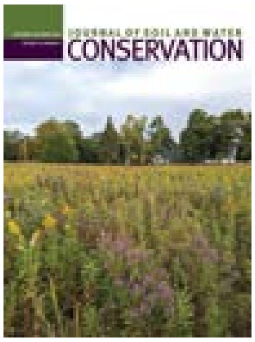

Journal of Soil and Water Conservation

ISSN: (Print) (Online) Journal homepage: www.tandfonline.com/journals/uswc20

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To cite this article: Jason S. Cavadini & Eileen J. Kladivko (21 Mar 2025): Oilseed radish/cereal cover crop bicultures and soil phosphorus distribution, Journal of Soil and Water Conservation, DOI: 10.1080/00224561.2024.2429960

To link to this article: https://doi.org/10.1080/00224561.2024.2429960

Published online: 21 Mar 2025.

JOURNAL OF SOIL AND WATER CONSERVATION
https://doi.org/10.1080/00224561.2024.2429960

RESEARCH ARTICLE

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Oilseed radish/cereal cover crop bicultures and soil phosphorus distribution

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ABSTRACT

Cover crops are implemented for many different goals including soil nutrient retention and cycling. Certain cover crop species are regarded specifically for nutrient cycling, like the ability of oilseed radish (*Raphanus sativus* L.; OSR) to scavenge and release soil nutrients such as phosphorus (P). Oilseed radish can cycle significant quantities of soil P through a sequence of rapid fall growth, winterkilling, and rapid spring decay. The quickness and unpredictability of this process has made managing OSR specifically for soil P retention challenging, creating a need to identify management strategies that support the goals of growing this cover crop. The objective of this project was to determine if mixing winter-surviving or slow-decaying species with OSR as a biculture can moderate the process of OSR nutrient cycling in a way that times the release of plant available P (available P) with the growth of subsequent crops. This study measured dry matter (DM) and P accumulation of cover crop biomass, as well as water soluble P (WSP) and Mehlich-3 P (MP) of soil to test how OSR and OSR bicultures acquire, retain, release, and distribute P. Three cover crop treatments of OSR, OSR+cereal rye (*Secale cereal* L.), and OSR+oats (*Avena sativa* L.), plus a no-cover control were compared across three times (fall, spring, and V6 growth stage of corn [*Zea mays* L.]), three soil depths (0 to 2.5, 2.5 to 10, and 10 to 20 cm), and two row positions (root zone: <5 cm, buffer zone: >5 cm from OSR tuber) to observe the distribution of available P. Treatment responses varied, but available P consistently accumulated in the root zone versus the buffer zone compared to when no cover crop was grown. Significant treatment effects occurred more frequently when using OSR alone than using OSR bicultures. Singular treatment effects were modest, but there were many interactions between treatment, time, soil depth, and row position suggesting the potential for cereal crops to complement OSR by adding P-retention functionality to the cover crop. The results show that OSR can heavily influence P distribution and suggest that planting OSR in bicultures can be a good measure to ensure positive nutrient management outcomes consistent with other ecosystem services of cover crops such as soil health and water infiltration.

ARTICLE HISTORY
Received June 3, 2024
Revised August 27, 2024
Accepted October 22, 2024

KEYWORDS
cover crop;
nutrient stratification;
oats; radish; rye;
soil phosphorus

INTRODUCTION

While the practice of cover crops has existed for centuries, a growing awareness of declining water quality, degrading soil health, and diminishing economic returns in agriculture has placed a new sense of urgency and greater interest in the integration of cover crops into farming systems (Hartwig and Ammon 2002). The amount of research, demonstration, and outreach around cover crops has boomed in the past two decades, and there is now a general understanding of the benefits associated with cover crops (Singer and Nusser 2007). Cover crops are commonly viewed as a crop that provides a specific set of functions or ecosystem services (Schipanski et al. 2014) during the nongrowing season. Cover crops were originally used in the US Corn Belt to retain soil and critical nutrients such as nitrogen (N) (Kaspar, Radke, and Laflen 2001; Kaspar et al. 2004) in corn (*Zea mays* L.)–soybean (*Glycine max* [L.] Merr.) rotations, but today's cover-cropping systems are being implemented with a much wider scope of ecosystem services in mind (Schoumans et al. 2014; Weil and Kremen 2007).

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The Brassicaceae (brassica) family has been one of the most popular plant types used as a cover crop. Oilseed radish (*Raphanus sativus* L.; OSR) is a brassica that has become a widely used cover crop species in the Corn Belt because of a large array of potential benefits it provides the soil. Characterized by a large tuber (taproot) belowground and abundant, fast-growing leaf tissue aboveground, OSR serves specific functions that can contribute to improved soil health. Despite these ecosystem services, the same characteristics that make OSR desirable as a cover crop also carry the potential for negative outcomes when not used properly, creating a tradeoff that must be considered (Duncan et al. 2019). Oilseed radish is known for biologically tilling the soil (Chen and Weil 2010), taking up high amounts of residual nutrients (Dean and Weil 2009), and rapidly decomposing in the spring after being winter-killed (White and Weil 2011). This has often been viewed as a desirable suite of characteristics, with OSR capturing soil nutrients and making them available to subsequent main crops. However, assessing the ecosystem services of OSR is more challenging than previously known, due to the episodic nature and time sensitivity of functions such as nutrient retention (Schipanski et al. 2014). Under certain conditions or management, an untimely release of nutrients could lead to unintended environmental consequences (Liu et al. 2019). For example, recent research has found that after biologically tilling the soil and taking up large amounts of nutrients in the fall, winter-killed OSR often releases nutrients as its residue is exposed to freeze-thaw cycles (Cober, Macrae, and Van Eerd 2018, 2019), followed by rapid springtime decomposition, potentially leaving soil susceptible to erosion during periods of spring snowmelt and rainfalls.

