Short Title: Mixtures versus monocultures

Are Cover Crop Mixtures Better at Suppressing Weeds than Cover Crop Monocultures?

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Abstract

Cover crops are increasingly being used for weed management and planting them as diverse mixtures has become an increasingly popular strategy for their implementation. While ecological theory suggests that cover crop mixtures should be more weed-suppressive than cover crop monocultures, few experiments have explicitly tested this for more than a single temporal niche.

We assessed the effects of cover crop mixtures (5- or 6-species and 14-species mixtures) and monocultures on weed abundance (weed biomass) and weed suppression at the time of cover crop termination. Separate experiments were conducted in Madbury, New Hampshire from 2014 to 2017 for each of three temporal cover cropping niches: summer (spring planting-summer termination), fall (summer planting-fall termination), and spring (fall planting-subsequent spring termination). Regardless of temporal niche, mixtures were never more weed suppressive than the most weed-suppressive cover crop grown as a monoculture, and the more diverse mixture (14 species) never outperformed the less diverse mixture. Mean weed suppression levels of the best performing monocultures in each temporal niche ranged from 97-98% for buckwheat (*Fagopyrum esculentum* Moench) in the summer niche and forage radish (*Raphanus sativus* L. var. niger J. Kern.) in the fall niche, and 83-100% for triticale (×*Triticosecale* Wittm.) in the winter-spring niche. In comparison, weed suppression levels for the mixtures ranged from 66-97%, 70-90%, 67-99% in the summer, fall, and spring niches, respectively. Stability of weed suppression, measured as the coefficient of variation, was two- to six-times greater in the best performing monoculture compared to the most stable mixture, depending on the temporal niche. Results of this study suggest that when weed suppression is the sole objective, farmers are more likely to achieve better results planting the most weed-suppressive cover crop as a monoculture than a mixture.

Introduction

Cover crops provide a variety of services to agricultural ecosystems, including soil nutrient retention, reductions in wind and water erosion, and improvements in soil structure and health (Blanco-Canqui et al. 2015; Snapp et al. 2005), and thus may be an important tool to counteract environmental pollution and sustainability challenges associated with agriculture (Hunter et al. 2017; Kladivko et al. 2014). In addition, cover crops are increasingly being promoted for their benefits to weed management, particularly the management of herbicide-resistant weeds (Creamer et al. 1996; Norsworthy et al. 2012; Price et al. 2011; Wallace et al. 2019). As a resistance management tool, cover crops can not only suppress weeds that are already herbicide-resistant, but also the abundance and biomass of weeds that are susceptible to herbicides, thereby reducing the intensity of selection for future resistance (Wallace et al. 2019; Wiggins et al. 2016). This is especially the case in no-till cropping systems where herbicides are used to terminate cover crops. In this context reductions in the abundance, biomass, or seed production of weeds growing within the cover crop, either due to competition or suppression of seedling emergence from the seed bank, would reduce selection pressure at the time of cover crop termination (Brainard et al. 2011; Wallace et al. 2019).

Cover crops have traditionally been planted as single-species monocultures or simple grass-legume bicultures (Snapp et al. 2005). A large body of work has demonstrated that mixing a grass and legume cover crop in biculture often results in greater productivity, resource capture, and weed suppression compared to growing either species alone, especially the legume (Brainard et al. 2011; Ranells and Wagger 1997; Thapa et al. 2018; but see Mohler and Liebman 1987). Increasingly, however, farmers and other agriculture professionals are promoting and

implementing cover crops as species-diverse mixtures (also known as 'cocktails' or 'blends') (Groff 2008; MacLaren et al. 2019; Murrell et al. 2017). As evidence, a recent survey of farmers from across the US indicated that a majority of the respondents reported having used cover crop mixtures, with the majority of those reporting using mixtures containing three to as many as eight or more species (CTIC 2017). The same survey indicated that farmers rated cover crop mixtures as "the best species" for controlling herbicide-resistant weeds (CTIC 2017), suggesting that many farmers view mixtures as offering superior weed suppression compared to cover crops grown in monocultures.

The rationale for planting cover crops as species-diverse mixtures comes primarily from ecological research demonstrating grassland plant communities with higher species diversity often exhibit greater levels of ecosystem functioning, including productivity, resource utilization and nutrient retention, and pest suppression (i.e., "biotic resistance"), compared to communities with fewer species (Fargione et al. 2003; Naeem et al. 2000; Tilman et al. 1996, 2014; Weisser et al. 2017). Plant diversity has also been shown to influence the stability of these ecosystem functions, with more diverse communities tending to be more temporally stable (less variable) compared to less diverse communities (Tilman et al. 2014). Extending these observations to cover crops, species-diverse cover crop mixtures should therefore be expected to provide these same functions in agroecosystems and at higher levels than cover crops grown as monocultures (Couëdel et al. 2018; Florence et al. 2019; MacLaren et al. 2019).

However, despite the compelling empirical evidence from grassland studies, relatively few agronomic studies have quantified weed suppression in species-diverse mixtures (i.e., > 2 species) or for more than a single temporal cover cropping niche (e.g., Creamer et al. 1997; Baraibar et al. 2018; Blesh et al. 2019; Bybee-Finley et al. 2017; MacLaren et al. 2019). For

example, in some of the earliest work on multi-species cover crop mixtures, Creamer et al. (1997) examined 13 different four-species winter cover crop mixtures made up of combinations of 23 species and reported wide variability in the productivity and weed suppressiveness of the individual mixtures. While none of the treatments included the individual species grown as monocultures, the researchers concluded none of the four-species mixtures performed optimally due to at least one species in the mixture always being maladapted to the growing conditions (Creamer et al. 1997). In a subsequent study, the same researchers examined the relative weed suppressive ability of a single four species mixture comprised of the best performing species from their previous study compared to each of the four component species grown as monocultures but found little evidence to support enhanced weed suppression in the mixture (Creamer et al. 1996).

