Nutritional composition and in vitro digestibility of grass and legume winter (cover) crops

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ABSTRACT

In dairy farming systems, growing winter crops for forage is frequently limited to annual grasses grown in monoculture. The objectives of this study were to determine how cropping grasses alone or in mixtures with legumes affects the yield, nutritional composition, and in vitro digestibility of fresh and ensiled winter crops and the yield, nutritional composition, and in vitro digestibility of the subsequent summer crops. Experimental plots were planted with 15 different winter crops at 3 locations in Virginia. At each site, 4 plots of each treatment were planted in a randomized complete block design. The 15 treatments included 5 winter annual grasses [barley (BA), ryegrass (RG), rye (RY), triticale (TR), and wheat (WT)] in monoculture [i.e., no legumes (NO)] or with 1 of 2 winter annual legumes [crimson clover (CC) and hairy vetch (HV)]. After harvesting the winter crops, corn and forage sorghum were planted within the same plots perpendicular to the winter crop plantings. The nutritional composition and the in vitro digestibility of winter and summer crops were determined for fresh and ensiled samples. Growing grasses in mixtures with CC increased forage dry matter (DM) yield (2.84 Mg/ha), but the yield of mixtures with HV (2.47 Mg/ha) was similar to that of grasses grown in monoculture (2.40 Mg/ha). Growing grasses in mixtures with legumes increased the crude protein concentration of the fresh forage from 13.0% to 15.5% for CC and to 17.3% for HV. For neutral detergent fiber (NDF) concentrations, the interaction between grasses and legumes was significant for both fresh and ensiled forages. Growing BA, RY, and TR in mixtures with legumes decreased NDF concentrations, whereas growing RG and WT with legumes did not affect the NDF concentrations of either the fresh or the ensiled forages. Growing grasses in mixtures with legumes

decreased the concentration of sugars of fresh forages relative to grasses grown in monoculture. Primarily, this decrease can be attributed to low concentrations of sugars of mixtures with HV (10.5%). Growing grasses in mixtures with legumes reduced the fiber digestibility of both winter crops (75.7% to 72.8% NDF). Growing grasses in mixtures with legumes did not affect estimated DM yield, nutritional composition, or digestibility of the succeeding summer crops. In conclusion, growing grasses in mixtures with legumes as winter forage crops can increase forage estimated DM yields and its nutritional quality in dairy farming systems.

Key words: cover crop, small grain, legume, silage, digestibility

INTRODUCTION

Cover crops are planted to increase the health and fertility of soils, to control weeds, and to benefit the environment (SARE, 2007; Myers and Watts, 2015). By covering the soil surface, the vegetative portion of the crops protects the soil from wind and rain erosion. In addition, the roots of the cover crops hold the soil in place, reducing soil erosion from wind and rain. For a corn and soybean rotation, Villamil et al. (2006) observed that soil aggregate stability was increased by 9 to 17% when cover crops were grown between crops compared with when the ground was left fallow and no cover crops were used during the rotation. Also, growing cover crops decreased bulk density and penetration resistance of the soil and increased total porosity and soil water retention (Villamil et al., 2006).

Different plant species are grown for cover crops. Grass species, such as rye (*Secale cereale*), annual ryegrass (*Lolium multiflorum*), or oats (*Avena sativa*), are the species grown most frequently (Myers and Watts, 2015). The legume species hairy vetch (*Vicia villosa*) or crimson clover (*Trifolium incarnatum*) are also used frequently for this purpose (Shipley et al., 1992). Grass species have an extensive and fine fibrous root system, whereas legumes have a tap root system (Stokes et al.,

Received May 30, 2017.

Accepted November 5, 2017.

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2009; De Baets et al., 2011; Vannoppen et al., 2015). As fibrous root systems reduce soil erosion more than tap root systems (De Baets et al., 2011), grass species are often recommended for cover crops. Also, as they scavenge more residual soil N than legume species, growing grass species is advisable to reduce soil nitrate leaching (SARE, 2007; Ketterings et al., 2015). Alternatively, legume species can fix N from the atmosphere through a symbiotic relationship between legume plants and bacteria in the soil (Hartwig and Ammon, 2002; Plaza-Bonilla et al., 2015). When left as a cover or mulch, the biomass of the cover crop can provide additional residual N for the following crop (Shipley et al., 1992). However, in dairy farming systems, where winter crops are harvested and ensiled for feed, this benefit could be marginal or nonexistent. Still, cover crops can contribute to below-ground biomass development (i.e., root growth). For instance, Kuo et al. (1997a) reported that a ryegrass cover crop increased below-ground biomass DM by 120 to 170% compared with a control treatment without cover crop. Similarly, Kuo et al. (1997b) reported that a hairy vetch cover crop increased below-ground N by 28% compared with a control treatment without cover crop.

In dairy farming systems, winter forage crops are most frequently annual grasses grown in monoculture (Kaiser et al., 2007; Long et al., 2012). However, based on the potential benefits on soil fertility, the interest in growing grasses in mixtures with legumes has increased over past years. Because grass winter crops can deplete soil N (Ketterings et al., 2015), growing grasses in mixtures with legumes could potentially increase residual soil N and therefore improve forage yields of the successive summer crops. Miguez and Bollero (2006) reported that rye grown in monoculture reduced the successive corn biomass when no N was added to the

corn, although this detrimental effect was not observed when rye was grown in mixture with hairy vetch.

