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Forage contribution of cool-season annuals as cover crops in warm-season pastures

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Cover crops (CC) can contribute to production in pastures, but the diversity of CC mixtures and defoliation frequency (DF) may alter productivity. A 2-yr experiment conducted at Raymond, MS, quantified CC × DF effects on forage mass (FM) and nutritive value of winter CC and subsequent summer hay production. Treatments were factorial combinations of 10 CC (using several species of grasses, legumes, and brassicas) and three DF (harvested every 4 or 8-wk or cut and left as mulch) in a split-plot arrangement of a randomized complete block design experiment with three replications. Generally, mixtures with legumes had greater FM and better nutritive value. Summer hay production did not respond to difference in CC composition, however, harvesting of CC reduced summer hay but increased year-long FM. These results suggest that CC when harvested can contribute to forage production with improved nutritive value and can increase year-long FM, but summer hay production can benefit when the CC is left as mulch.

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CHAPTER I

INTRODUCTION

Global environmental concerns have received extensive publicity and political attention in recent years with demands for food production and economic issues often perceived as constraints to more sustainable production approaches (FAO, 2017). Progress toward global sustainability has repeatedly failed to meet goals, as illustrated by missed targets for reduction in loss of species and ecosystem services adopted by the Global Strategic Plan for Biodiversity 2011-2020 interim targets. Both food security and environmental sustainability are priority goals for US agriculture, and multiple-species forage crops provide an opportunity for support of such combined goals. Perennial warm-season grass pastures and rangelands of the lower south-central USA are intensively managed, involving repeated use of nitrogen fertilizer throughout the year and chemical control of weeds. Environmental sustainability and productivity by improving the soil quality of these pastures and rangelands may be enhanced by overseeding with mixtures of multiple plant species that can improve herbage mass, soil health, soil fertility, plant biodiversity, and plant population stability (Scherr and McNeely, 2008).

Soil quality is the soil's ability to sustain plant and animal life in a managed or natural ecosystem and overall enhancing environmental quality (Singh et al., 2014). Some prefer the term "soil health" because it portrays the soil as a living system, whose functions are facilitated by a diversity of organism that needs management and conservation. Proper soil management will increase organic matter and enhance microbial activity (Mohammadi et al., 2011). Often this can be accomplished by reducing soil tillage, frequently adding organic matter, growing diverse species of plants, eliminating practices that may encourage salinization, and keeping the land covered with vegetation throughout the year (White et al., 2017).

Tilling soil repeatedly is a practice that is as old as agriculture itself. Tillage involves physical, chemical, and biological soil manipulation to optimize conditions for germination, seedling establishment, and crop growth. Such soil inversions, however, can result in decreased soil moisture, intensified compaction, and diminished organic material, all of which can lead to an upsurge in water and fertilizer usage, along with reduced drainage and increased erosion and runoff (Sharma and Abrol, 2012). Birkás et al. (2004) reported that annual disking and plowing resulted in subsoil compaction at the depth of tillage within three years and that the compacted layer expanded both in the surface and deeper layers after the five years. Hamza and Anderson (2003) reported that water infiltrated faster in soils that are less compacted and have well-aggregated soil particles than compacted soil structures. Mijangos et al. (2006) reported that indicators of soil quality, including mineralizable N, enzyme activity, basal respiration, substrate-induced respiration, and earthworm abundance, were greater in no-till plots than in conventionally tilled plots.

Plant diversity is one of the pillars that can improve soil health. It is often shown that diverse crop species increases productivity and stability in natural grasslands (Jing et al., 2017). In pastures, a diversified ecosystem has led to improved forage production and reduced weed invasion when compared to communities with few plant species (Tracy and Sanderson, 2004). Diverse mixtures will also enhance the amount and total biomass stability within grasslands because of niche complementarity in species-rich mixes that optimize the capture of light, water, and nutrient resources (Jing et al., 2017). Sanderson et al. (2007) reported that different mixtures produced more herbage mass than pure grass stands in rotationally stocked pastures but that in clipped small plot studies, there was no herbage production benefit from planting forage mixtures of four or more species. The advantage of diverse mixtures in a pasture may depend on the presence of the most productive species under different environmental conditions to which pastures are exposed across several seasons. Some have viewed species diversity as a sort of insurance policy where different species contribute in its own time or can take the place of species that fail from stress or mismanagement (Sanderson et al., 2007). As a result, more diverse grassland plant communities are more likely to maintain ecosystem functions during periods of climatic stress (Russell and Bisinger, 2015).

Greater diversity of plant communities has resulted in increased aboveground productivity and carbon sequestration as soil organic matter. Nitrogen fixed by legume species enhanced belowground biomass production by warm-season grass species (Quinn et al., 2013). A field experiment conducted by Elhakeem et al. (2018) looked at the variability of the biomass of ten species mixtures and ten pure stands obtained from six plant families, including crucifers, grasses, and legumes. They reported that diverse groups had greater average biomass yields (4.5 Mg ha^{-1}) compared to pure stands (3.7 Mg ha^{-1}). The diversity of species in a complex plant community will fulfill some positions within a landscape, reducing the chances for invasive species to establish (Xu et al., 2015). Florence et al. (2019) found that increases in species diversity correlated with increased weed suppression and biomass stability. Furthermore, diverse plant communities provide variation in feed materials and plant structure required to withstand a diverse population of animal species (Russell and Bisinger, 2015). The results of these benefits with plant diversity are considered a measure of rangeland health when prevailing vegetation can be associated with native vegetation (Ricklefs et al., 2008).

Cover crops are conventionally used as a soil-amendment tool. There are ancillary benefits, however, from growing cover crops such as weed management, improved water quality, nutrient cycling, moisture conservation, crop productivity, and source of feed for livestock (Hobbs et al., 2008). The ultimate impact of cover crops depends on many factors, but the most crucial factor is the cover crop species. Forage legumes are quite popular in many grassland farming areas around the world, and their importance has increased because of their ability to fix atmospheric N_2 biologically, and secondly because they increase protein concentration and digestibility of the sward (Iglesias and Lloveras, 1998; Rochon et al., 2004). Grass cover crops control soil erosion, produce high biomass, suppress weeds, and improve soil organic matter (Wilson and Slocum, 2017). Brassicas cover crops with their robust root systems help to reverse soil compaction and increase infiltration (Chen and Weil, 2010; Kaspar et al., 2001). Single species cover crops cannot deliver all the multiple ecosystem service benefits that diverse cover crop mixtures can provide. Thus, different mixes are essential in supplying more multifunctional benefits to the ecosystem (Tosti et al., 2014).

Cover crops can be viewed as a temporary pasture with a source of high–quality feed (Golden et al., 2016). Grass species such as annual ryegrass (*Lolium multiflorum* L.) (Lemus and White, 2017) and small grains such as cereal rye (*Secale cereale* L.) oat (*Avena sativa* L.), and wheat (*Triticum aestivum* L.), can be grown during Mississippi's mild winters (Snapp et al., 2005). Clovers that grows in the spring and early summer, have the benefit of fixing atmospheric nitrogen and increasing the nutritive value of subsequent crop (Snapp et al., 2005). Rye is often established in the fall as a cover crop after a grain or silage harvest. Harvesting rye, the following spring as hay or silage has the added benefit of supplying a high-quality feed and is more helpful to soil health by preventing erosion and improving soil structure and drainage, than leaving the field uncultivated during the winter periods (Weerasekara et al., 2017). Snapp et al. (2005) reported that small grain cover crops produced greater biomass and are useful for building soil organic matter. They also indicated that brassica/cereal mixtures or legumes/cereal might be better suited over a wide range of ecological niches. Franzluebbers and Stuedemann (2015) conducted a 7-year study with grazed and non-grazed cover crops in a tilled and no-tillage system. They reported that cover crops grazed did not affect the C and N active soil fractions. They also indicated that cover crop in the conventional tillage system provides significant C and N pool to the soil similar to that of the no-till system. Similar findings were presented by Sarrantonio (2007), who concluded that cover crops could contribute to increased C sequestration and improved soil quality.

Excessive application of fertilizers in agricultural fields is cause for concern in both developed and developing countries because of its impact on the soil environment and water quality. In contrast, there are situations where the over-extraction of nutrients has resulted in reduced crop yields (Turral et al., 2012). Crop diversity offers farmers potentially greater productivity and profits, especially in the long term, by helping farmers spend less money on synthetic fertilizers and pesticides that can potentially pollute waterways, and on fuel used during application (Pfiffner and Balmer, 2011). Although the many benefits obtained from using cover crop mixtures, there is still a growing concern in terms of seed costs, as well as higher water demand and difficulty in the establishment, management, and termination (Smith et al., 2014; Wortman et al., 2012). Therefore, for sustainable crop production, it is essential to evaluate the performance of different cover crops, whether pure stands or different diverse mixes.

The benefits of growing cover crops are many and varied and are beneficial to crop production systems, but there are few reports of traditional cover crop use in perennial pastures systems under different harvest management. Research is needed to determine if benefits can be accrued from growing winter cover crops on perennial warm-season pastures and hayfields. Many of the species used as cover crops are already traditionally used as cool-season forages. Thus, it is necessary to explore different harvest management to determine how they will function as cover crops in winter forage systems.

Using diverse mixtures of plant species will increase the biodiversity of forage crops in the south-central region and enhance interspersed forage crop fields. Rangeland and permanent pasture production based on an ecological process with reduced dependence on chemical inputs can improve the sustainability of regions and overall agriculture. We can be confident that such a system can contribute to biological N fixation, soil health, plant diversity, leading to an increase in forage biomass production. Springer (1997) reported that inter-seeding cool-season forage legumes into bermudagrass [*Cynodon dactylon* (L.) Pers.] extended the grazing season and increased forage quality and quantity. They indicated that there was a linear response in percentage legume ground cover with increasing bermudagrass height.

There is evidence that overseeding common bermudagrass with berseem clover or annual ryegrass can improve the yield of subsequent summer hay (Mclaughlin et al. 2005). In this study, the authors overseeded a diverse mixture of cool-season annual grasses, legumes, and brassica species into bermudagrass sod and quantified cover crop forage mass harvested at different frequencies and subsequent effects on summer hay production. It has been documented that plant species diversity in forage crop production has not been well explored (Sanderson et al., 2004). Additionally, the effect of harvest management requires study to determine an optimum forage production, forage quality, and beneficial effects of cover crop balance. While more frequent harvest would typically provide better quality forage, it may reduce forage mass production because it stands may thin, and weeds may invade (Putnam et al., 2000). Longer regrowth intervals may increase forage mass, but the quality of forage reduces with maturity.

In our study, the null hypothesis is that various mixtures of cover crop species and harvest frequencies will not have an effect on cover crop forage mass or subsequent summer hay production. The objectives of this study were to quantify the forage mass, nutritive value, and botanical composition of various combinations of diverse cover crop mixtures (CC) harvested at three different defoliation frequencies (DF) consisting of 4- or 8-wk harvest intervals, or cut and left as a mulch at the end of the winter growing season and (2) assess the effects of these mixtures × harvest management combinations on subsequent hay production.

CHAPTER II

LITERATURE REVIEW

Importance of Cover Crop Mixtures in Agriculture

Adopting methods that depend primarily on inputs that are renewable to maintain the current level of crop productivity, remains the chief goal of sustainable agriculture in the USA (Tittensor et al., 2014). One potential strategy of achieving this goal is through diverse mixtures of cover crops. Multispecies cover crops that have been recently gaining popularity in the United States due to its soil amending benefits and the ability to provide other ecosystem services such as erosion control, forage for animal feedstuff, and act as water filter (Clark 2007). Based on the national survey done on cover crop users, there was a 38% increase in farmers who have adopted the use of multi-species between 2012 and 2016 (CTIC and SARE, 2013; CTIC, 2017). Cover crops systems offer farmers potentially greater productivity and profits, especially in the long term, by helping farmers spend less money on synthetic fertilizers and pesticides that can potentially pollute waterways, and on fuel used during application (Pfiffner and Balmer, 2011). Moreover, cover crop practices will assist in reducing greenhouse gases that result in global climate change (Kaye and Quemada, 2017). The advantages of cover crop management might take several years to develop, however, depending on the crop management practices and the goal of the farm enterprise (Mapfumo et al., 2002).

There are different species and mixes that can be used to achieve cover crop goals in the southcentral United States. Some of these adapted species include grasses such as annual ryegrass, oat, wheat, and cereal rye, also legumes such as white clover (*Trifolium repens* L.), crimson clover (*T. incarnatum* L), ball clover (*T. nigrescens* L.), and red clover (*T. pratense* L), and brassicas such as radish *(Raphanus raphanistrum subsp. Sativus* L.), rape (*Brassica napus* L.), and turnip (*B. rapa* L.). Small grains such as oat, wheat, cereal rye, and grasses such as annual ryegrass grow actively during Mississippi mild winters. At the same time, cool-season legumes are more productive during the spring and early summer, making forage available year-round (Snapp et al., 2005).