Given that cover crops can be important tools for controlling weeds, and mixtures are increasingly being promoted as superior to monocultures, we sought to answer the following questions. Is a mixture of cover crops more effective at suppressing weeds compared to the most weed-suppressive cover crop grown in monoculture? Is mixture performance relative to monocultures dependent on the season cover crops are implemented? And, if cover crop mixture performance is expected to be less variable from year to year, are the weed-suppressive effects of mixtures less variable than the most weed-suppressive monoculture? To address these questions, we conducted three, three-year experiments, each involving a different suite of cover crop species grown as monocultures and mixtures appropriate to one of three temporal niches, in which we quantified weed biomass and suppression. Hence, we generated a total of nine site-years of data involving a wide variety of cover crop species and planting windows with which to address our questions.

Materials and Methods

Site Description

We conducted three field experiments from 2014 to 2017 at the University of New Hampshire Kingman Research Farm in Madbury, New Hampshire, USA (43.18N, 70.93W). Soils at the site are a Hollis-Charlton fine sandy loam (Hollis = loamy, mixed, mesic Entic Lithic Haplorthods; Charlton = coarse-loamy, mixed, mesic Entic Haplorthods) (Freyre and Loy 2000). Mean monthly temperature and precipitation data were collected during the study period at a weather station located approximately 5 km away from the study site (Figure S1, Supplemental Material). For several years prior to the study, the fields used for the experiments were part of a squash (*Cucurbita maxima* Duchesne) and pumpkin (*Cucurbita pepo* L.) breeding program and managed as a conventional vegetable-winter rye (*Lolium* spp.) cover crop rotation.

Overview of the Experiments

Each experiment corresponded to one of three temporal niches for cover cropping (Figure 1) and involved either five or six cover crop species appropriate for that growing period. The temporal niches were spring planting-summer termination (hereafter 'Summer Experiment'), summer planting-fall termination (hereafter 'Fall Experiment'), and fall planting-subsequent spring termination (hereafter 'Spring Experiment'). The cover crop species utilized across the three experiments included eight species of annual cool-season and warm-season grasses, three legumes, and three non-legume forbs (Table 1). Each experiment was a randomized complete block design with treatment levels replicated across four blocks. Treatment levels included in each experiment were each of the five or six cover crop species sown as monocultures at the full

rate recommended for that particular species and a mixture of all five or six species, depending on the experiment, each sown at 1/5 or 1/6 of their recommended rate, respectively (Table 1). All three experiments also included a 14-species mixture comprised of all the species utilized across all three experiments, each sown at 1/14 of their recommended rate, as well as a treatment in which no cover crop was sown (hereafter 'weedy fallow').

All cover crop monoculture and mixture treatments were planted with an ALMACO light duty grain drill. Prior to sowing each run of the experiments, the experimental site was plowed, harrowed, and then rolled to create a firm seedbed. The weedy fallow treatment was prepped as described above. No supplemental fertilizer was applied to any of the treatments. Individual replicate plots were 1.4 m wide and varied in length each year (year 1 = 6.1 m; year 2 = 12.2 m; year 3 = 9.9 m) based on field area. The experiments were conducted in a different field of the farm each year to avoid potential carry-over effects of the previous treatments. Planting for the Summer Experiment occurred on 11 June 2014, 12 June 2015, and 22 June 2016. The Fall experiment was planted on 8 August 2014, 20 August 2015, and 25 August 2016, while the Spring Experiment was planted on 1 October 2014, 7 October 2015, and 28 September 2016. The first year of the Spring Experiment did not include the 14-species mixture; however, this treatment was included in the second and third years of the Spring Experiment and in all three years of the Summer and Fall experiments. After planting, cover crop treatments were allowed to grow for approximately 1.5 or 2.25 months (Summer and Fall experiments, respectively) or 7.5 months (Spring Experiment), after which time cover crop and weed biomass were quantified as described below.

Data Collection

We quantified cover crop and weed biomass at the end of the cover crop growing period in each replicate plot in each of the three experiments each year (Figure 1). All plant material rooted within two 0.5-m by 0.5-m (0.25 m^2) quadrats placed in each plot were clipped at the soil surface. Harvested material was placed in a labeled paper bag, sorted into cover crop and weed components, dried at 65 ^{o}C for 48-72 hours, and weighed to the nearest 0.01 g. Cover crop material in the mixture treatments was sorted by cover crop species and weeds were sorted into grass and broadleaf weeds. Weed suppression in each cover crop treatment was calculated at the block-level as the percent reduction in total weed biomass in that cover crop treatment replicate relative to the total weed biomass in the weedy fallow treatment replicate of the corresponding block.

Statistical Analyses

We analyzed the Summer, Fall, and Spring experiments separately. For each experiment, we used a mixed factor ANOVA to assess how the mixture and monoculture treatments affected weed biomass and weed suppression. The statistical model included block, cover crop treatment, and year as factors, as well as the interaction between cover crop treatment and year. Treatment and year were considered fixed factors, while block was treated as a random factor. In cases where the treatment by year interaction was significant ($P < 0.05$), we analyzed each year separately with a reduced model that included only the block and treatment factors. For the Spring Experiment, because the 14-way mixture was not included in year 1, we analyzed that year separately from years 2 and 3. For all analyses, when the treatment effect was significant at the $P < 0.05$ level, we compared treatment means with least-squared (LS) means. The weedy fallow treatment was included in all analyses of weed biomass, and data from this treatment were

used in the calculation of weed suppression, as described above. Weed biomass data were log (x+1) transformed and percent weed suppression data were arcsine square-root transformed prior to analysis to satisfy normality and homoscedasticity requirements of ANOVA. We also examined the relationships between weed suppression and total cover crop biomass in each experiment using linear regression. Regression analyses were conducted across years for each experiment unless ANOVA indicated a significant treatment by year interaction for weed suppression, in which case years were analyzed separately. All analyses were conducted with JMP Pro (Version 14). Untransformed data are presented in the tables for ease of interpretation. Cover crop biomass data are also presented as Supplementary Materials for readers who wish to compare cover crop biomass levels between individual treatments.

