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Ecosystem service delivery by cover crop mixtures and monocultures is context dependent

Emily R. Reiss | **Laurie E. Drinkwater**

School of Integrative Plant Science, Horticulture, Cornell University, Ithaca, NY 14853, USA

Correspondence
Emily R. Reiss, School of Integrative Plant Science, Horticulture, Cornell University, Ithaca, NY 14853, USA.
Email: err76@cornell.edu

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Abstract

While cover crops and mixtures are increasingly used to provide ecosystem services in agroecosystems, some fundamental questions remain about how cover crop performance and composition vary in different conditions, limiting optimal cover crop use. We conducted a field experiment at a research farm in New York, including a subset of treatments in three working farm fields. We selected two common cover crops, hairy vetch (*Vicia villosa* Roth), a legume, and common wheat (*Triticum aestivum* L.), a grass, with multiple cultivars of each. We looked at the effect of cover crop composition spanning intraspecific and grass–legume mixtures on five ecosystem services: cover crop productivity, weed suppression, total biomass nitrogen, soil N retention, and long-term N supply via legume fixed N. We did not find intraspecific diversity to have an effect on any ecosystem services we measured, nor was that response context dependent. We did observe significant ecosystem service improvements in the grass–legume mixture, though this was context dependent and the performance of the mixture varied relative to the monocultures at different farm sites. Regardless of this interaction however, the grass–legume mixture was as good as or better than either monoculture for all services and sites, except soil N accrual at one site. Consequently, increasing complexity in cover crops through grass–legume mixtures is a low risk practice that may have the potential to deliver ecosystem service outcomes greater than those of monocultures across a range of growing conditions.

1 | INTRODUCTION

Cover crops are increasingly used by a wide range of farmers to support various ecosystem services from erosion control to pest regulation, with nutrient management and soil health as high priorities (Dunn, Ulrich-Schad, Prokopy, Myers, & Watts, 2016; Schipanski, Barbercheck, Douglas, Finney, & Haider, 2014; Wayman, Kissing Kucek, Mirsky, Ackroyd, & Cordeau, 2016). Cover crops can be any plant species established when ground is usually fallow between cash crops, with the goal of promoting certain ecosystem services to support cash crop yields or reducing externalities, such as nutrient leaching (Doltra & Olesen, 2013; Ritter, Scarborough, & Chirnside, 1998). Research on the benefits and management of cover crops has a long history, and has also been increasing rapidly in recent years along with grower adoption (Clark, Decker, & Meisinger, 1994; Creamer, Bennett, & Stinner, 1997; Schipanski et al., 2014). However, given the wide range of possible cover crop practices and on-farm context of soil and climate and management conditions, providing generalized guidelines and consistent delivery of desired ecosystem services remains a major management challenge (Myers & Watts, 2015).

Mixtures of cover crops, specifically those with grasses and legumes, are commonly used by farmers because of the multiple benefits they can provide (Appelgate, Lenssen, Wiedenhoeft, & Kaspar, 2017; Myers & Watts, 2015; Snapp, Swinton, Labarta, Mutch, & Black, 2005; Valkama, Lemola, Känkänen, & Turtola, 2015). When appropriately implemented, these mixtures can deliver the strengths of each functional group simultaneously, minimizing the tradeoffs of each group growing alone (Brainard, Bellinder, & Kumar, 2011; Kaye, Finney, White, Bradley, & Schipanski, 2019; Ranaldo, Carlesi, Costanzo, & Bàrberi, 2020; Schipanski et al., 2014; White, DuPont, Hautau, Hartman, & Finney, 2017). Because of these benefits, grass–legume mixtures, often referred to as intercrops, are widely used in agricultural systems from cover crops to pastures and forage and grain production (Baba, Halim, Alimon, & Abubakar, 2011; Bedoussac, Journet, Hauggaard-Nielsen, Naudin, & Corre-Hellou, 2015; Nyfeler, Huguenin-Elie, Suter, Frossard, & Connolly, 2009; Osman & Diek, 1982; Vandermeer, 1989). In symbiosis with root rhizobia, legumes fix nitrogen (N) contributing to long-term N supply, and lowering the carbon to nitrogen (C/N) ratio for fast residue decomposition and nutrient availability for subsequent crops (Brainard et al., 2011; Ruffo & Bollero, 2003). Conversely, grasses tend to take up soil N more efficiently, and have greater aboveground biomass which results in more organic matter accumulation and better weed suppression than legumes (Kruidhof, Bastiaans, & Kropff, 2008; Sainju, Whitehead, & Singh, 2005).

While grass–legume mixtures do generally deliver these multiple benefits and exhibit synergy when combined, the performance of these mixtures can be variable depending on environmental conditions and plant community composition (Alonso-Ayuso, Gabriel, García-González, Del Monte, & Quemada, 2018; Bybee-Finley, Mirsky, & Ryan, 2016; Poffenbarger, Mirsky, Weil, Maul, & Kramer, 2015b; Ranells & Wagger, 1997; Sainju et al., 2005). Critically, environmental and management conditions (e.g., manure applications) can each influence the dynamics of a grass–legume mixture, thus ultimately affecting the ecosystem service outcomes (Poffenbarger, Mirsky, Weil, Kramer, & Spargo, 2015a; Schipanski & Drinkwater, 2012). Given a certain context and growth conditions, the ecosystem service delivery of a species may be altered when in a mixture (Murrell, Schipanski, Finney, Hunter, & Burgess, 2017; Ranells & Wagger, 1997). For example, the rate of symbiotic N fixation by hairy vetch (*Vicia villosa* Roth) was less in monoculture compared to when mixed with a grass, but this was not consistent across cultivars (Brainard, Henshaw, & Snapp, 2012).

Core Ideas

- Overall, the cover crop response to increasing composition complexity was context dependent.
- Farm history and site conditions affected the performance of mixtures relative to monocultures.
- In general, the grass–legume mixture was the same or better than the best monoculture.
- The wheat–vetch mix produced equivalent fixed N as pure vetch at half the seeding rate and cost.

Understanding the potential interaction between community composition and the on-farm context for growth is critical for successfully managing cover crop mixtures on a farm, as well as understanding how to transfer research results to a wider range of conditions. While frequently included in plant breeding research, evaluating the treatment × environment interaction or the impact of context is not as common an objective in other fields (Helland & Holland, 2003; Sinebo, 2005). Background soil types and characteristics, weather, management histories, and current practices can vary across landscapes and farms (Asrat, Yesuf, Carlsson, & Wale, 2010; Blesh, VanDusen, & Brainard, 2019; Drinkwater, 2016; Schipanski & Drinkwater, 2011). All of these factors influence the growing environment and form the context for crop growth, including that of cover crop mixtures.