Forage legumes are quite popular in many grassland farming areas around the world, primarily because of their ability to fix atmospheric N_2 biologically and because of their high protein concentration and digestibility (Iglesias and Lloveras, 1998; Rochon et al., 2004). By improving the N status of soils, legumes contribute N without the risk of N leaching to the soil system associated with using inorganic fertilizers (Sharma and Bali, 2017). In addition to reducing N inputs costs and risk of N leaching at the farm level, another agronomic advantage is a better distribution of annual herbage production (Ryan et al., 2011). Grasses and small grains as cover crops help to reduce soil erosion, enhances weed suppression, and contribute to the improvement of soil organic matter. For example, rye has the potential of scavenging residual inorganic N from the previous crop, aids in reducing soil erosion and compaction, and suppressing weed emergence (Dabney et al., 2001). Brassicas grows rapidly and produces abundant biomass, which accounts for effective soil erosion control. Also, some have robust root systems that can help to reverse soil compaction and increase infiltration, and their high glucosinolate content makes them resistant against some fungi, soil-borne diseases, weeds, and nematodes (Alcántara et al., 2009).

Cover crops mixtures have the potential to improve soil organic matter by enhancing soil fertility and productivity (Wilhelm, 2004). Cover crops increase soil organic matter by decomposing their residues, which then enhances soil physical, chemical, and biological properties and, consequently, crop yields (Finney and Kaye, 2017). Soil organic matter aids in stabilizing soil aggregates, making the soil easier to cultivate. It also increases aeration and increases soil water holding and buffering capacities, and once it is broken down by soil microbe, it releases available nutrients to plants (Carter, 2002). Generally, cover crop mixtures contribute positively to agricultural soils by improving organic matter soil fertility, soil tilth crop production, and overall soil sustainability (Fageria et al., 2005).

Agricultural systems based on plant monocultures and chemical inputs have decreased the effectiveness of soil microbial populations, increasing the susceptibility of plants to biotic and abiotic stresses (Perez-Jaramillo et al., 2016). Both plant and microbial ecology research efforts are currently contributing to a new understanding of biodiversity's role in upholding ecosystem processes and services in natural and agricultural systems (Rodríguez-Ortega et al., 2014). Differences in functional traits and ecological niches allow multiple species to contribute to ecosystem functioning both above-ground and below-ground with contributions of microbial communities suggested to be critical variables for delivery of essential ecosystem services for human societies from both natural and agricultural systems (Rodríguez-Ortega et al., 2014). Complex interactions between plants and soils mediated by soil microbial communities supply unrealized opportunities for using ecological processes for ecosystem management. Direct effects of plant diversity are related to microbial community structure and diversity in the rhizosphere, which contributes to plant community effects on soil health (Kowalchuk et al., 2002).

Diverse cover crop mixtures have the benefit of recycling large amounts of nutrients while preventing their losses, reducing N leaching, and improving the yield of the following crop (Thorup-Kristensen et al., 2003). Barel et al. (2018) indicated that differentiation in a niche allows mixtures to be more productive than monocultures. Additionally, cover crop mixtures help in the build-up of soil C and N stocks and reduce pest pressure, in so doing, forming potential feedback to subsequent plant growth. Recent studies showed that the benefits of mixed cover crops are associated with the use of legumes and non-leguminous species (Hunter et al., 2019). It has been reported that these mixtures simultaneously reduce nitrate leaching and fix N from the atmosphere (Tribouillois et al., 2015). According to Tosti et al. (2014), the C: N ratio of a barley-vetch mixture allowed for faster mineralization compared to that of barley alone as a component crop. Cover crop mixtures may also help to reduce the deleterious effects of global warming by increased sequestration of atmospheric CO_2 and N (Sainju et al., 2002).

Extensive research in natural ecosystems indicates that increased plant diversity enhances biomass production (Cardinale et al., 2011) and has a positive effect on weed suppression (Mirsky et al., 2013). Diverse cover crop communities are more weed suppressive since less resource is left available to support the establishment and growth of weedy plants. Compared to monocultures, they may result in broader allelopathic activities toward the weedy species or other soil environment modifications that enhance weed suppression (Liebman and Davis, 2000). Bybee-Finley et al. (2017) showed that an increase in species number led to increased biomass, which was positively correlated to weed suppression, decreased N leaching, and increased biomass N accumulation. Wortman et al. (2012) conducted a study that looked at land equivalent ratio (LER) and stability indices for multi-species mixtures of legumes and brassicas cover crops. Results showed that the diverse combinations of cover crops were more productive than the component species grown as monocultures.

Some producers may forego grazing opportunities with the use of cover crop mixtures. Grazing land for cattle is a valuable asset, and forage needs lead some producers to graze their herds on winter crop crops such as winter wheat. Many cover crops are highly palatable and can increase the performance of livestock (Singer and Meek, 2004). Other advantages of using cover crops in the forage system include adding nutrients through manure from grazing livestock, adjust C: N ratios, and they can also serve as a critical component of integrated crop-livestock systems by offering high-quality forage during the winter periods (Delgado and Gantzer, 2015). This review is related to cover crop mixtures and harvest management effect on winter cover crop production and that of subsequent summer hay fields. Therefore, this review will focus on and discuss information related to cover crop mixtures such as the benefits of increasing forage mass and weed suppression, its impact on forage quality, and the effects of different harvesting intervals on cover crop mixtures.

Biomass Production of Cover Crops

Multi-species cover crop mixtures is one a strategy of increasing biomass production (Smith et al., 2014). Research in natural ecosystems demonstrated that an increase in plant diversity leads to greater biomass production (Cardinale et al., 2011). One way of optimizing the service provided by cover crop mixtures is by maximizing biomass production, and this is achieved when mixtures exhibit over yielding or transgressive overyielding (Schmid et al., 2008; Wang et al., 2012). When biomass production is greater than that of the average monoculture of the species contained in the mixture, it is referred to overyielding (Schmid et al., 2008), and when the productivity in a mix is larger than the maximal productivity of the component species, it is called transgressive overyielding. Hence, transgressive overyielding should be the primary goal relating to cover crop species (Gravel et al., 2012). Sturludóttir et al. (2014) investigated whether mixtures of grasses and legumes would have greater herbage yield compared to monocultures. Species used were red clover, white clover, smooth meadow grass (*Poa pratensis* L.), and timothy (*Phleum pratense* L.), grown in monoculture and eleven mixtures with systematically varying amounts of the four species. They reported a positive diversity effect, which led to greater herbage production in mixtures than from monocultures. Mixtures were 9 to 15% more productive than most monocultures, which occurred through transgressive overyielding. This result was consistent across three harvest years and was consistent for all sites.

Biomass Production Under Different Environmental Conditions

Different forage species have different resource requirements and physiological efficiencies. Due to these factors, different species will thrive and fail under different conditions (Griffin et al., 2009). In a study of early-planted (late August) or late-planted (late September) rye and annual ryegrass either in monocultures or in mixtures with crimson clover, and with other treatments that included monocultures of crimson clover and a mixture consisting of wheat-hairy vetch (Vicia villosa Roth.), Odhiambo and Bomkeb (2001) reported that early planting increased dry matter accumulation by 26 to 269% during the spring growth period. Dry matter accumulation ranged between 0.6 Mg ha^{-1} for clover and 10 Mg ha^{-1} for wheat, wheat–clover, and wheat–vetch treatments. Late planted cover crops produced 15 to 75% less biomass.

At final spring sampling, N accumulation was 3 to 74 kg ha^{-1} for early-planted compared to 3 to 47 kg ha^{-1} for late-planted crops.

Plant Competition and Botanical Composition in Cover Crop Mixtures

Plant competition is one of the chief disadvantages of cover crop mixtures if species are not carefully selected. Plant competition occurs between plants for limited resources such as light water, nutrients, and space (Malézieux et al., 2009). Competition among plants has a major effect on the species composition and longevity in cover crop mixtures. In pure stand cover crops, there is only intraspecific competition, i.e., competition between plants of the same species while in cover crop mixtures interspecific competition occurs, which is the competition of different species of plant with each other. Generally, intraspecific competition is more intense than interspecific competition, since two plants require the same resources, and is more dependent on the density of the plant (Anderson, 2015). During the early stages of plants, there is little no competition as there is plenty of space, water nutrients, and light. But as plants begin to develop and expand, other plants become shaded, and thus competition begins, which leads to the most competitive plants out-competing those plants that are less competitive, and these may not persist. Interspecific competition is essential in understanding how pasture mixtures establish and, consequently, the persistence of certain sown species. Even when legumes may have established earlier, grasses such as annual ryegrass have rapid germination, growth, and canopy development and will out-compete clovers for light and space, resulting in cover crop mixtures that are dominated by annual ryegrass with low clover proportions (Murray, 2012). Hence it is crucial to consider the compatibility of cover crop species before sowing them in combination (Anderson, 2015).

Substituting N fertilizer by exploiting symbiotic N_2 fixation in agricultural grasslands is considered as an important contribution to resource-efficient and sustainable agricultural systems (Gruber and Galloway, 2008). Symbiotically fixed N_2 in legumes ranged from 100 to 380 kg N ha^{-1} $year^{-1}$, but large amounts of more than 500 kg of N ha^{-1} $year^{-1}$ are also reported (Carlsson and Huss-Danell, 2003; Zanetti et al., 1997). In mixed grass-legume systems, N amounts of 10–75 kg of N ha^{-1} $year^{-1}$ may additionally be transferred from legumes to grasses (Nyfeler et al., 2011). For example, Nyfeler et al. (2011) reported that N from symbiosis from grass-clover mixtures containing orchardgrass (*Dactylis glomerata* L), perennial ryegrass (*Lolium perenne*), red clover, and white clover was increased in combinations with 60-80% and 40-60 % clovers as compared to pure stands clovers. The stimulatory effect is a typical example of a sink and source model of the regulation of symbiotic N_2 fixation. Symbiotic fixation was high in grass-dominated swards where low mineral N was available to clovers since the competitive grass component utilized most of the mineral N that was available in the soil, while there was minimal N uptake by clovers (Nyfeler et al., 2011). In pure clover stands, however, the activity of symbiotic N_2 fixation was down-regulated due to clover having adequate access to mineral N sources because of no grass abundance. The sensitivity of legume plants to regulate their percentage of N derived from symbiosis shows interspecific differences (Rasmussen et al., 2012). Overall, forage legumes grown in mixtures with grasses obtain most of their N (>80%) from symbiotic N_2 fixation (Oberson et al., 2013), which indicates that the amount of N derived from symbiosis usually depends on the dry matter production of the forage legume (Unkovich et al., 2010; Lüscher et al., 2011).

Cover crop mixtures may not always have adverse effects on each other, and this occurs when species use complementary resources without competing for the same resources, allowing for a more efficient resource capture of the mixture when compared to component crops (Brooker et al., 2016). For example, complementarity in the use of N sources has been documented primarily in associations of legume and non-legume species (Bedoussac and Justes, 2010; Cong et al., 2015; Li et al., 2016). In these associations, legumes increase their reliance on atmospheric N as the non-legume species are more competitive for soil N. Hauggaard-Nielsen et al. (2001) showed that complementarity could also occur within the soil profile, between species exhibiting complementary rooting depths, such as pea and barley, leading to a better soil exploration. For example, Maltais-Landry (2015) reported that cereals in mixtures with legumes had greater biomass and P and N uptake than as monoculture crops. Facilitation in soil nutrient uptake can result from mechanisms such as increased resource availability (Zhang and Li, 2003; Li et al., 2014), or a reduction in disease and pest attacks (Hauggaard-Nielsen et al., 2008).

Wortman et al. (2012) studied mixtures vs. single species cover crops to determine the level of productivity and stability each group provides and identify those components most active in contributing or detracting from mixture productivity. Using mixtures containing legumes or brassicas into groups of two, four, six, and eight species combinations, they found that brassica component crops were twice as productive (2428 kg ha^{-1}) as component crops in the legumes (1216 kg ha^{-1}). Daniel et al. (1999) evaluated winter cover crops for biomass production, ground cover, and aboveground N assimilation. Treatments were crimson clover, white lupin (*Lupinus albus* L.), rye, hairy vetch, and wheat in mixtures and pure stands. Results showed that monoculture rye produced more biomass (3048 kg ha^{-1}) than hairy vetch + rye (2601 kg ha^{-1}) crimson clover (2444 kg ha^{-1}), wheat, (2426 kg ha^{-1}), hairy vetch (1752.8 kg ha^{-1}), and white lupin (946.4 kg ha^{-1}). Crimson clover had the greatest N assimilation for legumes, and rye had the greatest for small grain at 78 kg ha^{-1} and 74 kg ha^{-1}, respectively. The authors indicated that N from the legume cover crops came from residual soil N ($NO_3 + NH_4$), and N_2 fixed by rhizobia associated with the legume. Nitrogen in non-legume cover crops comes from residual soil N supplies. Picasso et al. (2008) reported that well-adapted monocultures produced high biomass regardless of species richness in diverse mixtures. Similarly, Odhiambo and Bomkeb (2001) said that monoculture rye produced more biomass than mixtures with crimson clover. To improve biomass production, a better understanding of how species interact is vital (Brooker et al., 2016; Yu et al., 2015).