The tradeoff between ecosystem services and environmental considerations associated with cover crops is exemplified by the relationship between OSR and soil phosphorus (P). Phosphorus contamination of surface waters continues to be a leading environmental concern in the Midwest and much of the United States today (Macrae et al. 2024; Sharpley and Wang 2014). While cover crops have largely been viewed as a solution to nutrient transport, the relationship between cover crops and P is not straightforward (Bridgeman et al. 2012; Duncan et al. 2019; Hallama et al. 2019). Although cover crops often reduce losses of soil sediment and particulate P, they do not reduce losses of water soluble P (WSP) and in many cases increase WSP losses (Smith, Huang, and Haney 2017). Oilseed radish has been found to mobilize plant available P (available P) near its roots (Li et al. 2007), potentially making it more available to growing crops (White and Weil 2011). While one result of P mobilization is greater available P for a subsequent crop in the soil profile, another is the loss of P from the soil surface. Much of the previous research on OSR cover crops has focused on the aboveground transport of P from the field, but fewer studies have looked at the vertical and horizontal movement (accumulation) of available P in the soil profile and whether it is beneficial to a subsequent crop. While the ecosystem services of P retention and cycling by cover crops as a nutrient management tool have merit, management strategies that minimize the potential for unintended environmental outcomes must be identified to responsibly pursue these functions (Clark et al. 1997). For example, species of cereal crops such as oats (*Avena sativa* L.) and cereal rye (*Secale cereal* L.) have been used successfully as cover crops and have been shown to effectively retain soil sediment and nutrients (De Baets et al. 2011; Kaspar, Radke, and Laflen 2001) and decompose more slowly in the spring than OSR. The management strategy of mixing a cereal crop with OSR (biculture) could still allow the desired benefits of OSR while retaining soil and nutrients in the spring and available P for subsequent crops (White and Weil 2010). This biculture could be considered a best management practice for cover crops in areas where P is an environmental resource concern.

Various species of cover crops deploy a diversity of strategies to acquire different pools of P in the soil. The exact mechanism of P acquisition by a cover crop depends on the species of cover crops being grown, the forms of P available in the soil, and the microbial abundance and activity of the soil (Hallama et al. 2021; Honvault et al. 2021). The functional diversity of a cover crop can be improved by planting multiple, complementary species featuring an array of P acquisition strategies. Cover crop mixtures can affect P cycling through a collective process of changing the microbial and enzymatic dynamics of the soil, stimulating the mobilization and mineralization of P and resulting in a diverse array of organic and inorganic P with various degrees of plant availability (Dada, Armstrong, and Smith 2021; Hallama et al. 2019, 2021; Honvault et al. 2021; Lehman et al. 2012; Muhammad et al. 2021).

The objectives of this study were to (1) measure above- and belowground cover crop biomass to observe how OSR and OSR bicultures accumulate dry matter (DM) and P in the plant through the life of the cover crop, (2) measure vertical and horizontal distribution of available P in proximity to the cover crop and observe how OSR and OSR bicultures accumulate available P in the soil through the life of the cover crop, and (3) determine the status of available P following OSR and OSR bicultures while the subsequent cash crop becomes established. The hypotheses for this study were (1) OSR bicultures will contain more above- and belowground biomass DM and tissue P than OSR alone, (2) available P will accumulate at the soil surface and near the cover crop tuber after cover crop decomposition, and (3) available P will be greater following the decomposition of OSR bicultures than OSR alone.

MATERIALS AND METHODS

This study took place across four field sites at two locations near West Lafayette, Indiana: the Diagnostic Training Center (DTC) at the Purdue Agronomy Center for Research and Education, and the Throckmorton-Purdue Agricultural Center (TPAC). Three of the field sites were at DTC and one was at TPAC. The field sites at DTC were named according to the crop that preceded the cover crops; the DTC sites that contained soybeans, wheat, and corn previously were named DTCS, DTCW, and DTCC, respectively. Previous crop and soil information can be found in Table 1.

There were four treatments at each site: OSR alone, OSR mixed with oats (OSR/Oats), OSR mixed with cereal rye (OSR/Rye), and no cover crop (control). Treatments were replicated three times in a randomized complete block design for a total of 12 plots at each site. Plots at DTCS, DTCW, and DTCC were 4.3 m by 9.1 m, and plots at TPAC were 4.6 m by 30.5 m. Before cover crops were planted, wheat was harvested for grain from DTCW and TPAC with straw left in the field. Whole plant soybean was harvested from DTCS, and whole plant corn was harvested from DTCC. The DTCS, DTCW, and TPAC sites were disked prior to planting of the cover crops, while DTCC was not. All cover crop plots were established using a no-till drill with 19 cm row spacings. Oilseed radish (Groundhog variety, The Cisco Company, Indianapolis, Indiana) was seeded at 14 kg ha^{-1} when planted alone and 6.7 kg ha^{-1} when planted in a biculture with oats or cereal rye. Oats were seeded at 31 kg ha^{-1}, and cereal rye was seeded at 36 kg ha^{-1}. All seeding rates were adjusted for pure live seed. The biculture treatments at the DTC sites contained both species in every row, while at TPAC the cereal crop was in every other row and OSR was in every row. No fertilizer was applied to the cover crops

Table 1. Characteristics of the four field sites in Indiana.

	Site			
Parameter	**DTCS**	**DTCW**	**TPAC**	**DTCC**
Location	DTC	DTC	TPAC	DTC
Previous crop	Soybean	Wheat	Wheat	Corn
Soil series	Rockfield	Toronto	Toronto	Rockfield
Soil texture	Silt loam	Silt loam	Silt loam	Silt loam
Soil taxonomy	Typic Hapludalf	Udollic Ochraqualf	Udollic Ochraqualf	Typic Hapludalf
Cover crop planting date	Aug. 30, 2011	Aug. 30, 2011	Aug. 23, 2011	Sept. 17, 2012
SOM (%)	2.2	2.7	2.9	2.0
CEC (meq 100 g^{-1})	10.6	12.0	12.5	11.7

Notes: DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat; DTCC = Diagnostic Training Center, cover crops following corn; SOM = soil organic matter; CEC = cation exchange capacity.

at the DTC sites, and at TPAC polymer-coated urea was broadcast at a rate of 50 kg N ha^{-1}. The following spring OSR and oats were winter-killed and required no control prior to planting of the succeeding crop. The cereal rye continued growing in the spring and was terminated at DTCS and DTCW on March 29, 2012, with an application of glyphosate (N-(phosphonomethyl)glycine) (1.06 kg ha^{-1} active ingredient [a.i.]) and ammonium sulfate (AMS), and at TPAC on April 7, 2012, with an application of glyphosate (1.68 kg ha^{-1} a.i.). Corn was planted at all three sites on May 4, 2012, at a population of 84,000 plants ha^{-1} at DTCS and DTCW, and 79,000 plants ha^{-1} at TPAC. All three sites were no-till planted at 76 mm row spacings. Starter fertilizer was subsurface band-applied 5 cm below and 5 cm to the side of the corn seed with the corn planter at planting. No other fertilizer was applied to the succeeding crop. The starter fertilizer at the DTCS and DTCW sites contained 39.2 kg N ha^{-1} and 58.6 kg P ha^{-1} in the form of ammonium polyphosphate and at TPAC contained 43.5 kg N ha^{-1} in the form of urea ammonium nitrate. Weeds were managed in the succeeding corn crop with postemergence applications at TPAC of thiencarbazone-methyl (methyl 4-((((4,5-dihydro-3-methoxy-4-methyl-5-oxo-1H-1,2,4-triazol-1-yl)carbonyl)amino)sulfonyl)-5-methyl-3-thriophenecarboxylate) (0.015 kg a.i. ha^{-1}), tembotrione (1,3-cyclohexanedione, 2-(2-chloro-4-(methylsulfonyl) – ((2,2,2-trifluoroethoxy)methyl)benzoyl)) (0.076 kg a.i. ha^{-1}), glyphosate (isopropylamine salt of N-(phosphonomethyl)glycine) (1.06 kg a.i. ha^{-1}), and AMS. At DTCS and DTCW, only glyphosate (1.06 kg a.i. ha^{-1}) and AMS were used.