While grass–legume mixtures take advantage of different plant functional groups to balance potential tradeoffs, a similar moderating effect may be achieved by increasing the genetic and phenotypic diversity within a species. Mixing cultivars increases the intraspecific diversity within a monoculture of one species. The limited research on the effect of intraspecific diversity in agroecosystems suggests that there could be benefits for cover crops and their associated ecosystem services (Grettenberger & Tooker, 2015; Kiær, Skovgaard, & Østergård, 2009; Mundt, 2002; Smithson & Lenne, 1996). For example, in cash crops, cultivar mixtures have been found to increase yields as well as reduce the spread of disease (Mundt, 2002; Reiss & Drinkwater, 2018). Cultivars have long been developed and used in food crops to enhance specific desirable traits and to tailor a crop to regional conditions (Elzebroek, 2008; Fu, 2015). Given the wide range of cultivars available and the varying responses of these cultivars to different conditions, the identity of the cultivar used is likely a large driver of ultimate crop performance (Brainard et al.,

TABLE 1 Measured variables to evaluate corresponding ecosystem service

Collected metric	Ecosystem service
Soil N accrual in cover crop biomass (N_S)	Soil N retention
Shoot fixed N (N_F)	N supply, long-term
Total biomass N (N_T)	Potential N supply for the following crop
Weed biomass	Weed suppression
Cover crop biomass	C accrual

2012; Canali, Ortolani, Campanelli, Robacer, & von Fragstein, 2017; Helland & Holland, 2001; Mengistu, Baenziger, Nelson, Eskridge, & Klein, 2010; Ramirez-Garcia, Carrillo, Ruiz, Alonso-Ayuso, & Quemada, 2015). However, this cultivar diversity is not commonly utilized for cover crops.

We were interested in addressing these outstanding questions about the site context effect on cover crop performance and ways to moderate this effect while building practical management recommendations. To do this, we designed a nested set of experiments to evaluate the role of cultivars and grass–legume mixtures in maintaining cover crop performance across a range of contexts. We selected nine cultivars of two cover crops species, hairy vetch, a legume and common wheat (*Triticum aestivum* L.), a grass. We tested this set of cover crops at a university research farm as part of a larger experiment addressing diversity in cover crop mixtures, and established these treatments at three working farm sites as well. We looked at the effect of cover crop identity and composition in three ways on five ecosystem services that farmers seek to obtain from agroecosystems: soil carbon accrual, weed suppression, total biomass N, soil N retention, and long-term N supply using corresponding measurable metrics (Table 1; Millennium Ecosystem Assessment Program, 2005). We first evaluated the effect of increased intraspecific diversity by comparing the performance of the cultivar mixture to the mean of cultivar monocultures on the above-mentioned ecosystem services. Second, we tested for differences in the performance of the individual cultivars between each other and the cultivar mixture. Finally, we evaluated the effect of mixing vetch and wheat compared to each alone. We tested for an interaction between farm and each of these three characterizations of cover crop composition on the five ecosystem services. Overall, we expected there to be interactions between farm site conditions and cover crop identity, but that the strength and specific effect would be dependent on the context and the services evaluated.

2 | METHODS

2.1 | Plot establishment and management

We established a nested set of experimental trials at three organic, mixed vegetable farms (Farms 1–3) in the Finger Lakes region of New York State, as part of a larger trial at Cornell University Musgrave Research Farm (Farm 4) in Aurora, NY (42.73′ N, 76.66′ W). Details of the farm sites and management can be found in Table 2. Even though all the farm sites were within 25 mi of one another, some edaphic soil factors as well as management histories differed, resulting in varied conditions across the sites. The four farms were situated on three different soil series, though they were all classified as silt loams. When actual soil samples from each farm were evaluated for texture, all but Farm 2 (clay loam) were loam. The different background soil conditions along with varying management histories serve to create a diverse set of site conditions across these four farms (Table 2).

Farm 1 was a high fertility site, particularly in terms of inorganic N, phosphorus (P), and potassium (K) likely due to long-term compost additions over the past two decades of organic vegetable production. However, other important soil characteristics such as total carbon, N mineralization potential, and cation exchange capacity were not notably higher than other sites. Background weed pressure was generally low, though there was substantial variability in sampled biomass across the field due to large brassica weeds (*Barbarea vulgaris*) in the sampling area of one cover crop control plot that were not evenly dispersed through the field (range 25–356 g m^{-2} in control plots). This farm maintains permanent beds 6 ft wide with grass strips separating the beds. This allows for intensive cultivation in the growing zone without wheel compaction from equipment. Tillage is typically done with a power take-off (PTO)-driven rototiller implement at the appropriate depth for the task (i.e., terminating a cover crop, seed bed preparation). Cover crops are heavily used in rotation with annual vegetables.

Farm 2 had been in a conventional corn (*Zea mays*) and soybean (*Glycine max*) rotation for over 20 yr prior to conversion to organic hay and mixed vegetables 5 yr prior to the experiment establishment. This may explain the low soil carbon and less soil nutrients overall compared to Farm 1. Weed biomass in control plots was comparable to typical levels at Farm 1 (13–63 g m^{-2}). Tillage at this farm is minimal and accomplished using animal-powered equipment. Fertility at this farm comes primarily from cover crops and on-farm animal manure.

TABLE 2 Site conditions at the four farm sites. Mean and standard deviation (in parentheses) for all variables except for weed biomass (range in parentheses) are shown. Different uppercase letters for a given variable indicate significant differences among farms based on Tukey's HSD test ($p < .05$). Farms 1–3 are small-scale working farms, and Farm 4 is a university-managed research farm

