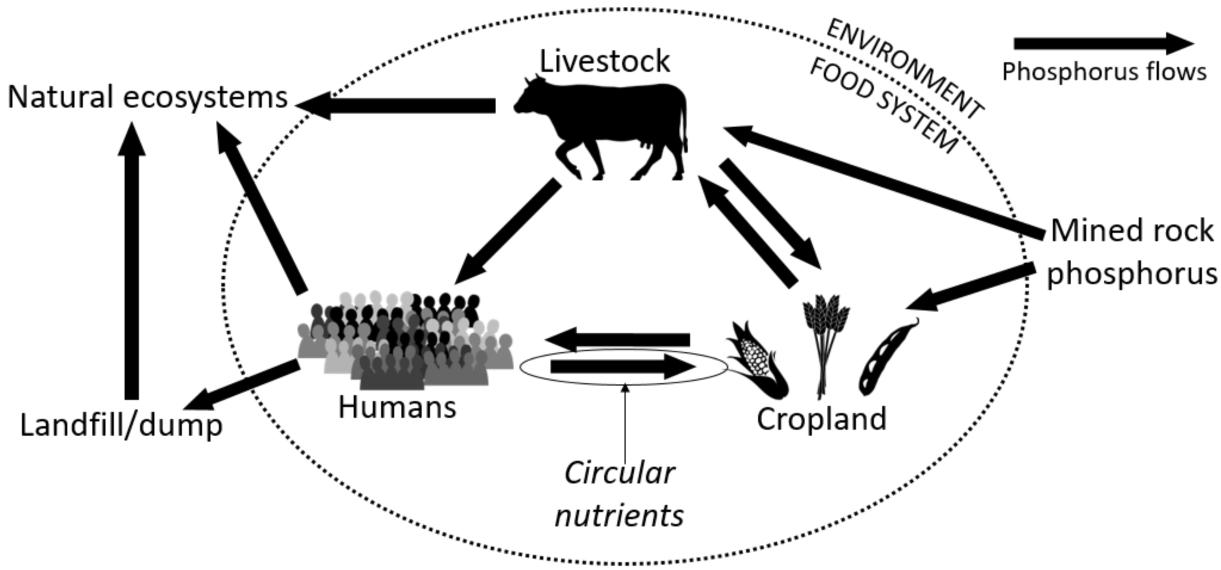

Figure 1.1 Conceptual food system model for phosphorus flows.

Concentration of animal production that depends on imported feed (and the associated P therein) can lead to excess P accumulation in soils. Manure and other organic amendments are usually applied based on crop nitrogen (N) requirements, which leads to increased soil P because crop offtake N:P ratios are higher than the N:P ratio in most organic amendments (Sharpley and Moyer 2000; Eghball 2002). High soil P levels, and resultant increased environmental P loss in runoff, is a challenge which plagues regions of high livestock density, like the Chesapeake Bay watershed on the American east coast (Beegle 2013). While runoff P losses are generally not agronomically important (e.g., <1 kg ha^{-1} $year^{-1}$ (Tiessen et al. 2010)), they are environmentally important as they can contribute significantly to P loading in water bodies (Schindler et al. 2012). The same problem exists where humans are concentrated in urban settings and produce substantial P waste (Jarvie et al. 2015), though urban P is more often sent to landfill or released to waterbodies directly rather than re-applied to agricultural land (Cordell et al. 2009).

While excess P plagues some regions, P deficiency is equally a threat to food system sustainability and security. A recent meta-analysis found that 49% of cropland globally is P-limited (Hou et al. 2020). P limitation is especially prevalent on organic farms, where options for nutrient import are limited. Organic farms often rely on biological N fixation with leguminous green manures, but this leaves them at risk of negative P balances, resulting in P "mining" and eventual P deficiency (Welsh et al. 2009; Reimer et al. 2020). A survey of organic farms on the Northern Great Plains found P deficiency on farms with a long history of organic management (Entz et al. 2001). Similarly, farms managed organically for over 15 years in England had lower soil P than conventionally managed farms (Gosling and Shepherd 2005). P "mining" on organic farmland contradicts the organic ethic of ecology, and undermines the long-term sustainability of organic farming.

Sir Albert Howard, a pioneer of the organic movement, recognized the importance of recycling organic wastes to the soil. His "Law of Return" advocated use of sewage sludge as a means of returning fertility and organic matter to the soil (Heckman 2006). This idea of designing systems for circularity was prescient, coming long before the recognition that humans were depleting non-renewable P reserves. Given this history, one might expect organic farmers and researchers to be at the forefront of using urban wastes as agricultural amendments. However, concerns about potentially toxic elements and organic compounds, particularly in human wastes, is in tension with the adoption of urban wastes streams in organic agriculture (Möller et al. 2018). P in human wastewater accounts for the largest proportion of urban P (representing 50%-60% of mined P fertilizer applied in Europe) and thus cannot be ignored by the organic community when considering sustainable P sources (Möller et al. 2018). The conversation is perhaps furthest advanced in Europe, where a survey of stakeholders in the organic industry including farmers and certifiers found that on average 60% viewed the use of human urine and sewage sludge in organic agriculture positively (Løes 2016), and struvite precipitated from municipal wastewater is in the process of gaining legislative approval for use in organic agriculture (Cuoco and Hermann 2020). Meanwhile, struvite from municipal wastewater has been reviewed but not approved by the Canadian Technical Committee on Organic Agriculture (Organic Federation of Canada 2020).

In order to ensure long-term sustainability of organic production systems, we must investigate technologies that not only produce safe nutrient sources, but also provide co-benefits within the food system and beyond. For example, anaerobic digestion of food waste produces methane biogas, which can be used in place of fossil fuels (Wainaina et al. 2020). Insects, which are used to process food waste, produce not only a circular nutrient source in their frass, but their biomass itself is a useful high protein food or feed source (Čičková et al. 2015). When struvite is precipitated from municipal wastewater systems, it not only produces a high P fertilizer but also removes P before it can enter waterways and contribute to eutrophication (Kumar and Pal 2015). Further, struvite has sufficiently high P content that it may be feasible to transport it to regions of P deficit.

Of course, in order for these nutrient sources to be a feasible component of a circular P cycle, they must also be effective sources of P for crop plants. For example, treatments which chemically remove P from wastewater streams prior to discharge, meeting environmental protection goals, may render that P unavailable in sewage sludge (Möller et al. 2018). Al and Fe treatments cause the precipitation of Fe and Al phosphates, as well as strong adsorption of P to Fe and Al hydroxides, limiting the P availability of the sludge compared to triple super phosphate to less than 25% (Torri et al. 2017) and rendering them less ideal as circular nutrient sources.

The research carried out in this thesis aims to evaluate the efficacy of three circular nutrient sources for P supply and yield improvement in a low P organically managed soil of the Prairies.

1.3 Phosphorus in Manitoba

Within Manitoba exist many of the P challenges described at the global level: an overall P deficit, especially on organic farms; P surpluses in some areas due to livestock concentration; and rapid eutrophication of the largest lake in Manitoba due to P loading.

While no recent comprehensive P flow assessment has been performed for Manitoba, Loro et al. (2013) estimated P budgets for agricultural soils in Rural Municipalities of Manitoba based on 2011 livestock numbers. Overall, Manitoba's agricultural land lost P at a rate of over 13 million kg P in 2011, or 2.0 kg P ha^{-1}. This includes the import of synthetic fertilizer, which accounts for 71% of P inputs; if synthetic fertilizer were not included, the P deficit would be 8.7 kg P ha^{-1}. Thus, Manitoba is currently heavily dependent on non-renewable P fertilizer imports for crop productivity. Insufficient manure resources pose an extra risk to Manitoba's organic farms, which cannot rely on synthetic fertilizer imports to balance their P budgets.

Manitoba's P deficit problem is exacerbated by inefficient use of manure P. Nine of 78 Rural Municipalities in Manitoba actually have P surpluses, due to concentration of livestock in these areas (Loro et al. 2013). Seven of the nine could reach P balance by removing synthetic P imports and instead relying exclusively on manure P. The other two (Hanover and La Broquerie) have insufficient cropland relative to livestock to reach P balance even if they halted use of synthetic fertilizer. Livestock densities have been well correlated to P concentration in runoff in Manitoba (Salvano et al. 2009). This highlights that even regions with an overall P deficit, like Manitoba, can have localized areas of surplus P and increased P runoff risk when livestock are concentrated rather than distributed across the landscape.

Human sources of P were not considered in the Loro et al. (2013) report. Based on Manitoba's 2019 population of 1,372,708 (Government of Manitoba 2019), and P in human and food waste of 0.49 and 0.39 kg P person^{-1} year^{-1} respectively (Metson et al. 2016), human P sources could theoretically contribute 1.2 million kg P year^{-1} to Manitoba's P cycle. This value is small compared to manure P produced in 2011 (17.1 million kg P) (Loro et al. 2013), but could slightly offset the P import requirements of the province.

Further, diverting Manitoba's human P pool to agricultural land with a P deficit has positive environmental implications. P in Manitoba wastewater is an important contributor to P loading in Lake Winnipeg, accounting for 9% of the total P load to Lake Winnipeg, and 20% of Manitoba P sources (Lake Winnipeg Stewardship Board 2006). Lake Winnipeg is the world's tenth largest freshwater lake, is shallow, and has the highest ratio of watershed area to lake surface area of any great lake worldwide, rendering it particularly susceptible to eutrophication (Lake Winnipeg Stewardship Board 2006). Lake Winnipeg has undergone rapid eutrophication since the mid-1990s, driven by P-loading from increases in livestock and human populations (Schindler et al. 2012).

Utilization of circular nutrient sources in Manitoba can slightly decrease the province's P deficit, while reducing the human contribution to P loading and eutrophication in Lake Winnipeg and other water bodies.

1.4 Amendments of interest

Two amendments sourced from novel processing techniques for urban food waste, digestate and frass, and one amendment sourced from municipal wastewater, struvite, were evaluated in this thesis. All amendments in this study are commercially available, and the frass and digestate are approved for use in organic production systems. Municipally-derived struvite was reviewed, but not approved, for use in organic agriculture in 2020; it will likely be reviewed again in 2025 (Organic Federation of Canada 2020). In the meantime, livestock- or plant-derived struvite has been approved for use in organic agriculture, and thus the inclusion of municipally-derived struvite is relevant for organic farmers in the short as well as long-term (Canadian General Standards Board 2018).

Interest in novel circular nutrient sources is increasing, but they remain understudied compared to conventional sources like manure and monoammonium phosphate (MAP). Results of a rudimentary Web of Science search reveal that publications on conventional P sources are orders of magnitude more numerous than publications on the circular nutrient sources evaluated in this study (Table 1.1). A greater body of literature is required to establish whether these nutrient sources can effectively supply P in organic systems.

Table 1.1 Results of a November 5, 2020 literature search in all Web of Science databases.

	Amendment Search Term	**# results**
Conventional sources	"*ammonium phosphate"	11760
	manure	72952
Circular sources	struvite	1005
	frass	61
	"anaerobic digestate"	171

* The full search term was the Amendment search term AND (fertilizer OR amendment). Note that these values are illustrative of the order of magnitude differences in the bodies of literature only, and do not represent the exact number of studies assessing agronomic or fertilizer potential of each product; many of the search results focus on the creation of the amendments and only mention fertilizer or amendment potential in passing, but were still found in this search. Likewise, some studies which used synonyms may have been missed in this search.

The frass (insect excreta) used in this study is derived from black soldier fly (BSF) (*Hermetia illucens*) larvae fed a diet of pre-consumer (restaurant and grocery) non-meat waste. Insects are gaining attention in the sustainable food sphere, due to their lower environmental impacts and feed to protein conversion ratio compared to conventional livestock (Fan et al. 2015; Wegier et al. 2018). Using insects as livestock feed reduces the cropland required to grow feed, and thus frees this land for production of human consumables (Wegier et al. 2018). In addition, insects can be eaten directly as sustainable protein sources for humans, though cultural norms often inhibit this use (Fan et al. 2015). BSF larvae have good potential as processors of urban food waste due to their ability to eat a wide variety of organic material (Čičková et al. 2015).

Few peer-reviewed publications exist on the fertilizer potential of BSF larvae frass, with the majority of the literature focussing on production of insect biomass (e.g., Pastor et al. 2015; Kierończyk et al. 2020), and no known studies focusing specifically on its properties as a P source. One pot study tested various proportions of frass (from insects grown with an artificial diet rather than food waste) with peat as a growing medium in basil, tomato, and lettuce (Setti et al. 2019). Rates up to 20% frass improved plant growth, but a high proportion of frass suppressed plant growth. Another pot study using a 2:1 ratio of soil to frass from food waste-fed BSF larvae found growth suppression of maize (*Zea* mays) in the frass treatment (Alattar et al. 2016). These studies indicate that high rates of frass may be phytotoxic, perhaps due to salinity or high ammonium concentration of the frass. Kebli and Sinaj (2017) found a positive response to food-waste-fed BSF frass applied based on N rate in a pot study using lettuce and ryegrass. In ryegrass, frass produced similar yields to a synthetic fertilizer in two of three soils. In lettuce, frass produced similar or greater yield compared to the synthetic fertilizer in the low-pH sandy soil, but lower yield in the two neutral to high pH soils. Frass consistently improved yields compared to the control for both lettuce and ryegrass. Choi et al. (2009) found similar growth rate of Chinese cabbage (*Brassica rapa*) with a BSF frass and a commercial fertilizer, though the identity of the commercial fertilizer is not specified. Gärttling et al. (2020) found lower N supply from BSF frass compared to a synthetic fertilizer in a maize pot study. These studies vary in whether the BSF larvae are fed urban food waste or another feed substrate, and the type of feed used may be important in determining frass fertilizer properties. Klammsteiner et al. (2020) found significantly different pH, electrical conductivity, and N content of BSF frass from larvae fed three different diets (P content not reported), though the three frass types resulted in similar growth of ryegrass to each other and to a mineral fertilizer when applied based on N rate. Frass efficacy may be dependent on BSF feedstock as well as pH or other soil properties, and further research is needed to delineate these relationships, particularly with respect to its P fertilization properties.