To assess whether the mixtures resulted in more stable (i.e., less variable) levels of weed suppression compared to the monocultures, we calculated the coefficient of variation (CV) of the untransformed weed suppression values for each treatment. Calculation of the CV for each treatment was based on the four replicates present in each of the three years (except for the 14-species mixture in the Spring Experiment, which was included in the second and third year only). Hence, the CV for each treatment was based on n = 12 (n = 8 for the 14-species mixture in the Spring Experiment) and reflects both the spatial and temporal variability of weed suppressiveness of that treatment. Note that because each treatment results in a single CV value, these data cannot be assessed statistically; therefore, for each experiment, we present each treatment ranked by its CV of weed suppression from lowest (most spatially and temporally stable) to highest (least stable).

Results and Discussion

Summer Experiment

The weed community present in the Summer Experiment was made up primarily of annual broadleaf species (making up 73-90% of the total weed biomass in the weedy fallow treatment each year), with horseweed (*Erigeron canadensis* L.), redroot pigweed (*Amaranthus retroflexus* L.), common lambsquarters (*Chenopodium album* L.), and large crabgrass [*Digitaria sanguinalis* (L.) Scop.] being the dominant species. The ANOVA conducted on the weed biomass data indicated a significant treatment by year interaction; therefore, treatment effects were assessed separately for each year (Table 2). In year 1, the treatments with the lowest weed biomass were the buckwheat and millet monocultures. In year 2, the buckwheat and oat monocultures were among the treatments with the lowest weed biomass, while in year 3 the buckwheat and millet monocultures and the 6-way mixture were among the treatments with the lowest weed biomass. The buckwheat monoculture resulted in lower weed biomass compared to the 14-way mixture in all three years and the 6-way mixture in years 1 and 2. Both mixtures resulted in lower weed biomass compared to the weedy fallow treatment in each of the three years.

Treatment effects on weed suppression also varied by year (Table 2). In general, in each of the three years, the highest levels of weed suppression were associated with the buckwheat monoculture, ranging from 97 to 98% depending on the year; however, these were not statistically different from the millet monoculture in year 1, the millet and oat monocultures or the 6-way mixture in year 2, or several of the monocultures and both the 6-way and 14-way mixture in the third year of the experiment. Weed suppression levels did not differ between the 6-way and 14-way mixtures in any of the years and ranged from 66.4% to 96.8%, depending on

the mixture treatment and year. The lowest levels of weed suppression each year, ranging from 8.5 to 26%, were observed in the chickling vetch monoculture. In general, those treatments with the highest levels of weed suppression were also those that produced the highest cover crop biomass, with the R^2 of the linear relationship between these two variables across all treatments ranging from 0.53 to 0.67 over the three years (Figure 2; Figure S2, Supplemental Material).

Fall Experiment

The weed community present in the Fall Experiment was dominated by annual broadleaf species (making up 99% of the total biomass in the weedy fallow treatments) and included many of the same weed species observed in the Summer Experiment. Like the Summer Experiment, the effects of the cover crop treatments on weed biomass depended on the year (Table 3). In year 1 the monocultures with the lowest weed biomass were forage radish and canola, both of which were significantly lower than either of the mixtures. In year 2, the forage radish and wheat monocultures and the 6-way mixture had the lowest weed biomass. In year 3, weed biomass did not differ between any of the monocultures or the mixtures, apart from the sunn hemp (*Crotolaria juncea* L.) monoculture. With the exception of the 14-way mixture in year 1, both mixtures resulted in lower weed biomass compared to the weedy fallow treatment in each of the three years.

In contrast to the Summer Experiment, we did not detect an interaction between treatment and year on weed suppression (Table 3). Among the monocultures with the highest levels of weed suppression, forage radish (97.5%) was more weed suppressive than oats (85.2%), triticale (81%), the 14-way mixture (70%), and the sunn hemp monoculture (55.3%). Differences in weed suppression between the 6-way (88.9%) and 14-way mixtures were not significant. The lowest

level of weed suppression was observed in the sunn hemp monoculture. Also, in contrast to the Summer Experiment, the treatments that had the highest levels of weed suppression each year were not necessarily the ones that produced the greatest cover crop biomass, as evidenced by the relatively low R^2 (0.05) of the regression analysis (Figure 3A; Figure S3, Supplemental Material).

Spring Experiment

Winter annual broadleaf mustard species, including shepherd's purse [*Capsella bursa-pastoris* (L.) Medik.], made up the majority (98-100% of total weed biomass in the weedy fallow) of the weed community each year. Cover crop treatment effects on weed biomass were significant only in the first year of the experiment (Table 4). While weed biomass was relatively low across all cover crop treatments in year 1, it was lower in the triticale monoculture compared to the monocultures of barley (*Hordeum vulgare* L.), cereal rye (*Secale cereale* L.), and wheat. We did not detect differences in weed biomass between the triticale monoculture and the 5-way mixture in year 1. All the cover crop treatments resulted in substantial reductions in weed biomass compared the weedy fallow treatment in year 1. No differences in weed biomass between the treatments were detected in years 2 and 3. Similarly, no differences in weed suppression between the treatments were detected in any of the years (Table 4). This lack of a detectable difference in weed suppression among the treatments cannot be explained solely by cover crop biomass, which did differ among treatments each year and was often much lower in the barley and hairy vetch monocultures compared to the other treatments (Figure 3B; Figure S4, Supplemental Material). Similarly, the linear regression between cover crop biomass and weed suppression in the Spring Experiment was not significant (P =0.57).