	Farm 1	Farm 2	Farm 3	Farm 4
Inorganic N, mg N kg^{-1} soil	46.5 (3.0) A	13 (3.4) B	6.5 (1.1) B	11 (1.4) B
N mineralization potential, mg N kg^{-1} soil $week^{-1}$	15.4 (2.9) B	17.6 (1.3) B	33.4 (1.8) A	8.8 (2.0) C
Soil N, %	0.197 (0.009) AB	0.164 (0.004) B	0.248 (0.036) A	0.187 (0.017) B
Soil C, %	1.76 (0.07) BC	1.48 (0.05) C	2.40 (0.40) A	2.05 (0.08) AB
C/N	8.9 (0.2) A	9.0 (0.1) A	9.7 (0.5) AB	11.0 (0.7) B
pH	6.9 (0) B	6.5 (0.1) C	5.8 (0.2) D	7.7 (0.1) A
P, mg kg^{-1}	437 (48) A	50 (4) B	37 (8) B	17 (1) B
K, mg kg^{-1}	196 (39) A	113 (19) B	70 (21) B	76 (8) B
Mg; mg kg^{-1}	237 (29) B	164 (18) C	107 (12) C	315 (26) A
Ca, mg kg^{-1}	2020 (362) A	1330 (37) B	1284 (126) B	2399 (74) A
CEC[a]	12.6 (2.1) AB	10.3 (0.3) B	12.0 (2.0) AB	14.8 (0.6) A
K, % saturation	4.0 (0.3) A	2.8 (0.5) B	1.5 (0.3) C	1.3 (0.1) C
Mg, % saturation	15.8 (0.8) A	13.3 (1.2) B	7.4 (0.5) C	17.7 (0.9) A
Ca, % saturation	80.3 (0.7) A	64.5 (1.2) B	54.0 (6.5) C	81.0 (1.0) A
Sand, %	40.7	37.2	36.7	44.6
Clay, %	25.5	27.9	25.3	22.7
Soil textural class	Loam	Clay Loam	Loam	Loam
Soil series	Honeoye silt loam (HnB)	Honeoye silt loam (HnB)	Erie silt loam (ErB)	Lima silt loam (LtA)
Field prep prior to planting	Rototill	Chisel plow	Moldboard plow	Disk
Field treatment after seeding	Shallow rototill	None	Field cultivate	Cultipack
Crop history	20 yr of mixed vegetables and cover crops	Conventional corn–soy for 20+ yr, then 5 yr of hay and mixed vegetables and cover crops	Hay for 10+ yr then 2 yr of mixed vegetables	Conventional corn–soy–wheat rotation as part of University research farm
GDD, °C (Spring, base 0 °C)	851	856	755	1,016
Background weed biomass (g m^{-2})[b]	191 (25–356)	29 (13–53)	158 (97–219)	46 (11–87)
Harvest date	2 June	3 June	29 May	10 June

[a]CEC, cation exchange capacity; GDD, growing degree days.
[b]At Farm 1, there was an uneven distribution of large *Brassica* weeds in sampling area resulting in an extremely large biomass range. See text for additional details.

Farm 3 had been hayed for 20 yr prior to conversion to mixed vegetables 2 yr before we established the experimental plots. This history is reflected in the high organic matter content specifically, and in the moderate levels of soil nutrients (Gregory, Dungait, Watts, Bol, & Dixon, 2016). The very high background weed pressure (97–219 g m^{-2}) may also be due to this history. The low pH at this site (5.8) may inhibit legume growth such as vetch cover crops (Clark, 2007). Most tillage at this farm is accomplished with a small horsepower tractor with implements attached to a rear three-point hitch and PTO driven. Low levels of off-farm poultry compost provided most of the fertility since transitioning to vegetable production, with no fertility added when managed for hay.

Farm 4, the research farm, is notable as the only conventionally managed site with no vegetable production (though the experimental plots were managed organically). Additionally, this is a relatively high pH soil, with low N mineralization potential and lower soil P levels. Together the lower P and higher pH may have resulted

TABLE 3 Description of the cover crop treatments applied at the four farm sites

Description	Treatment number	Cultivar or line
Vetch cultivar monocultures	1	'Purple Bounty'
	2	'Vetch VNS'
	3	'Ernst Vetch'
	4	'AL Vetch'
	5	'Purple Prosperity'
Wheat cultivar monocultures	6	'Cayuga'
	7	'Caledonia'
	8	'Houser'
	9	'Pride of Genesee'
Cultivar mixtures	10	all vetch cultivars planted together
	11	all wheat cultivars planted together
Grass–legume mixtures (1:1 vetch/wheat cultivar)	12	Vetch ('Purple Bounty')–wheat ('Caledonia')
	13	Vetch ('Purple Bounty')–wheat '(Houser')
	14	Vetch ('Albert Lea')–wheat ('Caledonia')
	15	Vetch ('Albert Lea')–wheat ('Houser')
Cover crop control	16	No cover crop planted, used to assess background weeds

in reduced plant-available P, which can also reduce cover crop growth (Clark, 2007). The background weed pressure was similar to that at Farms 1 and 2 (11–87 g m^{-2}). This farm has the most intensive tillage of any in the experiment, as the large tractors available allow for deeper and more frequent tillage. Fertility is typically applied to the cash crops in rotation as needed based on soil tests.

We selected a subset of treatments from the larger research farm trial to establish at these farms (Table 3). Specifically, we focused on one legume (hairy vetch), and one grass (common winter wheat). We selected hairy vetch as the legume for two main reasons. First, vegetable farmers in the region appreciate its winter hardiness and prolific N fixation potential in an easy to establish annual. While it does have drawback in terms of becoming a potential weed and being challenging to manage or terminate, it is one of a limited number of annual legumes that overwinter in the area. (The farmer at Farm 1 successfully mows the hairy vetch cover crop—alone or in mixture—to control growth and continue to gain benefits from the vetch well into the summer without unmanageable growth) Second, hairy vetch has the most well-established genetic diversity for an overwintering annual legume, with several commercially available cultivars many of which were developed through the hairy vetch breeding program based at USDA-ARS, Beltsville (Maul, Mirsky, Emche, & Devine, 2011). This allowed us the best opportunity to select disparate varieties. While rye is a more common overwintering grass used in cover crop mixtures in the area, winter wheat was selected as it has substantially greater genetic diversity available commercially, again allowing for a larger pool of varieties to mix. Wheat also has the benefit of a slightly lower C/N ratio and is not as vigorous making it slightly easier to manage.