The digestate used in this study was produced from food processing and food retail waste. Anaerobic digestion is the degradation of organic matter by natural microbial communities in the absence of oxygen, which produces a gaseous "biogas" mix predominantly composed of CH_4 and CO_2 (Wainaina et al. 2020). The raw biogas can be used directly in electricity and heat generation, or upgraded to be used as a vehicle fuel or injected into natural gas lines (Wainaina et al. 2020). Anaerobic digestion is a promising technology for processing food waste within the food-energy-water nexus because it produces clean energy and contributes to the recycling of nutrients (Kibler et al. 2018; Wainaina et al. 2020). In addition, anaerobic digestion has the potential to produce other valuable products like hydrogen gas and volatile fatty acids, though these processes are currently in the research stage (Ma and Liu 2019; Wainaina et al. 2020).

Liquid and solid fractions of wet digestate can be applied together or separately to agricultural land as nutrient and organic matter sources, with the solids often undergoing an aerobic composting phase prior to land application (Möller and Müller 2012; Kibler et al. 2018; Chojnacka et al. 2019). Solid fractions of anaerobic digestate tend to have higher P concentrations than liquid (Nkoa 2014). The digestate in the present study was the dried (at 90-100 °C) and pelletized solid fraction. Digestion often causes an increase in pH and formation of minerals like Ca or Mg phosphates and struvite within the digestate, though this may not necessarily hamper its efficacy as a P source (Möller and Müller 2012). For example, Haraldsen et al. (2011) found statistically similar P uptake and grain yield for a liquid food waste digestate and a synthetic fertilizer. The efficacy of P supply from food waste digestate may depend on the fraction (solid or liquid) and the pH of the soil. In a study of potential circular P fertilizers, Brod et al. (2015) found a greater fraction of recalcitrant species, and especially acid soluble Ca and Mg phosphates in solid compared to liquid food waste digestate. A bioassay using ryegrass in the same study found lower P uptake from the solid fraction compared to the liquid fraction and a synthetic fertilizer at soils of pH 5.5 and 6.9. In the higher pH soil, there was a significant relationship between P uptake and acid soluble recalcitrant fraction in the amendments, indicating that the presence of these low solubility precipitates is a greater hindrance to P uptake at higher pH (Brod et al. 2015).

Post-digestion processes may also impact P availability of anaerobic digestates. Ross et al. (2018) found that addition of a pelletized composted digestate did not increase soil test P compared to a control, while the unpelletized digestate did increase available P in an oat (*Avena sativa* L.) pot experiment. Further study of the impact of processes like drying and pelletization on P availability in digestate products is warranted.

The struvite used in this study was precipitated from a municipal wastewater stream. Struvite is an ammonium magnesium phosphate mineral ($NH_4MgPO_4 \cdot 6H_2O$) which precipitates naturally in sewage treatment facilities and can be an operational problem when it clogs pipes (Kumar and Pal 2015). However, it is possible to intentionally precipitate struvite in wastewater treatment facilities through manipulation of pH to around 9.0 and sometimes addition of Mg (Kumar and Pal 2015). Struvite has a high P content (NPK 5-28-0-10Mg) on the same order of magnitude as mined synthetic P fertilizers like MAP (11-52-0). This makes struvite a feasible option for shipping P from high-P to P-depleted regions (Metson et al. 2016). However, Möller et al. (2018) point out that struvite's lack of organic matter and micronutrients may make it a less effective overall soil amendment. From a circular food system perspective, struvite's most important role is in moving P from areas where it is in excess to where it is needed; it may not be the most appropriate amendment in regions where there are abundant organic-matter-based P sources available.

Struvite has been widely found to have similar P availability compared to conventional fertilizers (e.g., Cabeza et al. 2011; Katanda et al. 2016). However, many struvite studies have been conducted in pots with ground struvite instead of granules, as would be applied in the field. There appears to be an interactive effect between granule size and soil pH on struvite dissolution. Degryse et al. (2017) found that ground struvite dissolved well within the pH range of 5.9 to 8.5, but granular struvite dissolved much more slowly in high pH soils than low pH soils. Another complicating factor is crop species; buckwheat (*Fagopyrum esculentum*) has been shown to be more efficient at accessing struvite than wheat, perhaps due to buckwheat's ability to exude organic acids and induce struvite dissolution (Talboys et al. 2016). Two meta-analyses of struvite studies showed good performance of struvite compared to synthetic fertilizers, but one showed decreasing efficacy of struvite with increasing pH (Hertzberger et al. 2020) while the other did not (Möller et al. 2018). In the more recent and comprehensive meta-analysis by Hertzberger et al. (2020), only 8% of reviewed experiments were field studies, pointing to the need for more field research on struvite, especially across a range of soil pH.

1.5 Project Objectives

This project aimed to evaluate the P supplying power and yield improvements resulting from application of three circular nutrients (frass, digestate, and struvite) in a P-depleted organically managed soil of the Canadian Prairies (Table 1.2). The field site at Libau, MB, represents a typical P-depleted organically-managed system in Manitoba. Long term alfalfa-based forage removal with no nutrient addition has led to extremely low soil-test P levels. The high pH of the soil is typical of many fine-textured soils in Manitoba (Government of Manitoba 2008). High pH has been indicated as a possible inhibitory factor to P availability for all three nutrient sources (see Section 1.4), and testing the amendments under this condition is therefore important in determining their potential as circular fertilizers for Manitoba.

Three crops were used to evaluate the amendment effects under different conditions and over different time periods. Italian ryegrass was grown in a greenhouse setting with and without the addition of non-limiting synthetic N in order to differentiate P effects and N effects of the amendments (Chapter 2). Ryegrass is a fast-growing plant that can be harvested successively to evaluate nutrient uptake over time, and for this reason it is a very popular bioassay plant (Brod et al. 2015; Kebli and Sinaj 2017; Grigatti et al. 2020).

Table 1.2 Summary of experiments

	Wheat	**Alfalfa forage**	**Italian ryegrass**
Crop Type	Annual field	Perennial field	Greenhouse bioassay
Timeframe	May-Sep 2019 and 2020	May 2019 to Aug 2020	4 months
Moisture Regime	Rainfed	Rainfed	Hand-watered
Factors	1. Nutrient source (5 amendments and control) 2. Genotype (BJ08-IG & AAC-Brandon)	1. Nutrient source (5 amendments and control)	1. Nutrient source (5 amendments and control) 2. Nitrogen level (+N or -N)

Two genotypes of wheat (*Triticum aestivum*) were grown in the field to evaluate a within-crop genotypic effect on response to the amendments in an early successional (annual) cropping system (Chapter 3). Spring wheat is the second most widely grown crop in Manitoba, second only to canola, and is therefore of great relevance to farmers (Manitoba Agricultural Services Corporation 2018). One variety, AAC Brandon, is conventionally bred and the most widely seeded wheat variety among both organic and conventional farmers in Manitoba (Manitoba Agricultural Services Corporation 2018). By contrast, the second genotype, BJ08-IG, was developed through a cooperative breeding program which partnered professional breeders and farmers. Previous work at this field site had indicated a better response to struvite in a farmer selected line (Thiessen Martens & Entz, unpublished data), and we wanted to investigate this effect with a different farmer-selected line and a variety of amendments.

Amendments were also applied to a legacy alfalfa-based (*Medicago sativa*) perennial forage stand (Chapter 3). This allowed us to monitor longer-term effects of the amendments over two growing seasons. In addition, the forage represents a later-successional (perennial) system, contrasting with the early successional wheat system.

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2 RECYCLED NUTRIENT SOURCES SUPPLY PHOSPHORUS AND IMPROVE RYEGRASS YIELDS ON PHOSPHORUS DEPLETED SOIL

2.1 Abstract

Recycling phosphorus (P) within the food system is fundamental to long-term food system sustainability. This greenhouse study compared three recycled P sources –struvite precipitated from municipal waste water, black soldier fly frass from food waste, and anaerobic digestate of food waste – to monoammonium phosphate (MAP), livestock compost, and a control. Italian Ryegrass (*Lolium multiflorum*) was successively harvested four times from a P-depleted soil. In nitrogen (N) sufficient conditions, all amendments significantly increased cumulative ryegrass yields over the control, and were statistically similar to MAP. Relative P supply was in the order frass = MAP > struvite >= compost >= digestate >> control. The circular nutrients tested show promise as sustainable P sources.

2.2 Introduction

Phosphorus (P) plays a pivotal and paradoxical role at the food-energy-water nexus: it is an essential nutrient for the crop growth, but also a harmful pollutant causing eutrophication of water bodies when it leaves the agricultural system (Jarvie et al. 2015). Mined phosphate rock, used to make P fertilizers and applied directly in organic systems, is a non-renewable resource that may be depleted in 70-140 years (Li et al. 2018). Recycled, rather than mined, sources of P must become the norm in order to ensure long-term food system sustainability. Recycled, or "circular", nutrients may be especially important on Canadian organic farms, which often suffer from P depletion due to negative farm-gate P balances (Schneider et al. 2016). Circular fertilizers remain understudied compared to mined conventional fertilizers or manures, particularly in terms of their P properties.

The present pot study assesses three circular nutrient sources that are commercially available in Canada – struvite, frass, and anaerobic digestate – in a low-P Prairie soil. Struvite is a phosphate mineral which can be extracted from animal or municipal wastewater; the struvite in the study is municipal in origin, with a median granule size of 0.9 mm. Struvite is the best studied of the circular nutrient sources. Two reviews have found that it has good P availability compared to conventional fertilizers (Möller et al. 2018; Hertzberger et al. 2020), but questions remain about its efficacy in high-pH soils like those found in the Prairies because of its low solubility at high pH. Thiessen Martens and Entz (unpublished data, 2018) found significant responses of wheat to struvite fertilizer in one of two years in a high pH, P-depleted soil at Libau, MB, Canada, which is the soil collection site for this study. Struvite is currently allowed as an organic amendment in Canada only when it is sourced from animal or plant wastes, though it is expected that this will be reviewed in the future.

The frass used in this study was the waste product of black soldier fly (BSF) larvae (*Hermetia illucens*) fed a diet of pre-consumer urban food waste. Few studies exist on the fertilizer potential of BSF frass, and we are not aware of any focussing specifically on its P-supplying properties. Kebli and Sinaj (2017) conducted a pot study on three soils using lettuce and ryegrass, and found that frass applied based on a N rate consistently improved yields over the control, and in a sandy, low-pH soil, yields were similar to or greater than the synthetic fertilizer. Further study of frass as a P source is needed.

The anaerobic digestate in this study was produced by digesting food retail and food processing waste in the absence of oxygen. Liquid and solid fractions of digestate can be applied together or separately, with solid fractions tending to have higher P concentrations than liquid (Nkoa 2014). The present study used the dried and pelletized solid fraction of digestate. Most previous studies on the fertilizer properties of digestate have used animal waste digestates and/or focused on N supply, but Brod et al. (2015) characterized P fractions of liquid and solid food waste digestate, and used a ryegrass pot study to test P uptake. They found a higher proportion of low-solubility P in the solid digestate, lower P recovery of solid digestate compared to liquid digestate or synthetic P, and lower ryegrass yields for solid digestate compared to liquid digestate or synthetic P at the earliest harvest, though yields were similar at later harvests.

We hypothesized that the circular nutrient sources would improve ryegrass yields and P uptake compared to the control, and that they would have similar yields and P uptake compared to a conventional fertilizer under conditions of N-sufficiency. A secondary hypothesis was that nutrient sources with lower N content relative to P (e.g., struvite) would yield less than those with high N content (e.g., frass) when N was not supplied, because of N supply from the amendment itself. We assessed these hypotheses by comparing the nutrient sources with and without the addition of non-limiting N fertilizer.

2.3 Materials and Methods

2.3.1 Nutrient Sources

Struvite, frass, and digestate were compared with a common conventional (monoammonium phosphate, MAP) and a common organic (aerobic compost, predominantly horse manure and bedding) nutrient source. Total nutrient concentrations for MAP (110 g kg^{-1} N, 230 g kg^{-1} P) and struvite (50 g kg^{-1} N, 120 g kg^{-1} P) were based on manufacturer specifications, and for digestate (38 g kg^{-1} N, 28 g kg^{-1} P), frass (32 g kg^{-1} N, 8.7 g kg^{-1} P), and compost (7.1 g kg^{-1} N, 2.3 g kg^{-1} P) were determined by Agvise Laboratories, ND, USA. All amendments were applied at a rate of 20 kg P ha^{-1}, so that amendment addition per pot was: 0.16 g MAP, 15.6 g compost, 0.29 g struvite, 4.0 g frass, and 1.3 g digestate. Nitrogen to phosphorus (N:P) ratios of the amendments varied, such that different amounts of N were added to each pot via the amendment treatments. N added was the equivalent of 9.4 kg ha^{-1} for struvite, 73 kg ha^{-1} for frass, 28 kg ha^{-1} for digestate, 63 kg ha^{-1} for compost, and 10 kg ha^{-1} for MAP based on the surface area of the pots.

2.3.2 Experimental Setup

The pot study was conducted on a P-deficient soil using Italian Ryegrass (*Lolium multiflorum*) as the bioassay crop. The soil was a Gleyed Rego Black Chernozem, collected from the 0-15 cm layer at a site under long-term organic management, with a clay texture, 35 kg ha^{-1} nitrate-N, 3 mg kg^{-1} Olsen-P, 312 mg kg^{-1} K, 40 kg ha^{-1} Cl, 27 kg ha^{-1} S, 1.3 mg kg^{-1} B, 0.6 mg kg^{-1} Zn, 18.7 mg kg^{-1} Fe, 1086 mg kg^{-1} Mn, 0.96 mg kg^{-1} Cu, 1086 mg kg^{-1} Mg, 6622 mg kg^{-1} Ca, 54 g kg^{-1} organic matter, and a pH of 8.1 based on analysis by Agvise Laboratories, ND, USA. Soil was coarse-sieved (approx. 15 mm), homogenized in a cement mixer, and added to 15-cm diameter pots at a rate of 1914 g dry soil per pot.

The experiment was laid out as a randomized complete block design with four blocks as replicates. The treatment structure was factorial, with P source (5 amendments and unamended control) and N fertilization (+N pots receiving N fertilizer and –N pots receiving no N fertilizer). Amendments were thoroughly mixed with the top 5 cm of the soil at a rate equivalent to 20 kg P ha^{-1} based on the surface area of the pots and total P concentration of the amendments. The +N pots received the equivalent of 90 kg N ha^{-1} as urea dissolved in 100 mL reverse osmosis water at the beginning of the experiment and after each biomass harvest, which we assumed eliminated any N-deficiency. The –N pots received no N, but an equivalent amount of water. Based on the soil analysis, other nutrients were not expected to be limiting in this soil, and so no additional nutrient solution was added.