Weed Suppression of Cover Crop Mixtures

Weed control is another benefit of using cover crop mixtures. Cover crops compete with weeds for space, water, nutrients, and sunlight. Weeds are primarily affected by the species of cover crops, the management, and the weed community composition (Bàrberi and Mazzoncini, 2001). Some cover crop species naturally release allelopathic chemicals that suppress weed growth. Control of weeds is usually best from dense cover crops and when they are allowed to grow for the longest possible time. Cover crops such as rye and other high-biomass forage crops are good options that create an environment that steals growth requirements from weeds (Clark, 2007).

Weed prevention through competition, physical, and allelopathic effects are usually better when species from the Brassicaceae and Graminaceae families are sown together with legume cover crops (Blum et al., 2011). Interference by cover crops and their residue is mostly caused by sequestration of soil nutrients (especially N), allelochemicals being released, and modifications in the soil environment (Gallandt et al., 1999). For example, rye possesses great N uptake ability and also the potential to release allelopathic compounds. Kale (*Brassica* spp.), mustard (*Sinapis spp*.), and arugula (*Eruca spp*.) contain glucosinolates and have high allelopathic potential. In legumes such as crimson clover and subterranean clover, the weed-suppressive ability is usually less due to stimulatory effect on weed emergence of N released from cover crop residues, especially when they are ploughed down (Barberi, 2002). Teasdale and Mohler (2000) claimed that when cover crops are left as a surface mulch to decompose, suppression of weeds is mostly from physical effects rather than nutrients or allelochemicals. A study by Creamer et al. (1996) indicated that rye, crimson clover, hairy vetch, barley, along with mixtures, suppressed the emergence of eastern black nightshade (*Solanum ptychanthum Dun*.). They attributed crimson clover inhibition of eastern black nightshade emergence to physical suppression alone.

Living cover crop mulch is better at suppressing weed at all stages of the weed life cycle compared to surface mulch. Living mulch absorbs red light and will reduce the red; far red ratio sufficiently to inhibit phytochrome mediated seed germination, whereas cover crop residue has a minimal effect on this ratio (Teasfale and Daughtry, 1993). Living mulch competes with emerging and growing weeds for essential resources and inhibits weed seed production (Brennan and Smith, 2005). Studies by Davis and Liebman, (2003) and Gallandt et al. (2005) reported greater weed seed predation at the soil surface was greater with living cover crop vegetation, suggesting a role for living cover crops in enhancing weed seed mortality. This suggestion by Gallandt et al. (2005) was supported by results obtained from a study conducted by Reddy and Koger (2004), who indicated that cover crop surface mulch does not suppress weeds as consistently as live cover crops do. Generally, living mulch suppressed weeds more completely at more phases of the weed cycle compared to cover crop surface mulch.

Cover crop mixtures increase forage biomass that creates a mat that minimizes rainfall erosion, helps maintain constant soil temperature, and decreases weed emergence (Morton et al., 2006). Ch et al. (2016) found that mustard, fodder radish (*Raphanus sativus var. niger* J. Kern), and spring vetch (*Vicia sativa* L.) suppressed weeds by 60% and cover crop mixtures-controlled weeds by 66% during the fallow period across 3 yr. Holmes et al. (2017) assessed the productivity and weed suppressive capacity of 12 cover crop species (both cool and warm season) planted as sole crops and in mixtures. They reported that mustard and oat were among the most productive and were better at suppressing weeds. For the warm season cover crops, sudangrass and buckwheat (*Fagopyrum sagittatum* Moench) were among the most prolific and weed suppressive. Treatments that excluded mustard from mixture resulted in increased weed biomass. Forage radish produced little biomass but reduced weed biomass by 45 to 100%. Overall these studies showed that increases in cover crop mixtures diversity were correlated with increased weed suppression and biomass stability (Florence et al., 2019).

Weed control by cover crop mixtures can allow a producer to forego one to two herbicide treatments on the subsequent crop (Morton et al., 2006), but effects will not last the entire growing season of the crop (Lu et al., 2000). Nevertheless, cover crop weed control has the potential for subsequent crop production savings (Lu et al., 2000). Teasdale and Mohler (2000) found that increasing levels of biomass nearly eliminated light extinction resulted in exponentially decreasing rates of weed emergence. Reddy (2003) found that a rye cover crop in Mississippi reduced total weed density 9 to 27%, and total weed biomass 19 to 38% across different tillage systems. While this decrease in weed percent may not eliminate the need for an herbicide, it could lower the total costs spent on the herbicide. Herbicide savings will depend on the crop planted, the type of cover crop selected, and the management of cover-crop biomass production (SARE, 2013).

Performance of Subsequent Crops

Cover crop mixtures play a vital role in combating many agricultural issues (Fageria et al., 2005). Initially, they are used as soil amending tools, but they can provide temporary pasture and high-quality feed for livestock. Cover crops and perennial warm-season forages have the potential to improve the physical, chemical, and biological properties of the soil, and in so doing, improving the productivity of subsequent crops (Fageria et al., 2005). The increase in crop production varies for different crops and agroecological regions. Also, improvements in production are dependent on the management of cover crops as well as those of subsequent crops. For example, the subsequent cereal crop grown after a legume cover crop showed a positive response to N is due to the transfer of biologically fixed N and less immobilization of nitrate during the decomposition of legume residues (Fageria et al., 2005).

Creamer and Baldwin (2000) studied summer cover crops that included six legumes, two nonlegume broadleaf species, and five annual grasses and they found that biomass production from the legumes ranged from 1420 for velvet bean (*Mucuna pruriens*) to 4807 $kg \cdot ha^{-1}$ for sesbania and N content in the aboveground biomass ranged from 32 (velvet bean) to 97 $kg \cdot ha^{-1}$ (sesbania). Biomass for grasses varied from 3918 (*Echinochloa esculenta*) to 8792 $kg \cdot ha^{-1}$ (*Sorghum* × *drummondii*) and N content for the grasses ranged from 39 (*Echinochloa esculenta*) to 88 $kg \cdot ha^{-1}$ (*Sorghum* × *drummondii*). The C: N ratios were very high in the grasses and would require additional N application for subsequent crops to overcome N immobilization. Chu et al. (2017) reported that multispecies mixture of cool-season small grains, legumes, and brassica increased soybean (*Glycine max* L.) yield compared to monocultures or two-species mixtures of these crops.

Interseeding cool-season forage legumes into bermudagrass can extend the grazing season and increase forage quality. Mclaughlin et al. (2005) looked at extending the haying seasons by spring haying of overseeded cool-season annuals, including annual ryegrass, wheat, berseem clover (*Trifolium alexandrinum* L.), and crimson clover. They concluded that overseeding ‘Tifton 44’ bermudagrass with berseem clover increased the forage mass harvested of subsequent summer hay and suggested that the increase may have been due to increased N available fixed by the berseem clover. Sweeney and Moyer (1994) found that sorghum yields increased by 70 to 131% following cover crop mixtures consisting of winter legumes compared to no cover crops on eastern great plain soils. Blanco-Canqui et al. (2012) found that grain sorghum yields following sunn hemp (*Crotalaria juncea* L) were 1.18 to 1.54 greater that than yields harvested for grain sorghum in no-cover-crop plots. Several studies have revealed that the total N in the soil is primarily from the use of legume cover crops, which is the critical factor that improves subsequent crop yield (Lu et al., 2000; Snapp et al., 2005). Improvement of soil structure, breaking of pest and disease cycles, and allelopathic effects of crop residues have all been attributed in the yield response of subsequent crop (Fageria et al., 2008).

Cover Crops Effects on Forage Quality

The terms forage quality and forage nutritive value are often incorrectly used interchangeably. The quality of forage is best defined in terms of animal performance (daily gain, milk production, wool, etc.) when the animal potential is not limiting, forage availability is not limiting, and the forage is the sole source of energy and protein available to the animal (Mott and Moore, 1985) In practical terms, forage quality is the extent to which a forage has the potential to produce the desired animal response. Although forage quality and forage nutritive value are used interchangeably, forage nutritive value usually refers to the concentration of available energy and CP. Forage quality not only includes nutritive value but also forage intake (Ball et al., 2001). In species mixtures, forage quality is determined by maturity and variation (Eskandari et al., 2009). Maturity is considered the most important factor that determines forage quality. As the plant matures, their cell wall concentration increases, causing an accumulation of indigestible lignin, which results in decreased forage quality. Differences between grasses and legumes can be substantial. The CP concentration of legumes are usually greater than that of grasses, and the fiber of legumes tend to digest faster than grass fiber, allowing ruminant to eat more legumes. Including legumes into cool-season annual grasses has the potential for improving forage production and quality (Eskandari et al., 2009).

Legumes are known for high CP. Increased CP in the sward has long been recognized as one of the benefits when legume is included in forage mixtures (Eskandari et al., 2009). Compared to grasses, legumes typically produce less dry matter and are weak competitors against weeds (Corre-Hellou, 2011). Therefore, it seems that growing legumes as a sole crop is not ideal for forage production.

Lithourgidis et al. (2006) evaluated common vetch, triticale, and oat as monocultures and mixtures of common vetch with each of the cereals at two seeding ratios (55:45 and 65:35) and reported that all mixes had increased CP with an increased seeding rate of common vetch. They showed that monoculture vetch had greater CP (139.3 g kg^{-1}) than the 65:35 mixture of common vetch with oat (119.1 g kg^{-1}) and the two mixtures of common vetch with triticale (109.2 and 103g kg^{-1}, respectively). Triticale and oat monocultures had the least CP (63.2 and 78.4 g kg^{-1}, respectively). Lithourgidis et al. (2006) summarized that although the 65:35 mixture of common vetch-oat had lesser CP than monoculture common vetch, it produced more CP (1100 kg ha $^{-1}$) than all crops because of its forage mass. The CP per ha was the least for monoculture triticale (680 kg ha^{-1}) and the 55:45 mixture of common vetch-triticale (790 kg ha^{-1}).

The concentration of NDF and ADF are measures of forage nutritive value. As NDF increases, dry matter intake decreases (Arelovich et al., 2008). Lauriault et al. (2004) reported that intercropping winter pea with barley, wheat, triticale, or oat caused a decrease in NDF in all cereal + legume intercrops compared with the cereal monocultures. Ghanbari-Bonjar and Lee (2003) evaluated sole crops and mixtures of wheat and field bean and concluded that combinations led to an increased forage nutritive value compared with the wheat monoculture. Contreras-Gova et al. (2006) reported that NDF and ADF concentration improved with wheat-clover blends compared to the component wheat crop. Sleugh et al. (2000) found that NDF concentration decreased by 30% in Kura clover-wheatgrass mixtures compared to wheatgrass alone. Kantar et al. (2011) reported that the forage mass of three winter rye cultivars increased, but CP, NDF digestibility, and digestible dry matter decreased, and NDF increased with increasing maturity.

Harvest Management Effect on Cover Crop Mixtures

Defoliation has manifold consequences on plant growth and allocation (Ferraro and Oesterheld, 2002). Losing aboveground biomass of forage means losing photosynthetic tissues, which results in loss of carbon and nutrients. The loss of photosynthetic tissues may often lead to reduced forage biomass, but in some cases, it can be positive. Such a response is called compensatory regrowth because the defoliated plants partially or completely compensate for the removal of forage biomass. This compensatory response is associated with nutrient levels, flexible carbon allocation, light environment, recovery conditions, and evolutionary conditions. Defoliation may also have an effect on root growth and below ground carbohydrate reserves, decreasing root biomass, and belowground relative growth rate (Ferraro and Oesterheld, 2002). The impact of defoliation on forage growth was studied extensively, and the magnitude and generality of compensatory growth responses have received extensive discussion. The analyses of the evidence have so far been of a qualitative nature (Ferraro and Oesterheld, 2002).

Studies have shown that clover growth, morphology, and forage biomass respond to the severity of a cutting regime (Simon et al., 2004). Intensity (cutting height) and the frequency of defoliation are the two main parameters that describe the severity of clipping (Herbert et al., 2018), and both parameters can have an effect on clover persistence in pastures (Nolan et al., 2001). These two parameters are known to determine morphological characteristics, including residual leaf area, a number of growing points, and the size of storage organs, including root and stolon (Avice et al., 2001). Also, defoliation severity reduces C assimilation as a consequence of leaf removal (Briske and Richards, 1995). Under such conditions, photosynthesis is not sufficient to provide assimilates needed for the development of the regrowing organs and to sustain the energy demand required for N_2 fixation and soil-N assimilation (Gordon et al., 1990). Teixeira (2007) conducted a study on the effect of grazing frequency (38- vs. 28-d regrowth intervals between grazing events) on the shoot mass and accumulation of C and N reserve in alfalfa. Annual shoot dry matter at the long regrowth interval averaged 23t ha^{-1}, which was 5 to 60% greater than at the shorter regrowth interval. Also, the short regrowth intervals caused a reduction in the accumulation of crown and taproot. However, overall, C and N reserve status fluctuated more due to seasonal patterns of accumulation and depletion than to defoliation management.