All treatments used in the experiment are detailed in Table 3. For hairy vetch, we included five cultivars or lines (Treatments 1–5), and four cultivars of soft white winter wheat (Treatments 6–9). With these two species and their cultivars, we created mixtures of all cultivars to examine the effect of intraspecific diversity (Treatments 10 and 11), and mixtures of the two species to examine the effect of combining a grass and legume together (Treatments 12–15). We selected the cultivars and lines based on available trait and morphology information such as flowering time for vetch, wheat height, and fall vigor. When constructing the grass–legume mixtures (Treatments 12–15), we selected cultivars or lines with contrasting traits where possible. 'Purple Bounty' and 'Purple Prosperity' are early flowering vetch cultivars bred in a moderate climate, whereas 'AL Vetch' is later flowering vetch bred in a climate with lower average and minimum temperatures (Maul et al., 2011). 'Ernst Vetch' and 'Vetch VNS' are both "variety not stated" populations that are produced in different regions of the United States and may contain more genetic variability. 'Houser' wheat tends to have greater fall vigor than 'Caledonia' or 'Cayuga', while 'Pride of Genesee' is a tall statured, heritage cultivar in contrast to the three shorter, modern cultivars (M. Sorrells, personal communication,2015). 'AL Vetch' and 'Vetch VNS' were sourced from Albert Lea Seeds (Albert Lea, MN), while 'Ernst Vetch VNS' and 'Purple Bounty' were sourced from Ernst Conservation Seeds (Meadville, PA). Finally, 'Purple Prosperity' was sourced from the UDSA-NRCS National Plant Materials Center (Beltsville, MD). All wheat seed was generously provided by the Cornell Small Grains Breeding and Genetics Program.

Where treatments included multiple cultivars or species, the number of seeds was always split evenly, first by species (i.e., 50% wheat, 50% vetch), and then by cultivar (i.e., each vetch cultivar was 1/5 of the total seeding rate when all 5 cultivars were mixed). In addition to the cover crop treatment plots, one plot per replicate remained unplanted without any cover crops (Treatment 16) as a control to allow for assessment of the background weed pressure and variation across the fields (Table 2).

On all four farms we used a randomized, complete block replacement series with three blocks of the 16 treatments listed in Table 3. All plots were 2.5 by 2.5 m, with the exception of Farm 1, where the permanent bed size restricted the dimensions to 1.83 by 3.25 m. Regardless of plot dimensions, plots were planted at the same seeding density of 285 seeds m^{-2}, per the replacement series design. This seeding density is an appropriate seeding rate on a kg ha^{-1} basis for a wheat cover crop. The seeding rates as kg ha^{-1} varied by species and cultivar per differences in seed size, but were approximately 85 and 115 kg ha^{-1} for vetch and wheat monocultures respectively, and half that for each in the biculture composition (Table 1S). This resulted in a seeding rate higher (233%) than recommended rate for vetch. The design of this experiment is more similar to work on the biodiversity–ecosystem function relationship in unmanaged ecosystems (Tilman, Wedin, & Knops, 1996), than to proportional replacement series or additive designs used in crop diversity research in agroecosystems (Poffenbarger et al., 2015b).

We applied appropriate inoculant to all vetch seeds unless they were pretreated by the producer (N-Dure brand, Verdesian, Cary, NC). We planted all sites in mid-September 2014 within 10 d of one another (16–24 Sept.). The field was prepared by the farmers according to their typical practice for establishing cover crops, after which seeds were hand broadcast in the experimental plots. Following seeding, the field was treated according to farmer practice for seed incorporation. All sites were managed organically and received no inputs or other interventions such as irrigation during the trial.

2.2 | Data collection

We harvested aboveground biomass for all treatments once the vetch monocultures were at approximately 50% flowering (see Table 2 for specific dates). Farmers commonly terminate vetch at this time as it is unlikely to regrow, and at which time viable seed have not yet set. Biomass was cut 9 cm above the soil surface within a randomly selected quadrant area of 0.25 m^2 at least 20 cm from all edges. We recorded plant count for vetch, wheat, and weeds individually and separated biomass into paper bags which were oven-dried for at least 48 h at 60 °C before weighing to the nearest 0.01 g. We sampled weed biomass from cover crop control plots similarly but did not separate by species for weeds. All cover crop samples were ground to at least 2 mm. Wheat samples from the grass–legume mixtures were analyzed for total C and N on combustion using a LECO TruMac CN analyzer (Leco Corporation, St. Joseph, MO). Additionally, all vetch samples and wheat monoculture samples were processed for mass spectrometer isotopic analysis by first coarsely grinding, then grinding to 0.5 mm with a propeller mill (Cyclotec Sample Mill, Foss, Hillerød, Denmark). Samples were analyzed for ^{15}N natural abundance, total N content, and total C content using a continuous flow Isotope Ratio Mass Spectrometer (Stable Isotope Facility, University of California-Davis).

Using the ^{15}N natural abundance method, we estimated the symbiotic N fixation by legumes in monoculture and mixtures in unfertilized plots (Shearer & Kohl, 1986). For the following calculation for each legume sampled ($\delta^{15}N_{leg}$), we used the average ^{15}N signature of the wheat monoculture plots averaged by block, as the reference plant ($\delta^{15}N_{grass}$), where the B value, the isotopic fractionation of the N during fixation in the vetch, was determined as part of a previous study for each vetch cultivar (Supplemental Table S3).

$$\%\,\mathrm{N\,from\,fixation} = 100\left(\frac{\delta^{15}\mathrm{N\,grass} - \delta^{15}\mathrm{N\,leg}}{\delta^{15}\mathrm{N\,grass} - B}\right)$$

To characterize background soil conditions, we collected 10 soil cores to 20-cm depth for composite samples from each of the four replicates in the fall. A subsample of each composite sample was sieved to 2 mm, extracted for total inorganic N with 2 M KCl, incubated anaerobically for 7 d, then extracted with 2.67 M KCl for N mineralization. Total NH_4^+ and NO_3^- were analyzed via a colorimetric microplate technique (QuikChem, Lachet Instruments, Loveland, CO; Ringuet, Sassano, & Johnson, 2011). We dried each sample in the oven for 7 d at 65 °C to determine gravimetric water content. All samples were analyzed for total C and N on combustion (Leco Corporation, St. Joseph, MO). All fall soil samples were analyzed for water pH; cation exchange capacity; Mehlich buffer lime requirement; and for P, K, Mg, Ca, Zn, Cu, and S by the Mehlich 3 (ICP) test and particle size (Agricultural Analytical Services Laboratory, Penn State University, University Park, PA; Table 2).

Spring growth period growing degree days were calculated from the daily minimum and maximum air temperature as recorded by temperature loggers (Thermochron iButtons, Maxim, Sunnyvale, CA; http://www.maxim-ic.com/) at each site from 20 Mar. 2015 to the date of harvest for that site using 0 °C as the base temperature (Table 2).

3 | DATA ANALYSIS

3.1 | Ecosystem service calculations

We measured aboveground cover crop biomass (dry matter g m^{-2} as described above). Cover crop biomass directly contributes to soil organic matter levels and long-term soil C accrual, both of which are important for cash crop health and agroecosystem resilience. Aboveground biomass also indirectly influences a range of other services including weed suppression and total biomass N. We evaluated weed suppression as the total weed biomass harvested from the cover crop treatment plot.