Pots were seeded with 25-30 Italian Ryegrass seeds at an approximate depth of 0.5-1cm and were later thinned to 10 plants per pot. Pots were watered by weight up to a target of 80% free-drained container water capacity using reverse osmosis water. Subsequently, pots were watered every 1-3 days. Approximately once per week they were watered to their target weight and re-randomized within their blocks. Other waterings were done by weighing 3-5 pots from across the table and watering based on the lower end of water use. Following the second biomass harvest (70 days after planting) +N and –N pots were watered separately due to substantially higher water use of the +N pots. Pots were initially housed in a greenhouse with ventilation but no cooling system, and experienced water stress on sunny days. After the first biomass harvest, the decision was made to transfer the pots to a greenhouse with a cooling wall, which alleviated the water stress.

2.3.3 Sample Collection and Analysis

Four aboveground biomass harvests were conducted 42, 70, 95, and 123 days after planting (25-28 days between harvests) by cutting ryegrass plants just above the soil surface. Biomass was dried for a minimum of 48 h at 65°C and weighed. Biomass samples were ground using a coffee grinder for tissue P and Kjeldhal N determination. Sulfuric acid-peroxide wet-oxidation digestion was conducted on 0.4 g of each sample, and P was determined colorimetrically using the molybdate-blue ascorbic acid method, as described by Akinremi et al. (2003). Kjeldhal N in the digested samples was determined by a Technicon AutoAnalyzer II.

2.3.4 Statistical Analysis

A two-way analysis of variance (ANOVA) was conducted using PROC GLIMMIX and PROC MIXED of SAS software version 9.4, using the Tukey-Kramer multiple comparison procedure to compare means when the ANOVA indicated a significant treatment effect (SAS

Institute, 2017). Uniformity of variance was tested using COVTEST and comparison of AIC values for different models, and non-homogenous variances were accommodated as applicable using a Random _residual_/group statement and DDFM = Satterthwaite option. By default, PROC GLIMMIX was used, but in some cases PROC GLIMMIX was unable to run when non-homogenous variance was required. In these cases, PROC MIXED was used. Normality of residuals produced by the ANOVA was tested using PROC UNIVARIATE, with a threshold value of 0.9 for the Shapiro-Wilk statistic. Assessment of normality for N uptake revealed two outliers at biomass harvest #2, a frass –N pot with very high N uptake and a MAP +N pot with very low N uptake. These two pots were in the same block, and it was suspected that their N applications had likely been switched. Thus, these pots were removed from analysis. Regressions of tissue P concentration and biomass yield were conducted using PROC REG.

2.4 Results and Discussion

A significant amendment x nitrogen x harvest time effect was found for dry matter yield of ryegrass (Figure 2.1). Yields at the first harvest date were significantly greater overall for the +N treatments, though the difference within amendment was not significant for any of the amendments based on repeated measures analysis. After the first harvest, biomass yields in the –N pots plummeted for all treatments, presumably due to N depletion. Biomass yields in the +N amended pots remained steadily high or increased over time. In the +N pots, biomass yields were numerically higher for all treatments, and statistically higher for struvite and frass compared to the control at harvest times 1 and 2. By the third and fourth harvests, all amended +N pots had statistically greater yield than the control.

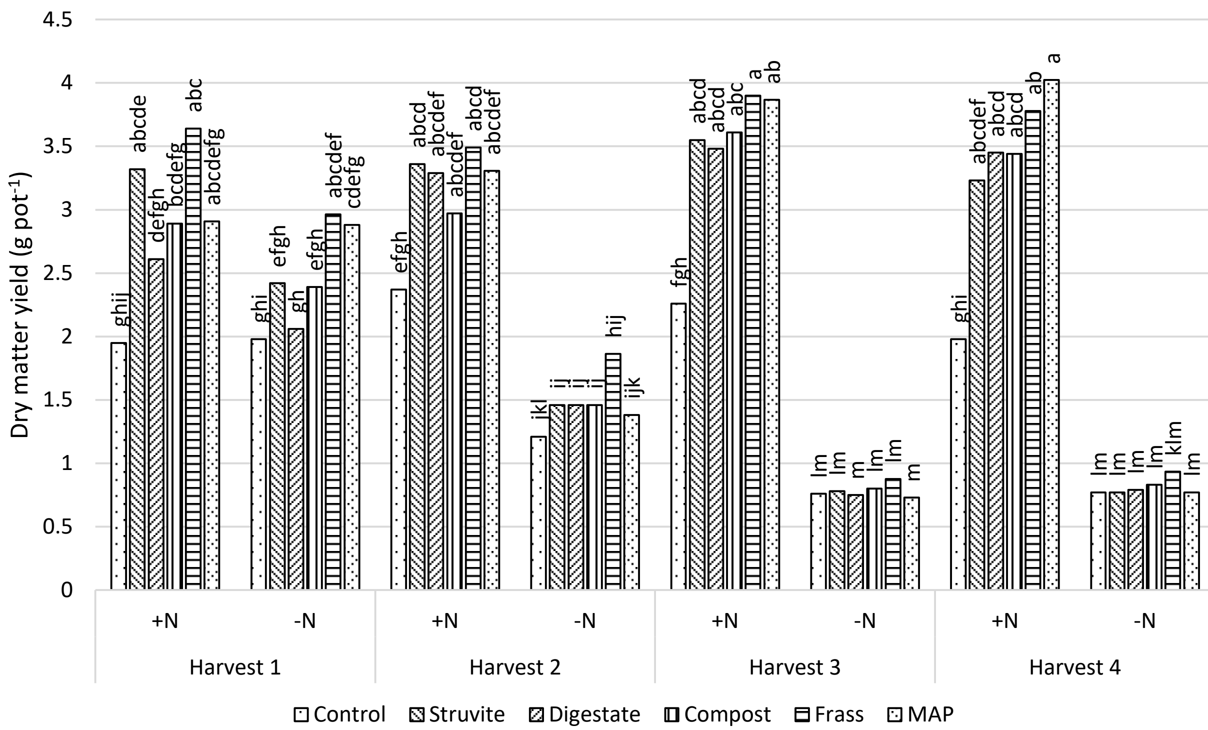

Figure 2.1 Ryegrass biomass at four successive harvests. Bars with at least one letter in common are statistically similar based on repeated measures analysis using Tukey-Kramer means separation and an alpha of 0.05.

Cumulative biomass yields, and cumulative P and N uptakes are presented in Table 2.1. All amendments significantly increased biomass yields relative to the control in the +N pots where N was non-limiting. The increase was between 49% for digestate and 72% for frass. Frass yielded significantly more than the digestate and compost treatments, while MAP and struvite were statistically similar to all amended treatments. P uptake was significantly greater in all +N amendments compared to the +N control, with increases between 70% in digestate and 130% in frass. Unlike biomass, where all amendments were similar to MAP, only frass was similar to MAP based on P uptake. Struvite and digestate had lower P uptake than MAP, and were similar to the compost comparison treatment. This aligns with previous studies that found solid food waste digestate had lower P availability than synthetic fertilizers (Brod et al. 2015) and that struvite P availability is reduced compared to conventional fertilizers at high pH (Hertzberger et al. 2020). These results support our hypothesis that circular nutrient sources would increase P uptake and improve yields compared to the control in a P-depleted soil, when N was non-limiting.

Table 2.1 Mean cumulative biomass yields, tissue P uptake, and Kjeldhal N uptake for ryegrass per pot.

		Cumulative biomass (g)		Total P Uptake (mg pot^{-1})		Total N Uptake (mg pot^{-1})	
+ Nitrogen	Control	8.6	c*	10.0	f	271	d
	Struvite	13.4	ab	19.5	b	387	abc
	Digestate	12.8	b	17.3	cd	379	bc
	Compost	12.9	b	18.3	bc	367	c
	Frass	14.8	a	23.2	a	426	a
	MAP	14.1	ab	21.6	a	424	ab
- Nitrogen	Control	4.7	g	11.6	f	89	g
	Struvite	5.4	ef	17.1	cd	98	f
	Digestate	5.1	fg	15.0	e	93	fg
	Compost	5.5	ef	16.7	d	97	f
	Frass	6.6	d	22.8	a	112	e
	MAP	5.8	e	17.3	cd	97	fg
ANOVA p-values	Amendment	<.0001		<.0001		<.0001	
	Nitrogen	<.0001		<.0001		<.0001	
	Amd *Nit	<.0001		<.0001		<.0001	

* Treatment means within a column followed by the same letter are not significantly different according to the Tukey-Kramer multiple comparison procedure at an alpha of 0.05

In the –N pots all amendments except digestate produced increased cumulative biomass relative to the control. Our secondary hypothesis was that high-N amendments would produce greater yield than low-N amendments in N-limited conditions. The result for frass, which had the highest N:P ratio and resulted in the greatest N uptake and yield among the –N treatments appears to support the hypothesis. However, as non-limiting P was not supplied to the –N pots, we cannot conclusively determine whether yield differences were due to N supply; they may also be the result of P supply during early growth before N was depleted. MAP and struvite, which had very low N:P produced similar yields and N uptake to compost, the other high-N:P amendment. Digestate had intermediate N:P but produced similar yields and N uptake compared to the control in the –N pots. The N in compost and digestate may have been in unavailable forms which the plants could not access.

The presence of adequate N in the +N pots promoted greater cumulative P uptake compared to the –N pots for all amendment treatments except frass and the control, which took up similar total P in both the +N and –N treatments (Table 2.1). Presumably the presence of adequate N allowed the amendments (except frass) to take up more of the available P due to more vigorous plant growth, as indicated by the much higher biomass produced by the +N treatments. The reason that this did not apply to frass is not clear; perhaps the P in frass is so readily available that it was taken up equally with or without sufficient N, or perhaps the N supplied by the frass itself was sufficient to promote P uptake equal to the +N frass treatment.

There is evidence that the P supply from the amendments drove yield differences in the non-N limited +N pots. Statistically significant regressions were found for biomass yields on P tissue concentration for the third and fourth harvests, with adjusted R^2=0.53 for harvest 3, adjusted R^2=0.59 for harvest 4, and $p<0.0001$ for both. Regressions were not significant for the first and second harvests ($p>0.05$). While other attributes of the amendments may also have driven yield differences (e.g., changes in soil properties, undetected micronutrient limitations), these significant regressions indicate that P supply played an important role.

2.5 Conclusions

All three circular nutrient sources in this study showed a capacity to increase P uptake and substantially increase biomass yields relative to the unamended control. In conditions of sufficient soil N, P supplying power decreased in the order frass = MAP > struvite >= compost >= digestate >> control. Notably, all circular amendments produced statistically similar ryegrass biomass yields compared to the soluble MAP fertilizer under greenhouse conditions. Frass may also play an important role as a N source under N-limiting conditions. This research supports the role of these circular nutrients as P sources. Further investigation and validation of these findings is needed under field conditions and with a variety of crop species.

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3 CIRCULAR NUTRIENT SOURCES SUPPLY PHOSPHORUS AND IMPROVE YIELDS IN WHEAT AND ALFALFA ON A LOW P ORGANICALLY MANAGED SOIL OF THE NORTHERN GREAT PLAINS

3.1 Abstract

Circular nutrient sources, which recycle urban nutrients back onto farmland, have the potential to improve phosphorus (P) supply and yields on P-depleted organic farms, reduce global reliance on non-renewable mined phosphate rock, and reduce environmental contamination with excess P. Three circular nutrient sources were compared with common conventional and organic P sources in field experiments in wheat (*Triticum aestivum*) and alfalfa (*Medicago sativa*) on a high pH (8.1-8.3) soil of the Canadian Northern Great Plains. Struvite is a mineral fertilizer precipitated from municipal wastewater, frass is the waste product of insects fed urban food waste, and digestate is anaerobically digested food waste. The common conventional P source was mono-ammonium phosphate (MAP) and the common organic P source was an aerobic compost. Experiments were conducted on an extremely low P site (3 mg kg^{-1} Olsen P) in 2019 and 2020. Conventional and organically-bred wheat genotypes were used and new experiments were established each year. Measurements included wheat growth, P uptake, and grain yield. In wheat, P uptake was on the order frass >= MAP >= compost >= digestate > struvite = unfertilized, and frass and digestate increased growth and yields compared to the control by 40% and 20%, respectively. Some genotype differences were observed, with the organically selected genotype producing more total biomass but similar yield. P treatments were applied to an established perennial alfalfa/grass hay crop and monitored in 2019 and 2020. In alfalfa, P supply was on the order frass >= MAP = digestate = struvite >= compost > unfertilized, and frass, digestate, and struvite increased 2020 alfalfa yields by 137%, 95%, and 126%

respectively. The circular nutrient sources varied in their efficacy in the two crops and relative to other nutrient sources, but all showed some potential to supply P and improve yields on P-depleted organic soils.

3.2 Introduction

Long-term organic management can lead to nutrient deficiency and system collapse over time, particularly due to phosphorus (P) depletion (Welsh et al. 2009). P depletion is apparent on many organically managed farms in the northern Great Plains (Entz et al. 2001) and elsewhere (Gosling and Shepherd 2005). P is a non-renewable resource, with estimates for phosphate rock depletion in as little as 70-140 years (Li et al. 2018). Jarvie et al. (2015) demonstrate P's important and paradoxical role in the food-energy-water nexus, as a necessary resource for food and biofuel production, but as a harmful contaminant to water quality and aquatic ecosystems, causing eutrophication when present in excess. Diverting more food and human waste products back to farms via "circular nutrients", rather than allowing them to enter landfills or waterways, may contribute to the amelioration of all three problems: P deficiency on organic farms, depletion of the phosphate rock resource, and damage to aquatic ecosystems via P-limited eutrophication. Cycling P within the food system is essential to long-term food security and sustainability.

Circular nutrients have received increasing attention from the general public and in academia, as evidenced by inclusion of "circular nutrients" in the European Union's action plan for a "Circular Economy" (European Commision 2015), and many recent publications on productively incorporating food and human organic wastes into the circular economy (e.g., Kibler et al. 2018; Ma and Liu 2019; Case and Jensen 2019). However, the literature addressing the agronomic potential and P characterization of circular nutrients lags far behind that dealing with conventional fertilizer and manure sources (see Table 1.1 in Chapter 1).