Ferraro and Oesterheld (2002) reported that the defoliation of grasses resulted in reduced plant growth and substantial variability in the responses of different plant components. There was a greater negative effect on aboveground biomass production but less of an effect on root biomass. Defoliation frequency and time for recovery from last defoliation both functioned in these responses measured. Those studies with frequent defoliation and short recovery intervals showed more negative effects. Contrastingly, the amount of canopy removed (i.e., defoliation intensity) did not have much effect on the response to defoliation. The result of these studies indicated that the magnitude of defoliation response for different plants might vary, and compensatory responses, modulated by factors such as recovery time after defoliation and nutrient availability, are a norm (Ferraro and Oesterheld, 2002). In summary, the findings from these studies showed that longer harvest intervals allow for maximum forage production but reduce the quality of the forage. Hence a balance between forage quality and quantity is necessary (Ball et al., 2001).

CHAPTER III

FORAGE MASS, BOTANICAL COMPOSITION AND NUTRITIVE VALUE OF COVER CROP MIXTURES UNDER DIFFERENT DEFOLIATION FREQUENCIES

Abstract

Overseeded winter cover crops (CC) can contribute to forage production in perennial pasture systems, but the diversity of CC mixtures and defoliation frequency (DF) may have effects on sward productivity. This 2-yr experiment conducted at Raymond, MS, quantified the forage mass (FM), nutritive value, and botanical composition of winter CC. Treatments were factorial combinations of 10 CC (eight CC mixtures using several species of grasses, legumes, and brassicas and two non-overseeded controls) and three DF (harvested every 4 or 8-wk or cut and left as mulch) in a split-plot arrangement of a randomized complete block design experiment with three replications. Mixtures with legumes and those harvested at the 4-wk DF generally had greater FM and crude protein levels and lesser ADF and NDF concentrations. Cover crop diversity can improve forage production, and legumes included in mixes can improve forage quality.

Introduction

Perennial warm-season grass pastures of the southeastern USA are intensively managed, typically involving repeated use of nitrogen fertilizer and chemical control of weeds throughout the year. Environmental sustainability and productivity of these pastures and rangelands can be enhanced by overseeding with mixtures of multiple plant species that can improve herbage mass, soil health, soil fertility, plant biodiversity, and plant population stability (Scherr and McNeely, 2007). Cool-season annuals used as cover crops have contributed substantially to forage production in the United States because of its biomass production and high nutritive value (Burns and Fisher, 2010). Biomass production and nutritive value may differ, however, due to different species of plants (Hunter et al., 2019), soil type, and also with a variation of the weather (Abdin et al., 1997).

Ecosystem studies have concluded that particular species, when mixed, are complementary in their pattern of resource use and can increase crop productivity and nutrient retention (Hooper et al., 2005). However, plant species diversity in forage crop production has not been well explored (Sanderson et al., 2004). Major benefits of forage legumes include the contribution of nitrogen through N_2 fixation and high-quality forage production (Nelson and Burns, 2006). Non-leguminous forages such as annual ryegrass, small grains, and brassicas produce large amounts of biomass. Annual ryegrass is a popularly used forage because it establishes quickly, produces abundantly in a short time, has a high nutritive value, and is adaptable to a wide range of soil types (Clark, 2007). Small grains such as oats perform well with other small grains, clovers, vetch, and peas. Brassicas are also known for their rapid fall growth, high biomass production (Clark 2007). Because of its accelerated growth, brassica forages can extend the grazing season late fall into early winter, providing substantial amounts of digestible nutrients to livestock (Reid et al., 1994). Utilizing complex mixtures of these species as cover crops in dormant sod of the widespread warm-season perennial grass crops of the region has the potential to transform forage crop production from chemical input-based systems to ecologically driven production. This is associated with reduced fertilizer requirements through improved soil fertility and health, reduced competition from weeds through complementary crop resource use, and enhanced sustainability of both the areas of forage crop production and the associated environment.

Plant species play a substantial role in the nutritive value of the sward. In diverse mixtures, variation in the nutritive value occurs with greater species diversity due to the differences in chemical composition and differences in the stages of maturity in the plant community (Bruinenberg et al., 2002; Huyghe et al., 2008). Protein and digestibility are greatest when plants are in the early stages of growth, but decreases as the plants mature (Tzialla et al. 2000; Ammar et al., 2004; Mountousis, 2008; Hejcman et al., 2010). This decline in forage nutritive value is more distinct and rapid in warm-season perennial grasses, especially in plants that are older than 35 to 40 days (Newman et al., 2006). Therefore, harvesting or grazing at the right stage is necessary to achieve high-quality forage (Koutsoukis et al., 2017). While more frequent harvest would typically provide better quality forage, it may reduce forage mass production because it stands may thin, and weeds may invade (Putnam et al., 2000). Conversely, longer regrowth intervals may increase forage mass, but the nutritive value of forage reduces with maturity. Hence, the effect of harvest management requires study to determine an optimum forage production, forage quality, and beneficial effects of cover crop balance. This study quantified the forage mass, nutritive value, and botanical composition of various combinations of diverse cover crop mixtures (CC) harvested at three different defoliation frequencies (DF) consisting of 4- or 8-wk harvest intervals or cut and left as a mulch at the end of the winter growing season.

Materials and Methods

Study location, soil, and weather description

This study was conducted at the Brown Loam Branch Experiment Station at Raymond, MS (32°12′ N, 90°30′ W) from 2017 to 2019. The predominant soil type at the experimental site is a Loring silt loam (fine-silty, mixed, active, thermic Oxyaquic Fragiudalfs), moderately well-drained with a fragipan and slopes ranging from 2 to 5%. Compared with the 30-yr normal monthly precipitation from October to January, May, and July was less, but in February, there was more than double the amount of rainfall during the 2017–2018 growing season (Table 3.1). In the 2018–2019 growing season, precipitation was much less than the 30-year average in October, December, and February. In should be noted that in both years, winter-spring monthly rainfall represented extremes of dry or wet conditions during the growing period.

Table 3.1 Monthly accumulated rainfall and mean air temperature at Brown Loam Experiment Station, Raymond, MS from October to September of 2017 to 2018 and 2018 to 2019.

	Monthly rainfall			Average monthly air temperature		
Month	2017-2018	2018-2019	30-yr avg.†	2017-2018	2018-2019	30-yr avg. †
	mm			°C		
Oct.	73.9	34.5	99.6	19.9	20.2	18.4
Nov.	18.5	196.6	120.9	15.3	10.9	13.2
Dec.	92.7	218.7	130.8	9.3	9.8	8.8
Jan.	102.4	205.7	126.2	5.5	8.4	7.6
Feb.	259.6	84.8	120.9	14.6	13.2	9.7
Mar.	162.3	101.9	128.0	15.0	13.1	13.8
Apr.	148.8	190.8	126.0	15.6	17.4	17.8
May	96.8	175.3	111.3	25.0	24.0	22.4
Jun.	103.6	134.1	104.6	27.6	25.7	26.1
Jul.	95.0	149.1	122.2	28.3	27.3	27.6
Aug.	103.9	126.0	107.7	27.3	28.0	27.4
Sep.	167.4	0.5	77.0	26.7	28.0	24.2
Season						
Total	1424.9	1618.0	1375.2			
Average	118.7	134.8	114.6	19.2	18.8	18.1

†The 30-year climate normals are provided by the National Climatic Data Center of the National Weather Service for the latest three-decade averages of climate variables. These data are from the period 1981-2010 for Jackson, MS, available at https://www.weather.gov/jan/climatenormals1981-2010. Accessed 9 Dec. 2019.

Treatments

Experimental variables were 10 cover crop (CC) treatments and three defoliation frequencies (DF). The 10 CC treatments included overseeding with eight combinations of diverse forage species, consisting of monoculture functional groups of cool-season annual grasses, legumes, and brassicas, or various mixtures of these crops, and two non-overseeded controls. Defoliation frequency treatments consisted of harvesting for forage at 4- or 8-wk harvest intervals or an end-of-season mulch (cut at the end of the cover crop growing season and left on the plot). This experiment was conducted using a split-plot arrangement of a randomized complete block design with three replications. The three DF treatments were assigned randomly as the main plot factor, and the subplot factor was CC treatments assigned randomly within each main plot. The subplots were 5 m × 1.5 m, separated by a 0.5-m alleyway to avoid the species that were specific to each treatment from spreading to adjacent subplots. There was a 2-m alleyway between main plots within blocks and a 3-m alleyway between blocks.

Grasses used in the study were 'Wrens Abruzzi' rye (*Secale cereale* L.), 'TAMO 606' oat (*Avena sativa* L.), 'EK 102' wheat (*Triticum aestivum* L.), and 'Marshall' annual ryegrass (*Lolium multiflorum* L.). Legumes used were 'Durana' white clover (*Trifolium repens* L.), 'Dixie' crimson clover (*T. incarnatum* L.), 'AU Don' ball clover (*T. nigrescens* L.*),* and 'AU Red Ace' red clover (*T. pratense* L.). Brassicas were 'Eco Till' radish (*Raphanus raphanistrum subsp. Sativus* L.), '*Brassica*' rape (*Brassica napus* L.), and 'Barkant' turnip (*Brassica rapa* L.). The eight overseeding treatment combinations were mixtures of 1) all 11 species of the three functional groups: grasses, legumes, and brassicas, 2) grass functional group, 3) legume functional group, 4) brassica functional group, 5) grasses and legumes, 6) grasses and brassica, 7) legumes and brassica, and 8) monoculture annual ryegrass. The two control treatments were none overseeded plots either with winter weeds controlled or no weed control. Weed control was done using glyphosate (Roundup Ultra Max) at 1.68 kg a.i. ha^{-1} applied in late January of both years.

Overseeded treatments were no-till seeded (using a Hege plot seeder, Kincaid Equipment Manufacturing, Haven, KS) in seven rows spaced 20 cm apart into existing bermudagrass [*Cynodon dactylon* (L.) Pers.] hayfields during mid-October each year. Seeding rates (for functional groups and multi-group mixtures, respectively) were: annual ryegrass, 34 and 22 kg ha^{-1}; rye, 100 and 56 kg ha^{-1}; oat, 100 and 67 kg ha^{-1}; wheat 100 and 67 kg ha^{-1}; white clover, 5.6 and 3.4 kg ha^{-1}, red clover 13.4 and 9.0 kg ha^{-1}; crimson clover, 16.8 and 13.4 kg ha^{-1}; ball clover, 5.6 and 3.4 kg ha^{-1}; turnip 3.4 kg ha^{-1}; rape, 4.5 kg ha^{-1}; and radish, 6.8 kg ha^{-1} (brassicas were seeded at the same rate regardless monoculture or mixture). The same treatment arrays were applied to the same plots every year. No fertilizer or herbicides were applied to the cover crops, but during the summer growing season, fertilizer was applied to plots similar to the adjacent hayfields. Because of management constraints, planned fertilizer application was not done in 2017. In 2018, 316 kg ha^{-1} of 19-19-19 (N-P-K) fertilizer was applied in early July and 60 kg N ha^{-1} as urea was applied in late August.

Sampling

For the cool-season diverse forage mixtures, forage mass harvested was done for each plot according to the treatment. The first harvest was April 11 in Year 1 and April 10 in Year 2, and the final harvest and mulch treatment were June 6 in Year 1 and June 12 (one week late because of rainy weather) in Year 2. For the 4- and 8-wk DF, the center 3 × 0.6 m of each overseeded subplot was harvested to a 5-cm residual stubble using a push mower with a bag attachment to catch the harvested forage. For the end-of-season mulch plots, a 1.0 × 0.6 m of each overseeded subplot was harvested using hand-held clippers. Non-overseeded control plots were not harvested. The total harvested material was weighed fresh, and an approximately 1-kg subsample was taken and dried at 55°C for 72 hours in a forced-air oven to determine dry matter (DM) concentration to calculate forage mass DM harvested. For botanical composition (BC) determinations, a hand-clipped sample was taken adjacent to the forage mass harvested strip. Depending on the species mixture in the treatment, samples were separated into the components of the functional groups, that is, grasses, legumes, and brassicas if they were present. All other plants, including species used in the study but not belonging to the particular treatment, were considered weeds. If the treatment did not include grass in the mixture, then volunteer annual ryegrass was considered as a weed. The separated samples were dried as for DM determinations and weighed. For each harvest, the proportion of the BC components was calculated after summing all the components of that sample and dividing each component by the total. To calculate the weighted BC averages for the season, the content of each BC component in each harvest was determined by multiplying the percent of each component by the forage mass harvested, then summing these contents for all harvests in the season, and finally, dividing the season total of each component by the total harvested mass for the season. Season-long BC data reported are the weighted averages. After harvesting and sampling were completed on the 4- and 8-wk DF, the rest of the subplot cut to a uniform 5-cm stubble height, and the forage was removed. After sampling the end-of-season mulch treatment, a lawnmower was used to cut the forage and leave it on the plot.