The levels of weed suppression provided by the cover crops prior to termination in our study were similar to levels reported in previous research (Hayden et al. 2012; Hodgdon et al. 2016; Teasdale et al. 2007). For example, buckwheat has been demonstrated to be especially weed suppressive compared to other summer-sown cover crop species (Falquet et al. 2015; Smith et al. 2014; but see Bicksler and Masiunas 2009), likely due to its rapid growth and canopy development; however, other factors, such as allelopathy may also play a role (Falquet et al. 2015; Weston 1996). The high degree of weed suppression provided by the forage radish monoculture in the Fall Experiment was also congruent with previous research conducted in the Northeast and Mid-Atlantic regions (Hodgdon et al. 2016; Lawley et al. 2011; but see Baraibar et al. 2018), likely due to its early and competitive fall growth and rapid canopy development (Lawley et al. 2012). For example, Lawley et al. (2011) observed nearly complete suppression of winter annual weeds in fall in forage radish monocultures in Maryland and suggested that when growth was optimal it could replace a pre-plant burndown herbicide application for a subsequent corn crop. Our observation that the legume monocultures provided only minimal weed suppression services in the Summer and Fall experiments is congruent with previous research showing that legume cover crop monocultures are often less competitive with weeds in the fall compared to other grassy and broadleaf cover crops (Baraibar et al. 2018).

Spatio-temporal Variability in Weed Suppression

Even if cover crop mixtures are not more weed suppressive than the most suppressive monoculture, they could be considered beneficial if their weed suppression levels are more consistent or dependable from location to location or season to season than cover crop monocultures. We used the coefficient of variation to quantify the degree to which weed

suppressive effects of the cover crop treatments in each experiment were spatially and temporally stable. In all three experiments, we found that the treatment with the lowest CV for weed suppression was always a monoculture, never a mixture (Table 5). The buckwheat monoculture was the least variable treatment in the Summer Experiment from a weed suppression standpoint, while in the Fall and Spring experiments, the forage radish and triticale monocultures were the least variable treatments, respectively. Triticale was also included in the Fall experiment, where it was substantially more variable relative to several of the other monocultures, as well as the 6-species mixture, suggesting that the temporal niche is an important determinant of a cover crop's relative stability. While never the most stable treatments overall, the 5- or 6-way mixtures were always ranked in the top two or three treatments in each experiment and these were always ranked as being more stable than the 14-way mixture (Table 5).

Taken as a whole, these data do not support the hypothesis that cover crop mixtures provide greater weed suppression benefits than the most suppressive cover crop grown as a monoculture. In fact, in many instances one or both mixtures performed substantially worse in terms of weed biomass or suppression than the best monoculture. Nor was it the case that we observed evidence that a greater number of species within a mixture enhanced weed suppression, rather, we observed that the two mixtures rarely differed from one another for either weed biomass or suppression. In the one case where the two mixtures did differ (year 1 in the Fall Experiment), the 6-way mixture outperformed the 14-way mixture. And in no cases did the 14-way mixture provide more stable weed suppression than the 5- or 6-way mixture (Table 5). The fact that the 14-species mixture contained both cool- and warm-season species, and therefore always featured some proportion of species that were likely not well adapted to any given

temporal cover cropping niche, may help explain this observation. A similar conclusion was drawn by Creamer et al. (1997) to explain the suboptimal performance of many of their four-species mixtures, which were composed from a suite of 23 cool and warm season legumes and grasses.

These results are congruent with recent research showing cover crop mixtures are rarely if ever more productive or weed-suppressive than the best performing monoculture (Baraibar et al. 2018; Blesh et al. 2019; Creamer et al. 1996; Finney et al. 2016; Florence et al. 2019; MacLaren et al. 2019; Nelson et al. 2011; Osipitan et al. 2018; Schappert et al. 2019; Smith et al. 2014; Wortman et al. 2012). The majority of these studies examined mixture performance for a single temporal cover cropping niche. For example, in a previous study conducted in New Hampshire, Smith et al. (2014) found that a summer-sown 5-species mixture that included buckwheat and other grass, legume, and mustard cover crops provided weed suppression and suppression stability in the fall comparable to a buckwheat monoculture. Researchers in Ohio examined the relative weed suppressive ability of a fall-sown mixture of crimson clover, hairy vetch, barley, and cereal rye compared to each of the four species grown as a monoculture and concluded that weed suppression varied by species and the mixture did not result in broader-spectrum weed control compared to the individual monocultures (Creamer et al. 1996). Baraibar et al. (2018) found that in Pennsylvania, weeds were equally well suppressed in the spring in a fall-sown cereal rye monoculture and 3-, 4-, and 6-species mixtures containing cereal rye. Similarly, a large-scale cover crop mixture study in Nebraska concluded that biomass production, rather than the diversity of late summer/fall-sown mixtures, was the primary determinant of weed suppression (Florence et al. 2019; see also MacLaren et al. (2019) for an example from South Africa). Our study indicates that the comparable or reduced weed

suppression potential of mixtures relative to high performing monocultures is not restricted to a single temporal cover cropping niche and is therefore likely a property of cover crop mixtures more generally.