Struvite is a hydrated magnesium ammonium phosphate mineral, which in this study was precipitated from municipal wastewater and had a median granule size of 3mm. Struvite is precipitated under alkaline conditions, and has greater solubility with decreasing pH (Bhuiyan et al. 2007). Soil pH was the main determinant of granule dissolution rate in soils with pH between 5.9 and 8.5, where the high pH soils had dissolution rates of less than 1/8 those of the acidic soils (Degryse et al. 2017). A meta-analysis found a decreasing crop response ratio of struvite relative to ammonium phosphates and superphosphates with increasing pH in both field and pot experiments, and that response ratios were greater than 1 in acidic soils and less than 1 in alkaline soils (Hertzberger et al. 2020). However, the pot experiment described in Chapter 2, using high-pH soil from the field site in the present study, found similar Italian ryegrass (*Lolium multiflorum*) yields from struvite and MAP, though cumulative P uptake was slightly lower for struvite. Theissen Martens & Entz (unpublished data, 2017) found a significant response of wheat to struvite fertilizer in one of two years in the high-pH, P-depleted soil of the field site used in the present experiments.

The frass in these experiments is the waste product of black soldier fly larvae (*Hermetia illucens*) that have been used to process urban food waste. Black soldier fly larvae have been shown to be effective processors of organic waste due to their ability to feed on a wide variety of organic material (Čičková et al. 2015). Research into biodegradation of organic wastes by insect larvae is generally focussed on production of larval biomass for use in livestock and pet foods; insect frass is poorly studied as a soil amendment. In fact, we are aware of no studies which have specifically studied the P supplying properties of frass; previous studies have evaluated frass on a nitrogen (N) basis or as a component of a potting medium. Kebli and Sinaj (2017) conducted a pot study on three soils using lettuce and ryegrass, and found that frass from food waste consistently improved yields over the control. However, it was only in the sandy, low-pH soil that yields were similar to or greater than the mineral fertilizer. The pot study we conducted on high pH clayey soils from this experiment's field site revealed similar yields and P uptake of frass compared to MAP (Chapter 2). Setti et al. (2019) evaluated black soldier fly processing residue (frass and bedding) mixed with peat moss as a greenhouse growing medium at rates between 10% and 40% by volume. High rates of frass reduced emergence in lettuce, basil, and tomato, and the best yields were generally found for 10% frass/90% peat moss and for 100% peat moss with mineral fertilizer.

The anaerobic digestate in these experiments was industrially produced through the decomposition of food retail and food processing waste in the absence of oxygen. Anaerobic digestion produces methane biogas, which can be used directly in electricity and heat generation, or upgraded to be used as a vehicle fuel or injected into natural gas lines (Wainaina et al. 2020). Liquid and solid fractions of digestate can be applied together or separately, with solid fractions tending to have higher P concentrations than liquid (Nkoa 2014). The dried and pelletized solid fraction of the digestate was used in this study. P characterization of the solid fraction of food waste digestate is limited to one study to the best of our knowledge, which conducted P speciation of alternative fertilizers including a solid and a liquid food waste digestate. A higher proportion of chemically recalcitrant species, and especially Ca-associated phosphates, was present in solid digestate compared to liquid digestate. The solid digestate had lower P recovery in a pot experiment compared to liquid digestate and mineral fertilizer in both acidic (pH 5.5) and near-neutral (pH 6.9) soils (Brod et al. 2015). In the higher pH soil, P recovery from several circular nutrient sources was negatively related to the acid-soluble Ca-associated fraction of P. The pot study we conducted found similar yield but lower P uptake from digestate compared to mineral fertilizer (Chapter 2).

The current study investigated the agronomic potential, and especially the P-supplying ability, of struvite, frass, and digestate for a high-pH, P-depleted organic soil on the Canadian Prairies, and compared them with common conventional and organic P sources. The typical organic P-source was aerobically composted organic wastes, with horse manure and bedding as the primary substrates, along with shredded wood and bark, hatchery waste, yard waste, and waste protein powder. The typical conventional P-source was a soluble synthetic fertilizer, MAP. An unfertilized control was included for reference. P content and availability of organic wastes varies widely with substrate source and processing (Sharpley and Moyer 2000), but composted organic waste usually has good P availability compared to synthetic fertilizers. A pot study with five soils showed similar or greater yields from composted chicken manure than MAP, where nutrients were applied at a P rate, and N and potassium were corrected for with synthetic fertilizer (Ramphisa et al. 2020). Another study found similar P uptake from composted poultry manure and triple super phosphate (Sikora and Enkiri 2005). A pot study with canola found greater cumulative P uptake from composted dairy cattle manure than soluble monopotassium phosphate (Zvomuya et al. 2006). A contrasting experiment found less than 1/3 the apparent P recovery from a composted garden waste compared to triple super phosphate in three soils (Christiansen et al. 2020). We hypothesized that the circular nutrient sources would increase P uptake and crop yields compared to the control, and that they would be similarly effective compared to the conventional MAP fertilizer and the organic compost in the study site's high pH soil. Testing the amendments in an annual (wheat) and a perennial (forage) system allows them to be evaluated in an early succession versus a later succession cropping system. We hypothesized that circular nutrients would supply P to crops equally well in annual wheat (early successional) and perennial forage (later successional) cropping systems.

3.3 Methods

3.3.1 Materials

Properties of the organic-matter based amendments (frass, digestate, and compost) were determined by Agvise Laboratories, ND, USA; the N and P content of the mineral struvite and MAP were based on manufacturer specifications (Table 3.1). The P content of the amendments varied by two orders of magnitude, with the organic matter amendments containing less P than the mineral amendments. Among the organic amendments, digestate contained approximately 12 times, and frass contained almost 4 times, the P contained in the organic comparison, compost. Struvite contained approximately half the P contained in the conventional comparison fertilizer, MAP. The amendments also varied in N content and N:P ratio, with frass and compost having the greatest N:P, the mineral amendments struvite and MAP having very low N:P, and digestate being intermediate. All amendments were applied based on a P rate of 20 kg ha^{-1} P, resulting in varied rates of N addition.

Table 3.1 Amendment properties. For frass, digestate, and compost, values are “as received”.

Measurement	Unit	Frass	Digestate	Compost	Struvite	MAP
Total Nitrogen	g kg^{-1}	32	38	7.1	50	110
Total Phosphorus	g kg^{-1}	8.74	27.53	2.27	120	230
N:P ratio		3.7	1.4	3.1	0.4	0.5
Potassium	g kg^{-1}	8.7	4.0	5.0	-	-
Sodium	g kg^{-1}	4.4	4.3	1	-	-
Calcium	g kg^{-1}	2.5	69	63	-	-
Magnesium	g kg^{-1}	5.2	12	21	-	-
Zinc	mg kg^{-1}	84	360	140	-	-
Iron	mg kg^{-1}	871	18125	5852	-	-
Manganese	mg kg^{-1}	87	369	295	-	-
Copper	mg kg^{-1}	12	96	21	-	-
Sulfur	g kg^{-1}	3.5	14	1.4	-	-
Water Content	g kg^{-1}	90	80	180	-	-
Carbon	g kg^{-1}	400	350	89	-	-

3.3.2 Field Site

The experiments were conducted in 2019 and 2020 at an organically-managed field site near Libau, MB (50°24’01” N, 96°72’95” W), which has a cool subhumid continental climate. The soil is a Gleyed Rego Black Chernozem with a clay texture. Spring soil test results were determined by Agvise Laboratories, ND, USA (Table 3.2). All fields had very low Olsen-P concentrations of 2-3 mg kg^{-1} soil, caused by the site history of alfalfa hay removal since 2006 with no external nutrient addition. In 2017 the wheat experiment areas were converted to annual cropping. Legume-cereal green manures were grown in the year prior to the wheat experiments in order to fix and supply N to the wheat crop through mineralization. The 2018 green manure was terminated and incorporated in the fall of 2018 prior to the 2019 wheat experiment. Heavy fall rains prevented incorporation of the 2019 green manure until the spring of 2020, also leading to clodding and poor seedbed quality. Spring incorporation of green manures may account for the lower nitrate values in 2020, as the green manure had not had time to mineralize.

Table 3.2 Soil test properties

Soil Characteristic	Unit	Alfalfa (2019-2020)	Wheat (2019)	Wheat (2020)
Nitrate 0-15cm	kg ha^{-1}	19	35	22
Nitrate 15-60cm	kg ha^{-1}	20	40	24
Nitrate 0-60cm	kg ha^{-1}	39	75	46
Olsen Phosphorus 0-15cm	mg kg^{-1}	2	3	3
Potassium 0-15cm	mg kg^{-1}	269	312	248
Chloride 0-60cm	kg ha^{-1}	40	40	45
Sulfur 0-15cm	kg ha^{-1}	20	27	29
Sulfur 15-60cm	kg ha^{-1}	74	54	61
Sulfur 0-60cm	kg ha^{-1}	94	81	90
Boron 0-15cm	mg kg^{-1}	1.6	1.3	1.3
Zinc 0-15cm	mg kg^{-1}	0.59	0.6	0.51
Iron 0-15cm	mg kg^{-1}	17.7	18.7	22.5
Manganese 0-15cm	mg kg^{-1}	2.8	1086	3.1
Copper 0-15cm	mg kg^{-1}	1.07	0.96	0.76
Magnesium 0-15cm	mg kg^{-1}	1381	1086	1013
Calcium 0-15cm	mg kg^{-1}	5800	6622	6858
Sodium 0-15cm	mg kg^{-1}	16	20	25
Organic Matter (0-15cm)	g kg^{-1}	52	54	40
pH 0-15cm		8.3	8.1	8.1

The alfalfa trial was established on a 13-y-old alfalfa/grass hay stand. Hay was usually harvested 1-2 times per year, and no external nutrient addition occurred during this period. Grass species included orchardgrass (*Dactilus glomerata*) and quackgrass (*Elymus repens*).

A rain gauge was installed and monitored between planting and harvest of the wheat crop in both years (Table 3.3). “Normal” precipitation was based on 30-year averages from Selkirk weather station (8.3 km from field site, 50°17’71” N, 96°79’28” W). 2019 was extremely dry, with precipitation during the wheat growing season only 39% of the long-term average, and even lower in the early season. 2020 was also drier than average, though it had close to normal precipitation early in the season.

Table 3.3 2019 and 2020 precipitation summary, based on sampling dates for wheat and alfalfa. Cumulative values are recorded from the wheat planting date, May 16 in 2019 and May 29 in 2020. Selkirk is the nearest long-term climate monitoring station at 8.3 km away.

2019

Date	Cumulative Rainfall from May 16 (mm)	Selkirk Normal from May 16 (mm)	% of Normal	Sample Taken
27-Jun-19	34	112	30%	Wheat Biomass 1
04-Jul-19	34	131	26%	Alfalfa Biomass 1
16-Jul-19	70	159	44%	Wheat Biomass 2
07-Aug-19	82	209	39%	Wheat Biomass 3
20-Aug-19	94	244	39%	Alfalfa Biomass 2
23-Aug-19	98	250	39%	Wheat Harvest

2020

Date	Cumulative Rainfall from May 29 (mm)	Selkirk Normal from May 29 (mm)	% of Normal	Sample Taken
21-Jun-20	68	66	103%	Alfalfa Biomass 3
02-Jul-20	82	96	86%	Wheat Biomass 1
16-Jul-20	102	129	79%	Wheat Biomass 2
28-Jul-20	102	153	67%	Alfalfa Biomass 4
24-Aug-20	168	221	76%	Wheat Biomass 3
10-Sep-20	204	255	80%	Wheat Harvest

3.3.3 Wheat Field Experiment

A randomized complete block design with four blocks (replications) was used for the wheat experiments in 2019 and 2020. The treatment layout was factorial with amendment (struvite, frass, digestate, MAP, compost, unfertilized control) and wheat genotype (AAC Brandon and BJ08-IG) as the two factors. AAC Brandon is conventionally bred and is the most popular wheat variety in Manitoba by area planted (Manitoba Agricultural Services Corporation 2018). BJ08-IG is a genotype selected through a partnership between professional plant breeders and organic farmers in a participatory plant breeding program. Farmers grew out and selected lines from crosses made by professional breeders (Entz et al. 2018). BJ08-IG was selected by a farmer under low-P conditions. A previous study, which included a comparison of BJ08-IG and AAC Brandon, showed that BJ08-IG produced taller plants with greater biomass and similar days to maturity, as well as greater yields in 2 of 3 years (Entz et al. 2018). Previous experiments at this field site had suggested that a line selected on-site (not BJ08-IG) responded more to struvite application than a conventional line (Thiessen Martens and Entz, unpublished data). We hypothesized that BJ08-IG, a line selected under low-P organic conditions, would respond more to circular nutrient addition than the conventionally bred AAC Brandon.

Fields were tilled with a light-duty cultivator (2019) or a heavy-duty cultivator followed by light-duty (2020) depending on field conditions within 5 d prior to seeding. Amendments were broadcast by hand into 2 m x 10 m plots and incorporated with a rototiller to approximately 10 cm immediately prior to wheat planting. Wheat was planted May 16 in 2019 and May 29 in 2020 at a target depth of approximately 5 cm with 15 cm row spacing using a Fabro plot seeder equipped with a seed distribution cone (Fabro Industries, Swift Current, SK, Canada). In 2019 a target seeding rate of 350 plants m^{-2} was used. In 2020 the target seeding rate was increased to 450 plants m^{-2} to compensate for poor seedbed quality due to clay clods.

Plant population density measurements were taken in a 0.3 m^2 area (2 rows x 1m) after full emergence. A 0.3 m^2 (2 rows x 1m) biomass sample was taken at stem elongation and anthesis on June 27 and July 16 in 2019 and July 2 and July 16 in 2020. A 0.9 m^2 (6 rows x 1m) biomass sample was taken at physiological maturity on August 7 in 2019 and August 24 in 2020. Biomass samples were oven-dried at 65°C for a minimum of 48 hours before weighing. Wheat grain was harvested on August 23 in 2019 using a Wintersteiger plot combine (Wintersteiger Model “Classic”, Saskatoon, Canada) and on September 10 in 2020 using a Hege plot combine

(Hege model 125, Hege Company, Waldenburg, Germany). Grain samples were air dried on forced-air beds for a minimum of 72 h before weighing.