Nutritive value analysis

A second-hand clipped subsample harvested adjacent to the forage harvested strip and dried similarly to the DM concentration samples was used for the nutritive value analysis. Samples were analyzed for total N concentration, neutral detergent fiber (NDF) and acid detergent fiber (ADF). Total N was measured by rapid combustion using a LECO FP-528 Protein Analyzer (LECO Corp., St Joseph, MI), and crude protein (CP) was calculated as total N × 6.25. Concentrations of NDF and ADF were determined with an Ankom Model 200 fiber analyzer (Ankom Technology, Macedon, NY) using a sodium sulfite procedure (Robertson and

Van Soest, 1981). All laboratory analyses were done at the Louisiana State University's forage testing laboratory.

Statistical analysis

Analysis of variance was done by fitting mixed models using PROC MIXED and PROC GLIMMIX in SAS (SAS Institute Inc., Cary, NC). The best model fit was determined using the Schwarz' Bayesian Criterion. Whole plots were DF, and CC were subplots. Forage mass harvested for treatments with multiple harvests within a year were considered repeated measures. Means separation was done using the PDIFF option, and responses were considered different at the 0.05 probability level.

Results and Discussion

In the presentation and discussion of all results of this study, it should be noted that in both years, the brassica crops we seeded germinated but did not survive past the early seedling stage. We suspect that excessive rainfall that led to waterlogged conditions, along with record cold temperatures during the establishment of the crops may have resulted in the lack of survival of brassicas. Thus, the results must be considered in light of this. It should then be understood that mixtures with all functional groups are essentially grass-legume mixtures, and the grass-brassica and legume brassica mixtures are more or less pure all grass or all legumes stands but at a reduced seeding rate compared to the monoculture functional group treatments. Also, the brassica alone monoculture, in essence, became a third non-overseeded control.

It is known that the quantity and distribution of precipitation and air temperature can have an effect on forage production (Mouriño et al., 2003). During the growing season, record cold temperatures in January and the heavy rainfall in February (Table 1) may have restricted early-season forage growth of all crops generally and possibly killed all brassica plants that germinated. Singh et al. (2008) indicated that winter-grown canola (*Brassica napus* L.) production is limited mostly by frost and winter-kill in the southern canola-growing regions of the United States. Low temperature caused a reduction in germination rate and seedling emergence of *Brassica* species (Zheng et al., 1994). An inverse response was observed among the cultivars of brassica between freeze damage seed yield in an experiment conducted by Cebert and Rufina (2007). The lack of survival of brassica in our study suggests that brassica as a cover crop or as a forage crop in similar soil types are not likely to be successful in years with above-normal rainfall. In a study conducted on a Loring soil south of Woodville, MS, it was observed in a cool-season forage mixture seeded on perennial pastures with variations in topography, grasses dominated most of the landscape with isolated areas of legume dominance and only sparse and varied populations of brassica species (Bridges et al., 2019).

Forage mass

There was a CC × year interaction effect ($P = 0.04$) on annual forage mass of cool-season annuals (Table 3.2). In 2018, the grass alone functional group had greater forage mass (2880 kg ha^{-1}) than the legume-brassica mixture (2166 kg ha^{-1}). However, all other treatments were intermediate and not different from either of these treatments. The pattern of differences among CC was different in 2019, which partially is the cause of the interaction effect. The legume function group CC had greater forage mass than the grass functional group, and the grass-brassica (1400 kg ha^{-1}) and pure stand monoculture annual ryegrass (1559 kg ha^{-1}) had the least. All species, grass-legume, and legume brassica CC mixtures were intermediate but not statistically different between the legume and the grass alone functional groups, and the grass alone functional group was not different from the two treatments with the least forage mass.

Overall, all species mixtures, legume-grass mixtures, grass and legumes alone functional group perform exceptionally well in both years compared to the other cover crop mixtures. Our results were similar to that of Haughey et al. (2018), Hector et al. (2010), and Isbell et al. (2009), who showed a positive correlation between diversity and biomass production. Cover crops mixtures containing legumes benefited from the legume's ability to make use of atmospheric N_2 through symbiotic N_2 fixation, thus reducing their requirements for soil and fertilizer N (Carlson and Huss-Danell, 2003; Lüscher et al., 2014). Picasso et al. (2008) investigated species composition, species richness, and harvest management effects on crop and weed biomass in perennial herbaceous polycultures, using 49 combinations of seven species of legumes and C_3 and C_4 grasses, including all monocultures and selected two to six species polycultures. They reported that polycultures produced more biomass than monocultures by an average of 73%. Wendling et al., 2017 reported that biomass production from mixtures depended on the species involved and the competitiveness of the species involved. Several studies have reported the importance of functional differences between species for the positive outcome of mixture performance (Diaz et al., 2001). Differences in functional traits lead to the complementarity between species, especially for mixtures of legume and non-legume species (Creissen et al., 2016).

Table 3.2 Cover crop (CC) × year interaction effect on annual total forage mass of CC treatments across three defoliation frequencies (harvested at 4- or 8-wk intervals or cut and left as mulch at the end of the season) at the Brown Loam Branch Experiment Station, Raymond, MS.

	Year		
CC Mixture	2018	2019	*P*-value†
	------------ kg ha^{-1} ----------		
All species	2387 AB‡	2319 AB	0.827
Grass	2880 A	1858 BC	0.001
Legumes	2761 AB	2693 A	0.826
Grass-legume	2636 AB	2407 AB	0.460
Grass-brassica	2378 AB	1400 C	0.003
Legume-brassica	2166 B	2327 AB	0.602
Monoculture annual ryegrass	2299 AB	1559 C	0.019
SEM§	229.13	229.13	

†Probability values to compare CC means between years.
‡Within columns, CC means followed by the same upper-case letters are not different ($P > 0.05$).
§ Standard error of mean.

Monoculture annual ryegrass and the grass alone functional group performed well in 2018 but declined in 2019. Furey (2015) reported that monoculture annual ryegrass was the best for carbon fixation and produced biomass production similar to mixtures containing species from grass and legumes functional groups. In our study, brassicas germinated but did not grow, resulting in low forage biomass harvested. These results may be due to low temperatures and heavy rainfall that occurred in the early spring of both years.

There was also a DF × year interaction effect (*P* = 0.036) on annual cool-season forage mass (Fig. 3.1). Between years, both 4- and 8-wk DF had greater forage mass in 2018 than in 2019, but there was no difference with end-of-season mulch treatment. During Year 1, forage mass at the 8- (2615 kg ha^{-1}), and end-of-season mulch (2754 kg ha^{-1}) was greater than at the 4-wk DF (2121 kg ha^{-1}). In Year 2, the 4- (1672 kg ha^{-1}) and 8-wk DF (1741 kg ha^{-1}) had similar forage mass, but both were less than the end-of-season mulch forage mass (2827 kg ha^{-1}). Where forage is a priority, harvest management that achieves better forage quality (greater nutritive value) is necessary (Richner et al., 2014). Across both years, end-of-season mulch produced more forage biomass compared to 4- and 8-wk DF (Fig. 3.1). The mulch treatment can cause greater forage biomass due to benefits such as the build-up of soil organic matter and improved water and nutrient use efficiency (Alharbi, 2015). In our study, however, the greater forage mass may be mainly because this treatment was not defoliated during the growing season, allowing for maximum biomass production.

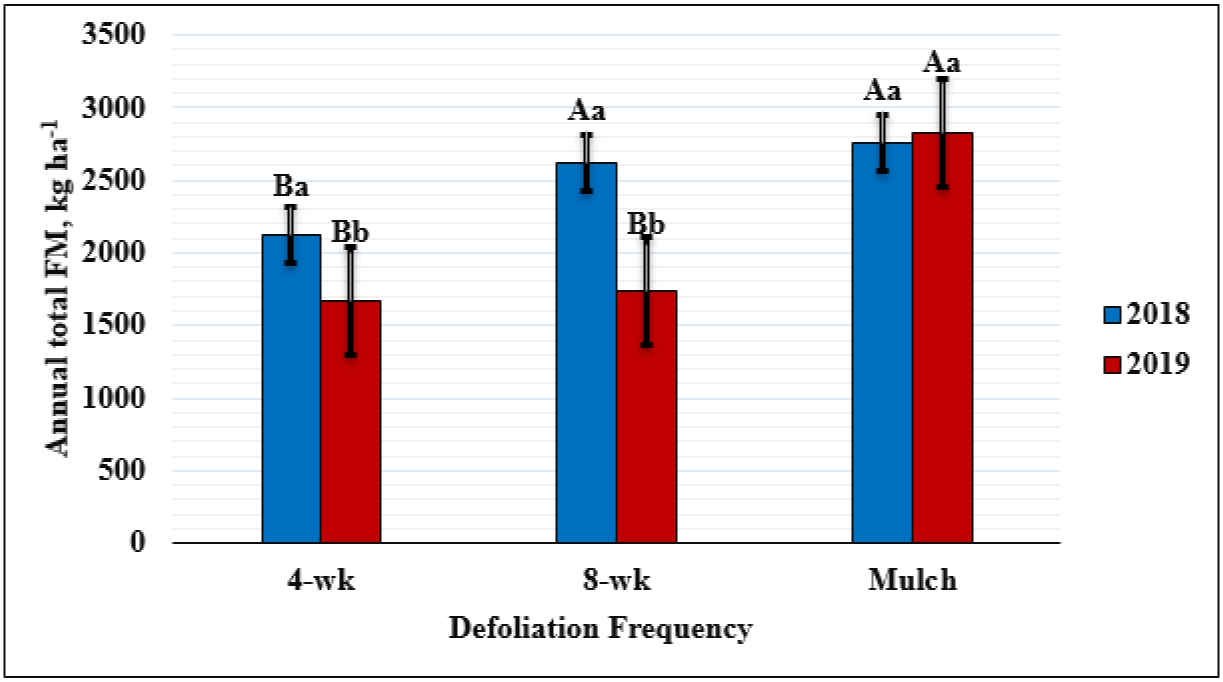

Figure 3.1 Defoliation frequency (DF) × year interaction effect on annual total forage mass (FM).

In Fig. 3.1, the annual total forage mass proportion is represented by blue bars in 2018 and red bars in 2019. Data are from across winter cover crops harvested at 4- or 8-wk intervals or cut and left as mulch at the end of the season at the Brown Loam Branch Experiment Station, Raymond, MS. Within years, means followed by different uppercase letters, and between years means followed by different lower-case letters are different ($P < 0.05$). These responses are discussed on Page 37.

Losing photosynthetic tissues may often lead to reduced forage biomass (Herbert et al., 2018), which may explain why the 4-wk DF had the least forage mass compare to compared to 8-wk and end-of-season mulch DF. Several studies have found that frequent harvesting during the growing season generally results in less forage mass harvested than infrequent harvests (Bittman et al. 1991; Foster et al., 2014). Foster et al. (2014) found that greater forage mass accumulated under two harvest cuts than three harvest cuts system in all years of a 4-yr study. Similarly, Belesky et al. (1999) reported that clipping of every 6-wk produced 26% more herbage than clipping at 3-wk harvest intervals. Rinne and Nykanen (2000) reported that the biomass of primary growth increased by 116 kg ha^{-1} d^{-1} by harvesting in the later stages of plant growth, which was partly compensated by a reciprocal effect in the regrowth.

Botanical composition

There was a DF × CC interaction effect ($P < 0.02$) on proportion grass in the total FM harvested of the five CC treatments that included grasses (Table 3.3). There were no differences in grass composition among CC at the 4-wk DF. At 8-wk DF, there were no differences in the grass composition among the all species mixture, grass-brassica mixture, and monoculture annual ryegrass CC, but the grass-legume (423 g grass kg FM^{-1}) and grass alone functional CC (387 g grass kg FM^{-1}) had the least. At the end-of-season mulch DF, there were no differences in grass composition among grass alone, monoculture annual ryegrass, grass-brassica, and grass-legume CC, but the all species CC had less proportion of grass in the total FM than grass alone and grass-brassica. Across DF, there were no differences in grass composition for monoculture annual ryegrass, grass-brassica, grass-legume, and grass CC, but the all species CC had less annual weighted grass when not defoliated during the season compared to the 4- and 8-wk DF

(Table 3.3). Within the 4-wk DF, there were no differences among CC, indicating that there was no effect of the more frequent harvest schedule in this study on grass composition. Rinne and Nykanen (2000) reported that harvesting schedules did not change the botanical composition of herbage within harvests. The small grass proportion in the mixture with grass-legume and grass alone functional group CC could have been due to reduced N fertility. During the summer of Year 1, the N fertilizer application was not carried out as planned. Also, no fertilizer was applied during the cover crop growing season, so N deficiency possibly occurred. This partially explains less percent grass possibly due to slower grass growth rates and an advantage to the legumes in the CC that included both grasses and legumes.

Table 3.3 Defoliation frequency (DF) × cover crop (CC) effect on annual weighted proportion of grass in the total forage mass harvested (FM) of the five CC treatments that included grass harvested at 4- or 8-wk intervals or cut at the end of the season and left as mulch at the Brown Loam Branch Experiment Station, Raymond, MS.