If cover crop mixtures are not more weed suppressive than monocultures, how do we reconcile this with the compelling empirical evidence for enhanced ecosystem functioning commonly observed in grassland diversity studies? One explanation could be the facilitative and complementarity effects thought to be the mechanistic drivers of many of the diversity effects documented in perennial grassland communities do not manifest to the same degree in annual plant communities and/or over the relatively short timescales that cover crops are typically grown (Jiang et al. 2007; Mohler and Liebman 1987; but see Fridley 2003). Another explanation could be related to the fact that in most cover crop mixture studies, the mixtures are constructed using a replacement design, where the rate of each species in the mixture is reduced, usually based on the full rate for that species divided by the total number of species in mixture. This means the seeding rates, and therefore densities, of the individual species in the mixture are lower than in their respective monocultures. Hence, if one or a few individual species are particularly weed suppressive, their effects could be "diluted" by being sown at a lower density and with less competitive species. While we cannot rule out this explanation, at least two studies we are aware of that compared a mixture constructed with a replacement design to the same mixture in which all species sown at 100% of their full monoculture rate (Mohler and Liebman 1987; Smith et al. 2015) found little evidence that the higher seeding rates increased the competitive ability of the mixture.

An alternative to these explanations could be that grassland diversity studies are not the appropriate context for interpreting (or extrapolating) how crop diversity will affect ecosystem

functions in annual cropping systems. This primarily stems from how diversity effects are typically assessed in such studies—by comparing the mean response of species monocultures to the mean response of the higher-diversity treatments (e.g., Fargione et al. 2003; Tilman et al. 1996, 2014). In this case, the metric of performance at the monoculture level of diversity, be it productivity or biotic resistance, is somewhere between the performance of the best and the worst performing monocultures (Florence et al. 2019). However, while this makes sense for ecological studies aimed at identifying plant diversity effects more generally, farmers do not plant the "mean monoculture", nor are they likely to plant the worst-performing crop, rather, they plant what they expect to be the optimally-performing species. Hence, a more appropriate benchmark in an agronomic context is the best performing monoculture. This criterion sets a particularly high bar for concluding mixture superiority, particularly for metrics such as weed suppression, which can approach 100% depending on the species of cover crop (Hodgdon et al. 2016; Lawley et al. 2011).

Proponents of cover crop mixtures rightly point out that mixtures represent a strategy for implementing crop diversity into cropping systems that might otherwise lack opportunities for diversification and that mixtures support a wider array of ecosystems services than any single cover crop monoculture can provide (Finney and Kaye 2017; Storkey et al. 2015). While both points may be true, our study indicates that when it comes to the service of weed suppression, cover crop mixtures are not inherently better, and in fact can be substantially less weed-suppressive than the best cover crop grown as a monoculture, and therefore their potential benefits in this regard should likely not be oversold. This is especially critical if the intended role of the cover crop is to reduce weed abundance or the size of individual weeds at burndown as part of an herbicide resistance management strategy (Norsworthy et al. 2012; Price et al. 2011;

Wallace et al. 2019). If farmers are adopting cover crop mixtures based on unrealistic expectations for weed control and then not experiencing sufficiently positive outcomes, this may ultimately undermine not only the efficacy of their herbicide resistance management, but also longer-term adoption rates of cover crops more broadly (MacLaren et al. 2019). Rather, recent research suggests that a better understanding and communication to farmers of the potential risks associated with cover crops, and how those risks can be managed or mitigated, would likely increase their adoption (Arbuckle and Roesch-McNally 2015).

In cases where the best cover crop species for weed suppression is unknown, cover crop mixtures may be a means to hedge one's bets. Indeed, while the mixtures we examined where never better than the best monoculture they also were also rarely among the worst-performing cover crop treatments. That said, both species that provided superior weed suppression in the Summer and Fall experiments, buckwheat and forage radish, respectively, have previously been documented to be among the most consistently weed-suppressive cover crop options for those temporal niches (Hodgdon et al. 2016; Lawley et al. 2011; Smith et al. 2014). Similarly, cereal rye is perhaps the most widely used winter cover crop in US cropping systems (Blesh et al. 2019; Snapp et al. 2005), and our study indicated that it along with any of the other four species examined in the Spring Experiment can provide weed suppression at levels comparable to either of the mixtures. Therefore, the need to bet hedge with mixtures for the purpose of weed suppression may be less in some cropping systems and growing areas than in others.

Use of cover crop mixtures, as with any agricultural practice, involves trade-offs (Blanco-Canqui et al. 2015; Blesh et al. 2019; Snapp et al. 2005). Given that weed suppression appears to be a tradeoff associated with species-diverse cover crop mixtures, future research should be aimed at better understanding and alleviating this trade-off, so as to maximize the utility of

mixtures for weed suppression and the myriad other benefits that diverse plant communities may provide to agriculture. An improved understanding of how weed suppressive traits vary across cover crop species, and whether and how these traits are affected by mixture composition, using an approach similar that recently employed by Tribouillois et al. (2015), would also likely lead to more effective cover crop mixture designs.

Conflicts of Interest

No conflicts of interest have been declared.

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Tables

Table 1. Cover crop species and seeding rates used in the monocultures and mixture treatments in the Summer, Fall, and Spring Experiments.