3.3.4 Alfalfa Field Experiment

A one-factor randomized complete block design with four blocks (replicates) and 2 m x 10 m plots was used for the alfalfa experiment. Amendment was the only factor and included 6 levels, as in the wheat experiment. All amendments except struvite were surface broadcast on May 16 and 17 of 2019. Struvite was subsurface banded into the soil at a depth of approximately 5 cm using the Fabro seeder on May 16 due to its low solubility in near-neutral pH rainwater. Struvite broadcast on the surface would be unable to dissolve and infiltrate the soil to a significant degree over the duration of the experiment.

Four biomass samples were taken over the course of the 2019 and 2020 growing seasons to roughly correspond with farmer hay harvests. A 0.75 m^2 biomass sample was taken on July 4, 2019 using a 0.25 m^2 quadrat and harvesting all plant material within the quadrat with a hand sickle. Samples were oven-dried at 65°C for a minimum of 48 h and weighed. After observing substantial variability in the first biomass, a Kohler Lawn Boy 0.5 m wide push mower with a bag attachment was used for all subsequent biomass samples, which allowed a greater sample area to be harvested. Some plots in the first and third replicates had areas of winter-kill with no alfalfa plants on one side. In these cases, the plots were trimmed to exclude the winter-killed areas. The distance between the trimmed areas was measured. Two passes were made up and down the plots with the push mower, avoiding the edges of the plots. The total wet biomass from each plot was weighed, mixed to homogenize, and subsampled. The subsample was weighed and oven-dried at 65°C for a minimum of 48 h before weighing again to determine moisture content. Biomass samples were taken using the push mower on August 20, 2019, June 21, 2020, and July

28, 2020. On August 20, 2019, the lowest mower setting was used in order to cut biomass near the soil surface. However, this resulted in the blade occasionally cutting into soil where the ground was uneven. 2020 biomass samples were taken with the blade raised to approximately 5 cm off the ground to avoid interaction with the soil.

3.3.5 P and N Determination

Biomass samples were ground using a Wiley mill with a 2 mm mesh and wheat grain samples were ground using a Cyclone sample mill with a 1 mm mesh. A 0.4 g subsample of each ground sample was hot-digested using a sulfuric acid-hydrogen peroxide solution, and P was determined colorimetrically using the molybdate-blue ascorbic acid method, as described by Akinremi et al. (2003). Kjeldahl N of the digested samples was determined on a Technicon Autoanalyzer II. For wheat grain samples only, an approximate protein content was determined by multiplying Kjeldahl N content by a protein factor of 5.7 (McDonald 1977). P and N uptake were determined by multiplying nutrient concentration by biomass/grain yield. Apparent P recovery from amendments was calculated as:

$$P\ recovery\ (\%) = \frac{P\ uptake_{amendment} - P\ uptake\ _{unfertilized}}{P\ applied}\ x\ 100$$

P applied was 20 kg P ha^{-1} for all amendments.

P tissue concentration and uptake from the physiological maturity biomass were measured, but the data are excluded from this evaluation. The calculated whole plant maturity biomass P uptakes were consistently lower than those calculated for P uptake in grain, which is a logical impossibility. We assume that this was caused by oversampling of less-dense low-P straw and under-sampling of more-dense high-P grain in imperfectly homogenized ground samples.

This resulted in a systematic underestimation of tissue P and P uptake for the whole-plant analysis. This issue limits our ability to calculate whole-plant P recovery.

3.3.6 Statistical Analysis

Analysis of variance was conducted using PROC GLIMMIX in SAS software version 9.4 (SAS Institute, 2017). For the wheat experiments, amendment, genotype, and amendment xgenotype were fixed effects, and block(year) and year were random effects. Each sampling time was analysed independently. For alfalfa, amendment was a fixed effect and block was a random effect. Normality of residuals produced by the model were tested with PROC UNIVARIATE, with Shapiro-Wilk values of greater than 0.9 considered indicative of near-normal data. Homogeneity of variance was tested by comparing Akaike Information Criterion (AIC) values of models with and without heterogenous variance; heterogenous variance was included in the model when it produced the lowest AIC. Analysis of variance for P recovery as a percentage of applied P was conducted using a beta distribution. Regressions between tissue nutrient concentration at each sampling time and grain or biomass yield were performed using PROC REG. Despite year being considered a random variable in analysis of variance for response variables, separate regressions were performed for each year because samples were at slightly different days after planting between years. P concentration naturally decreases through the growing season (Tomasiewicz 2000), so that if crops aren’t at precisely the same stage, combining the regressions confounds results, as the more advanced plants have higher biomass but lower P concentration due simply to their more advanced growth.

3.4 Results and Discussion

3.4.1 Wheat

Plant counts were analyzed separately for 2019 and 2020, due to the poor seedbed quality observed in 2020 compared to 2019. In 2019 there was no significant amendment, variety, or amendment x variety effect on seedling emergence, and mean plant counts were 321 plants m^{-2} with a coefficient of variation (CV) of 15%. These results show that none of the circular nutrient sources negatively affected crop establishment. In 2020, there was a significant effect of variety on plant count, with higher plant counts in BJ08-IG (302 plants m^{-2}, CV 16%) compared to Brandon (271 plants m^{-2}, CV 21%). Brandon, as a semi-dwarf variety, may have a shortened coleoptile (Tang et al. 2009), which could cause reduced emergence in the poor seedbed conditions in 2020. In fact, screening conducted in our lab of farmer-selected lines compared to conventionally-bred checks revealed that Brandon's coleoptile was significantly shorter than that of BJ08-IG by an average of 1.3cm (unpublished data). Mean wheat yields were 3071 kg ha^{-1}, CV 20% in 2019 and 2106 kg ha^{-1}, CV 13% in 2020. Previous small-plot research studies in this region found organic wheat grain yields ranging widely between 441 kg ha^{-1} and 3430 kg ha^{-1} (Welsh et al. 2009; Cicek et al. 2014; Halde et al. 2015). Therefore, wheat grain yields in this study represent moderate to high organic wheat yields for small-plot studies in this region, despite below-average growing season precipitation in both years.

For wheat biomass and grain yield, a significant genotype effect occurred at all biomass samplings but not at grain yield. A significant amendment effect occurred at all sampling times, and a significant genotype x amendment effect occurred at stem elongation (Table 3.4). A significant amendment effect was found for both tissue P concentration and P uptake at every sampling time (Table 3.4). A significant genotype effect occurred in tissue P concentration at stem elongation, and grain yield, while a significant effect of P uptake occurred for anthesis biomass and grain yield (Table 3.4).

3.4.1.1 Genotype effect on biomass and phosphorus uptake. BJ08-IG produced similar grain yield to Brandon, but greater biomass at all biomass sampling times. Greater biomass at physiological maturity but similar grain yield indicates greater straw production by BJ08-IG compared to Brandon. BJ08-IG also had significantly greater plant height (81 cm, CV 6%) than Brandon (64 cm, CV 7%) at physiological maturity.

The results for genotype effect align with a previous survey of farmer-selected lines compared to conventional checks which included BJ08-IG and Brandon (Entz et al. 2018). That study showed that BJ08-IG was an average of 11 cm, 19 cm, and 25 cm taller than Brandon in three of three site-years, and that BJ08-IG produced 25% and 32% greater biomass at maturity compared to Brandon in two of two site years. BJ08-IG also produced 35% and 52% greater yields at two of three site-years, and statistically similar yields at the third site-year.

BJ08-IG had lower tissue P concentration but similar P uptake to Brandon at stem elongation, which is consistent with a dilution effect as BJ08-IG also had greater biomass at these times. However, BJ08-IG had higher P concentration and uptake at anthesis and in the grain. Greater P acquisition by BJ08-IG may have helped to promote higher biomass yields, but high P levels in grain may not be a desirable trait.

Adequate P is necessary for seedling vigour if the crop is replanted as seed (Bolland and Baker 1988), but seed P stored as phytate is not digestible by humans or other mono-gastric animals, and most P exported from cropland is released to the environment (Veneklaas et al. 2012). It may be that the selection environment of BJ08-IG in a low-P soil induced greater P allocation to the grain to promote future seedling vigour. This adaptation, while possibly beneficial for seedling vigour in the short term, would hasten P-depletion from soils. Further investigation is needed to determine whether genotypes selected in low-P conditions allocate a greater proportion of P to the grain compared to conventionally bred varieties.

Table 3.4 Yield, tissue P concentration, and P uptake for 3 biomass samples and grain harvest sample. Standard error of the mean (SEM) is given in brackets.

		Stem Elongation									**Anthesis**								
		Biomass kg ha^{-1}			**Tissue P mg kg^{-1}**			**P uptake kg ha^{-1}**			**Biomass kg ha^{-1}**			**Tissue mg kg^{-1}**			**P uptake kg ha^{-1}**		
	Brandon	840	(110)	B	2160	(430)	A	1.8	(0.2)		2930	(110)	B	1490	(250)		4.4	(0.7)	B
	BJ08-IG	940	(110)	A	1930	(430)	B	1.8	(0.2)		3200	(110)	A	1460	(250)		4.7	(0.7)	A
	Unfertilized	620	(110)	D	1720	(430)	D	1.0	(0.2)	D	2410	(140)	E	1260	(260)	C	3.0	(0.7)	D
	Struvite	730	(110)	CD	1870	(430)	CD	1.3	(0.2)	CD	2700	(140)	DE	1370	(260)	BC	3.7	(0.7)	CD
	Digestate	770	(110)	C	1990	(430)	BC	1.5	(0.2)	C	2880	(140)	CD	1480	(260)	B	4.2	(0.7)	BC
	Compost	980	(110)	B	2060	(430)	BC	2.0	(0.2)	B	3170	(140)	BC	1460	(260)	B	4.6	(0.7)	B
	Frass	1120	(110)	A	2500	(430)	A	2.8	(0.2)	A	3470	(140)	AB	1770	(260)	A	6.1	(0.7)	A
	MAP	1100	(110)	AB	2160	(430)	B	2.3	(0.2)	AB	3750	(140)	A	1490	(260)	B	5.6	(0.7)	A
Brandon	Unfertilized	530	(110)	E	1840	(430)		1.0	(0.2)		2390	(180)		1310	(260)		3.1	(0.7)	
	Struvite	700	(110)	DE	1970	(430)		1.3	(0.2)		2480	(180)		1370	(260)		3.4	(0.7)	
	Digestate	720	(110)	DE	2140	(430)		1.5	(0.2)		2780	(180)		1470	(260)		4.0	(0.7)	
	Compost	900	(110)	BCD	2180	(430)		1.9	(0.2)		3060	(180)		1520	(260)		4.6	(0.7)	
	Frass	1020	(110)	ABC	2570	(430)		2.6	(0.2)		3160	(180)		1730	(260)		5.4	(0.7)	
	MAP	1160	(110)	A	2290	(430)		2.6	(0.2)		3690	(180)		1520	(260)		5.5	(0.7)	
BJ08-IG	Unfertilized	700	(110)	DE	1600	(430)		1.1	(0.2)		2430	(180)		1200	(260)		2.9	(0.7)	
	Struvite	760	(110)	D	1760	(430)		1.3	(0.2)		2910	(180)		1380	(260)		3.9	(0.7)	
	Digestate	810	(110)	CD	1840	(430)		1.4	(0.2)		2970	(180)		1480	(260)		4.4	(0.7)	
	Compost	1050	(110)	AB	1940	(430)		2.0	(0.2)		3280	(180)		1410	(260)		4.6	(0.7)	
	Frass	1230	(110)	A	2430	(430)		3.0	(0.2)		3780	(180)		1820	(260)		6.9	(0.7)	
	MAP	1050	(110)	AB	2030	(430)		2.1	(0.2)		3820	(180)		1470	(260)		5.6	(0.7)	
p-Value	Genotype	0.0004			<.0001			0.92			0.0021			0.39			0.03		
	Amendment	<.0001			<.0001			<.0001			<.0001			<.0001			<.0001		
	Geno*Amd	0.0103			0.93			0.13			0.3903			0.36			0.06		

*Means followed by the same letter, within the same column and factor, are not significantly different based on the Tukey multiple comparison procedure at an alpha of 0.05

Table 3.4 continued

		Maturity Biomass kg ha^{-1}			Yield kg ha^{-1}			Grain Tissue P mg kg^{-1}			P uptake kg ha^{-1}		
	Brandon	5310	(290)	B	2590	(480)		3170	(220)	B	8.4	(2.2)	B
	BJ08-IG	5670	(290)	A	2600	(480)		3360	(220)	A	8.9	(2.2)	A
	Unfertilized	4550	(310)	D	2100	(480)	D	2940	(230)	C	6.3	(2.2)	D
	Struvite	5030	(310)	CD	2340	(480)	CD	3100	(230)	C	7.3	(2.2)	D
	Digestate	5350	(310)	BC	2520	(480)	BC	3310	(230)	B	8.4	(2.2)	C
	Compost	5780	(310)	AB	2710	(480)	AB	3330	(230)	B	9.1	(2.2)	BC
	Frass	6090	(310)	A	2950	(480)	A	3570	(230)	A	10.7	(2.2)	A
	MAP	6140	(310)	A	2940	(480)	A	3330	(230)	B	9.9	(2.2)	AB
Brandon	Unfertilized	4570	(340)		2050	(490)		2860	(230)		5.9	(2.2)	
	Struvite	4710	(340)		2280	(490)		2950	(230)		6.8	(2.2)	
	Digestate	5180	(340)		2470	(490)		3200	(230)		8.0	(2.2)	
	Compost	5540	(340)		2830	(490)		3290	(230)		9.4	(2.2)	
	Frass	5920	(340)		2910	(490)		3450	(230)		10.1	(2.2)	
	MAP	5930	(340)		2990	(490)		3230	(230)		9.8	(2.2)	
BJ08-IG	Unfertilized	4520	(340)		2160	(490)		3030	(230)		6.7	(2.2)	
	Struvite	5350	(340)		2390	(490)		3240	(230)		7.8	(2.2)	
	Digestate	5530	(340)		2560	(490)		3410	(230)		8.8	(2.2)	
	Compost	6020	(340)		2590	(490)		3370	(230)		8.8	(2.2)	
	Frass	6270	(340)		3000	(490)		3700	(230)		11.3	(2.2)	
	MAP	6360	(340)		2880	(490)		3420	(230)		10.0	(2.2)	
p-Value	Genotype	0.0009			0.8547			<.0001			0.0134		
	Amendment	<.0001			<.0001			<.0001			<.0001		
	Geno*Amd	0.5696			0.3863			0.62			0.1634		

*Means followed by the same letter, within the same column and factor, are not significantly different based on the Tukey multiple comparison procedure at an alpha of 0.05

3.4.1.2 Amendment effect on biomass and phosphorus uptake. All amendments except struvite produced greater biomass and grain yields compared to the unfertilized control at every sampling time; yield from the struvite treatment was numerically greater but statistically similar to the control at all sampling times (Table 3.4). Digestate gave numerically lower yield but was statistically similar to the compost organic comparison treatment at anthesis, maturity, and grain yield; digestate yield was statistically lower than compost yield at the earliest sampling time, stem elongation. Digestate always yielded statistically less than the conventional comparison treatment, MAP. Frass produced statistically greater early-season stem elongation biomass than compost, and similar biomass/grain yield at subsequent samplings. Frass was the only amendment that was consistently statistically similar to the conventional MAP treatment.