We also evaluated cover crop N use as total biomass N, and then partitioned this into soil derived N (soil N retention) and fixed N by legumes (long-term N supply). Total mass of biomass N per m^2 (N_T) was calculated for all plants as %N times g of cover crop biomass m^{-2}. Nitrogen derived from the atmosphere through biological nitrogen fixation (N_F) in vetch was partitioned using the %N from fixation result from the ^{15}N natural abundance method. The mass of fixed N in vetch was calculated as %symbiotic N fixation times N_T. The soil N accrual (N_S) for vetch was calculated as $N_T - N_F$. For wheat, all biomass N is derived from the soil, so $N_S = N_T$. These N partitions are relevant for farmers when identifying goals from cover crops. For instance, on an integrated animal–crop farm, new N for crops may be easily supplied by manure. As such, N_F from legumes is less important than retaining the available soil N (N_S) in cover crop biomass until the next crop is ready for it. For an organic vegetable farm, N_F from legumes would the most likely foundation of the long-term N supply for that system (Drinkwater & Snapp, 2007). Regardless of the system or farmer's goals for cover crops, the N_T is an important measurement—often included in a residual N value—when evaluating N fertilizer needs for a subsequent crop, as that biomass N has the potential to be available to crops upon decomposition.

3.2 | Statistical analysis

Differences in soil characteristics by farm (Table 2) were tested with a mixed model (block as a random effect nested in farm and farm as a fixed effect) with differences in least squares method assessed by Tukey's Honestly Significant Difference (HSD) at $\alpha = .05$. Variables were assessed for homogeneity of variance and other assumptions for analysis of variance (ANOVA). Mixed models were used to test the effects of composition on ecosystem service outcome. We determined the effect of composition with three separate cover crop effect estimates. The three composition–diversity effects are as follows: (a) overall increased intraspecific diversity; (b) individual cultivar and cultivar mixture performance; and (c) functional group diversity in the grass–legume mixture. First, we evaluated the overall effect of increased intraspecific diversity by comparing the mean of the ecosystem service performance of cultivar monocultures to that of the cultivar mixture for each of the two species. Specifically, the mean of Treatments 1–5 was compared to Treatment 10 for vetch and the mean of Treatments 6–9 compared to Treatment 11 for wheat (Table 3). Second, we assessed the effect of the individual cultivar and cultivar mixture performance by comparing each cultivar monoculture to the others as well as to the cultivar mixture for each species. In other words, Treatments 1–5 and 10 were compared to one another for vetch and Treatments 6–9 and 11 for wheat. Third and finally, for the grass–legume mixture effect, we compared the performance of the four vetch–wheat mixes (Treatments 12–15) to the monocultures of each of the vetch and wheat cultivars alone (Treatments 1 and 4 for vetch and 7 and 8 for wheat). Treatment means by replicate were used to avoid pseudoreplication for the separate diversity–composition levels used in the analysis. For example, the mean of Treatments 1 and 4 together was calculated for each block representing monoculture vetch, and the mean of Treatments 12–15 together by block was also calculated for biculture of vetch and wheat. Consequently, for means presented in figures, the *n* for each treatment type–diversity level is equal to the number of blocks (3 at Farms 1–3, 4 at Farm 4). The mixed models included the main effect being tested (e.g., grass–legume mixture), the farm and their interaction along with block nested in farm as a random effect. Tukey's HSD at $\alpha = .05$ was used to test for differences between multiple levels, while two sample t-tests were used to compare two levels. Data was transformed to meet model assumptions. Variables and models where data was transformed are identified in figure legends and supplemental materials. For most analyses unless otherwise stated, a constant (1) was added to weed biomass and then transformed using natural log. All analyses were conducted using JMP v.11 software (SAS Institute, Cary, NC).

4 | RESULTS AND DISCUSSION

4.1 | Cover crop performance by farm

There were significant differences between vetch and wheat monocultures for the delivery of the five ecosystems among the four farms (Figure 1). Overall, Farm 1 had high or the highest ecosystem services performance measured by the performance of the vetch and wheat monocultures. Cover crop seed at this farm was incorporated

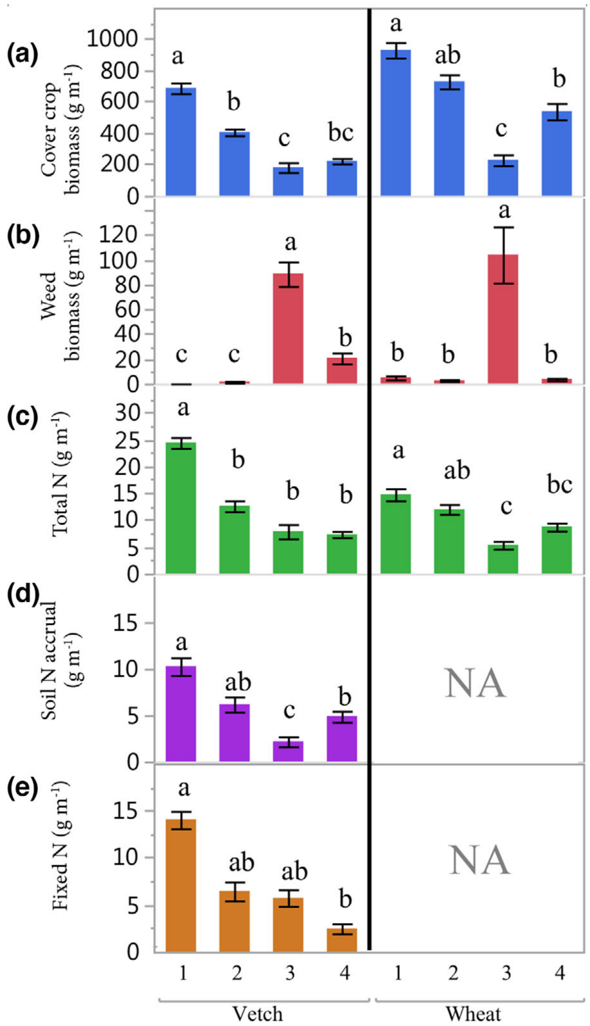

FIGURE 1 Average (a) aboveground cover crop biomass, (b) weed biomass, (c) total N in biomass, (d) soil N accrual, and (e) fixed nitrogen (mean ± standard error, SE) by farm (1–4) for all vetch and wheat monocultures. Different lowercase letters within a species and metric indicate significant differences based on Tukey's HSD test ($p < .05$). Analysis on transformed data was done (square root for a, c, d, and e; natural log for b), but untransformed data presented. See Supplemental Table S7 for more details