For this study, we hypothesized that growing grasses in mixtures with legumes, compared with grasses grown in monoculture, would increase forage yield and improve forage quality of winter crops as well as the following summer crop. Therefore, the objectives of this study were to determine how growing grasses alone or in mixtures with legumes affects (1) the estimated yield, nutritional value, and in vitro digestibility of fresh and ensiled winter crops and (2) the estimated yield, nutritional value, and in vitro digestibility of the following summer annual crop (fresh and ensiled).

MATERIALS AND METHODS

Animal Care and Use

The Institutional Animal Care and Use Committee of Virginia Tech approved all procedures involving dairy cows for collecting rumen contents (protocol 15-234).

Experimental Sites and Climate Data

This study was performed from September 2014 to September 2015 at 3 research stations (hereafter referred as sites) located near Blacksburg (37°11'35" N; 80°34'42" W), Blackstone (37°05'10" N; 77°58'34" W), and Orange (38°13'03" N; 78°07'38" W), Virginia. Average monthly temperature and total precipitation data during the experimental period are presented in Table 1. Weather data were collected from weather stations at each site using the National Centers for Environmental Information of the National Oceanic and Atmospheric Administration (NOAA, US Department of Commerce; www.noaa.gov).

Table 1. Average temperature (°C) and total precipitation (mm) during the growing season of 2014–2015

Month	Blacksburg		Blackstone		Orange	
	Temperature	Precipitation	Temperature	Precipitation	Temperature	Precipitation
Sep 2014	18.4	97	20.8	107	14.3	30
Oct 2014	12.2	104	15.2	74	14.4	109
Nov 2014	3.18	76	6.1	91	5.9	69
Dec 2014	3.4	69	5.4	91	3.8	51
Jan 2015	−0.7	33	1.6	51	0.1	51
Feb 2015	−3.4	53	−0.9	61	−2.4	36
Mar 2015	5.5	117	6.5	89	5.4	89
Apr 2015	11.3	132	13.6	74	12.9	112
May 2015	17.7	61	20	38	19.9	91
Jun 2015	21.8	89	NA ¹	NA	23.1	168
Jul 2015	22.6	109	24.6	112	24.2	150
Aug 2015	21.4	104	23.6	94	23.4	15
Sep 2015	18.6	218	21.6	112	21.2	178

¹NA = not available.

Table 2. Planting and harvesting dates for winter and summer crops

Item	Blacksburg		Blackstone		Orange	
	Planting	Harvesting	Planting	Harvesting	Planting	Harvesting
Winter crops ¹	Sep 19, 2014	Apr 25, 2015	Oct 8, 2014	Apr 24, 2015	Sep 11, 2014	Apr 27, 2015
Corn	May 8, 2015	Aug 27, 2015	May 6, 2015	Aug 21, 2015	May 12, 2015	Aug 24, 2015
Sorghum	May 8, 2015	Sep 11, 2015	May 6, 2015	Sep 1, 2015	May 12, 2015	Sep 5, 2015

¹Barley, ryegrass, rye, triticale, and wheat in monoculture or mixed with crimson clover or hairy vetch.

Treatments and Experimental Design

For the first objective of this study, 5 annual grasses were planted in monoculture (i.e., no legumes; **NO**) or in mixture with 1 of 2 annual legumes resulting in 15 treatments. Annual grasses included barley (**BA**; *Hordeum vulgare* ‘Thoroughbred’), ryegrass (**RG**; ‘Marshall’), rye (**RY**; ‘Huron’), triticale (**TR**; *Triticosecale* ‘Trical 815’), and wheat (**WT**; *Triticum aestivum* ‘Merl’). Annual legumes included crimson clover (**CC**; *Trifolium incarnatum* ‘Dixie’) and hairy vetch (**HV**; *Vicia villosa* ‘Purple Bounty’). All seed, except BA and WT, was provided by a regional supplier (King’s Agriseeds Inc., Ronks, PA). Barley and WT seed was provided by the Virginia Tech small grain breeding program. Planting dates are presented in Table 2. Planting densities for treatments (Table 3) were selected following guidelines from Brann et al. (2009). Within each site, 4 plots for each of the 15 treatments were planted according to a randomized complete block design (i.e., 60 plots per site) in 9.2-m-long plots consisting of 7 rows separated by 15 cm. For each mixture, grass and legume seeds were combined, mixed, and planted using a no-till experimental drill (Hege 1000, Colwich, KS). Fertilizer was applied before planting at 22 kg/ha of N, 56 kg/ha of P₂O₅, and 45 kg/ha of K₂O in Blacksburg and Orange. Fertilizer was applied before planting at 30 kg/ha of N and 32 kg/ha of K₂O in Blackstone. Winter crops were not fertilized during the spring.