Frass consistently had higher tissue P concentrations than all other amendments, and greater P uptake than all amendments except MAP, which it is similar to at all stages. Digestate produced greater P concentration and uptake than the unfertilized control at all sampling times, and was statistically similar to compost at all times after stem elongation. Struvite had numerically greater, but statistically similar tissue P concentration and P uptake compared to the control at all sampling times.

Previous experiments at this site had found a significant yield response of wheat to 20 kg P ha^{-1} of struvite in one of two years. (Thiessen Martens & Entz, 2017, unpublished data). Previous experiments utilized struvite banded with the seed, whereas in the present study struvite was broadcast. While the authors are unaware of any experiments comparing banded versus broadcast struvite, it is well established that banding conventional P fertilizers improves their efficiency compared to broadcast (Grant and Flaten 2019). It may be that banding of struvite is required to enhance wheat access, particularly in high-pH soils where the struvite dissolution rate is much lower than in acidic soils (Degryse et al. 2017).

The lower yield response and P uptake of wheat by digestate compared to compost (at stem elongation only), frass, and MAP may be explained by a high proportion of the P existing in poorly soluble fractions. Brod et al. (2015) characterized several circular P sources, including liquid and solid food waste digestate, based on a modified Hedley fractionation, XRD, and ^{31}P MAS_NMR. Only 21.9% of the solid digestate fraction P was H_2O or $NaHCO_3$ extractable, compared to 38.3% in the liquid digestate. The solid digestate had a particularly high proportion of acid (HCl)-soluble recalcitrant P, which may be less available in high soil pH like that found in the present study. This led to lower P uptake of the solid digestate compared to the liquid digestate in an associated ryegrass pot study. A contrasting study found that the soluble fraction of P based on Hedley fractionation was not different between liquid, solid, and solid-dried fractions of dairy slurry/maize silage and mixed crop silage digestates (Bachmann et al. 2016). Further work is needed to determine whether food waste is consistent in having less available P in its solid fraction, or whether this property is simply variable among all digestates.

The statistical similarity between frass and MAP in terms of biomass/grain yield and P uptake at all sampling times indicates that the P in frass and the P in MAP have similar availability to wheat, at least over one growing season. Insufficient research has been done on the P properties of food waste-derived frass to compare this experiment to previous findings. This suggests a need for future research characterizing P in frass in relation to its fertilizer properties.

3.4.1.3 Genotype x Amendment effect on biomass, plant height, and phosphorus uptake. A significant genotype by amendment effect on biomass was found at stem elongation, the earliest sampling time. The moderately conservative Tukey multiple comparison procedure found no significant differences between the two varieties within the same amendment. However, all amendment treatments produced numerically higher biomass with the BJ08-IG genotype compared to Brandon, except for the conventional comparison, MAP, which may explain the significant interaction effect found by the ANOVA. However, this genotype x amendment interaction did not persist later in the season in this experiment.

In the relatively cold soils of the Northern Great Plains, early-season P uptake is particularly important to obtaining optimum yields (Grant et al. 2001). While the interaction was observed only in early-season biomass production and not P uptake or final grain yield, this suggests a need for further investigation of how organically-selected genotypes may respond differently to amendments compared to conventionally bred varieties. There is some early evidence for wheat genotypes selected under different management having different yield responses to compost in an organically managed system (Schmidt et al. 2018), suggesting the potential to breed genotypes which access alternative nutrient sources more efficiently.

3.4.1.4 Phosphorus recovery. P recovery was significantly affected by amendment at stem elongation, anthesis, and grain harvest (maturity biomass not analysed due to unreliability of P uptake values). The performance of the amendments relative to each other was fairly consistent at each sampling time, though the means separation changed somewhat (Figure 3.1). P recovery in grain was on the order frass >= MAP >= compost >= digestate >= struvite, with uptake efficiencies ranging between 5% and 22%. Except for struvite, these recoveries are in line with or higher than those typically found for soluble P fertilizers in the year of application of 10-20% (Chien et al. 2011), especially when considering that this P recovery includes only the grain and not total aboveground biomass or roots.

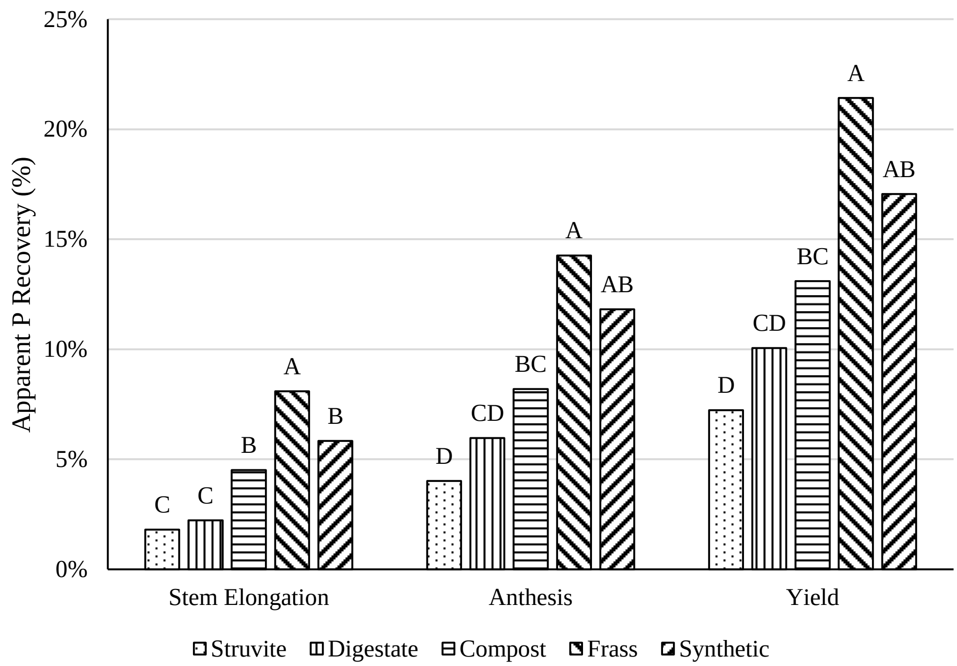

Figure 3.1 Phosphorus recovery of each amendment (mean of two genotypes), based on 20 kg ha^{-1} applied P. Note that at Anthesis BJ08-IG had significantly greater P recovery than Brandon. No amendment by genotype interaction occurred at any time. Bars at the same sampling time with the same letter are not significantly different based on the Tukey multiple comparison procedure at a α of 0.05.

The genotype effect was significant at anthesis only, where BJ08-IG (11% recovery) had a 49% greater P recovery than Brandon (7.4% recovery). This could be associated with greater above-ground biomass of BJ08-IG (Table 3.4) and perhaps greater root biomass. P uptake is diffusion mediated, with P moving <0.5 mm to the plant root, and is therefore related to root surface area (Barber 1977 in Grant et al. 2001), such that a larger root system in BJ08-IG could explore a greater volume of soil and access more of the amendment P. Root biomass was not directly measured in this study, but above-ground biomass is highly related to root biomass (Qin et al. 2016).

A genotype x amendment interaction was not present at any sampling time, indicating that the two genotypes responded similarly to amendment type.

3.4.1.5 Attribution of yield differences to nutrient supply. While the previously presented data show that the amendments significantly impacted yield, tissue P concentration, and P uptake (Table 3.4), differences in yield could not be directly attributed to the amendments' impact on P nutrition. The importance of P nutrition in driving yields was established by regression relationships between tissue P concentration and grain yields (Figure 3.2). Significant positive relationships between biomass tissue P and grain yield were observed at stem elongation and anthesis in both 2019 and 2020. In addition, a significant positive relationship existed between grain P concentration and grain yield in 2019. This establishes P supply from the amendments as an important factor driving the observed yield differences.

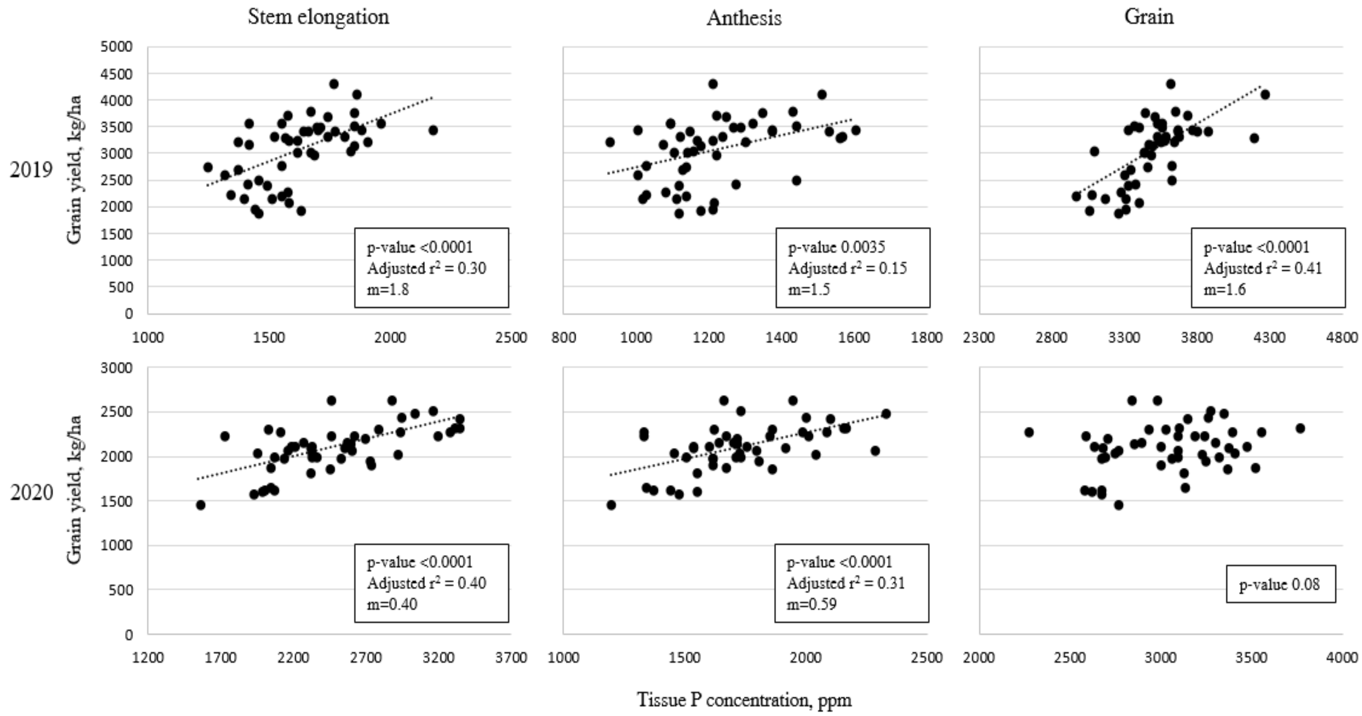

Figure 3.2 Regressions between grain yield and grain/tissue P concentration in 2019 and 2020 (both genotypes). Where the model is significant, as indicated by a p-value <0.05, adjusted r^2 and slope (m) of the trendline are given.

These results align with a previous Manitoba study which showed that final wheat yields were well correlated to P concentration between mid-tillering and flag leaf stage in P-deficient soils at varying fertilizer levels, with the strength of the relationship between whole-plant P concentration and yield declining afterwards (Tomasiewicz 2000). Tissue P concentration decreases over the growing season in wheat, so that the "critical" tissue P concentration required for optimal yield also decreases. Our stem elongation biomass samples in 2019 and 2020 were taken about 35 d after emergence (DAE) and 25 DAE respectively, which meant that the critical P threshold in 2019 was lower than in 2020 because the plants were older. Tomasiewicz (2000) determined critical shoot P concentrations at 35 DAE of 2712 mg kg^{-1}, and at 25 DAE of 3586 mg kg^{-1}. These critical thresholds were not met by any individual sample in either year, though 7 frass samples came close (>3000 mg kg^{-1} P) in 2020. In addition, none of the regression graphs have an obvious "plateau" at higher P concentrations, which would indicate P sufficiency above a critical concentration. Amendment addition of 20 kg P ha^{-1} may have been insufficient to reach maximum yields for any of these amendments in this extremely P-depleted soil, though a plateau would be difficult to detect given the scatter of the data.

In addition to establishing P as a driving factor in yield differences, we needed to establish whether differences in N supplied by the amendments contributed to yield differences. Based on the limited native P supply, and the growth of a N-fixing green manure the year before wheat planting, N was not expected to be the limiting nutrient in this system. This was tested by using regression analysis between the N tissue concentration (converted to % protein for grain) and the biomass yield produced at the same sampling time. Lower tissue N concentration was consistently associated with higher yields, indicating dilution of N (Table 3.5). This supports the conclusion that N did not limit grain yield in this system. Low grain yield is commonly associated with high grain N concentration when yield is limited by another factor, such as drought (Woodard and Bly 1998). In this case the non-N factor limiting yield was P supply rather than drought. Therefore, the N applied in the amendments at varying levels is not expected to have impacted yield differences.

Table 3.5 Summary statistics for regressions of biomass/grain yield (kg ha^{-1}) on N concentration (in mg kg^{-1} for biomass and in % protein for grain) and biomass/grain yield at the same sampling time. All sampling dates have a significant negative relationship between N concentration and yield.