Biomass sampling: Fall

Cover crop observations and samples were obtained from within 0.25 m^2 measuring frames at TPAC, DTCS, and DTCW on November 1, 17, and 21, 2011, respectively, and from DTCC on November 14, 2012. Two samples were obtained from each plot, and frames were arranged so that every sample included three rows of cover crop. Oat and cereal rye shoots were cut approximately 2.5 cm above the soil surface with hand shears, and OSR shoots were removed at the top of the tuber. Aboveground portions of OSR tuber were not included in the shoot biomass samples.

Belowground cover crop samples were also obtained from each frame at DTCS and TPAC. Every OSR tuber inside the frame was counted, measured for length and width, and collected. A spading fork was used to extract the tubers from the soil to ensure no part was broken off and left behind. Oat and cereal rye roots were sampled from the center row of each frame as opposed to from the entire frame to ensure precise and consistent samples of the main root mass. A flat shovel was used to remove the volume of soil approximately 19 cm wide by 50 cm long and 10 cm deep. The main root mass was collected from the loosened soil and stored in a plastic bag. To account for additional small roots that remained in the soil after collecting the main root mass, subsamples of soil containing the fine roots were obtained from the loosened soil of the center row. The loosened soil was collected, mixed, weighed, and subsampled for further analysis. All tubers, main root masses, and fine root subsamples were thoroughly washed over 0.8 mm sieves with tap water to remove all soil.

Biomass sampling: Spring

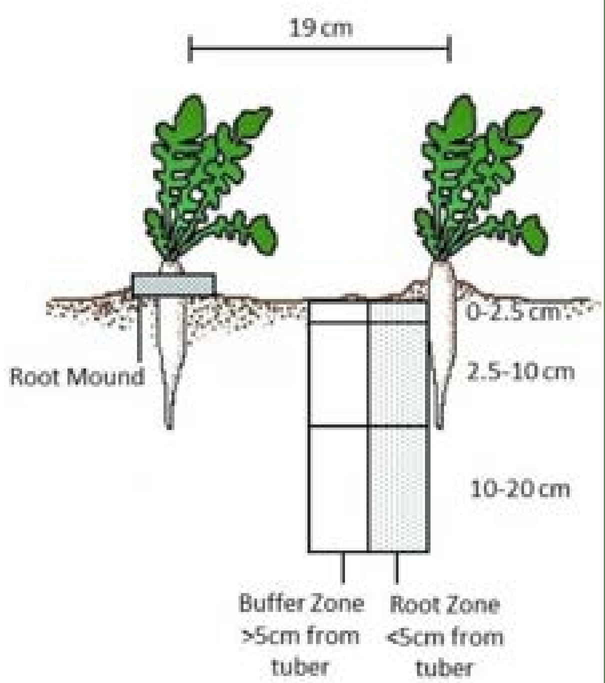
Cereal rye was the only cover crop that was not winter-killed. Aboveground samples of cereal rye biomass were sampled from DTCS, DTCW, and TPAC on March 29, 2012. Two samples were collected from each plot from within 0.25 m^2 frames. Extended leaf height measurements were also recorded. All above- and belowground biomass samples were dried at 60°C, ground, and analyzed by A&L Great Lakes Laboratories (Fort Wayne, Indiana).

Soil sampling

Fall soil samples were collected from DTCS on November 17, TPAC on December 3, and DTCW on December 8, 2011, and from DTCC on November 14, 2012. Samples were collected from two row positions in relation to the OSR tuber. The root zone (<5 cm from tuber) was the row position near the OSR tuber. The buffer zone (>5 cm from tuber) was the row position further away from the tuber (White and Weil 2011). Nine soil cores were collected from each row position in each plot. Cores were then separated into three depth increments for each row position: 0 to 2.5 cm, 2.5 to 10 cm, and 10 to 20 cm (Figure 1). In addition, the mound of soil (called the root mound) formed around the base of the OSR tuber from biological tillage was sampled at DTCS, TPAC, and DTCW. The root mound is of interest because the biological tillage of OSR noted in previous work (Chen and Weil 2010) may leave the soil susceptible to erosion and nutrient losses. Nine root mounds were collected from each plot.

Spring soil samples were obtained from DTCS, DTCW, and TPAC on March 9, 16, and 19, 2012, respectively, and from DTCC on March 15, 2013. The sampling procedure remained the same as in the fall: nine cores from each of two row positions, separated into three depth increments in each plot. At that time only traces of OSR tubers remained and root mounds were no longer present for sampling. Soil samples were obtained

Figure 1. Soil sampling diagram including the root mound, two row positions (root zone and buffer zone), and three soil depths relative to the oilseed radish tuber.



from the same plots at three of the sites when the succeeding corn crop reached the sixth vegetative growth stage (V6) but were no longer specified by row position since the cover crop rows were no longer evident. Samples were collected from DTCS, DTCW, and TPAC on June 8, 11, and 12, 2012, respectively.

All soil samples were air-dried and ground to pass through a 2 mm sieve in preparation for analysis. Samples from DTCS, DTCW, and TPAC were analyzed for WSP and MP, and samples from DTCC were analyzed for MP only. Water soluble P was extracted by adding 1 g of ground soil, 10 mL of deionized water, and 1 drop of chloroform ($CHCl_3$) to a 50 mL centrifuge tube, shaking for 1 hr, centrifuging for 10 min, and filtering through a Whatman no. 42 ashless filter (Whatman International, Maidstone, United Kingdom) (Murphy and Riley 1962; Self-Davis et al. 2009). Mehlich-3 P was extracted by adding 1 g of soil and 10 mL of MP extraction solution to a 50 mL centrifuge tube, shaking for 5 min, and filtering through a Whatman no. 42 ashless filter (Mehlich 1984). Phosphorus concentrations of the extractants were measured using inductively coupled plasma (ICP). Two replicates of each sample were analyzed.