For the second objective of this study, after harvesting the winter crops each plot was split equally in half (i.e., 4.6 m × 1.30 m), and each half was planted with either corn (*Zea mays* Mycogen TMF2L874; Mycogen, Indianapolis, IN) or forage sorghum (*Sorghum bicolor*

Alta Seeds AF7401; Alta Seeds, Irving, TX) in a direction perpendicular to the winter crop plantings. Both sorghum and corn were planted in 76-cm rows using a no-till planter.

Harvesting Winter Crops

Due to the complexity of the experiment (i.e., multiple and remote sites and many treatments), a single harvest, regardless of the different winter crops, was conducted at each site (Table 2). Thus, not all winter crops were harvested at the same developmental stage. As a reference, all sites were harvested when BA had inflorescences fully emerged. This stage was chosen so the corn and sorghum could be planted at dates typical for the region (Table 2). Of the remaining winter grasses, RY and TR had three-fourths of inflorescences emerged, whereas RG and WT were harvested at late vegetative stages (i.e., boot stage). Of the legumes, CC was harvested in early- to mid-flowering stage, whereas HV was harvested in bud to early-flowering stage.

Forage biomass of the whole plot was harvested using a Cibus F plot forage harvester (Wintersteiger Inc., Salt Lake City, UT) at Blackstone and Orange and a Swift Current Forage Harvester IV (Thompson, 1972) in Blacksburg. After weighing harvested biomass, samples of fresh forages (~1 kg) were collected in plastic bags, immediately placed in a cooler with dry ice, and transferred to the laboratory for storage at -20°C.

Summer Crops

Within 15 d after harvesting the winter crops (Table 2), corn and sorghum were planted at seeding rates

Table 3. Planting densities (kg/ha) of winter crops

Item	Grass (monoculture)	Grass + crimson clover	Grass + hairy vetch
Barley	108	54 + 11	54 + 11
Ryegrass	28	14 + 11	14 + 11
Rye	126	63 + 11	63 + 11
Triticale	126	63 + 11	63 + 11
Wheat	135	68 + 11	68 + 11

of 70,000 and 220,000 plants/ha, respectively, using a 6-row John Deere 7200 no-till planter (Deere and Co., Moline, IL) with 76 cm between rows. Urea was applied at 35 kg/ha of N at planting and 155 kg/ha of N sidedressed at postemergence (i.e., 6 fully expanded leaves) in Blacksburg and Orange. At Blackstone, fertilizer was applied at planting only at a rate of 168 kg/ha of N, 168 kg/ha of P_2O_5 , and 168 kg/ha of K_2O . Harvest date (Table 2) was targeted to occur at the one-half milk-line stage of maturity for corn.

At harvest, 5 consecutive plants from 2 center rows (from a total of 6 rows) and at 2 randomly selected spots within each plot (i.e., 10 plants per plot) were cut by hand at 15 cm above ground level. Whole plants were weighed and chopped with a Stanley CH2 wood chipper (GXi Outdoor Power LLC, Clayton, NC). After mixing thoroughly within a barrel, samples of fresh forages were collected in plastic bags, immediately placed in a cooler with dry ice, and transferred to the laboratory for storage at $-20^{\circ}C$. The number of plants per row was counted, and estimated DM yield was determined as plant biomass multiplied by the number of plants per hectare.

Sample Processing

For the winter crops, samples were thawed and then chopped using a commercial lettuce cutter (Vollrath Redco Lettuce King I; Vollrath Co. LLC, Sheboygan, WI). The length of cut was set at 19 mm. One subsample of the chopped material (hereafter named fresh forage) was dried to a constant weight at $55^{\circ}C$ in a forced-air oven. The resulting DM concentration was used to determine estimated DM yield of the winter crop. The dried fresh forage was then ground to pass through a 1-mm screen of a cyclone mill (Udy Corp., Fort Collins, CO) before conducting chemical analyses.

A second 400- to 500-g subsample of the chopped material was placed in 620-cm² aluminum pans and wilted using space heaters (Sunbeam Products Inc., Boca Raton, FL) and standing fans (Lasko Products Inc., West Chester, PA). After wilting for approximately 30 h (until target 30% of DM), a 400- to 500-g subsample (hereafter named ensiled forage or silage) was inoculated using Biotol Plus II (Lallemand Animal Nutrition, Milwaukee, WI) following the manufacturer's instructions. The inoculated material was placed into MR-1014 polyethylene embossed pouches (Doug Care Equipment, Springfield, CA) and double sealed anaerobically with a minipack FastVac vacuum sealer (Doug Care Equipment) as described by Der Bedrosian et al. (2012). After 60 d of fermentation in the dark, mini silos were opened and pH was determined by blend-

ing (Waring Commercial, Torrington, CT) 10 g of corn silage with 90 mL of deionized water for 5 min and immediately measuring pH (Accumet AB 150 pH meter, Fisher Scientific Co., Suwanee, GA). The remaining winter crop silage was dried at $55^{\circ}C$ in a forced-air drying oven for 48 h and ground to pass through a 1-mm screen of a cyclone mill.

For corn and sorghum, chopped samples were thawed and a first subsample (hereafter named fresh corn or sorghum) was dried in a forced-air drying oven to a constant weight at $55^{\circ}C$. The resulting DM concentration was used to estimate DM yield. The dried corn and sorghum were then ground to pass through a 1-mm screen of a cyclone mill for nutritional analyses. A second 400- to 500-g subsample of the thawed sample (hereafter named corn or sorghum silage) was ensiled and then processed as the winter crops except no inoculants were added to enhance fermentation.