	Defoliation frequency			
CC mixture	4-wk.	8-wk.	Mulch	SEM
	- - - - - - - - g grass kg FM^{-1} - - - - - - - - - -			
All species	431 Aa†	495 Aa	387 BCb	
Grass alone	477 Aa	375 Ca	550 Aa	
Grass-legume	486 Aa	423 BCa	405 Aba	
Grass-brassica	469 Aa	465 Aba	472 Aba	
Monoculture annual ryegrass	467 Aa	552 Aa	506 Aba	
SEM‡				71.0

†Within columns, CC means followed by the same upper-case letters, and within rows, DF means followed by the same lower-case letter are not different (*P* > 0.05).
‡Standard error of means.

There was a DF × year interaction effect ($P < 0.03$) on annual weighted proportion of grass in the total FM harvested across CC (Fig 3.2). In Year 1, total grass proportion in the total

FM harvested was greater at 4- compared to 8-wk DF, but in 2019, the reverse occurred. In both years, grass proportion in the end-of-season mulch treatment was intermediate and not different from either of the harvested DF. Between years, the annual weighted proportion of grass across CC was greater in Year 1 than Year 2, possibly because of fluctuations in weather (Table 3.1) and also low N fertility because fertilizer was not applied during the previous summer as planned.

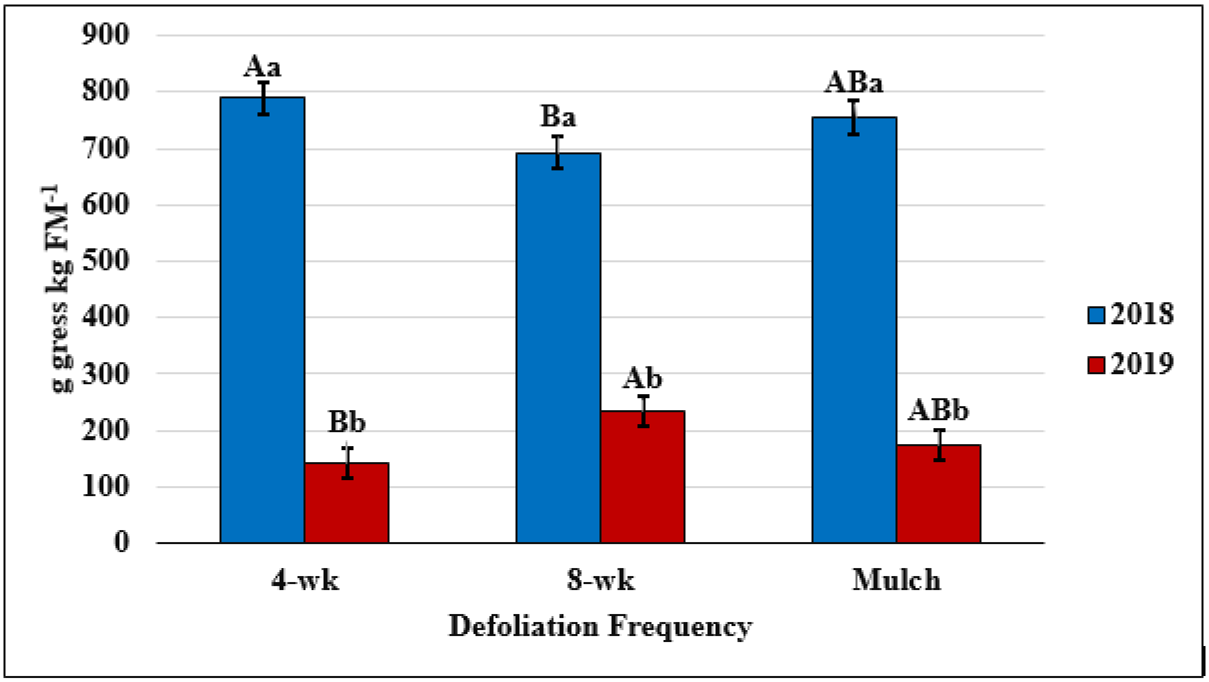

Figure 3.2 Defoliation frequency (DF) × year effect on annual weighted average grass proportion in the total forage mass harvested (FM).

In Fig. 3.2, the annual weighted average grass proportion is represented by blue bars in 2018 and red bars in 2019. As noted on Page 40, data are from across diverse mixtures of winter cover crops, harvested every 4 or 8-wk or cut at the end of the season and left as mulch, during 2018 and 2019 at the Brown Loam Branch Experiment Station, Raymond, MS. Within years, means followed by different upper-case letters are different among DF, and within DF, means followed by different lower-case letters are different between years ($P < 0.05$).

Among those CC that included legumes, there was a CC effect ($P < 0.004$) on annual weighted average proportion of legumes in the total FM harvested. Those CC with no grasses, that is, legume-alone (323 g legume kg FM^{-1}), and legume-brassica mixture (219 g legume kg FM^{-1}), had similar proportions of legume (data not shown). Brassicas did not survive in the experiment, so the latter CC essentially became similar to legume-alone but at less seeding rate than the monoculture plots. In CC with both grasses and legumes, that is, the all species (163 g legume kg FM^{-1}) and legume-grass (135 g legume kg FM^{-1}), proportion of legume in the total FM was not different. Between years, proportion of legume in the total FM was different ($P <$ 0.001; 113 g legume kg FM^{-1} in Year 1 and 307 g legume kg FM^{-1} in Year 2). Less proportion of legumes in the total FM in those CC with grasses possibly was due to species competition. Typically, grasses grow more vigorously than legumes (Clark, 2007). Also, increased weed suppression in CC with grasses may reflect allelopathic effects (Clark, 2007).

There was a CC × year interaction effect ($P < 0.04$) on the annual weighted average proportion of weeds in the total FM harvested (Table 3.4). In Year 1, proportion of weed was greater in legume-brassica than legume alone CC, and both were greater than the all species, the grass-legume, and the monoculture annual ryegrass CC, which were CC that contained grasses. Proportion weed was not different among the CC with grasses in the mixture. The abundance of weeds in the legume plots during Year 1 was due possibly to poor establishment and growth of legumes and death of the brassicas, most likely caused by waterlogged conditions. In Year 2, the grass-alone, grass-brassica, and annual ryegrass monoculture CC generally had greater proportion weed than legume alone. This may be due to poor N fertility causing reduced growth rate in grasses. During the summer of Year 1, N fertilizer was not applied as planned, and N deficiency possibly occurred. Legumes will not suffer as much from lack of N because of their inherent ability to fix N from the atmosphere through their association with rhizobia bacteria. Because of these differences in responses across years with CC that were predominantly grasses or included legumes, those with legumes generally had less weeds in Year 2 than Year 1, while the reverse occurred in those that were predominantly grasses. These results indicated that the composition of the mixture in the CC has the potential to inhibit the establishment and growth of weeds, depending on growing conditions. Sanderson et al. (2013) reported that the average weed percentage in the harvested herbage mass across three years was consistently greater in cover crop monocultures than mixtures, indicating that mixtures were successful in suppressing weeds. Picasso et al. (2008) reported that weed biomass decreased exponentially with species richness. Florence et al. (2019) concluded that increases in cover crop mixtures diversity were correlated with increased weed suppression and biomass stability. While species diversity reduces weed abundance, it is greatly dependent on the type of species in the mix (Frankow-Lindberg et al. 2009). Sanderson et al. (2013) found that greater amount of legume reduced weed abundance, in agreement with the findings of our study.

Table 3.4 Cover crop mixture (CC) × year effect on annual weighted average proportion of weeds in the total forage mass harvested (FM)of seven CC across three defoliation frequencies (harvested every 4 or 8-wk or cut at the end of the season and left as mulch) at the Brown Loam Branch Experiment Station, Raymond, MS.

	Year		
CC mixture	2018	2019	*P*-value†
	- - - - - g weed kg FM^{-1} - - - - -		
All species	254 C‡	544 C	< 0.001
Grass alone	224 C	839 A	< 0.001
Legume alone	749 B	604 C	0.093
Grass-legume	258 C	594 C	< 0.001
Grass-brassica	214 C	848 A	< 0.001
Legume-brassica	921 A	640 BC	< 0.002
Monoculture annual ryegrass	199 C	785 AB	< 0.001
SEM§	113.8	48.6	< 0.001

†*P*-value to compare CC means between years.
‡Within columns, CC means followed by the same upper-case letters are not different ($P > 0.05$)
§Standard error of means.

There was a DF × year interaction effect ($P < 0.02$) on annual weighted average of weed in the total FM harvested across CC (Fig. 3). In Year 1, proportion of weeds were greater at 8-wk than 4-wk or end-of-season mulch DF, and the latter two were not different. In Year 2, the reverse occurred where 8-wk DF had less weeds, while there was no difference between 4-wk and end-of-season mulch. Between years, there was greater proportion of weeds in Year 2 than Year 1. Less proportion of weeds at the 4-wk compared to the other DF in Year 1 occurred possibly because more frequent defoliation suppressed weed growth. Herbert et al. (2018)

reported that the loss of photosynthetic tissues caused by frequent defoliation can reduce plant growth, including weeds. Greater amount of weed at the 8-wk DF occurred possibly because the longer defoliation interval allowed more time for weed growth. Belesky et al. (1999) found that longer harvest intervals allowed more vegetation growth, including weeds. The mulch DF possibly suppressed weeds because the CC was undefoliated, and their growth outcompeted the weeds (Brennan and Smith, 2005).

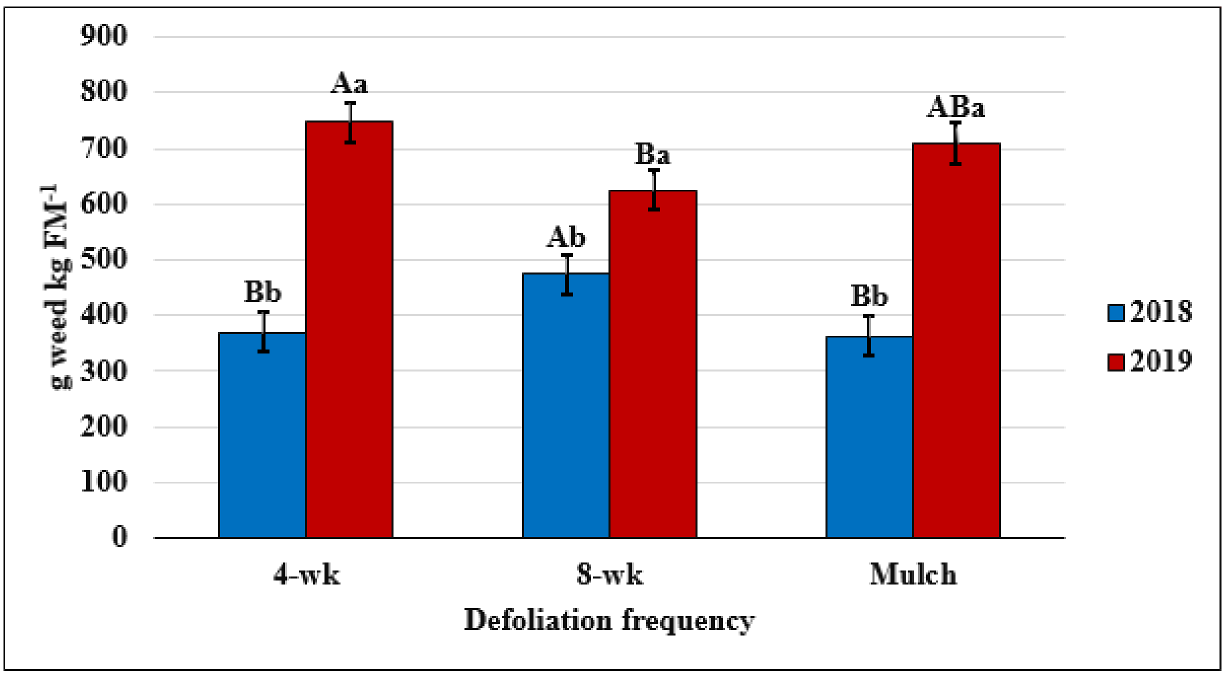

Figure 3.3 Defoliation frequency (DF) × year effect on the annual weighted average proportion of weeds in the total forage mass harvested (FM).

In Fig. 3.3, the proportion of weed in the total forage mass is represented by blue bars in 2018 and red bars in 2019. As noted on Page 45, data are from across cover crop mixtures harvested at the Brown Loam Branch Experiment Station, Raymond, MS. Within years, means followed by different upper-case letters are different among DF, and within DF, means followed by different lower-case letters are different between years ($P < 0.05$).