Species	Experiment	Treatments			
		Monoculture	5-sp.	6-sp.	14-sp.
		kg seed ha^{-1}			
Barley	Spring	123.29	24.66	----	8.81
BMR Sorghum	Summer	39.23	----	6.54	2.80
Buckwheat	Summer	100.88	----	16.81	7.21
Canola	Fall	11.21	----	1.87	0.80
Cereal rye	Spring	134.5	26.90	----	9.61
Chickling vetch	Summer	78.46	----	13.08	5.60
Forage radish	Fall	11.21	----	1.87	0.80
Hairy vetch	Spring	44.83	8.97	----	3.20
Millet	Summer	33.63	----	5.61	2.40
Oats	Summer, Fall	123.29	----	20.55	8.81
Sunn Hemp	Fall	44.83	----	7.47	3.20
Teff	Summer	8.97	----	1.50	0.64
Triticale	Fall, Spring	168.13	33.63	28.02	12.01
Wheat	Fall, Spring	168.13	33.63	28.02	12.01

Table 2. Effects of cover crop monoculture and mixture treatments on weed biomass and weed suppression in the Summer Experiment. Data are means (SE), n = 4.

Treatment	Weed biomass			Weed suppression		
	Year 1[a]	Year 2	Year 3	Year 1	Year 2	Year 3
	kg ha^{-1}			%		
Buckwheat	79 (37) F	41 (22) E	10 (9) F	98.4 (0.7) A	97.2 (1.5) A	98.4 (1.4) A
BMR Sorghum	788 (308) D	535 (79) B	73 (20) D	82.3 (9.1) BC	61.1 (6.7) C	91.0 (4.2) A
Chickling vetch	4626 (264) AB	1147 (194) AB	1213 (323) A	12.0 (4.2) E	26.0 (11.1) D	8.5 (7.5) C
Millet	479 (351) E	164 (66) CD	19 (9) EF	87.6 (9.9) AB	86.3 (6.7) AB	98.2 (0.8) A
Oats	1870 (385) ABCD	77 (31) DE	297 (46) C	58.7 (12.4) D	93.6 (3.1) AB	70.6 (3.2) B
Teff	1989 (333) ABC	678 (162) AB	541 (184) BC	57.2 (10.1) D	53.3 (7.0) C	48.9 (19.1) B
6-sp. mixture	1429 (561) CD	170 (24) C	31 (10) DEF	72.4 (8.5) CD	87.5 (2.0) AB	96.8 (1.5) A
14-sp. mixture	1600 (232) BCD	186 (18) C	58 (29) DE	66.4 (7.1) CD	86.0 (2.9) B	92.6 (5.1) A
Weedy fallow	4979 (608) A	1415 (150) A	1082 (264) AB	----	----	----
ANOVA[b]						
Year (Y)		****			NS	
Treatment (T)		****			****	
Y*T		****			**	

[a]Within a column, means sharing the same letter are not significantly different at P < 0.05 (LS Means). [b]Significance levels are *, P < 0.05; **, P < 0.01, ***, P < 0.001; ****, P < 0.0001.

Table 3. Effects of cover crop monoculture and mixture treatments on weed biomass and weed suppression in the Fall Experiment. Data are means (SE), n = 4; weed suppression, n = 12.

Treatment	Weed biomass			Weed suppression
	Year 1[a]	Year 2	Year 3	
	kg ha^{-1}			%
Canola	42 (28) D	175 (81) BC	11 (8) C	91.5 (3.1) AB
Forage radish	137 (134) D	11 (4) E	3 (3) C	97.5 (1.4) A
Oats	506 (282) C	92 (29) CD	26 (15) BC	85.2 (5.1) B
Sunn hemp	1814 (396) AB	585 (228) AB	61 (29) B	55.3 (8.1) C
Triticale	498 (167) C	94 (16) BC	43 (36) BC	81.0 (7.9) B
Wheat	1057 (511) BC	22 (7) DE	8 (7) C	87.3 (5.4) AB
6-sp. mixture	507 (170) C	34 (13) CDE	17 (4) BC	88.9 (2.6) AB
14-sp. mixture	2028 (899) AB	80 (26) CD	3 (2) C	70.0 (10.7) B
Weedy fallow	3455 (482) A	1235 (384) A	199 (77) A	----
ANOVA[b]				
Year (Y)		****		NS
Treatment (T)		****		****
Y*T		***		NS

[a]Within a column, means sharing the same letter are not significantly different at $P<0.05$ (LS Means). [b]Significance levels are *, $P<0.05$; **, $P<0.01$, ***, $P<0.001$; ****, $P<0.0001$.

Table 4. Effects of cover crop monoculture and mixture treatments on weed biomass and weed suppression in the Spring Experiment. Data are means (SE), n = 4 in year 1 and n = 8 in years 2-3.

Treatment	Weed biomass		Weed suppression	
	Year 1[a]	Years 2-3	Year 1	Years 2-3
	kg ha^{-1}		%	
Barley	21 (8) BC	234 (96)	98.5 (0.5)	57.5 (10.6)
Cereal rye	21 (4) B	157 (93)	98.4 (0.3)	70.6 (15.3)
Hairy vetch	11 (4) BCD	269 (157)	99.1 (0.3)	50.0 (13.4)
Triticale	4 (2) D	93 (49)	99.7 (0.2)	82.9 (6.2)
Wheat	16 (4) BC	136 (73)	98.8 (0.2)	70.6 (11.8)
5-sp. mixture	7 (2) CD	266 (138)	99.4 (0.2)	72.0 (11.4)
14-sp. mixture	----	194 (121)	----	67.3 (11.4)
Weedy fallow	1322 (121) A	780 (346)	----	----
ANOVA[b]				
Year (Y)	----	****	----	NS
Treatment (T)	****	NS	NS	NS
Y*T	----	NS	----	NS

[a]Within a column, means sharing the same letter are not significantly different at P<0.05 (LS Means). [b]Significance levels are *, P<0.05; **, P<0.01, ***, P<0.001; ****, P<0.0001.