Nutritional Composition and Digestibility

Ash concentration was determined after combusting samples in a furnace for 3 h at $600^{\circ}C$ (method 942.05; AOAC International, 2016). Crude protein concentration was calculated as percentage N $\times$ 6.25 after combustion analysis (method 990.03; AOAC International, 2016) using a Vario El Cube CN analyzer (Elementar Americas Inc., Mount Laurel, NJ). Neutral detergent fiber and ADF concentrations were determined using the Ankom200 Fiber Analyzer (Ankom Technology, Macedon, NY). Sodium sulfite and α -amylase (Ankom Technology) were included for NDF analysis. Acid detergent fiber and ADL concentrations were determined sequentially. After determining ADF, the residual fiber was incubated for 3 h in 72% sulfuric acid within a 4-L jar that was placed in a DaisyII Incubator (Ankom Technology). Starch concentrations were determined using the acetate buffer method of Hall (2009) with α -amylase from *Bacillus licheniformis* (Ankom Technology) and amyloglucosidase from *Aspergillus niger* (E-AMGDF; Megazyme International, Wicklow, Ireland). Sugar concentrations were determined as total ethanol/water-soluble carbohydrates as described by Hall et al. (1999).

In vitro DM digestibility, in vitro true DM digestibility (IVTDM), and in vitro NDF digestibility (IVNDFD) were determined using a DaisyII rotating jar in vitro incubator (Ankom Technology) following the procedures described by Ferreira and Mertens (2005). A composited inoculum was prepared with rumen fluid and rumen solids collected from 3 ruminally cannulated lactating dairy cows (1 Holstein and 2 Jerseys) that were fed a diet containing 43% corn silage, 6% triticale

Table 4. Yield, nutritional composition (DM basis), and digestibility of fresh forage from winter crops

Item	Legume ¹ (L)			Grass ² (G)					SEM	<i>P</i> <		
	NO	CC	HV	BA	RG	RY	TR	WT		L	G	L × G
Estimated DM yield, Mg/ha	2.40 ^B	2.84 ^A	2.47 ^B	3.11 ^a	2.47 ^{bc}	2.50 ^{bc}	2.22 ^c	2.79 ^b	0.25	0.01	0.01	0.53
DM, %	19.9 ^A	17.7 ^B	17.3 ^B	19.5 ^a	18.9 ^a	17.5 ^{bc}	17.0 ^b	18.7 ^c	0.59	0.01	0.01	0.31
Ash, %	8.8	8.8	9.1	8.8	9.2	8.8	8.7	8.8	0.53	0.13	0.20	0.23
CP, %	13.0 ^C	15.5 ^B	17.3 ^A	14.2 ^b	14.3 ^b	16.1 ^a	16.2 ^a	15.7 ^a	1.06	0.01	0.01	0.44
ADL, %	2.4 ^C	2.7 ^B	2.9 ^A	2.7 ^b	2.6 ^b	2.4 ^b	2.5 ^b	3.0 ^a	0.21	0.01	0.01	0.33
Sugars, %	14.3 ^A	13.2 ^B	10.5 ^C	10.8 ^c	9.9 ^c	13.2 ^b	13.4 ^b	15.9 ^a	1.98	0.01	0.01	0.09
Starch, %	3.2 ^B	3.6 ^A	3.2 ^B	3.0 ^c	2.7 ^c	3.5 ^b	3.5 ^b	4.0 ^a	0.25	0.01	0.01	0.37

^{a-c}Values for grass main effects with different lowercase letters within a row differ statistically ($P < 0.05$).

^{A-C}Values for legume main effects with different uppercase letters within a row differ statistically ($P < 0.05$).

¹Main effects of legume: NO = average of all grasses grown in monoculture; CC = average of all grasses grown with crimson clover; HV = average of all grasses grown with hairy vetch.

²Main effects of grass: BA = (barley grown in monoculture + barley grown with crimson clover + barley grown with hairy vetch)/3; RG = (ryegrass grown in monoculture + ryegrass grown with crimson clover + ryegrass grown with hairy vetch)/3; RY = (rye grown in monoculture + rye grown with crimson clover + rye grown with hairy vetch)/3; TR = (triticale grown in monoculture + triticale grown with crimson clover + triticale grown with hairy vetch)/3; WT = (wheat grown in monoculture + wheat grown with crimson clover + wheat grown with hairy vetch)/3.

silage, 4% alfalfa hay, and 47% concentrate mix (DM basis).

Statistical Analyses

All variables were analyzed using the MIXED procedure of SAS (version 9.4; SAS Institute Inc., Cary, NC). For the winter crops, data were analyzed as a randomized complete block design using a model that included the effects of site [random, degrees of freedom (df) = 2], block within site (random, df = 9), grass (fixed, df = 4), legume (fixed, df = 2), the grass × legume interaction (fixed, df = 8), and the residual error (random, df = 154).