	2019				2020			
	Stem Elongation	Anthesis	Maturity	Grain	Stem Elongation	Anthesis	Maturity	Grain
Model p-value	0.0008	0.0027	0.0005	<0.0001	<0.0001	<0.0001	<0.0001	0.0001
Adjusted r^2	0.2	0.16	0.22	0.69	0.44	0.37	0.28	0.27
Slope estimate, m	-0.049	-0.28	-0.21	-357	-0.053	-0.18	-0.34	-142

The negative relationship between yield and N concentration is also reflected in a significant amendment effect on grain protein (Figure 3.3). Treatments which produced the greatest yields (frass, MAP, compost) had statistically lower protein content than the lower-yielding treatments (unfertilized, struvite, digestate). Most amendment treatments resulted in wheat quality below the optimum for marketing purposes (13.5% protein), though protein contents were higher in 2019 (mean 13.5% protein, CV 10%) than in 2020 (mean 11% protein, CV 9%). This aligns with higher soil-test nitrate (75 kg nitrate-N ha^{-1} from 0-60 cm) and mean N uptake in grain (72 kg N ha^{-1}, CV 13%) in 2019 compared to 2020 (46 kg nitrate-N ha^{-1} from 0-60 cm and 41 kg N uptake ha^{-1}, CV 12%). While this appears to contradict the regressions (Figure 3.2, Table 3.5) which indicate P rather than N limitation, this may be a result of different nutrient limitations driving yield versus protein concentration at different times in the season. P nutrition is most important in driving <u>yield</u> early in the season (Grant et al. 2001), whereas increased <u>protein concentration</u> relies on the presence of excess N <u>above</u> that required for yield late in the season (Woodard and Bly 1998). Thus, for treatments where increased P supply substantially increased yield above the control (frass, MAP, compost), N was not present in sufficient excess late in the season, particularly in 2020, in order to raise protein concentrations.

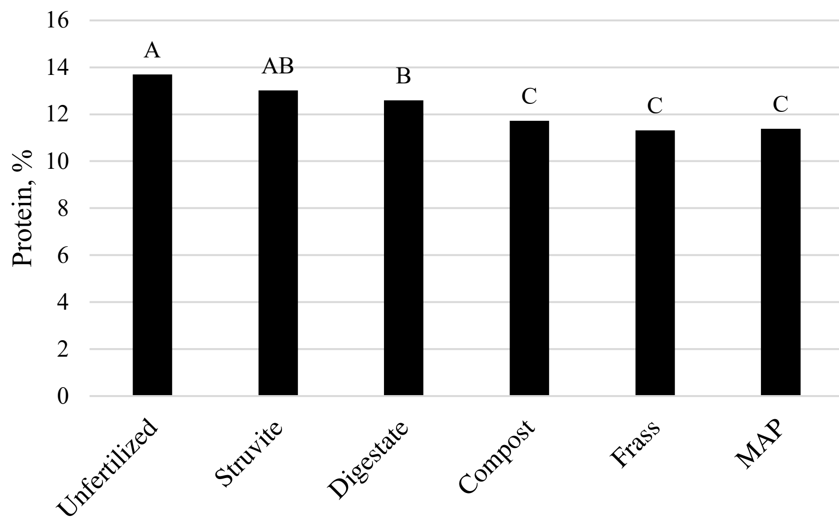

Figure 3.3 Wheat grain protein content by amendment treatment. Bars with the same letter are not significantly different based on the Tukey multiple comparison procedure at a α of 0.05.

Despite growing legume-based green manures in the season prior to the wheat experiments, soil N levels were low, particularly in 2020. This may have been due to a P-limitation on N fixation by the green manures. In a field study in this region, Cicek et al. (2014) found that a pea-oat green manure similar to that used in this study fixed sufficient N for a subsequent grain crop. They found comparable wheat grain yields (2903 kg ha^{-1} to 3603 kg ha^{-1}), and higher mean protein concentrations of 14.1% - 15.9% compared to the present study. The soil in that experiment had much higher (>15 mg kg^{-1} Olsen P) levels than the present wheat experiment. As P is required for nodulation and N fixation in legumes (Israel 1987), the severe P limitation in our experiment may have resulted in poor N fixation by the preceding green manure cover crop, and subsequently in low wheat-seed protein.

3.4.2 Perennial forage

3.4.2.1 Amendment effect on biomass and phosphorus uptake. The effect of amendment on alfalfa hay field production was not significant at either harvest in 2019, but it was significant at both harvest times in 2020 (Table 3.6). This is in contrast to the wheat experiments, where yield impacts of the amendments were observed in the year of application. Tissue P concentration and P uptake were significantly affected by amendment at every sampling time (Table 3.7). Mean biomass produced per year in this experiment (1973 kg ha^{-1}) is much lower than that reported for a typical grass hay field in Manitoba of 7530 kg ha^{-1} (Manitoba Agriculture 2007). It is in line with the lower end of yields reported for unfertilized degraded rangeland systems in Alberta, Canada, which ranged between 1850 kg ha^{-1} and 4540 kg ha^{-1} at five sites over three years (Lardner et al. 2001). This is reflective of the age and severe P depletion of this stand. Counterintuitively, mean yields were lower in 2020 than 2019, after the amendments had started statistically improving yields compared to the control. This may be, at least partially, an artifact of sampling, as the blade of the mower used to harvest was raised in 2020 to avoid contact with soil.

At the first biomass sample, struvite and frass had significantly greater tissue P concentration than the unfertilized control, and frass also had greater P uptake than the control. By the second biomass sample on August 20, 2019, all amendments had greater tissue P concentration than the control, and frass and MAP had greater tissue P than compost, digestate, and struvite. Only frass and MAP demonstrated statistically greater total P uptake than the control. At the third biomass sample on June 21, 2020, all amendments had greater biomass, tissue P, and P uptake than the control, except for compost P uptake which was similar to the control. At the final biomass sample, all amendments except compost had greater biomass than the control, all amendments except digestate and compost had greater tissue P concentration, and all amendments except compost had greater P uptake.

There are two possible explanations for the non-significant increase in forage biomass in the first year after amendment application, in spite of significant increases in P uptake. First, the time gap between increases in P concentration and increased biomass production may have been the result of a physiological delay between increased P uptake and the translation of the nutritional benefit into increased biomass due to drought in 2019. This is supported by the lack of a significant relationship between tissue P concentration and biomass yield in 2019; water may have been the primary limiting factor to yield, and greater P uptake in amended plots could not be translated into a yield benefit. Second, because of the high variability in the alfalfa plant stand, sampling error may have been too great to detect significant yield differences. Our results may be due to a combination of both explanations: drought in 2019 meant that P uptake differences were more pronounced than yield differences, and heterogeneity of the alfalfa hay stand caused sufficient variation to mask less-pronounced yield differences. This idea is supported by an alfalfa pot study conducted by Fan et al. (2015) on a high-pH, low-P soil with varying levels of P fertilization and water stress. Under severe water stress, P application increased shoot dry weight production slightly, but the differences were less pronounced than yield differences produced by P fertilization under adequate moisture conditions.

Table 3.6 Alfalfa biomass harvested at each of 4 harvest dates, cumulative alfalfa harvested. Values in the same column followed by the same letter are not significantly different based on the Tukey multiple comparison procedure at a α of 0.05. SEM is given in brackets.

	Individual Harvest Dates										Cumulative Biomass			2020 Total Biomass		
	14-Jul-19		20-Aug-19		21-Jun-20			28-Jul-20								
	kg ha^{-1}															
Unfertilized	880	(120)	1180	(130)	390	(80)	C	440	(90)	B	2900	(360)	B	840	(160)	C
Struvite	910	(120)	1260	(130)	1000	(80)	AB	760	(90)	A	4050	(360)	AB	1890	(160)	AB
Digestate	1100	(120)	1420	(130)	870	(80)	AB	890	(90)	A	4150	(360)	A	1630	(160)	AB
Compost	1140	(120)	1250	(130)	850	(80)	B	650	(90)	AB	3890	(360)	AB	1500	(160)	B
Frass	1290	(120)	1320	(130)	1100	(80)	A	880	(90)	A	4590	(360)	A	1980	(160)	A
MAP	1020	(120)	1290	(130)	910	(80)	AB	870	(90)	A	4100	(360)	AB	1780	(160)	AB
ANOVA p-value	0.12		0.67		<.0001			0.0008			0.01			<.0001		

Table 3.7 Phosphorus tissue and uptake values for four harvest dates, and cumulative uptake and acquisition efficiency based on 20kg ha^{-1} P application for alfalfa. Values in the same column followed by the same letter are not significantly different based on the Tukey multiple comparison procedure at a α of 0.05. SEM is given in brackets.

	14-Jul-19						20-Aug-19						21-Jun-20					
	Tissue P mg kg^{-1}			P uptake kg ha^{-1}			Tissue P mg kg^{-1}			P uptake kg ha^{-1}			Tissue P mg kg^{-1}			P uptake kg ha^{-1}		
Unfertilized	750	(47)	B	0.66	(0.12)	B	730	(35)	C	0.86	(0.13)	B	1110	(53)	C	0.44	(0.13)	C
Struvite	920	(47)	A	0.82	(0.12)	AB	920	(35)	B	1.15	(0.13)	AB	1460	(53)	AB	1.46	(0.13)	AB
Digestate	830	(47)	AB	0.92	(0.12)	AB	880	(35)	B	1.26	(0.13)	AB	1330	(53)	B	1.17	(0.13)	B
Compost	830	(47)	AB	0.95	(0.12)	AB	910	(35)	B	1.15	(0.13)	AB	1340	(53)	B	1.14	(0.13)	B
Frass	970	(47)	A	1.25	(0.12)	A	1100	(35)	A	1.47	(0.13)	A	1490	(53)	A	1.64	(0.13)	A
MAP	910	(47)	AB	0.93	(0.12)	AB	1080	(35)	A	1.40	(0.13)	A	1580	(53)	A	1.44	(0.13)	AB
ANOVA p-value	0.0076			0.0255			<.0001			0.0046			<.0001			<.0001		

Table 3.7 continued

	28-Jul-20						Cumulative					
	Tissue P mg kg^{-1}			P uptake kg ha^{-1}			P uptake kg ha^{-1}			Recovery (%)		
Unfertilized	960	(35)	B	0.43	(0.10)	C	2.39	(0.42)	C	-		
Struvite	1150	(35)	A	1.01	(0.10)	A	4.14	(0.42)	AB	10.3%	1.8%	AB
Digestate	1050	(35)	AB	0.79	(0.10)	AB	4.45	(0.42)	AB	8.7%	2.0%	B
Compost	1060	(35)	AB	0.69	(0.10)	BC	3.93	(0.42)	B	7.7%	3.0%	B
Frass	1130	(35)	A	0.99	(0.10)	AB	5.35	(0.42)	A	14.8%	2.3%	A
MAP	1150	(35)	A	1.00	(0.10)	AB	4.76	(0.42)	AB	11.9%	2.5%	AB
ANOVA p-value	0.0023			0.0001			<.0001			0.014		

Higher P concentration in plant tissue of struvite and frass at the earliest biomass sample indicates rapid access to these amendments by the plants. Struvite was notably the only amendment which was banded below the surface, and was therefore already in the plant root zone, which may explain this early result. The concentration of struvite in a band may also have contributed to its rapid availability. Soluble synthetic P fertilizers have greater availability when concentrated in a band than when broadcast due to lower interaction with and retention by the soil (Grant and Flaten 2019); however, it is unclear whether this would also be the case with struvite, as a low solubility P source. In addition, many plants can concentrate root growth in areas of high P concentration, which may improve P acquisition compared to diffuse P (Strong and Soper 1974; Rose et al. 2009). By contrast, frass was surface applied, and the P therein must have dissolved and infiltrated the soil in rainwater in order to be available to plant roots. There was only 34 mm of rainfall between amendment application on May 16/17 and the initial biomass samples on July 14, with only one rainfall event greater than 10 mm. This suggests rapid solubility and plant availability of P in frass.

All amendments increased biomass production compared to the control in this legacy alfalfa stand in 2020, the second season after amendment application. Yield increases compared to the control were 137% for frass, 126% for struvite, 113% for MAP, 95% for digestate, and 79% for compost. An Alberta study found forage yield improvements with broadcast application of conventional fertilizer (including N) to degraded grasslands of between -1% and 117%, with 7 of 15 site years registering statistically significant improvements (Lardner et al. 2001). The improvements in our study were in line with the greatest improvements found in the Alberta study, though notably our study found an improvement only in the second year after application, and had only one site.

Frass produced statistically greater biomass yield than compost in 2020, while struvite, digestate, and MAP were similar to both frass and compost. In combination with the demonstration of P concentration driving biomass production (below, Section 3.4.2.2), this demonstrates the capacity of these circular nutrient sources to revitalize P-depleted legacy alfalfa hay stands by improving P nutrition.

3.4.2.2 Attribution of yield differences to nutrient supply. Significant relationships between were found using regression analysis of forage biomass production on tissue P concentration at the two 2020 biomass samplings, establishing P as an important driving factor in biomass differences the second year after amendment application (Figure 3.4). In 2019 there was no significant relationship between biomass yield and tissue P concentration at either sampling date.

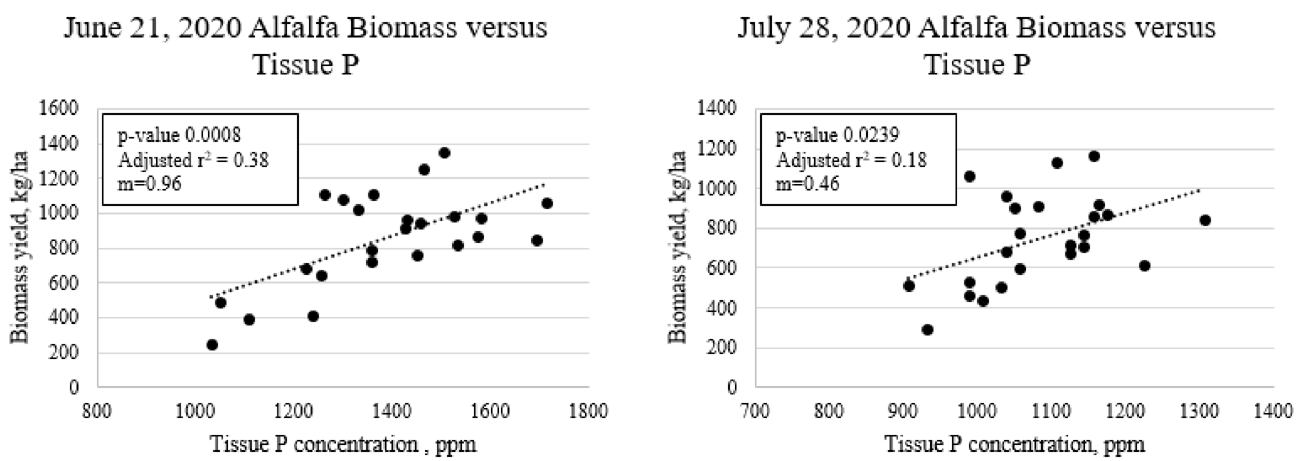

Figure 3.4 Relationships between forage biomass produced and tissue phosphorus content for the two 2020 harvest dates.