Table 5. Spatio-temporal variability in weed suppression of cover crop monocultures and mixtures. For each experiment, treatments are ranked from least to most variable and the mixtures are bolded.

Experiment	Treatment	Variability of weed suppression (CV)
		%
Summer		
	Buckwheat	2.4
	Millet	15.1
	6-species mixture	**16.4**
	14-species mixture	**18.5**
	BMR Sorghum	23.2
	Oats	27.5
	Teff	45.1
	Chickling vetch	107.7
Fall		
	Forage radish	5.1
	6-species mixture	**10.2**
	Canola	11.8
	Oats	20.8
	Wheat	21.4
	Triticale	34
	Sunn Hemp	50.8
	14-species mixture	**53.1**
Spring		
	Triticale	18.3
	5-species mixture	**35.8**
	Wheat	37.7
	Barley	44.1
	Cereal rye	46.4
	14-species mixture	**47.9**
	Hairy vetch	58.2

Figure 1. Temporal duration of the Summer, Fall, and Spring cover cropping experiments, indicating approximate timing of cover crop planting, P, and biomass harvest/termination, T. Each experiment was replicated over three years.
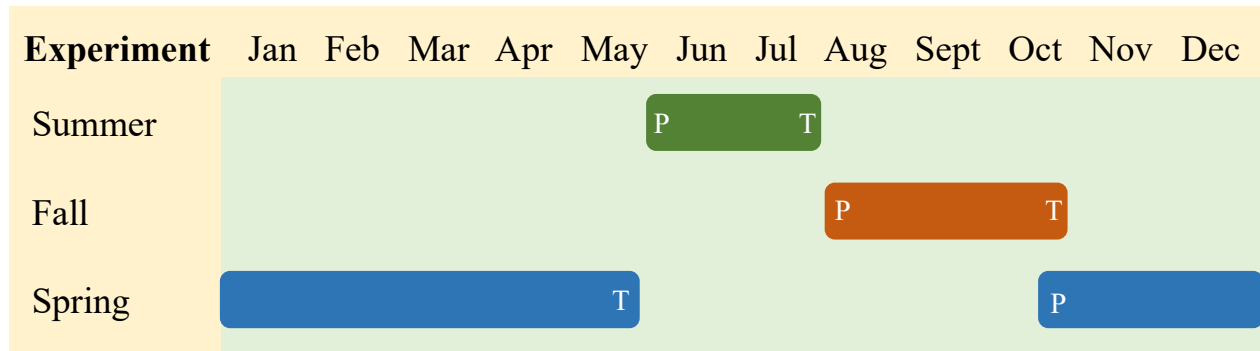
Experiment	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sept	Oct	Nov	Dec
Summer						P	T					
Fall								P		T		
Spring	T				T					P		P

Figure 2. Relationships between cover crop biomass and weed suppression in monoculture and mixture treatments in the Summer Experiment in (A) Year 1, (B) Year 2, and (C) Year 3. Data are means ± 1 SE; n = 4. Linear regression analyses: (Year 1) Y = 34.73 + 0.08417X, R^2 = 0.533, P < 0.0001; (Year 2) Y = 36.52 + 0.1493X, R^2 = 0.652, P < 0.0001; (Year 3) Y = 27.36 +