For corn and sorghum, data were analyzed as a randomized complete block design in a split plot. The model included the effects of site (random, df = 2), block within site (random, df = 9), grass (fixed, df = 4), legume (fixed, df = 2), the grass × legume interaction (fixed, df = 8), the site × block × grass × legume interaction as the whole plot error (random, df = 154), the effect of summer crop (fixed, df = 1), the interaction of summer crop × grass (fixed, df = 4), the interaction of summer crop × legume (fixed, df = 2), the interaction of summer crop × grass × legume (fixed, df = 8), and the residual error (random, df = 165). Significant differences and tendencies to differ were declared at $P < 0.05$ and $P < 0.10$, respectively.

RESULTS AND DISCUSSION

Winter Crop Yield, Quality, and Digestibility

One objective of this study was to determine the effect of growing grasses in mixtures with legumes on for-

age yield, nutritional quality, and digestibility. Growing grasses in mixtures with legumes tended to increase forage estimated DM yields (Table 4). This increase was attributed to the yield of mixtures with CC (2.84 Mg/ha) only, as yields of mixtures with HV (2.47 Mg/ha) were similar to those of grasses grown in monoculture (2.40 Mg/ha). Among grasses, BA had the greatest estimated DM yields (3.11 Mg/ha) and TR, RG, and RY had the lowest (mean = 2.40 Mg/ha) estimated DM yields. Carr et al. (2004) reported respective DM yields of 2.91 and 3.84 Mg/ha for barley and oats grown in monoculture. However, DM yields increased to 3.53 and 4.56 Mg/ha, respectively, when barley and oats were grown in mixtures with peas. Similarly, growing oats in mixtures with vetch increased forage yield 10 to 35% relative to oats grown in monoculture (Kaiser et al., 2007). However, depending on harvesting time, the increase of forage yield ranged from 0 to 18% when ryegrass was grown in mixtures with vetch relative to ryegrass grown in monoculture (Kaiser et al., 2007).

Forage DM yields were lower than those observed for triticale and wheat (4.6 and 3.7 Mg/ha, respectively) in Virginia during 2015 (Thomason et al., 2015). There are 2 major reasons for these differences. First, we harvested the winter crops in early spring (Lawson et al., 2015) as we prioritized planting the following summer crop at early dates to maximize forage yield. Second, under the assumption that growing grasses with legumes would provide additional N, we fertilized with N only at planting (fall 2014) and not during the growing season (spring 2015).

It is worth mentioning that the low fertilization rates and the single and early dates for harvesting different winter crops were intentional. Under the concept that legumes add fertility to the soil, we challenged

the system for the legumes to reflect their potential to increase residual fertility for the following summer crop. In regards to harvesting, early and late harvesting dates have positive and negative effects. Delaying harvesting would likely increase forage yields of the winter crop, although it may deplete moisture and fertility for the following summer crop. As a secondary effect, a delayed planting date of the summer crop can also expose silking time to periods of heat stress, which may be detrimental to forage yield and quality (Ferreira et al., 2015). In contrast, harvesting winter crops early may improve the growing conditions for the following summer crop, albeit at the expense of DM yield of the preceding winter crop. Although somewhat unlikely, dairy farmers may elect to kill the winter crop rather than harvesting it as silage. Finally, the single harvest date for all winter crops was decided based on a single (optimum) planting date for the summer crops; different planting dates would have confounded the growth responses of the summer annual forage.

Growing grasses in mixtures with legumes increased the CP concentration of the fresh forage (Table 4) from 13.0% to 15.5% for CC and to 17.3% for HV. After ensiling, the CP of silages containing legumes also was greater than silages from grasses only (Table 5). As for the fresh forages, the concentration of CP in silages was also greater for HV than for CC (18.3 and 16.6%, respectively). Among grasses, the concentration of CP in fresh forage (Table 4) was lower for BA and RG (14.3%) than for the other grasses (16.0%). Generally, similar trends were observed for ensiled forages (Table 5).

For NDF concentration, the interaction between grasses and legumes was significant for both fresh ($P <$

0.01; Figure 1) and ensiled (data not shown) forages. Growing BA, RY, and TR in mixtures with legumes decreased NDF concentrations, whereas growing RG and WT with legumes did not change NDF concentrations of either the fresh or the ensiled forages. Grasses usually contain greater fiber concentrations than legumes (Fulkerson et al., 2007; Kammes and Allen, 2012; Ergon et al., 2016). Therefore, growing grasses in mixtures with legumes containing lower fiber concentrations likely reduces the NDF concentrations in the harvested forage. Similar to our study, Carr et al. (2004) reported lower NDF concentrations of fresh forage when barley and oats were intercropped with peas compared with barley and oats grown in monoculture (58.4% vs. 50.8% for barley and 61.8% vs. 55.2% for oats). The lower NDF concentrations in our grass-legume mixtures indicate that adding legumes to grasses may increase the energy concentration of winter crop forages (Carr et al., 2004).

Among grasses, NDF concentrations of fresh forages were lowest for RG (43.1%) and greatest for BA (57.4%; Figure 1). We relate the greatest estimated DM yield and NDF concentration observed for BA to the more advanced growth stage (i.e., inflorescence fully emerged) relative to the growth stage of other grasses. The lower concentrations of NDF in RG and WT explain the interaction observed between grasses and legumes. Even though we did not measure it in pure legumes, the concentrations of NDF for these 2 grasses would be proximate to the concentration of NDF of the legumes (Figure 1).