with a shallow rototill pass, which resulted in very consistent and high density establishment (Supplemental Figure S1). Farm 2 had relatively high cover crop biomass and ecosystem services performance, often equivalent to Farm 1, even though seeds were not mechanically incorporated after hand broadcasting, per typical farm practice. Farm 3 generally had some of the lowest performing cover crops across all ecosystem services measured. The success of weeds at this farm indicates that site productivity did not broadly inhibit cover crop performance. While we did not measure this, the seed may have been incorporated too deep for optimal establishment due to the use of a field cultivator for seed incorporation. Additionally, the low pH may have inhibited the legume growth as well as N mineralization from the large stores of organic matter under cooler temperatures (Curtin, Campbell, & Jalil, 1998; Fu, Xu, & Tabatabai, 1987). The research farm, Farm 4, was also slightly lower performing than Farms 1 and 2, perhaps also due to soil conditions, such as low P and high pH (Table 2). The substantial differences in cover crop and ecosystem services performance among the farms reiterates how important and influential site conditions can be for both practical and research outcomes.

4.2 | Intraspecific diversity

While there were substantial differences between farms in terms of overall cover crop performance (Figure 1), there was no effect of either measure of intraspecific diversity, and consequently, no interaction with farm (Supplemental Tables S4, S5). Specifically, we did not detect any differences in the mean cultivar monoculture performance compared to the cultivar mixture, nor did any of the cultivars differ from one another or from the cultivar mixture at a given site.

As we did not detect any differences among the cultivars (Supplemental Table S5), it would suggest that the range of phenotypic and trait diversity in the cultivar mixtures may have been too narrow. As such, it may not have been sufficient to result in a measurable effect on the target ecosystem services (Cadotte, Cardinale, & Oakley, 2008). The limited trait information available for the cultivars, especially for vetch, made this difficult. Additionally, vetch is an outcrossing species. Therefore, greater genetic diversity is contained within a given line or cultivar, making the relative increase in genotypic diversity lesser when multiple lines are planted together (Maul et al., 2011; Yeater, Bollero, Bullock, Rayburn, & Rodriguez-Zas, 2004). Three of the four wheat cultivars were modern cultivars developed for the New York region, and the fourth was a heritage variety long grown in this region as well. More diverse wheat cultivars from other areas could have been selected, but perhaps would not have been as representative of cultivars suited to the conditions.

The lack of discernable differences in cultivars or cultivar mixtures may also have been due in part to the average or above-average biomass production at all sites (Clark, 2007). As noted previously, our seeding rates were very high and may have contributed to this high biomass. Vetch was seeded as a monoculture at over 200% the recommended rate, whereas the wheat was seeded very close to recommended rates in terms of kg ha^{-1}. Despite these higher seeding rates, both the vetch and wheat established at much lower populations (Supplemental Figure S1;

Supplemental Table S2). Self-thinning and other biotic and abiotic pressures may have reduced the final stands from the high density at seeding (Park, Benjamin, & Watkinson, 2003). In fact, the final established plant density in vetch is nearly the same as the revised recommended rate for the region by Mirsky, Ackroyd, Cordeau, Curran, and Hashemi (2017). These authors suggest that in general, optimal biomass may be attained with lower than recommended rates for vetch (Figure 1; Supplemental Tables S1, S2), but of course there is a variable relationship between the seeding rate and final plant stand (Mirsky et al., 2017). Ultimately, the final outcome of these cover crops, such as productivity, is highly dependent on seeding date and time of termination (Baraibar, Hunter, Schipanski, Hamilton, & Mortensen, 2018). Given that these plots were planted at the later end of the fall cover crop window, a somewhat higher seeding rate would be appropriate, though our seeding rate was still excessive from an agronomic perspective (Mirsky et al., 2017). Especially given the large difference between seeded density and final density, our experimental rates and associated costs are not justified for a working farm, as the biculture cost of \$330 ha^{-1} would be prohibitive for most farms (Supplemental Table S1). While costs are an important concern, surveys suggest that cover crop use and adoption is not necessarily inhibited by the upfront costs (Dunn et al., 2016). Instead, evaluating the goals from a cover crop and working to estimate the benefits in return, such as new N from fixation instead of compost, is a logical step for determining appropriate seeding rates on-farm (Snapp et al., 2005).

4.3 | Increased complexity in grass–legume mixtures

Overall, the grass–legume mixture was as good as the best monoculture for three metrics: cover crop biomass, total N, and soil N accrual, whereas there were no differences for weed biomass or fixed N (Figure 2). Those three metrics with differences between the monocultures and mixtures, were each context dependent, with an interaction between the effect of grass–legume mixing and farm site (Table 4). Consequently, there were significant differences between the vetch monoculture; wheat monoculture; and the biculture at Farms 1, 3, and 4 for these metrics (Figure 3).

Even though the biomass response of grass–legume mixture was context dependent, there were some consistent patterns across the farms (Table 4). On three out of the four farms, the mixture had significantly greater biomass than the legume monoculture, and at Farm 3 mixture biomass was also significantly greater than the grass monoculture (Figure 3a). It is very common to grow grass–legume mixtures in large part because of the potential increase in