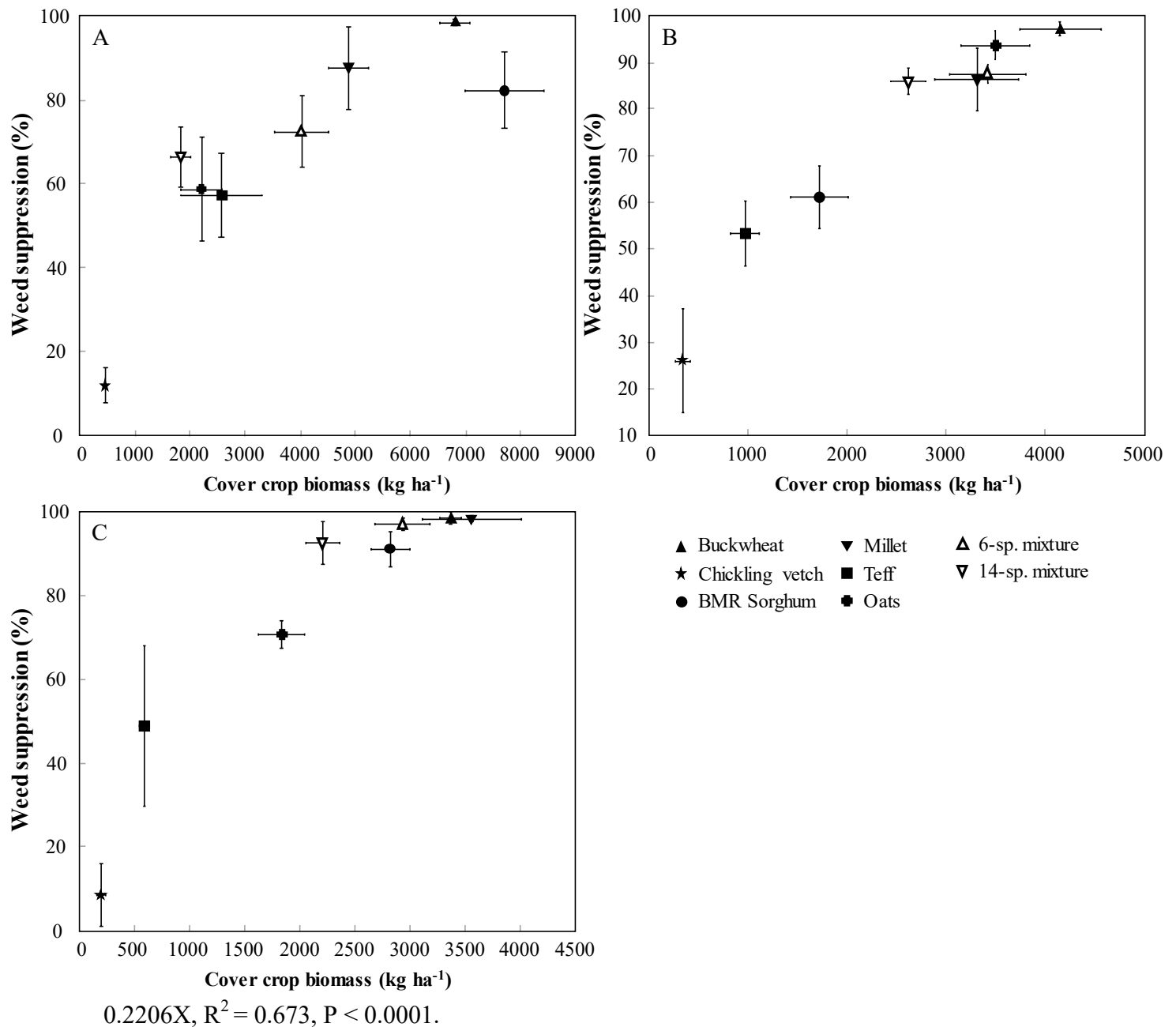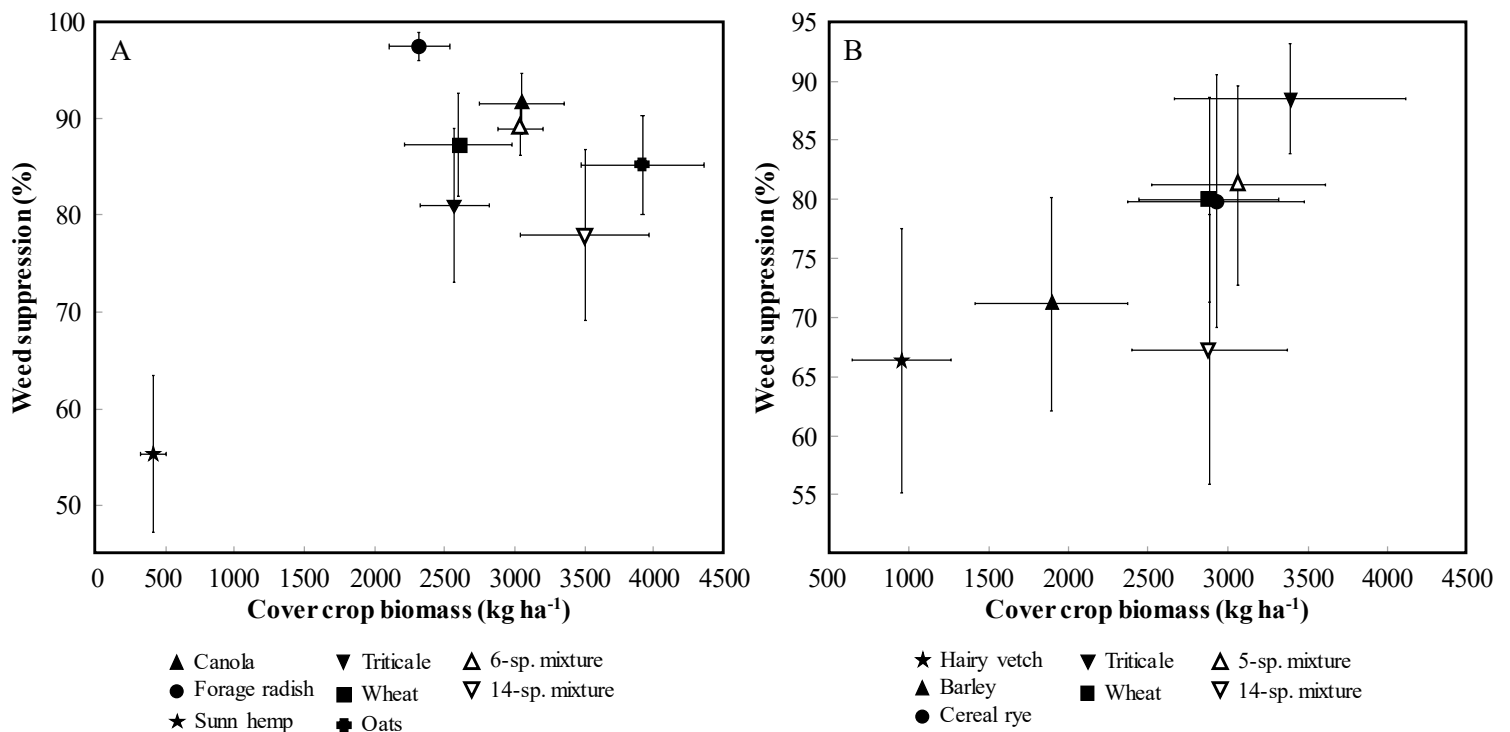0.2206X, R^2 = 0.673, P < 0.0001.

Figure 3. Relationships between cover crop biomass and weed suppression in monoculture and mixture treatments in the (A) Fall Experiment and (B) Spring Experiment. Data are means ± 1 SE; n = 12, except the 14-species mixture in the spring experiment, n = 8. Linear regression analyses: (Fall Experiment) Y = 73.9 + 0.03431X, R^2 = 0.046, P = 0.036; (Spring Experiment)

no linear relationship, P = 0.57.