Statistical analysis

Statistical analyses were performed using SAS version 9.2 (SAS Institute Inc., Cary, North Carolina). Some data were transformed to improve the homogeneity of the variance components. For soil data, WSP values were log transformed and MP values were square root transformed. All transformed data are presented in back-transformed units. Variances were pooled for analysis of variance (ANOVA) models that had more than one error term, with most of the variances not significant at $P = 0.25$, to simplify the model and increase degrees of freedom. The SAS GLM procedure was used to determine pooling. The SAS MIXED procedure was used for the ANOVA and least-squared mean (LSM) separation test. Least-squared means were compared where treatment effects were significant at $P \leq 0.05$.

The cover crop above- and belowground biomass data were all analyzed using a randomized complete block design. Root mound WSP and MP data were also analyzed using a randomized complete block design. Multiple ANOVA models were used to make all other comparisons of soil data. To test the main effect of row position with respect to cover crop rows, a split-plot, split-block model was used to compare the three cover crop treatments (OSR, OSR/Oats, and OSR/Rye), row position (root zone and buffer zone), time (fall and spring), and soil depth. The control was not included in this analysis because it did not have cover crop rows and thus no row position. To test the effects of cover crop treatments versus the control, another model tested cover crop root zone values versus the control and then cover crop buffer zone values versus the control. This split-plot, split-block model was used to compare all four treatments (cover crops and control), time, and depth within each row position. A split-block model was also used to compare all four treatments and depth during the V6 sampling time, as there was no longer a row position variable.

RESULTS AND DISCUSSION

Cover crop biomass

The cover crops were planted into very dry conditions at three sites in late summer 2011 but benefitted from timely rainfalls with abundant growth before going dormant or being winter-killed in late November. With a mild winter and early spring, the OSR and oat cover crops decomposed very early in spring 2012. When the succeeding corn crop was planted, very little sign of OSR remained in the field. The corn crop struggled through the historic drought of 2012. Corn was significantly limited by moisture all summer, making the observation of treatment effects very difficult. Furthermore, the extreme heat and dryness left the corn crop vulnerable to pest and disease infestation. Despite the extreme drought, the cover crops at DTCC were planted in September 2012 into favorable conditions following a few early fall rain events. The cover crops established quickly, but growth was halted by late October due to freezing temperatures.

Fall cover crop observations are presented in Table 2. The OSR treatment had the highest population of tubers because OSR was seeded at a lower rate in the biculture treatments. There were no significant differences in tuber populations between OSR/Rye and OSR/Oats. The TPAC site had the highest tuber population for all sites, which likely emphasizes the benefit of the fertilizer applied to the cover crops when they were planted. There was not much variation in OSR canopy height among treatments within a site except for the DTCW site, where OSR/Rye was shorter than OSR. Most sites had significant canopy height differences for the cereal crops (Table 2). Oats consistently had a taller canopy height than cereal rye, indicating the aggressive growth habit of oats in the fall that has also been noted in previous studies (Kabir and Koide 2002). All three species were the tallest at TPAC, which can likely be attributed to the one week earlier planting date and the addition of fertilizer at planting.

There were not consistent significant differences in OSR tuber sizes, but tubers tended to be the largest in length and diameter in the OSR/Rye treatment (Table 2). The larger tuber sizes may be attributed to the fact that the OSR/Rye treatment tended to have the lowest OSR population, resulting in less competition and more growth. This is consistent with previous data showing that lower populations allow for OSR tubers to grow larger (Amini 2011).

Fall cover crop shoot DM and P accumulation values for each treatment are presented in Table 2. In 2011, shoots tended to have the highest DM in the OSR/Oats treatment but were only significant at TPAC. The DM accumulation in fall 2012 was much lower than in fall 2011 due to later planting and a shorter fall growing season. There were no significant differences among treatments in shoot P accumulation. The highest P accumulation values were not always associated with the highest DM values, due to individual species in the bicultures having different P concentrations. Average shoot P concentrations for each species were 0.30, 0.22, and 0.33 (% P) for OSR, oats, and rye, respectively.

Fall values for cover crop root DM and P accumulation are presented in Table 3. The OSR treatment tended to have the highest tuber DM and P accumulation, although the differences in DM were not significant at any site. The highest tuber

Table 2. Cover crop biomass observations, dry matter (DM), and phosphorus (P) accumulation by site for fall 2011 and fall 2012.

Site	OSR population (plants ha^{-1})	Canopy height (cm)		OSR tuber size		Shoots (kg ha^{-1})	
		OSR	Cereal	Length	Dia.	DM	P
DTCS (2011)							
OSR	562,900 a*	24 a	—	12 ab	2.0 b	1,813 a	6.03†
OSR/Oats	355,500 b	24 a	41 a	11 b	1.9 b	2,032 a	5.38
OSR/Rye	278,500 b	25 a	24 b	15 a	2.5 a	1,984 a	6.43
DTCW (2011)							
OSR	557,000 a	34 a	—	16 a	2.3 a	2,302 a	6.34
OSR/Oats	479,900 a	29 ab	46 a	15 a	1.8 a	2,966 a	6.81
OSR/Rye	414,700 a	25 b	28 b	17 a	2.3 a	2,101 a	5.67
TPAC (2011)							
OSR	793,900 a	44 a	—	13 a	2.0 a	3,487 b	10.64
OSR/Oats	533,200 a	48 a	74 a	13 a	2.1 a	5,201 a	15.28
OSR/Rye	485,800 a	43 a	53 a	14 a	2.3 a	4,406 ab	13.59
DTCC (2012)							
OSR	—	9 a	—	—	—	988 a	3.85
OSR/Oats	—	9 a	19 a	—	—	592 a	2.37
OSR/Rye	—	9 a	11 b	—	—	667 a	2.16

Notes: OSR = oilseed radish; Dia. = tuber diameter; DM = dry matter; P = tissue phosphorus; DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat; DTCC = Diagnostic Training Center, cover crops following corn.
*Values within the same column and site that contain similar lowercase letters are not significantly different at the $P \leq 0.05$ level.
†No significant difference in P content among treatments at any site at $P \leq 0.05$.

Table 3. Cover crop root biomass dry matter (DM) accumulation and tissue phosphorus (P) content for oilseed radish (OSR) tubers, cereal roots, and total roots during fall 2011.