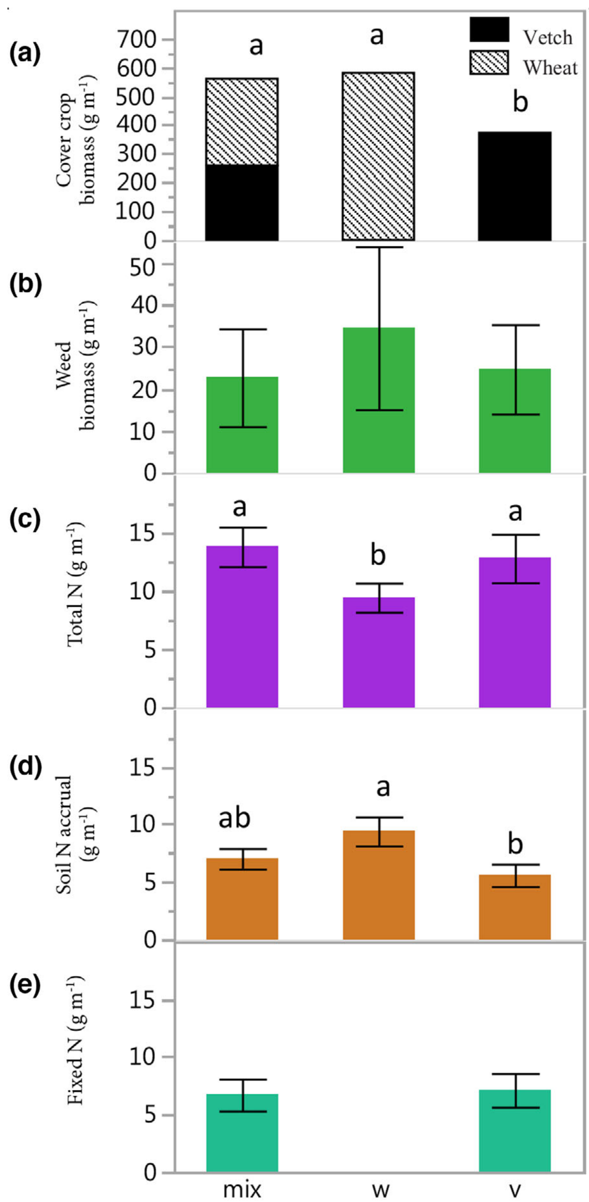

FIGURE 2 Effect of grass–legume mixture (mix) compared to wheat (w) and vetch (v) monocultures on (a) aboveground cover crop biomass, (b) weed biomass, (c) total N in biomass, (d) soil N accrual, and (e) fixed N (mean ± SE). Different lowercase letters within a metric indicate significant differences based on Tukey's HSD test ($p < .05$). Data was transformed for weed biomass (natural log) and soil N accrual (square root), but untransformed data are presented. Also see Table 4

total biomass from mixing a grass and a legume together (Bedoussac et al., 2015; Sainju et al., 2005). We generally did observe this biomass improvement, but there are clearly differences in the way the cover crop treatments responded to the site. There is often a concern that in soils with greater levels of N availability the grass might outcompete the legume in grass–legume mixtures, substantially reducing its biomass (Brainard et al., 2011; Poffenbarger

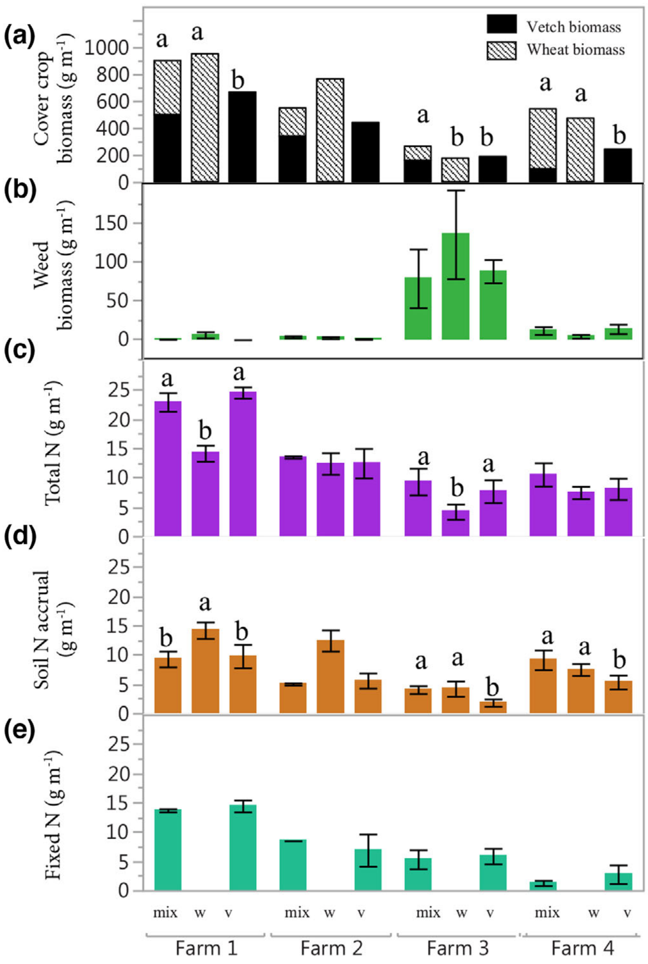

FIGURE 3 Mean effect of grass–legume mixture (mix) compared to wheat (w) and vetch (v) monocultures by farm on (a) aboveground cover crop biomass, (b) weed biomass, (c) total N in biomass, (d) soil N accrual, and (e) fixed N. Different lowercase letters within a farm and metric indicate significant differences based on Tukey's HSD test ($p < .05$). Data was transformed for weed biomass (natural log) and soil N accrual (square root), but untransformed data are presented. Also see Table 4

et al., 2015a; Staniforth, 1962). Given that vetch is a highly competitive legume, it's not surprising that across the four farms, vetch consistently made up 50–60% of the mixture (by density and biomass) with the exception of Farm 4 where it was closer to 50% by density and 20% by biomass. This 50–60% proportion is consistent across the three farms with varying overall grass–legume mixture responses (Figure 3a). More specifically, when the weeds are included as part of the nonlegume portion of the cover crop community, this difference is more dramatic (nonlegume: 60% by biomass for Farms 1–3, 83% at Farm 4). This distribution of legume to nonlegume ratios, along with the significant farm interaction, suggests that the context under which the cover crop is propagated can influence the effect of mixing grasses and legumes even when the ratio of legume to nonlegume in a mixture remains the same. With only four sites and a limited set of environmental variables measured, it is difficult to isolate potential factors driving these different mixture responses given similar mixture compositions, but it is an area that demands additional research.

We found that total N in the cover crop biomass and soil N accrual was context dependent in the response to the mixing of a grass and a legume (Figure 3; Table 4), and closely followed the pattern of biomass as expected. Using the natural abundance method (Shearer & Kohl, 1986), we were able to partition total N into soil N and fixed N. For wheat, total N is equal to soil N, but the vetch and mixture treatments have both soil N and fixed N.