Growing grasses in mixtures with legumes decreased the concentration of sugars within fresh forages relative to grasses grown in monoculture (Table 4). Lower con-

Table 5. Postfermentation pH, nutritional composition (DM basis), and digestibility of ensiled forage from winter crops

Item	Legume ¹ (L)			Grass ² (G)					SEM	$P <$		
	NO	CC	HV	BA	RG	RY	TR	WT		L	G	L × G
pH	4.14 ^B	4.19 ^B	4.42 ^A	4.39 ^a	4.17 ^c	4.28 ^b	4.30 ^{ab}	4.11 ^c	0.12	0.01	0.01	0.91
DM, %	28.4 ^A	24.6 ^B	24.8 ^B	29.3 ^a	27.4 ^{ab}	24.1 ^c	23.5 ^c	25.3 ^{bc}	1.07	0.01	0.01	0.33
Ash, %	9.0 ^B	10.2 ^A	10.4 ^A	9.3 ^b	10.6 ^a	9.5 ^b	10.3 ^a	9.5 ^b	0.95	0.01	0.01	0.91
CP, %	14.2 ^C	16.6 ^B	18.3 ^A	15.1 ^d	15.8 ^{cd}	16.5 ^{bc}	17.3 ^a	17.0 ^{ab}	0.80	0.01	0.01	0.32
ADF, %	29.9 ^B	29.3 ^B	31.5 ^A	32.6 ^a	28.7 ^b	31.9 ^a	29.2 ^b	28.9 ^b	1.45	0.01	0.01	0.27
ADL, %	3.0 ^B	3.5 ^B	3.9 ^A	3.3	3.5	3.3	3.2	4.1	0.59	0.02	0.11	0.81
Sugars, %	3.1 ^A	3.6 ^A	2.0 ^B	2.2	3.5	2.3	3.4	3.0	0.70	0.01	0.09	0.31
IVNDFD, ³ % of NDF	74.4 ^A	71.5 ^B	71.1 ^B	69.0 ^c	74.8 ^a	71.4 ^b	73.3 ^{ab}	73.2 ^{ab}	2.02	0.01	0.01	0.73

^{a-d}Values for grass main effects with different lowercase letters within a row differ ($P < 0.05$).

^{A-C}Values for legume main effects with different uppercase letters within a row differ ($P < 0.05$).

¹Main effects of legume: NO = average of all grasses grown in monoculture; CC = average of all grasses grown with crimson clover; HV = average of all grasses grown with hairy vetch.

²Main effects of grass: BA = (barley grown in monoculture + barley grown with crimson clover + barley grown with hairy vetch)/3; RG = (ryegrass grown in monoculture + ryegrass grown with crimson clover + ryegrass grown with hairy vetch)/3; RY = (rye grown in monoculture + rye grown with crimson clover + rye grown with hairy vetch)/3; TR = (triticale grown in monoculture + triticale grown with crimson clover + triticale grown with hairy vetch)/3; WT = (wheat grown in monoculture + wheat grown with crimson clover + wheat grown with hairy vetch)/3.

³IVNDFD = in vitro NDF digestibility.

concentrations of sugars in legumes relative to grasses were reported previously (Kaiser et al., 2007; Ergon et al., 2016). The lower concentration of sugars was counterbalanced by greater protein and, to a lesser extent, by changes in the concentration of NDF. Because sugars are the substrate for lactic acid bacteria during the ensiling process, sugar concentrations decreased substantially for all silages after the ensiling process (Table 5). Sugar concentration of ensiled samples followed a similar trend observed in fresh samples, as including HV in the winter crop mixture reduced sugar concentrations in the silage. However, the magnitude of this difference has minimal nutritional implications.

Among grasses, WT had the greatest (15.9%) and BA and RG had the lowest concentrations of sugars in fresh forage (10.4 and 9.9%, respectively; Table 4). Despite this, the concentration of sugars was similar in the ensiled forage (Table 5). As sugars are the substrates for microbes during the ensiling process, the concentrations of sugars in fresh forage were negatively correlated with silage pH ($-0.41 < r < -0.23$; data not shown).

The silage pH of grasses grown in mixtures with HV (pH = 4.42) was greater than the silage pH of the grasses grown in monoculture (pH = 4.14; Table 5). The silage pH of grasses grown in mixtures with CC (pH = 4.19) was similar to the silage pH of the grasses grown in monoculture. A lower pH usually indicates a more adequate ensiling process. In our study, differences in

pH were linked to differences in sugar concentrations of the fresh forage. However, as sugar concentrations were adequate to ensure appropriate silage fermentation, changes in pH may also relate to differences in buffering capacities of the forages or a combination of buffering capacity and concentration of sugars (Kaiser et al., 2007; Coblenz and Muck, 2012; Coblenz et al., 2016). Overall, ensiling grasses in mixtures with HV may be more challenging to lower silage pH than ensiling monocultures or mixtures of grasses with CC.