Site	Cover crop root biomass (kg ha^{-1})					
	OSR tubers		Cereal roots		Total roots	
	DM	P	DM	P	DM	P
DTCS						
OSR	1,271 a*	6.18 a	—	—	1,271 b‡	6.18 a
OSR/Oats	808 a	3.45 b	1,320 a	1.83 a	2,128 a	5.28 a
OSR/Rye	1,196 a	5.58 a	1,165 a	1.68 a	2,361 a	7.26 a
DTCW						
OSR	1,559 a	5.37 a	—	—	—	—
OSR/Oats	986 a	3.15 a	—	—	—	—
OSR/Rye	1,301 a	4.26 a	—	—	—	—
TPAC			—			
OSR	1,560 a	5.04 a	—	—	1,560 a	5.04 a
OSR/Oats	1,205 a	4.12 a	386 a†	0.428 a	1,591 a	4.55 a
OSR/Rye	1,460 a	4.35 a	207 b	0.262 b	1,667 a	4.61 a

Notes: DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat.
*Values within the same column and site that contain similar lowercase letters are not significantly different at $P \leq 0.05$.
†The cereal crop portion of the biculture treatments was planted only in every other row at TPAC.
‡Additional tuber fine roots were observed in the OSR treatment in 2 out of 18 samples taken from the three sites, but because no fine roots were found in the majority of the OSR samples, the two could not be confidently included in the total root values.

DM values were consistently associated with the highest tuber P accumulation values. Unlike the biculture shoots that were influenced by varying P concentrations of different species, the OSR tubers consisted of one type of tissue, resulting in consistent P concentrations across treatments. There was less variation in tuber DM and P accumulation among sites than with the shoots. Even with N fertilizer applied to the cover crop at TPAC, the tuber DM and P accumulation values were not much different than at the other sites. This may suggest that the fertilizer had a greater effect on shoot growth than root growth. The OSR/Oats treatment tended to have the lowest DM and P accumulation values in the tubers. This is due in part to the lower OSR seeding rate in the biculture treatments but is most likely a result of oats having more fall growth and a greater canopy height creating competition and reducing OSR tuber growth and P uptake.

The cereal root DM and P accumulation values from the biculture treatments at DTCS and TPAC in fall 2011 varied between the two sites. This most likely reflects different planting practices. The cereal crop was planted in each row at DTCS and every other row at TPAC. The OSR/Oats treatment tended to be greater in DM and P accumulation than OSR/Rye. This is consistent with previous findings that oats grow more in the fall than cereal rye (Kabir and Koide 2002). The data underscore that although there is a large amount of biomass in the shoots and radish tubers, the presence of cereal roots in the bicultures also adds significant amounts of DM and P accumulation to the system (Figure 2).

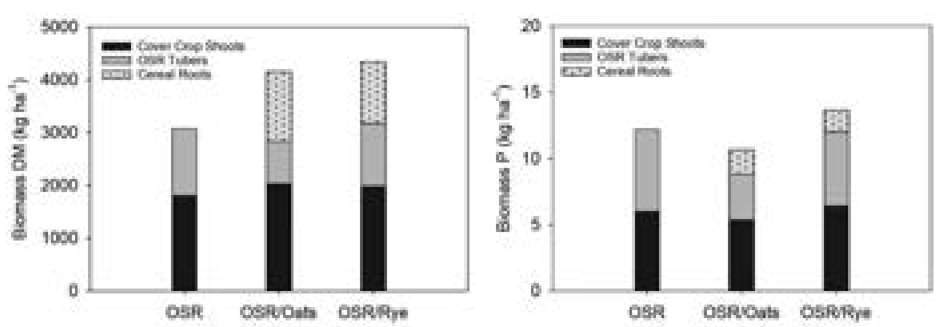
Cereal rye was the only cover crop species that did not winter-kill and resumed growth the following spring. The DM and P accumulation values for cereal rye shoots in spring 2012 are presented in Table 4. The spring values are similar to the fall values, despite the fact that the OSR was in the fall samples but not the spring samples after winter-killing, and the spring samples consisted solely of cereal rye. This shows that through winter-hardiness, cereal rye is able to ensure continued cover in the spring by making up for the amount of cover lost after OSR winter-kills and decomposes. While oats showed the ability to excel at providing cover in the fall, cereal rye showed the ability to ensure continued cover in the spring.

Soil phosphorus

The OSR root mounds (see diagram in Figure 1) were sampled from each treatment during fall 2011 at DTCS, DTCW, and TPAC. There were no significant differences among cover crop treatments in WSP or MP at any of the three sites in fall 2011. Water soluble P for the three sites ranged from 5.2 to 10.7, 4.7 to 8.6, and 4.6 to 8.6 mg kg^{-1} for OSR, OSR/Oats, and OSR/Rye, respectively. Mehlich-3 P ranged from 20 to 61, 20 to 46, and 18 to 38 mg kg^{-1} for OSR, OSR/Oats, and OSR/Rye, respectively. The root mound values tended to be similar in magnitude to the available P values at the soil surface of the root zone. While this may sound inconsequential, it should be noted that the root mound may be susceptible to soil and nutrient losses after the biological tillage of OSR observed in previous work (Chen and Weil 2010).

The next sections discuss the analyses of available P as affected by the combined factors of cover crop treatment, depth, time, and row position, as appropriate. As described in the Materials and Methods section, soil samples were collected from three cover crop treatments at three times

Figure 2. Total dry matter (DM) accumulation and phosphorus (P) content of different biomass fractions for each cover crop treatment during fall 2011 at the DTCS site. OSR is oilseed radish.



at DTCS, DTCW, and TPAC, and two times at DTCC across two row positions and three soil depths. In the no-cover control treatment, as there was no cover crop present, samples were collected by depth with no regard to row position. During the V6 stage of the succeeding corn crop, there was no cover crop present in any treatment, so bulk samples were collected with no regard to row position. All available P data from fall and spring were analyzed using two main statistical models, as described in the Materials and Methods section. The model in Table 5 tested the significance of each comparison made within the three cover crop treatments in fall and spring. The model in Table 6 tested the significance of each comparison made within each cover crop treatment, including the no-cover control, in fall and spring.

The statistical results in Table 5 show that there was no significant main effect of the three cover crop treatments on available P. This indicates that, contrary to what was hypothesized, the presence of cereal rye or oats did not affect the overall available P accumulation of OSR. Soil depth, however, had a significant effect on available P in all cases, with the greatest concentrations near the soil surface and decreasing with

Table 4. Cover crop shoot biomass accumulation and P content with site in the OSR/Rye treatment during spring 2012.

	OSR/Rye	
Site	**DM (kg ha^{-1})**	**P (kg ha^{-1})**
DTCS	1,926	6.44
DTCW	1,787	5.09
TPAC	3,345	9.78

Notes: OSR = oilseed radish; DM = cover crop dry matter; P = cover crop tissue phosphorus; DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat.