No significant relationships between biomass production and tissue N concentration were found at any sampling time (data not shown).

3.4.2.3 Amendment effect on nitrogen. In legume species like alfalfa, improving access to P can also contribute to improved N nutrition through N fixation, as adequate P is required for nodulation (Israel 1987). All amendments except compost significantly increased total N uptake in 2020 (Figure 3.5). Differences in N uptake may have resulted from amendment N application, increased soil N uptake, increased biological N fixation, or, a combination of these factors.

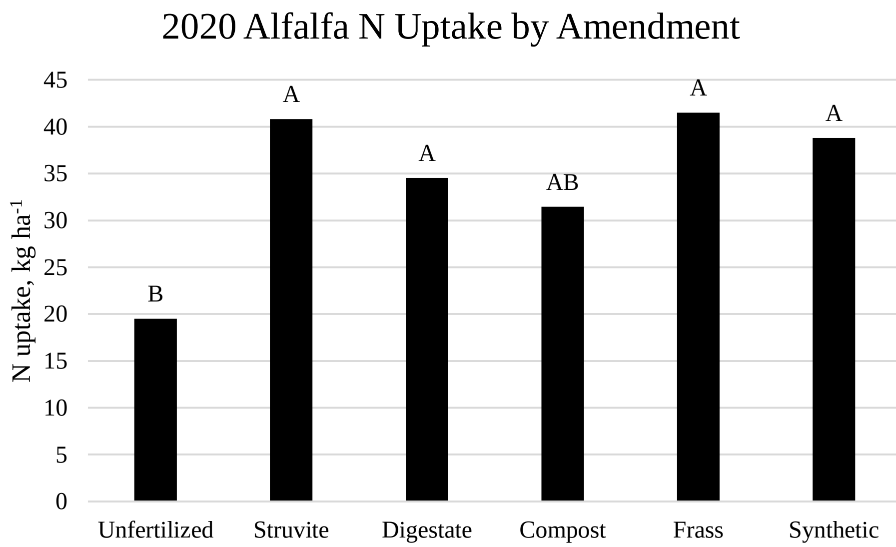

Figure 3.5 Cumulative nitrogen uptake by alfalfa plots for 2020. Bars with the same letter are not significantly different based on the Tukey multiple comparison procedure at an α of 0.05.

3.5 Conclusions

We hypothesized that the circular nutrient sources would supply more P and improve yields compared to the control, and that they would perform similarly to the organic and conventional comparison treatments.

In wheat, frass performed as well or better compared to compost and MAP in terms of P supply and yield improvement. Digestate was usually statistically similar to compost, but consistently underperformed compared to MAP. Struvite was consistently numerically greater, but statistically similar to the control. Therefore, our hypothesis was supported in the wheat system for frass, and digestate to an extent, but not for struvite.

In alfalfa in 2020, all three circular nutrient sources met expectations by supplying similar P compared to MAP, and frass supplied more P than compost. Therefore, our hypothesis was supported for all amendment types in the perennial alfalfa grass system. Thus, the circular nutrient sources considered here were effective in both an early successional annual crop and a later successional perennial crop.

The superior performance of struvite in alfalfa compared to wheat may be due to placement (subsurface banding in alfalfa versus broadcast and incorporation in wheat), the longer timeframe of the alfalfa experiment (two seasons versus one season), and/or physiological differences between wheat and forage plants in accessing P.

Frass stood out in both experiments for particularly high P uptake and yield improvements, which were greater than or similar to those of conventional MAP in both crops at all sampling times. Therefore, frass performed well when broadcast applied to both a low successional and higher successional plant community.

The dried solid fraction of food waste digestate used in this experiment had similar P supplying power to the organic comparison treatment, compost, in both wheat and alfalfa, and was also similar to the conventional comparison (MAP) in alfalfa. Solid digestate may not be consistently competitive with fully soluble fertilizers like MAP in terms of P supply, but compares well with a common organic amendment in both of these crops.

A key component of sustainable systems is that the waste product of one process is a valuable input to another. This research helps to establish circular nutrient sources as valuable inputs to dryland organic wheat and perennial forage cropping systems. Circular nutrients improve both organic crop productivity and food system sustainability by improving P cycling from urban areas to agricultural production fields.

3.6 References

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4 SYNTHESIS

Circular nutrients, which move phosphorus (P) from urban back onto rural landscapes, are an important part of a sustainable P cycle. In order to be effective components of the circular P cycle, the P in circular nutrients must be available to crop plants. The circular nutrients frass, digestate, and struvite vary in their capacity to supply P and improve yields in different crops and over different timescales in a low-P Manitoba soil (Figure 4.1). Interestingly, the comparative P supplying capacity of the amendments was not consistent across experiments: the alfalfa forage and the ryegrass experiments appear to be similar to each other (numerically if not based on means separations), whereas the wheat experiment diverged. All three circular nutrients supplied more P and significantly increased yields compared to the control in ryegrass and alfalfa. In wheat, only digestate and frass did so and struvite was not effective. Frass consistently had the numerically highest P uptakes, with MAP close behind and statistically similar to frass. Digestate and compost were statistically similar to each other, and numerically lower than frass and MAP in all crops. In wheat, struvite P uptake was numerically greater than the control, but the difference was not significant. By contrast, in alfalfa and ryegrass struvite had good P uptake, numerically greater than either digestate or compost.

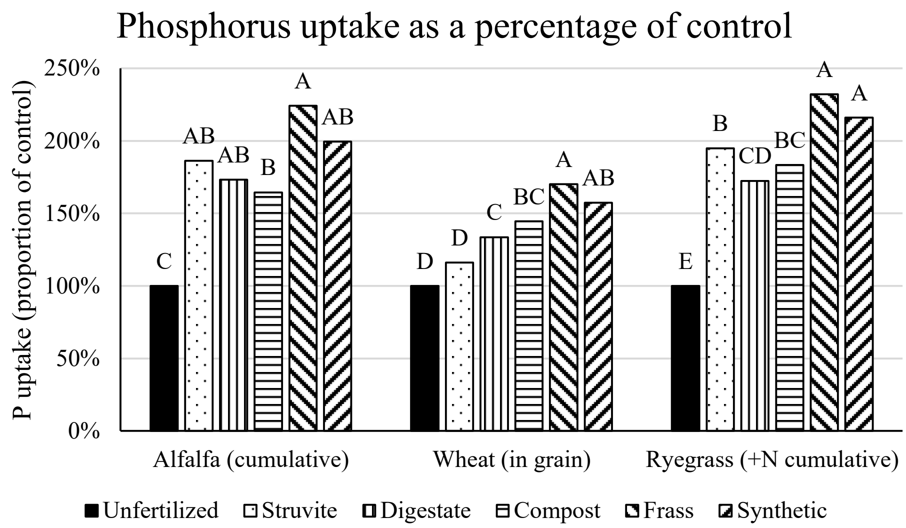

Figure 4.1 P uptake responses in 3 different crops in this study. Within the same crop, amendments that share a letter are not significantly different based on the Tukey means comparison procedure at an α of 0.05.

This difference is curious because the alfalfa and ryegrass systems seem to have the least similarity. The ryegrass experiment was short term, with adequate moisture provided consistently over the course of four months. By contrast the alfalfa experiment was rain fed (with below average precipitation in both years) and monitored over the course of two growing seasons. The wheat experiment was rain fed like the alfalfa, but occurred over the course of only one growing season of approximately three to four months, like the ryegrass experiment. Perhaps the longer monitoring period in the alfalfa experiment compensated for droughty conditions that reduced struvite uptake in wheat. However, it is unclear why low moisture would be more limiting for struvite P release, than for P release from digestate, frass, or compost, which performed well in wheat as well as alfalfa and ryegrass. If anything, one might hypothesize that moisture would be less important for struvite, which is water insoluble and considered to be released by organic acid exudation (Talboys et al. 2016), than for other amendments with a higher content of water-soluble P.

Another possibility is that the placement of struvite in wheat inhibited uptake. Struvite was broadcast and incorporated in the wheat trials, resulting in infrequent granules evenly distributed through the soil volume. By contrast, struvite was concentrated in subsurface bands in alfalfa. In the pot study, struvite was incorporated near the surface, which would have been rapidly and densely colonized by the ryegrass root system. Perhaps the concentration of struvite in the subsurface bands in alfalfa, and in the small soil volume of the pot study, allowed greater concentration of root growth, and thus more efficient uptake, compared to broadcast and incorporation of struvite in wheat.

Finally, crops themselves play an important role in P uptake, with different crops having different P mobilizing abilities and mechanisms. Wheat has been shown to mobilize less struvite compared to buckwheat (Talboys et al. 2016), and has less capacity to concentrate roots in high P zones compared to canola and buckwheat (Strong and Soper 1974). To my knowledge, there are no studies directly comparing the P acquisition strategies and capacities of wheat with alfalfa forages or ryegrass, but it can be inferred that these crops will have different P acquisition methods and capacities. Perhaps ryegrass and the forages had a greater capacity to acidify the rhizosphere and induce struvite dissolution than wheat. There is some evidence that polycultures facilitate P acquisition in alfalfa grown with corn as a result of interspecific competition (Sun et al. 2019), so that the polyculture nature of the alfalfa forage stand may have enhanced P uptake from struvite.

An important limitation of the research is that it was conducted in only one soil type (a high pH, high-clay, calcareous soil) and under one climate regime (cool subhumid continental). High-pH, clayey soil is common in the Prairie region, making this a good initial evaluation of these nutrients for the Prairies. Soil properties, particularly pH, are an important factor in regulating P availability (Brady and Weil 2008). In order to determine the consistency of amendment effects, and the factors driving those effects, future work should be conducted across a range of soil types and climates.

Opportunities for future work in determining the driving factors for P availability of struvite abound. Different crops could be directly compared for their root morphological and exudate adaptations that facilitate struvite uptake - this could also include an investigation of how placement of struvite influences P uptake in different crops. The potential interaction of moisture level and time in regulating struvite uptake could also be directly investigated with pot experiments. Soil properties, and particularly pH, should be kept in mind in all investigations of struvite.

Abundant questions remain about the P supplying properties of frass and digestate as well. Frass has been so little studied as a fertilizer that almost any additional research on this amendment would be novel. For example, frass could be compared with MAP and other potential P sources in a range of soils of varying pH, clay content, organic matter, and other properties in order to determine its consistency in competing with MAP as a P source. Characterization of the P within frass would also be interesting; a combination of fractionation into functional pools, x-ray diffraction to determine inorganic P species, and nuclear magnetic resonance imaging to determine organic P species has been used for a number of organic-matter-based circular products (Brod et al. 2015), and would be equally interesting for frass. P characterization could then be related to the performance of frass as a P source in soils with various properties.

The digestate tended to have lower P availability compared to frass and MAP, and further work could investigate what drove this difference in this soil. It may be that this digestate would supply more P in a lower-pH soil, given the high proportion of acid-soluble recalcitrant P species found in the solid fraction of food waste digestate (Brod et al. 2015). Drying and pelletization may also have reduced the P availability, as suggested by Ross et al. (2018). Characterization of a dried and pelletized product compared to the associated whole digestate, liquid digestate, and solid but undried digestate could provide some of these answers.

Farmers may prefer to apply P only once in a 3- to 4- year rotation for logistical reasons, and will therefore be interested in understanding the long-term/ residual P benefits of these amendments. The alfalfa forage experiment demonstrated that these amendments supply P in the second season after application for a perennial cropping system, but what about in an annual or mixed rotation over a longer period? P recovery in wheat grain was less than 25% in all amendments; the majority of the P applied remains in the soil, but its availability to future crops remains an important question. Future research could also investigate whether blends of these amendments might provide both rapidly available P (e.g., frass) and slowly available P (e.g., struvite) so that only one P application could supply a 3- to 4-year crop rotation.

The impacts of these circular nutrients go beyond P supply, particularly for frass and digestate which are organic-matter based. Their capacity to supply nitrogen (N) and other nutrients should also be considered. Frass and digestate supply organic matter to the soil and may contribute to the accumulation of soil carbon and changes in associated soil properties like aggregation. Their capacity for carbon sequestration, as well as nitrous oxide emissions and whole lifecycle greenhouse gas effects should be considered, and compared to alternative waste management strategies. While struvite does not supply organic matter, it may be interesting to evaluate its effect on soil microbiota. The N and other nutrients supplied may also be of interest in human nutrition. P deficiency is very uncommon in humans, with diets adequate in other nutrients being automatically adequate in P (Heaney 2012). However, other nutrients supplied by organic-matter-based amendments (e.g. Zn, Fe) may contribute to higher nutrient contents of the resulting foods (Zhang et al. 2015), and therefore be of interest to human nutrition.

Absent from this thesis has been any comparison of the economics for farmers of using these amendments, but of course the economics will be important drivers of adoption. All three circular nutrient sources considered in this thesis are commercially available, but awareness of them and adoption by farmers is low, particularly because they are generally more expensive than their linear counterparts. Organic farmers have an opportunity to be on the forefront of circular nutrient adoption, as organic premiums and prohibition of synthetic fertilizers encourage these farmers to use alternative nutrient sources. Increased production of these circular nutrients may result in economies of scale to allow them to better compete with linear nutrient sources. Evaluation of the cost-benefit for farmers, and the economic transport distance of each amendment based on P concentration and predicted yield increases, could reveal what regulatory or financial aid is required in order to make these amendments feasible P sources for more farmers.

The overall conclusion of this thesis is that while their specific efficacy in different situations varies, all three nutrient sources have good potential to supply P to organic crops. This makes them practical components of a circular P system.

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