For both fresh ($P < 0.01$) and ensiled (data not shown) forages, an interaction existed between grass species and the addition of legumes for IVTDMD (Figure 2). For the fresh forage, the addition of legumes to RG and WT reduced the IVTDMD. However, with the addition of legumes to BA, RY, and TR, IVTDMD remained the same. The ratio of lignin to fiber is greater for legumes than for grasses (Kuoppala et al., 2009). For RG and WT, the lower IVTDMD would be related to a greater lignin: fiber ratio and, therefore, lower digestibility of NDF, as adding legumes did not affect the concentration of NDF.

Similar to IVTDMD, the interaction for IVNDFD between grass species and the addition of legumes was significant for fresh forages ($P < 0.03$; Figure 3). The addition of legumes to RG, TR, and WT lowered the IVNDFD of the fresh forages. However, with the addition of legumes to BA and RY, IVNDFD remained the same.

Figure 1. Concentration of NDF in fresh forages (DM basis) harvested from different winter crops. Five grasses (barley [BA], ryegrass [RG], rye [RY], triticale [TR], and wheat [WT]) were grown in monoculture or in mixtures with 2 legumes (crimson clover [CC] and hairy vetch [HV]). NO = no legumes. Treatment means with different letters (a–e) differ ($P < 0.05$). Error bars are SEM = 1.33.

Figure 2. In vitro true DM digestibility (IVTDMD) in fresh forages (DM basis) harvested from different winter crops. Five grasses (barley [BA], ryegrass [RG], rye [RY], triticale [TR], and wheat [WT]) were grown in monoculture or in mixtures with 2 legumes (crimson clover [CC] and hairy vetch [HV]). NO = no legumes. Treatment means with different letters (a–d) differ ($P < 0.05$). Error bars are SEM = 1.84.

Fiber digestibility of legumes is usually lower than fiber digestibility of grasses (Voelker-Linton and Allen, 2008). Legumes have more lignin than grasses on a DM basis (Kuoppala et al., 2009). The lignin of legumes

is all concentrated in the xylem, whereas the lignin of grasses is spread out evenly among all tissues except the phloem (Wilson and Kennedy, 1996). The xylem of legumes is almost completely indigestible, but the

Figure 3. In vitro NDF digestibility (IVNDFD) in fresh forages (% NDF) harvested from different winter crops. Five grasses (barley [BA], ryegrass [RG], rye [RY], triticale [TR], and wheat [WT]) were grown in monoculture or in mixtures with 2 legumes (crimson clover [CC] and hairy vetch [HV]). NO = no legumes. Treatment means with different letters (a–c) differ ($P < 0.05$). Error bars are SEM = 3.43.

Table 6. Yield, nutritional composition (DM basis), and fiber digestibility of fresh summer crops and postfermentation pH, nutritional composition, and fiber digestibility of fresh and ensiled summer crops as affected by preceding winter crops

Item	Legume ¹ (L)			Grass ² (G)					SC ³			<i>P</i> <			
	NO	CC	HV	BA	RG	RY	TR	WT	C	S	SEM	L	G	L × G	SC
Fresh															
Estimated DM yield, Mg/ha	15.6	15.0	14.6	16.7 ^a	14.0 ^b	15.3 ^{ab}	15.5 ^a	14.0 ^b	14.9	15.3	1.00	0.28	0.01	0.21	0.47
DM, %	27.8	27.2	27.5	27.8	26.7	28.0	27.5	27.4	28.5	26.5	1.62	0.10	0.01	0.03	0.01
Ash, %	4.3	4.3	4.3	4.4	4.4	4.2	4.3	4.3	3.5	5.1	0.12	0.59	0.09	0.15	0.01
CP, %	10.5	10.7	10.5	10.6	10.6	10.3	10.6	10.6	10.0	11.1	0.37	0.26	0.41	0.46	0.01
NDF, %	47.4	47.2	47.5	46.4	49.1	46.6	47.7	47.0	42.8	52.0	0.99	0.94	0.06	0.07	0.01
IVNDFD, ⁴ % of NDF	42.0	42.1	42.2	41.9	42.5	41.9	42.0	42.1	39.9	44.4	0.79	0.72	0.53	0.03	0.01
Silage															
pH	3.72	3.70	3.72	3.72	3.70	3.71	3.71	3.72	3.65	3.78	0.02	0.46	0.82	0.80	0.01
DM, %	28.0	27.8	27.8	28.1	27.2	28.3	28.1	27.7	28.7	27.0	1.89	0.78	0.05	0.06	0.01
Ash, %	4.5	4.5	4.5	4.5	4.5	4.4	4.5	4.5	3.5	5.4	0.07	0.93	0.71	0.04	0.01
CP, %	10.4	10.5	10.4	10.4	10.3	10.4	10.5	10.5	10.2	10.7	0.45	0.87	0.73	0.91	0.01
NDF, %	44.4	45.8	45.7	45.2	46.1	44.7	44.8	45.7	41.3	49.3	1.31	0.11	0.49	0.11	0.01
IVNDFD, % of NDF	44.2	44.2	44.3	44.3	44.9	43.9	43.9	44.2	42.6	45.9	0.74	0.97	0.43	0.16	0.01

^{a,b}Values for grasses with different superscripts within a row differ ($P < 0.05$).