Table 5. Statistical significance for the split-plot, split-block model used to analyze the three cover crop treatments, time (fall and spring), row position, and soil depth at each site.

	DTCS		DTCW		TPAC		DTCC*
Source of variation	**WSP**	**MP**	**WSP**	**MP**	**WSP**	**MP**	**MP**
Treatment							
Position		X		X		X	
Treatment x position							
Time	X			X			X
Treatment x time		X					
Position x time						X	X
Treatment x position x time							
Depth	X	X	X	X	X	X	X
Treatment x depth							
Position x depth		X		X		X	
Treatment x position x depth				X			
Time x depth						X	
Treatment x time x depth							
Position x time x depth				X		X	
Treatment x position x time x depth							

Notes: DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat; DTCC = Diagnostic Training Center, cover crops following corn; WSP = water soluble phosphorus; MP = Mehlich-3 phosphorus.
*Comparisons of MP only were made at the DTCC site.

Table 6. Statistical significance for the split-plot, split-block model used to analyze all four treatments (OSR, OSR/Oats, OSR/Rye, and no-cover control), time (fall and spring), and depth within each row position at each site.

	DTCS		DTCW		TPAC		DTCC*
Source of variation	**WSP**	**MP**	**WSP**	**MP**	**WSP**	**MP**	**MP**
Root zone							
Treatment							
Time	X			X			X
Treatment x time		X		X		X	
Depth	X	X	X	X	X	X	X
Treatment x depth				X			
Time x depth				X		X	
Treatment x time x depth							
Buffer zone							
Treatment							
Time	X			X		X	X
Treatment x time				X			X
Depth	X	X		X	X	X	X
Treatment x depth							
Time x depth					X		
Treatment x time x depth							

Notes: DTCS = Diagnostic Training Center, cover crops following soybeans; DTCW = Diagnostic Training Center, cover crops following wheat; TPAC = Throckmorton-Purdue Agricultural Center, cover crops following wheat; DTCC = Diagnostic Training Center, cover crops following corn; WSP = water soluble phosphorus; MP = Mehlich-3 phosphorus.
*Comparisons of MP only were made at the DTCC site.

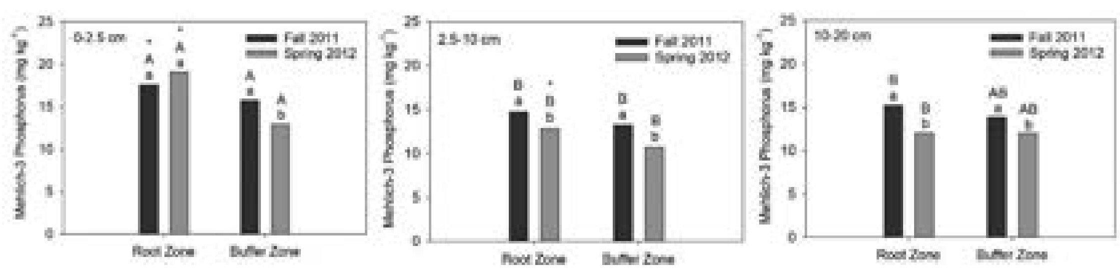
depth. The depth effect of available P and P-stratification have been well documented (Sharpley 2003; Smith, Huang, and Haney 2017), and our results do not add new insight, so we will not focus on the metric of soil depth by itself. Water soluble P generally had no significant differences other than depth, suggesting WSP may not be impacted by cover crops to the extent of MP. Therefore, further discussion will be limited to MP results.

Cover crop row position had a significant effect on MP, with the root zone having higher MP than the buffer zone. This supports our hypothesis and is also consistent with results from White and Weil (2011). There were also several interactions between row position and other parameters. There was a significant position × time interaction for MP at two sites (TPAC and DTCC), indicating that the difference between the root zone and buffer zone was different at different times. There was a significant position × soil depth interaction at three sites (DTCS, DTCW, and TPAC), indicating the available P differences between the root zone and buffer zone were different at different depths. There was also a significant three-way interaction for MP between row position, soil depth, and time at two sites (DTCW and TPAC). An example of the three-way interaction is shown in Figure 3 for DTCW. The most pronounced differences in MP occurred at the soil surface, where MP was greater in the root zone than in the buffer zone in both fall and spring. Mehlich-3 P decreased with depth within each row position at each time, and it decreased from fall to spring at all depths and row positions except for in the root zone at the 0 to 2.5 cm depth. These interactions reflect that the root zone had higher MP than the buffer zone primarily in the 0 to 2.5 cm depth, and that this position effect persisted in both fall and spring at this depth only. This three-way interaction highlights the tendency of cover crop roots to mobilize available P and accumulate it near the OSR roots and the soil surface. The results were relatively consistent at the three sites seeded in August 2011. The DTCC site seeded in September 2012 had fewer significant effects and interactions, likely due to much lower growth of the cover crop that year.

There was one case (DTCW) of a significant interaction between treatment, row position, and soil depth, indicating the row position × soil depth interaction varies by treatment (Figure 4). As expected, MP decreased with soil depth for all treatments and row positions. Mehlich-3 P decreased with distance from the row (position) primarily at the soil surface, and the effect of row position became less prominent lower in the soil profile. Cover crop treatment did not have a significant effect except for in the root zone of the 2.5 to 10 cm depth, where there was less MP under OSR/Oats than under OSR alone. This may be due in part to greater root and shoot growth with oats included than in the OSR alone, although the higher shoot biomass (2,966 versus 2,302 kg ha^{-1}) (Table 2) was not statistically significant.

In summary, when comparing the three cover crop treatments shown in Table 5, the amount of MP in the root zone versus the buffer zone

Figure 3. Position by time by depth interaction for Mehlich-3 P across three cover crop treatments at the DTC wheat site. Times within a row position containing similar lowercase letters are not significantly different. Depths within time and row position containing similar uppercase letters are not significantly different. Times within a row position containing an asterisk are significantly different than the same time in the other row position.



varied with time, soil depth, and occasionally cover crop treatment. In general, there was more MP in the root zone and near the soil surface than in the buffer zone and lower depths at different times. The MP generally decreased from fall to spring except in the root zone near the soil surface. These results highlight the effect that time can have on the vertical and horizontal distribution of available P from cover crops, which has been observed by other investigators (Schipanski et al. 2014).