Forage nutritive value

Across the years, there was a CC effect ($P < 0.0001$) on the annual weighted average CP (Table 3.5). The legume alone and legume-brassica had greater CP concentration than all other CC treatments. Grass alone, grass-brassica mix, and monoculture annual ryegrass had the least CP levels, and all species and grass-legume had intermediate CP. Across years, there was a DF effect ($P < 0.0001$) on CP, with greater CP at the 4-wk DF (101 g kg^{-1}) compared to 8-wk (86 g kg^{-1}) and end of season mulch (91 g kg^{-1}). The latter two were not different. There was a year effect ($P < 0.0001$) on CP also. In Year 1, CP concentration (112 g kg ha^{-1}) was greater than in Year 2 (73 g kg ha^{-1}). The greater CP at the 4-wk DF was expected because CP in plants is usually greater at the early stage of growth than at the mature stage (Roukos et al., 2011). These results also explain the lesser CP in 8-wk DF and end-of-season mulch. The greater CP found in the legume alone and legume-brassica CC and the lesser CP in the grass alone, grass-brassica mix, and monoculture annual ryegrass CC are explained by the greater CP typically found in legumes (Kandrelis, 2016). Studies have shown that legumes complement grasses by increasing forage production and the protein concentration of the mixtures (Berdahl et al., 2001).

Table 3.5 Cover crop (CC) main effect on annual weighted average crude protein (CP) and neutral detergent fiber (NDF) across the two years and three defoliation frequencies (harvested every 4 or 8-wk or cut at the end of the season and left as mulch) at the Brown Loam Branch Experiment Station, Raymond, MS.

	CP	NDF
CC mixture	- - - - - - - - - - - - g kg^{-1}	- - - - - - - - - - - - - - -
All species	96 B†	580 B
Grass alone	83 C	600 A
Legume alone	107 A	559 C
Grass-legume	96 B	581 B
Grass-brassica	83 C	606 A
Legume-brassica	105 A	561 C
Monoculture annual ryegrass	81 C	608 A
SEM‡	4.88	7.14

†Within columns mean followed by the same upper-case letters are not different ($P > 0.05$)
‡Standard error of mean.

There was a DF × year ($P < 0.0001$) on total NDF and ADF (Table 3.6.). In both years, NDF was least at the 4-wk DF, but in Year 1, NDF concentration was greater at 8-wk than at the end-of-season mulch DF, and in Year 2, ADF was greater for mulch than 8-wk DF. Possibly due to the fluctuations in ranking between the 8-wk and mulch DF, the interactions were caused by a reverse in differences between years with Year 1 being greater than Year 2 at the mulch DF and Year 2 greater than Year 1 at the 8-wk DF. Also, there was no difference between years at the 4-wk DF. The DF × year interaction effect on ADF was due partially to differences in ranking among DF treatments within years but was always least at the 4-wk DF. Between the years, ADF was greater in 2019 regardless of DF. The difference in ADF and NDF concentrations at the various DF across cover crops occurred possibly due to changes in growth stages and changes in the composition of the species in the mixtures during the growing season (Albayrak et al., 2011).

Table 3.6 Defoliation frequency (DF) × year effect on annual weighted average acid detergent fiber (ADF) and neutral detergent fiber (NDF) across cover crop treatments during 2018 and 2019 at the Brown Loam Branch Experiment Station, Raymond, MS.

	ADF		NDF	
	2018	2019	2018	2019
	- - - - - - - - g kg^{-1} - - - - - - -		- - - - - - - - g kg^{-1} - - - - - - -	
4-wk	293 Bb†	374 Ca	543 Ca	557 Ca
8-wk	344 Ab	409 Ba	610 Aa	582 Bb
Mulch	334 Ab	445 Aa	591 Bb	628 Aa
SEM‡	5.57	5.57	6.70	6.70

†Within columns, means followed by the same upper-case letters, and between years for each response variable, means followed by the same lower-case letters are not different ($P > 0.05$)
‡ Standard error of mean.

Summary and Conclusions

Results from this study showed that forage mass production and weed suppression benefited from having diverse mixtures of cover crop species. Overall, CC treatments of all species mixtures, legume-grass mixtures, and legumes alone functional group performed well in both years compared to the other cover crop mixtures, possibly because of the N fixation benefits from legumes. In both years of this study, CC under mulch treatments had greater forage biomass compared to 4-wk and 8-wk DF. These results were mainly because the mulch treatment was undefoliated prior to the end of the growing season and had a longer growth interval compared to the treatments that were harvested during the season. The 4-wk DF had the least forage mass production, and this was possibly due to a reduced growth rate from the loss of photosynthetic tissues.

Crude protein was greater at 4-wk DF compared to 8-wk and end of season mulch mainly because CP in plants is usually greater at the early stage of growth than at the mature stage. The CP levels were greater in legume alone and legume-brassica mixtures compared to the grass alone, grass-brassica mixture, and monoculture annual ryegrass, because of the inherent better nutritive value of legumes. The legume-grass mixture and all species mixtures had intermediate levels of CP, possibly due to the small legume percentage and/or the rapid growth of the grass species in the combination. The difference in ADF and NDF concentrations at the various DF across cover crops were due possibly to changes in growth stages and also the composition of the species in the mixtures occurring during the growing season. Overall, the differences in forage nutritive value among the cover crop mixtures could be related to species composition, competition among species with the combinations, and time of harvesting at the different growth stages.

Producers desire to have pasture systems that will produce high quantity forages. Cover crop systems gained popularity because of benefits to soil health and ecosystem services. Hence, the continuation of these studies will evaluate the treatment effects on soil microbial community structure and soil biological activity. It also may be of some interest to conduct grazing studies, as ruminant livestock may prefer grazing particular species, and animal performance may respond differently to different CC and grazing management. Additionally, the effect of animals grazing may have an effect on the expected benefits of the cover crops that may be different than when grown and harvested mechanically.

CHAPTER IV

COOL-SEASON COVER CROP AND DEFOLIATION FREQUENCY EFFECTS ON SUMMER HAY AND SEASON-LONG FORAGE PRODUCTION

Abstract

Overseeding cool-season cover crops (CC) into existing bermudagrass hayfields may benefit subsequent hay production. If the cover crop is harvested, the defoliation frequency (DF) may play a role in subsequent hay production also. A 2-yr experiment conducted at Raymond, MS quantified summer hay and total year-long forage mass of hayfields overseeded with cool season annuals. Treatments were factorial combinations of 10 CC (eight mixtures using several species of grasses, legumes, brassicas, and two none-overseeded controls) and three DF (harvested every 4 or 8-wk or cut and left as mulch) in a split-plot arrangement of a randomized complete block design with three replications. Generally, cool-season CC increased total year-long forage mass when harvested, and the effects of CC on summer forage growth could only be detected when the cover crops were not harvested.

Introduction

Perennial pastures, predominantly warm-season grasses such as bermudagrass [*Cynodon dactylon* (L.) Pers.] and bahiagrass (*Paspalum notatum* Flüggé), represent long-term land use on about 75% of the total pasture area, approximately 25 million ha, of the humid lower southeastern USA across the states of Alabama, Arkansas, Florida, Georgia, Louisiana, Mississippi, and South Carolina plus the eastern portions of Oklahoma and Texas (Ball et al., 2001; Nickerson et al., 2011). These forage crops respond to high rates of fertilizer, particularly nitrogen, and near monoculture stands are often maintained with repeated applications of selective herbicides. A number of factors indicate the potential to improve both the economic returns and environmental effects of these grasslands. One such factor is the use of cool-season annuals used as cover crops are that can provide a source of N for subsequent crops reducing the need for chemical fertilizers (Evers, 2011). Cool-season annuals have the potential to reduce erosion and contamination of surface waters (Schils et al., 2013, improve soil physical properties, suppress nematode populations (McSorley et al., 1994; McSorley and Dickson, 1995; Mojtahedi et al., 1993), and suppress weeds (Creamer and Baldwin, 2000).

Bermudagrass and bahiagrass are commonly used during the summer periods when traditional cool-season perennial pasture growth decreases to make forage available throughout the growing season (Fontaneli et al., 2001). These warm-season forages often have less nutritive value, with increased fiber and lignin concentrations and reduced leaf-to-stem ratios, which results in decreased digestibility compared to cool-season forages (Dillard et al., 2018). Overseeding combination of cool-season forage such as legumes, grasses, and small grains as cover crops into existing bermudagrass can help offset increased frequency of summer drought conditions and increased diversity of forage crops allowing for well-distributed forage production for a more extended period and reduces susceptibility to short-term drought (Rouquette, 2017). These cover crops can provide ecosystem benefits to perennial-based pasture systems and make available forage of high-quality at opportune times of the year. It can create a more diverse farm habitat and provides opportunities to renovate overused or underutilized areas of the farm (Macoon et al., 2016). Often, crop species mixtures increase productivity and stability in natural grasslands (Jing et al., 2017). In pastures, a diversified ecosystem has led to improved forage production and reduced weed invasion when compared to communities with few plant species (Tracy and Sanderson, 2004). These diverse mixtures enhance the amount and total biomass stability within grasslands as a result of niche complementarity in species-rich mixes that optimize the capture of light, water, and nutrient resources (Jing et al., 2017).

Substituting N fertilizer by exploiting symbiotic N_2 fixation in agricultural grasslands is considered as an essential contribution to resource-efficient and sustainable agricultural systems (Gruber and Galloway, 2008). Symbiotically fixed N_2 in legumes ranged from 100 to 380 kg N ha^{-1} $year^{-1}$, but large amounts of more than 500 kg of N ha^{-1} $year^{-1}$ are also reported (Carlsson and Huss-Danell, 2003; Zanetti et al., 1997). In mixed grass-legume systems, N amounts of 10–75 kg of N ha^{-1} $year^{-1}$ may additionally be transferred from legumes to grasses (Nyfeler et al., 2011). These leguminous cover crops have the potential of improving the productivity of subsequent crops (Fageria et al., 2005). Mclaughlin et al. (2005) concluded that overseeding ‘Tifton 44’ bermudagrass with berseem clover (*Trifolium alexandrinum* L.) increased the forage mass harvested of subsequent summer hay and suggested that the increase may have been due to increased N available fixed by the berseem clover. Also, Fageria et al. (2005) reported that cereal crops planted after a legume cover crop showed a positive yield response due to the transfer of biologically fixed N and less immobilization of nitrate during the decomposition of legume residues. Cover crop effects on subsequent crops may not be noticeable until an extended period, however (Blanco-Canqui, 2015). Most studies have reported cover crops and year interaction, which suggests that long term studies longer than 3 yr are needed to understand the effects of monocultures and cover crop mixtures better on subsequent crop yields and soil properties. Some studies have reported that while cover crops did not increase crop yields in the first year, positive effects were observed as time progresses (Clark et al., 1994; Andraski and Bundy, 2005).

Forage crops are grown to be defoliated, whether by cutting or by grazing animals. Defoliation can have an effect on the amount of harvested biomass as well as the proportion of legume in grass-legume mixtures and can alter the ability of legume to fix atmospheric N_2 (Swift et al., 1994; Elgersma and Schlepers, 1997; Schils et al., 2000; Unkovich et al., 1998). Vinther (2006) reported that DM production of the infrequent and frequent cutting treatments of perennial ryegrass– white clover mixtures produced a total yield of 7000 and 5700 kg DM ha^{-1}, respectively. Also, the proportion of N derived from N_2 fixation (pNdfa values) in the harvested biomass were in the range of 0.78 to 0.96 in the infrequent-cutting treatment, whereas pNdfa values in the frequent-cutting treatment decreased from 0.83 to 0.60. Farnham and George (1994) reported that herbage mass for birdsfoot trefoil (*Lotus corniculatus* L.) and orchardgrass (*Dactylis glomerata* L.) were greater under the three-cut system, averaging 8.8 Mg ha^{-1} compared with 6.4 Mg ha^{-1} for the six-cut system. It is also known that frequent cutting increases the production of root exudates and reduces the proportion of fine roots, which also increases the availability of N in the soil, and thereby reduces the dependency on atmospheric-N (Menneer et al., 2004). There are not many studies on cutting frequency effects on the biomass of diverse cover crop mixtures and that of subsequent summer hayfields, hence such studies are needed. This study quantified summer hay and total year-long forage mass when cool-season annuals were overseeded as cover crops into existing bermudagrass hayfields and managed at three defoliation frequencies (harvested every 4 or 8-wk or cut at the end of the season and left as a mulch).

Materials and Methods

In the previous chapter, a detailed description was given for study location, treatment, and cool-season management.

Sampling

For summer hay harvests, all subplots were harvested to coincide with harvest of adjacent hayfields. In 2018, harvesting was done July 23, August 13, September 13, and October 30. In 2019, harvesting dates were July 24, August 29, and October 9. The center 3 × 0.6 m of all 90 subplots was harvested to a 5-cm residual stubble using a push mower with the bag attachment to catch the harvested forage. Weighing, drying, and DM determination procedures were the same as described for cool-season harvests. Also, after harvest sampling was completed, the rest of the plot was cut to a uniform stubble height, and the forage removed as done with the cool-season harvests.