Although total N is an important consideration for the nutrition of subsequent crops, soil N accrual and fixed N each have critical roles in the long and short-term nutrient management strategy for a farm. By mixing the legume and grass we hoped to take advantage of both of their strengths and minimize tradeoffs (Aronsson, Hansen, Thomsen, Liu, & Øgaard, 2016; Ranells & Wagger, 1996). Though competition from wheat resulting in suppressed vetch growth and reduced fixed N is a typical concern, three of the four farms had close to 60% vetch in the mixtures, indicating that the vetch was not outcompeted by the wheat (Poffenbarger et al., 2015a; White et al., 2017). The high seeding rate overall may have allowed the vetch in the mixture to establish at that final density despite the seedling mortality (Supplemental Table S2). Additionally, at only one of the four farms (Farm 1), the mixture was worse at soil N retention than the best monoculture. This was true even though the mixtures were generally evenly mixed with the vetch and wheat. The exception at Farm 1 was likely due to the high level of available nutrients and N, which the legume was not able to take up at the same rate as the more efficient grass. In contrast to total N and soil N accrual, there was no interaction and no difference between vetch and the mix for the amount or rate of fixed N (Table 4; Figure 3e; Supplemental Figure S2). Consequently, for all farms, we found that the amount of fixed N, as well as the percentage of N from fixation was not different in the mixture compared to the vetch monoculture. While other research has shown how site conditions, such as soil fertility, can dramatically alter these N service outcomes, we ultimately found that the majority of the mixtures succeeded at balancing the tradeoffs of the legume and grass across the farms (Blesh, 2019; Schipanski & Drinkwater, 2011; West, HilleRisLambers, Lee, Hobbie, & Reich, 2005). The mixture of the two functional groups maintained good soil N retention while contributing new N equivalent to that of legume monocultures through symbiotic N fixation (Kaye et al., 2019).

There was no interaction between farm and the effect of mixing grasses and legumes for weed suppression, even

TABLE 4 Mixed model results for effect of mixing grasses and legumes (mix) on five metrics of ecosystem service performance including interaction with farm site (also see Figure 2). When an interaction between mix and farm exists, farm sites with significant or nearly significant differences are presented. Data was transformed for weed biomass (natural log) and soil N accrual (square root). There was no interaction with farm and no diversity effect for either measure of intraspecific diversity (cultivar mixture effect or cultivar differences). *df* = degree of freedom in the numerator; *df*Den = degrees of freedom in the denominator

Ecosystem service	Effect of grass/legume mixing Biculture compared to wheat and vetch in monoculture				
	Source	*df*	*df*Den	F Ratio	**Prob > F**
			Cover crop biomass		
	Farm	3	9	19.074	.0003*
	mix	2	18	23.2569	<.0001*
	Farm × mix	6	18	4.0732	.0094*
	Farm 1	F(2,4) = 50.8383; *p* = .0014			
	Farm 2	F(2,4) = 5.8129; *p* = .0655			
	Farm 3	F(2,4) = 38.3787; *p* = .0025			
	Farm 4	F(2,6) = 9.4535; *p* = .014			
			Weed biomass		
	Farm	3	9	77.6326	<.0001*
	mix	2	18	1.098	.3549
	Farm × mix	6	18	2.4652	.0643
			Total N		
	Farm	3	9	17.2387	.0005*
	mix	2	18	16.8699	<.0001*
	Farm × mix	6	18	4.3524	.0069*
	Farm 1	F(2,4) = 111.7145; *p* = .0003			
	Farm 3	F(2,4) = 25.1934; *p* = .0054			
			Soil N accrual		
	Farm	3	9	8.0648	.0064*
	mix	2	18	25.0479	<.0001*
	Farm × mix	6	18	5.9612	.0014*
	Farm 1	F(2,4) = 16.26; *p* = .012			
	Farm 2	F(2,4) = 6.9661; *p* = .0498			
	Farm 3	F(2,4) = 20.3943; *p* = .008			
	Farm 4	F(2,6) = 13.9829; *p* = .0055			
			Fixed N		
	Farm	3	9	18.6615	.0003*
	mix	1	9	0.032	.862
	Farm × mix	3	9	0.4451	.7267
	No interaction between mix and farm				

*Significant at the .05 probability level.

given the large range of background weed pressure across the farms, as we might have predicted (Table 4; Figure 3b). It appears that the cover crops generally reduced weed biomass compared to the background pressure, regardless of cover crop composition. The high seeding rate may have contributed to the smothering effect overall and reduced any treatment differences. While we did not measure light penetration, it would be a useful parameter to consider to evaluate the mechanism for suppression in these cover crop compositions at these high seeding densities (Liebman, Mohler, & Staver, 2001; MacLaren, Swanepoel, Bennett, Wright, & Dehnen-Schmutz, 2019).

4.4 | Practical management implications

Across all the farms, the grass–legume mixture was as good as or better than the best monoculture across ecosystem services with only one exception, soil N accrual at Farm 1 (Figure 3d). Similarly, while we did not find a positive effect from increased intraspecific diversity as we expected, we also did not find any negative effects. From our results, we can conclude that while increasing complexity, either through cultivar mixtures or grass–legume mixtures, may not regularly provide substantial benefits, the risk of a substantial downside to mixing grasses and legumes or cultivars is low. It does appear that the cover crop response to increasing composition complexity is context dependent, and as such a good practice might be to try a mixture alongside the monocultures at a given site and observe any differences, where possible. Of course, weather and other variable site factors might influence the outcome in a given year.

Management considerations are also critical to include when making decisions about cover crop composition, as the outcome alone may not justify the costs or additional time and complexity of adding a cover crop into a rotation. When N fixation is a priority, the mixture of wheat and vetch is a better choice than vetch alone as it produced an equivalent amount of fixed N at half the seeding rate of vetch (Table 4; Figure 3e). This biculture is approximately \$25–50 ha^{-1} cheaper than the monoculture of vetch (Supplemental Table S2). For farmers with a low-input approach to profitability, the additional cost of establishing a cover crop with any legume component could be restrictive (Blesh et al., 2019; Dunn et al., 2016). Identifying what other input costs could be offset from a biculture cover crop could help to justify these costs and make a cover crop more attractive. For instance, with higher rates of N fixation in mixtures, the cost of new N from fixation may be cheaper than off-farm sources of N such as manure. The mixture has the additional benefits of generally greater biomass and equivalent weed suppression compared to the vetch alone (Figure 3).

We only assessed a selection of ecosystem services from cover crops, and there may be other management considerations which might cause a farmer to make a certain decision about cover crop selection (e.g., pest or pollinator management). For instance, the preceding crop may make it difficult to establish a less cold-tolerant legume compared to a more tolerant grass like cereal rye (*Secale cereale* L). However, given these results compared to their monocultures, an even mixture of vetch and wheat appears to be an economical choice and provides good biomass production, N fixation and retention, and weed suppression across a range of on-farm contexts.

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ORCID

Emily R. Reiss https://orcid.org/0000-0002-2226-8206
Laurie E. Drinkwater https://orcid.org/0000-0002-0746-6765

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SUPPORTING INFORMATION

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