The statistical results in Table 6 show that there were no significant main effects among the cover crop treatments, including the no-cover control, within each row position. This indicates that, contrary to what was hypothesized, the presence of cover crops did not affect overall available P accumulation compared to where no cover crop was grown. However, there were some significant interactions between the cover crop treatments and other parameters. Within each row position, there were some significant interactions between treatment and time. To illustrate the treatment × time interaction for each row position, see Figure 5 for DTCW. In fall there were no treatment differences for MP in the root zone or buffer zone. In spring there were significant treatment effects with similar trends in each row position. Mehlich-3 P was higher for OSR than the no-cover control, while there were no differences between the bicultures and the control. Between fall and spring, MP decreased for six of eight comparisons: OSR/Rye and the no-cover control in the root zone, and all four treatments in the buffer zone. Though it could be logical to attribute the decrease of MP over time to cover crop uptake of available P between fall and spring in the OSR/Rye treatment, a similar trend in the no-cover

Figure 4. Treatment by position by depth interaction for Mehlich-3 phosphorus (P) across fall and spring at the DTC wheat site. Treatments within a row position containing the same lowercase letters are not significantly different. Treatments within a row position containing the same uppercase letters across depths are not significantly different. Treatments within the root zone row position containing an asterisk are significantly greater than the same treatment in the buffer zone row position.

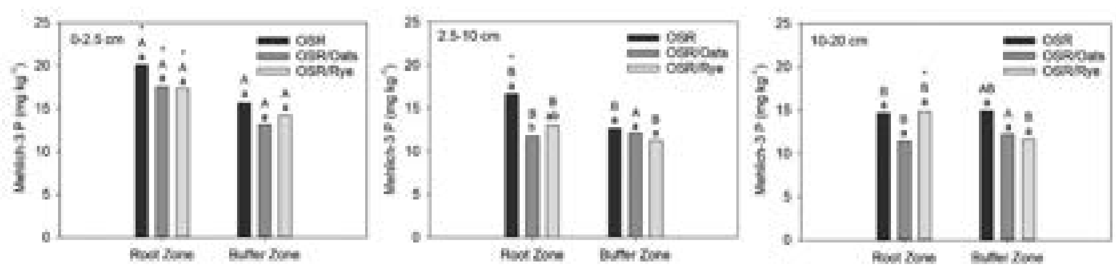
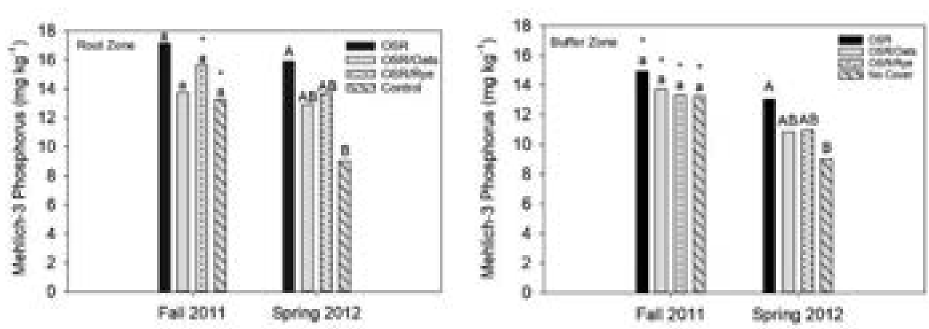


Figure 5. Treatment by time interaction for Mehlich-3 phosphorus averaged across depths in the root zone (left) and buffer zone (right) at DTC wheat site. Treatments within a time containing the same letters are not significantly different. Treatments containing asterisks are significantly different than the same treatment at the other time. OSR is oilseed radish.



control suggests cover crop uptake is not the sole factor. The fact that the MP is different among treatments in the spring but not in the fall indicates the extent of cover crop P uptake and soil P loss between fall and spring is not equal across treatments. A change in MP over time in the absence of cover crops could be attributed to environmental factors such as surface losses, leaching of P through the soil profile, biological activity, or a combination of all of these (Hallama et al. 2019). The two instances where MP did not decrease between fall and spring, the root zone of OSR/Oats and OSR, were similar in that they are the two winter-kill treatments consisting solely of dead cover crop material in spring. It is possible that in the root zone near the decaying radish tuber the nonliving OM provided sorption sites to retain nutrients, including P, thus retaining available P from fall to spring in the vicinity of the tuber. Lehman et al. (2012) found oats to be particularly effective at increasing arbuscular mycorrhizal fungi, thereby increasing P availability, and brassicas to have no negative effect. So, a stimulated microbial community and subsequent shift in P dynamics as observed by Hallama et al. (2021) is likely at play here as well.

When comparing the cover crop treatments to the no-cover control, there were several interactions for MP with soil depth in the root zone (Table 6). A treatment × depth interaction at DTCW (Figure 6) showed that the depth effect was significant for all treatments, but the treatment effect was only significant in the top two depths, where OSR had more MP than the no-cover control. A time × depth interaction at DTCW (Figure 7) shows there was more MP at the soil surface than in the bottom two depths during both fall and spring. There was no change between fall and spring at the soil surface, but there was a decrease in MP at the bottom depths between fall and spring. As mentioned earlier, the root zone MP may have remained elevated at the soil surface over time as a result of P being sorbed and retained by nonliving OM, or it could be a result of P being released by decomposing plant material. Shifting microbial dynamics likely played a role as well (Hallama et al. 2021; Honvault et al. 2021; Lehman et al. 2012). Regardless of the mechanism, the decrease in MP at the bottom depths over time suggests the contribution of available P from cover crops primarily affects the soil surface.

In summary, when comparing the three cover crop treatments and the no-cover control shown in Table 6, the amount of MP in the root zone and buffer zone varied with time and soil depth and changed even in the absence of cover crops. These results underscore the dynamic nature of

Figure 6. Treatment by depth interaction for Mehlich-3 phosphorus (P) averaged across fall and spring in the root zone at the DTC wheat site. Treatments within a depth containing the same lowercase letters are not significantly different. Treatments containing the same uppercase letters at different soil depths are not significantly different than the same treatment at the other depths. OSR is oilseed radish.