Statistical analysis

Total annual hay production was the sum of all summer harvests each year (four in 2018 and three in 2019). Year-long forage mass (FM) was the sum of total annual hay production and annual cool-season FM. Analysis of variance was done by fitting mixed models using PROC

MIXED and PROC GLIMMIX in SAS (SAS Institute Inc., Cary, NC). The best model fit was determined using the Schwarz' Bayesian Criterion. Whole plots were DF, and CC were subplots. Means separation was done using the PDIFF option, and responses were considered different at the 0.05 probability level.

Results and Discussion

As mentioned in Chapter 3, in both years of the study, the brassica crops germinated but did not survive past the early seedling stage, possibly because of the excessive rainfall that led to waterlogged conditions, along with record cold temperatures during the establishment of the crops (Table 3.1). It should be understood then that mixtures with all functional groups are essentially grass-legume mixtures, and the grass-brassica and legume brassica mixtures are more or less monoculture grass or legume stands but at a reduced seeding rate compared to the monoculture functional group treatments. Also, the brassica alone monoculture, in essence, became a third non-overseeded control.

There was no main effect of CC or any interactions with CC ($P > 0.10$) on summer hay production, but there was a CC × DF interaction effect ($P < 0.0001$) on total year-long FM (Table 4.1). Within the 4-wk DF, the grass-legume and grass-brassica CC treatments had the greatest year-long FM but were not different from the monoculture grass and legume functional groups, and legume-brassica CC treatments, which all had similar FM. Annual ryegrass year-long FM was not different from the latter three treatments but was less than the grass-legume and grass-brassica treatments. The all species CC had less year-long FM than the above-mentioned CC treatments but was greater than the controls and monoculture brassica functional group CC (essentially a control because no brassica survived in the plots).

Table 4.1 Defoliation frequency (DF) × cover crop effect on total year-long forage mass during 2018 and 2019 at the Brown Loam Branch Experiment Station, Raymond, MS.

CC Mixture	Defoliation frequency		
	4-wk harvest	8-wk harvest	End-of-season mulch
	- - - - - - - - - - - - - - - - - kg ha^{-1} - - - - - - - - - - - - - - - - -		
All species	5390 Ca†	5810 Ba	3210 Ab
All grasses	6480 Aba	6740 Aa	3560 Ab
Legumes	6500 Aba	6550 Aa	3650 Ab
Brassicas	3070 Da	3130 Ca	3520 Aa
Grass-legume	7090 Aa	6810 Aa	3620 Ab
Grass-brassica	6960 Aa	6740 Aa	3590 Ab
Legume-brassica	6650 Aba	6550 Aa	3800 Ab
Annual ryegrass	6250 Ba	6400 Aba	3670 Ab
Control (weed control)	3510 Da	3500 Ca	3720 Aa
Control (no weed control)	3300 Dab	3030 Cb	3820 Aa
SEM‡		113.4	

†Within columns, least square means followed by different upper-case letters are different, and within rows, means followed by different lower-case letters are different ($P < 0.05$).
‡Standard error of mean.

Within the 8-wk DF, all grasses, legumes, grass-legume, grass-brassica, and legume-brassica functional groups had the greatest year-long FM and were not different from each other. All species and annual ryegrass functional groups had similar year-long FM but were less those mentioned above, while, as with the 4-wk DF, the controls and the brassica monoculture had the least. Within the end-of-season mulch DF, there we no differences among CC treatments, no doubt because these are summer production alone with no contribution from the cool-season cover crops. Among DF, season-long FM was generally similar among the controls and the brassica monoculture treatments, but those CC that was harvested typically had greater year-long FM than the end-of-season mulch, with no differences between the 4- and 8-wk DF. As mentioned previously, CC treatments that included legumes benefited from the contributed of converted atmospheric N_2 through symbiotic N_2 fixation (Carlson and Huss-Danell, 2003;

Lüscher et al., 2014). Reduced FM for annual ryegrass at 4-wk DF is possibly due to frequent harvesting, which may result in less forage growth than infrequent harvests (Bittman et al. 1991; Foster et al., 2014). Least FM recorded for all species CC treatment may be due to inter-plant competition among the different species within the combination.

There were DF × year interaction effects on summer hay ($P = 0.002$) and total year-long FM ($P < 0.0001$). In Year 1, summer hay FM ranked end-of-season mulch > 8-wk DF > 4-wk DF. In Year 2, there was no difference in summer hay FM between mulch and the 4-wk DF, but both had greater FM than the 8-wk DF. As expected, the end-of-season mulch had the least year-long FM among DF treatments in both years, mainly because there was no contribution to the total annual FM from the cool-season annuals. Among the harvested treatments, greater FM was recorded at 8-wk than the 4-wk DF in Year 1, but the reverse occurred in Year 2.

In the summer of Year 1, the N fertilizer application was not carried out as planned. Also, no fertilizer was applied during the cover crop growing season, so N deficiency possibly occurred and likely had an effect on summer hay production. Although the mulch treatment can cause greater forage biomass due to benefits such as the build-up of soil organic matter, improved water, and nutrient use efficiency (Alharbi, 2015), in our study, the greater forage mass may be due mainly because this treatment was not defoliated during the growing season, which allowed for maximum biomass production.

Table 4.2 Defoliation frequency (DF) × year effect on summer hay and total year-long forage mass (FM) across 10 cover crop treatments at the Brown Loam Branch Experiment Station, Raymond, MS.

DF	Summer hay FM 2018	Summer hay FM 2019	Year-long FM 2018	Year-long FM 2019
	- - - - - - - - - - - - - - - - - kg ha^{-1} - - - - - - - - - - - - - - - - -			
4-wk	2870 Cb	3770 Aa	4810 Bb	6220 Aa
8 wk	3110 Bb	3510 Ba	5280 Ab	5790 Ba
Mulch	3360 Ab	3870 Aa	3360 Cb	3870 Ca
SEM‡	91.1		113.4	

†Within columns, least square means followed by different upper-case letters are different, and for each parameter, means between years followed by different lower-case letters are different ($P < 0.05$).
‡Standard error of mean.

As stated earlier, there was no main effect of CC or any interactions with CC ($P > 0.10$) on summer hay production, but often the presence of cover crops during winter may have a suppression effect on hay emergence. To explore this further, summer hay FM data were analyzed by harvest within and across years. When analyzed for 2018 alone, the was a harvest time × DF effect ($P < 0.001$), but there was no effect of CC or any interactions with CC ($P > 0.9$). The harvest time × DF interaction occurred mainly because at the first harvest, the mulch DF had greater FM than treatments harvested during the winter, but at the second and third harvests, there were no differences in summer hay FM among DF treatments (data not shown). When analyzed for 2019 alone, there was a harvest time main effect ($P < 0.001$) and a DF main effect ($P = 0.004$) with no two- or three-way interactions ($P > 0.26$). Generally, hay FM was less at the first harvests that at the second and third harvests (data not shown). Although there was no effect of CC ($P = 0.132$), there may be a trend that should be examined, so these data will be presented by harvest time for both years (Table 4.3). When the data were analyzed with years combined, there was a year × harvest time × DF effect ($P = 0.0002$), but there was no effect of

CC ($P = 0.65$) or any interactions involving CC ($P > 0.5$). The year × DF effect for the total summer FM data was discussed earlier.

Table 4.3 Summer hay forage mass for each harvest within years among 10 cover crop (CC) treatments at the Brown Loam Branch Experiment Station, Raymond, MS.

	2018				2019			
	Harvest Date				Harvest Date			
CC Mixture	7/23	9/13	10/30	Season total	7/24	8/29	10/9	Season total
	- - - - - - - - - - kg ha^{-1} - - - - - - - - - - - - - - -				- - - - - - - - - - - - kg ha^{-1} - - - - - - - - - - - - - - -			
All species	1109	1561	388	3058	914	1449	1264	3627
All grasses	1138	1666	390	3194	825	1511	1250	3585
Legumes	1106	1643	406	3155	911	1563	1151	3626
Brassicas	1029	1542	422	2993	816	1354	1318	3488
Grass-legume	1216	1698	410	3324	964	1527	1233	3724
Grass-brassica	1142	1632	457	3230	882	1502	1347	3732
Legume-brassica	1066	1528	414	3009	902	1645	1376	3923
Annual ryegrass	1131	1541	393	3065	807	1463	1326	3596
Control (weed control)	1075	1656	333	3065	1107	1562	1419	4088
Control (no weed control)	1109	1564	380	3053	840	1495	1438	3773
SEM†		99.6		143.7		93.6		143.7

†Standard error of mean. The was no CC effect in 2018 ($P > 0.9$) or 2019 ($P = 0.132$), but it may be useful to show these data to allow examination of the results, especially since the 2019 response may be considered a trend.

Summary and Conclusions

Results from this study showed that generally, the different CC mixtures did not have an effect on subsequent summer hay forage mass. However, when harvested, it contributed to the total year-long FM, indicating that the cool-season cover crop was important to the forage contribution. It is likely that the effects of different CC mixtures on summer forage growth could not be detected because there was not sufficient time for any benefits contributed by changes in soil quality to be realized. Hopefully, such benefits can become noticeable in the longer term. When CC was not harvested during the winter growing season, that is, cut at the end of the season and left as a mulch, there was greater summer hay production compared to when the CC was harvested for forage. The fluctuations in year-long forage mass may be due to a combination of environmental and weather-related factors such as low N fertility in the soil, cold winter temperatures, and fluctuations in precipitation, including excessive rainfall. Loss of the brassicas CC, along with the botanical compositions of the different species, and DF also had an effect on the year-long forage production. Cover crop mixtures that included legumes performed well in both years, possibly as a result of the N contribution to the soil. Overall, overseeded CC into bermudagrass sod has the potential of increasing forage production and extending the time the forage is available.

Continuation of this experiment will be valuable to gain long term data on cover crop and defoliation management effects on summer production because it was unlikely that benefits can be accrued in only a 2-yr period. Additionally, soil analysis to determine changes to soil chemical composition and ecology are planned to help explain observed plant responses. Economic analysis to determine the cost and benefits of cover crop systems may also be warranted. It also may be of interest to conduct studies to quantify responses of animals grazing cover crops as well as the perennial forages that were overseeded with cover crops.

CHAPTER V

SUMMARY AND CONCLUSIONS

To evaluate the potential benefits of diverse mixtures of cool-season annual grasses, legumes, and brassica species as cover crops (CC) under different defoliation frequency (DF), this study quantified forage mass, nutritive value, and botanical composition of various combinations of CC mixtures harvested at three different DF consisting of 4- or 8-wk harvest intervals, or cut and left as a mulch at the end of the winter growing season, and assessed the effects of these cover crops × harvest management combinations on subsequent hay and year-long forage production. Forage mass production benefited from having diverse mixtures of CC species. Forage mass harvested from the treatments containing all species, legume-grass, and mixtures containing legumes alone was greater than the other CC mixtures. For DF, the mulch treatment had greater forage mass than 4- and 8-wk DF because this treatment was not defoliated during the growing season.

Reduced N fertility in the soil resulted in slower grass growth rates, allowing grasses in legumes mixtures to be more productive than grass alone. The legume alone and the legume-brassica mixture had a greater percent legume compared to mixtures with grasses. Percent weed tended to be greater in those CC treatments that included brassicas since brassicas did not survive. Overall, these results suggest that, compared to monocultures, diverse mixtures as cover crops can be beneficial to weed suppression. Among DF, the results showed that the greatest weed suppression occurred when cover crops were not harvested but cut and left as a mulch at the end of the growing season. The lack of survival of brassica in our study suggests that brassica as a cover crop or as a forage crop in similar soil types are not likely to be successful in years with above-normal rainfall.

Our results also showed that the crude protein (CP) levels were greater in mixtures that contained legume, and at 4-wk defoliation frequency compared to 8-wk and end of season mulch. The ADF and NDF concentrations at the various defoliation frequency increased with maturity and when there was more grass in the mixtures. These results imply that the differences in forage nutritive value among the CC mixtures was due to species composition in the combinations, and time of harvesting at the different growth stages.

In these preliminary results from this study, different CC mixtures did not have an effect on subsequent summer hay forage mass but did had an effect on total year-long FM. More than likely, two years of cover crop is not sufficient to cause changes to soil characteristics that can cause immediate effects on the growth of the succeeding crop. When CC was not harvested during the winter season, that is, cut at the end of the season and left as a mulch, summer hay production was greater than when the CC was harvested for forage. These results suggest that the mulch is beneficial for hay growth. Total year-long forage mass was greater for the harvested CC plots than the controls and the brassica alone treatment, simply because there was no forage production from the latter treatments to be added to the summer hay for the year-long total. These results imply that the cool-season CC was important to the forage contribution, and the effects of CC on summer forage growth could be detected only when the cover crop is not harvested as forage.

Overall, our results imply that diverse CC under good harvest management practices can contribute to land stewardship and the overall long-term sustainability of perennial warm-season pasture systems. Continued research should include long-term field trials to measure the effects among CC mixtures on soil health and productivity and the interactions between species in mixtures. Since the brassicas used failed to establish, it may be useful to evaluate others that may fit Mississippi weather patterns. In addition, studies to quantify responses of animals grazing on CC systems are recommended. Also, economic analysis to determine the cost and benefits of CC systems may be warranted.

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