¹Main effects of legume: NO = average of all grasses grown in monoculture; CC = average of all grasses grown with crimson clover; HV = average of all grasses grown with hairy vetch.

²Main effects of grass: BA = (barley grown in monoculture + barley grown with crimson clover + barley grown with hairy vetch)/3; RG = (ryegrass grown in monoculture + ryegrass grown with crimson clover + ryegrass grown with hairy vetch)/3; RY = (rye grown in monoculture + rye grown with crimson clover + rye grown with hairy vetch)/3; TR = (triticale grown in monoculture + triticale grown with crimson clover + triticale grown with hairy vetch)/3; WT = (wheat grown in monoculture + wheat grown with crimson clover + wheat grown with hairy vetch)/3.

³Main effects of summer crop (SC): corn (C) vs. sorghum (S).

⁴IVNDFD = in vitro NDF digestibility.

remaining tissues are very digestible; therefore, the passage rate of legumes is faster than that of grasses because the lower concentration of indigestible fiber inhibits the passage rate of grasses (Kuoppala et al., 2009). The greater concentration of lignin and faster passage rate decreases the fiber digestibility of legumes. However, legumes usually have a lower fiber concentration and higher NFC concentration compared with grasses. Therefore, even though in some cases IVNDFD was reduced with the addition of legumes (Figure 3), the concentration of highly digestible nonfibrous components was greater in the mixtures than in the monocultures (Figure 1), thereby maintaining the nutritive value of the silage.

Summer Crops Yield, Quality, and Digestibility

The second major objective of this study was to determine the effect of growing different winter crops on forage yield, nutritional quality, and digestibility of the following summer crop. Relative to growing grasses in monoculture, growing winter-annual grasses in mixtures with legumes did not change estimated DM yields of corn or sorghum (Table 6), indicating no measurable benefits for growing grasses in mixtures with legumes. Gabriel and Quemada (2011) reported that when barley or hairy vetch were used as a cover crop for corn, corn yields were similar to a bare fallow control; however,

corn took up more N following vetch cover compared with the bare fallow control. In our study, the winter crops were harvested for silage and the above-ground biomass was not left as cover for the following summer crop; therefore, any advantage of adding legumes to the grasses may have been lost due to the small amount of residue remaining. Alternatively, cover crops can promote below-ground biomass development (Kuo et al., 1997a, b).

In addition to the similar estimated DM yields, in our study the nutritional composition and digestibility of the fresh and ensiled summer crops remained the same with the addition of legumes (Table 6). This was the case for both corn and sorghum crops.

Plots containing RG and WT reduced yield of both corn and sorghum (14.0 Mg/ha) compared with other grasses (16.7 and 15.5 Mg/ha for BA and TR, respectively; Table 6). Despite herbicide application after harvesting the winter crops, we observed a regrowth of RG after emergence of summer crops, which resulted in high weed pressure during early growth, therefore reducing DM yields. Although ryegrass can be an excellent cover crop, its resilience to herbicide applications can make ryegrass termination extremely difficult (Legleiter et al., 2015).

There were differences in nutrient composition but not yield between the 2 summer crops (corn vs. sorghum). As the interactions of summer crop × grasses,

summer crop $\times$ legumes, and summer crop $\times$ grasses $\times$ legumes were not significant (data not shown), no effects of winter crops on yield, nutritional composition, and digestibility of the fresh and ensiled forages were observed for the following summer crops (Table 6).

CONCLUSIONS

In this 1-yr study within the Mid-Atlantic region, growing grasses in mixtures with CC as winter crops for forage increased forage estimated DM yields in dairy farming systems, although this was not the case for HV. Also, growing grasses with legumes increased the concentration of CP, whereas it decreased the concentrations of NDF and sugars. As tissues from legumes have more lignified cell walls, there can be a decrease in fiber digestibility when grasses are grown with mixtures. Despite the lower fiber digestibility, the energy can be maintained by the lower fiber concentration when growing grasses in mixtures with legumes. Finally, under the conditions of this study, the inclusion of legume species in the winter crop for forages did not affect the yield, nutritional composition, or digestibility of the succeeding corn or sorghum.

ACKNOWLEDGMENTS

We thank Steve Gulick and Elizabeth Rucker (Department of Crop and Soil Environmental Sciences) for their assistance in preparing the experimental plots. We also thank Yang Yang (Department of Dairy Science) for assistance with harvesting plots. Finally, we thank King's Agriseeds (Ronks, PA) for donating the seeds for winter crops (i.e., ryegrass, rye, triticale, crimson clover, and hairy vetch), Advanta Seeds (Irving, TX) for donating the seeds for forage sorghum, and Lallemant Inc. (Milwaukee, WI) for donating the silage inoculant. This project was funded mainly by the USDA Natural Resources Conservation Virginia Conservation Innovation Grant program and partially by USDA National Institute of Food and Agriculture Hatch Project VA-160025 and USDA National Institute of Food and Agriculture Multistate Project VA-136291 (NC-2042, Management Systems to Improve the Economic and Environmental Sustainability of Dairy Enterprises).

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