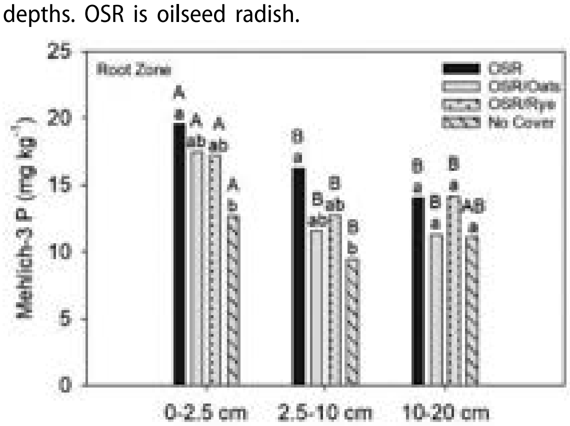
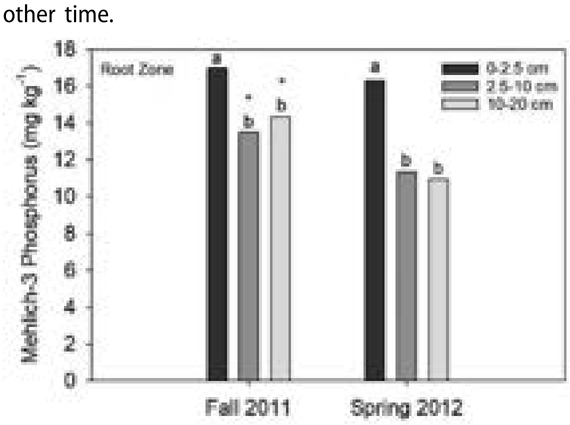


Figure 7. Time by depth interaction for Mehlich-3 phosphorus averaged across the cover crop root zones and the no-cover control at the DTC wheat site. Depths within a time containing the same letters are not significantly different. Depths containing asterisks at one time are significantly different than the same depth at the other time.



cover crops and their effects on nutrients and available P over time, in addition to the inherently dynamic nature of soil P that exists despite the crop.

Soil samples taken at the V6 stage of corn in 2012 did not show any significant differences among the cover crop treatments including the no-cover control. The historic drought of 2012 severely impacted the corn growth and nutrient uptake and likely the soil microbial processes, such that findings would likely not be indicative of typical behavior in these soils.

Overall, after comparing cover crop treatments to each other and to the no-cover control across row position, soil depth, and time, a few key points can be taken away regarding the effects of OSR and OSR bicultures on available P. First, the effect of the cover crops on available P was not always as hypothesized. The effects tended to be small and difficult to detect due to the dynamic nature of soil P and the episodic nature of P release from OSR. The inclusion of cereal crops along with OSR had only minor effects. Second, the dynamic nature of soil P is further exemplified by the comparison of the cover crop treatments to the no-cover control, where treatments had different relationships with each other at different times. While there were occasional differences between the winter-killing and winter-surviving cover crops compared to where there was no cover crop, the inclusion of oats and cereal rye with OSR did not consistently affect available P. Third, horizontal accumulation of available P (row position) had different relationships with time and depth, meaning that available P in the root zone versus the buffer zone varied by time and depth. The root zone tended to have higher available P than the buffer zone primarily in the surface soil, and it maintained that higher level from fall to spring, whereas deeper depths did not. Fourth, vertical accumulation of available P (depth) in this study was not different than would normally be expected for agricultural soils despite the presence of cover crops, matching observations from previous work (Sharpley 2003; Smith, Huang, and Haney 2017) and underscoring what little influence management has on soil P stratification. The occasional treatment differences observed suggest that although including cereal crops with OSR can impact the horizontal or vertical accumulation of available P, it is highly dynamic and unpredictable.

The results suggest some intriguing questions for nutrient management and future research. Standard soil sampling typically composites multiple soil probes taken without strict regard to row position or the presence/absence of cover crops in the field. A major finding of this study is the greater available P near the soil surface in the root zone near the OSR tuber compared to the buffer zone. Standard soil sampling would not detect this small-scale spatial variability in available P because it mixes samples from all row positions. Although overall P availability at a bulk level may not be affected by OSR or the OSR bicultures, it is possible that the succeeding cash crop roots might be able to take advantage of the “hot spots” (Kuzyakov and Blagodatskaya 2015) of available P near the decomposed tuber. Some producers intentionally seed OSR in rows where they will then plant their corn the following year, primarily for the biotilling effect (John Pike, personal communication, August 15, 2024), but it’s possible the corn could also gain nutrients from the OSR root zone. Future research could study microbial activity in the root zone versus buffer zone of the OSR (with or without cereal biculture) and more frequent sampling, especially in the spring as the tuber starts to decompose. Linking microbial activity, soil carbon pools, and available P dynamics in close proximity to the OSR tuber, could provide insight on the potential added benefits of precision planting of cover crops.

SUMMARY AND CONCLUSIONS

There are many well-documented benefits to growing any species or mix of cover crops, including OSR and cereal crops such as oats and cereal rye. While research has shown that cover crops are effective at retaining soil nutrients and have the ability to improve the cycling of available P through an array of mechanisms, there is still not strong evidence that the nutrient cycling functions of cover crops are predictable enough to make concrete nutrient management recommendations. We found this to be especially true of P and OSR. This is consistent with previous work aimed at developing sound recommendations around the P retention and cycling ecosystem services of cover crops, which overwhelmingly concluded that P is difficult to measure because of its dynamic nature. This study set out to address that challenge with a rigorous experimental design and statistical model focused on capturing the vertical and horizontal accumulation of P over time, and the results seem to underscore what previous efforts had found. This may highlight that soil P is even more dynamic than what was understood in the past, which cover crops alone are unlikely to disrupt. This is not surprising, as previous work identified numerous mechanisms constantly at play affecting available P in a cover crop system (Hallama et al. 2019; Honvault et al. 2021). Hallama et al. (2019) stated that effects of cover crops on available P are difficult to detect and cautioned against focusing on singular functions and overemphasizing particular ecosystems services such as P cycling. Our conclusion is in agreement with that statement. It is likely that impactful environmental P reduction strategies will require multiple conservation practices that are complementary to cover crops and that P retention and cycling alone should not be the driving factors for planting cover crops. At minimum, this work highlights the tradeoff that must be considered when managing cover crops in areas where P is a major resource concern.

Future research on P cycling with cover crops could investigate the potential importance of "hot spots" near the OSR tubers for subsequent P uptake by the cash crop. Precision planting subsequent crops on OSR rows is already done by some producers and might provide a practical P management strategy, in addition to the biotilling effect.

ACKNOWLEDGMENTS

Thank you to Kaylissa Halter for discussions about project ideas and assistance with data collection. Thanks to Judy Santini for help with statistical analyses.

DISCLOSURE STATEMENT

No potential conflict of interest was reported by the author(s).

FUNDING

This research was part of a regional collaborative project supported by the USDA National Institute of Food and Agriculture, Award No. 2011-68002-30190, Cropping Systems Coordinated Agricultural Project: Climate Change, Mitigation, and Adaptation in Corn-based Cropping Systems.

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