

NUTRITIONAL EVALUATION OF PERENNIAL PEANUT, ANNUAL PEANUT, AND
TIFTON 85 BERMUDAGRASS HAYS IN HORSES

By

JULIET V. ECKERT

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To Eden and Grandma Sally, may you both rest in peace.

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Abstract of Thesis Presented to the Graduate School
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Juliet V. Eckert

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Three experiments were conducted to evaluate the nutritional quality and acceptance of perennial peanut (PP), annual peanut (AP), and Tifton 85 (T85) bermudagrass hays in horses. In Experiment 1, *in vivo* digestibilities of PP, Coastal bermudagrass (CB), and T85 were compared. Six mature horses were used in a 3X3 Latin square design with three periods, each consisting of a 10 d adjustment phase and a 4 d total fecal and urine collection phase. Samples of hay offered were analyzed for dry matter (DM), ash, crude protein (CP), nitrogen (N), neutral (NDF) and acid (ADF) detergent fibers, calcium (Ca), and phosphorus (P). Hay refusals were analyzed for DM. Feces samples were analyzed for DM, ash, CP, N, NDF, ADF, Ca, and P. Urine samples were analyzed for nitrogen, Ca and P. Results show PP is a nutritionally superior forage to both CB and T85. Perennial peanut hay had greater nitrogen balance (NB), and DM, OM, and CP digestibilities than the mean of the bermudagrasses. Coastal bermudagrass and T85 were similar in all digestibility measurements except NB, where CB was greater than T85. Intake of T85 was lower than CB, probably causing a decreased NB. There were no differences between the three hays in Ca balance. The PP hay had a greater P balance than the mean of the bermudagrasses; however, the two bermudagrasses did not differ.

In Experiment 2, *in vitro* true digestibilities (IVTD) were determined for dried fresh forage or hay samples of AP (AP–Fresh1, AP–Fresh2, AP–hay), dried fresh forage or hay samples of PP (PP–Fresh, PP05–hay, and PP06–hay), alfalfa (A–hay), CB–hay, T85–hay, and six varieties of PP (Arbrook, AR; Floragrazer, FG; Tom Burton Selection, TB; Ecoturf, EC; Elite Line, EL; and Good Line, GL). Horse feces were used as the source of microbial inoculum and samples were incubated in a closed system fermentation apparatus (ANKOM Technology Corp., Fairport, NY). The IVTD of AP–Fresh1 and –Fresh2 were greater than that of PP–Fresh; but AP–hay was similar to PP06–hay. The IVTD of A–hay is similar to PP06–hay and greater than AP–hay. Perennial peanut (PP05–hay) from the *in vivo* trial had a numerically greater IVTD than the dry matter digestibility calculated in the *in vivo* trial. Of the six varieties tested, EC and EL had the greatest IVTD; however, all six varieties had similar IVTD as PP05–hay, PP06–hay and A–hay. The IVTD of T85–hay and CB–hay were numerically smaller than *in vivo* data; but still reflected the results found in the *in vivo* trial.

In Experiment 3, six yearling horses were used to assess the palatability of PP, AP, and A hays. Horses were offered a choice of two hays presented in all possible combinations for a total of six treatments. Results were largely inconclusive. In general, the horses preferred A to PP and AP, and PP to AP. However, it was difficult to collect all refused and uneaten hay, and during a portion of the trial, the PP hay offered was visually of less quality than what would normally be fed.

We concluded that 5 wk T85 is just as digestible or less than 4 wk CB, however, horses did not eat enough to sustain BW, even when offered at 2% BW. Mature T85 may not be suited as the only forage offered to horses; it will likely need to be supplemented with concentrate. Annual peanut is best offered as pasture because of the decreased nutritional quality and digestibility

from haying and storage, as well as the horses' reluctance to eat it. Finally, PP is a high-quality hay that can replace alfalfa in any horse diet.

CHAPTER 1 INTRODUCTION

The primary forages for horses in the south include alfalfa (*Medicago sativa*) and Coastal bermudagrass (*Cynodon dactylon*). High quality alfalfa does not grow well in the acidic sandy soils of Florida and the Southeast; therefore it must be transported in, which adds to its cost (Ball et al., 2002). Feeding a horse is generally the most expensive part of maintaining a horse; therefore, an alternative high quality legume that grows well in Florida and the Southeast is desirable. Perennial peanut and annual peanut hays could be an alternative to alfalfa. Perennial peanut is fed to horses already; however, there is limited information about its digestibility in horses. In addition, no research has been conducted to assess the acceptability and digestibility of annual peanut in horses.

Perennial peanut (*Arachis glabrata*), also known as rhizomal peanut, is a warm-season legume that is well adapted to the climate and soil conditions of the southeastern United States and eastern Texas. Annual peanut (*Arachis hypogaea*) also grows well in the Southeast United States. Both of these plants have the potential to become major legumes for horses in the south. Perennial peanut is already being grown for hay to feed to horses in the Southeastern United States. It has fine stems and is very leafy, two characteristics of forage that horses prefer. The only drawback to perennial peanut is that it is hard to establish, often taking two to three years before the first optimal harvest can be obtained (Hill, 2002). Annual peanut is easier to establish than perennial peanut but the hay has thicker stems with fewer leaves. Nothing is known about the nutritional quality of annual peanut hay for horses; however, annual peanut hay after pod harvest was found to be nutritionally similar to alfalfa in ruminants (Ronning et al., 1953; McBrayer et al., 1981). However, this hay is unsuitable for horses because it is too dusty due to the soil that adheres to the vines during harvest of the pods. Annual peanut hay grown

specifically for hay has not been compared to alfalfa, but both *in vitro* and *in situ* data have shown annual peanut hay to be a good quality forage for ruminants (Gorbet et al., 1994; Larbi et al., 1999). Both annual peanut and perennial peanut hays could be economical alternatives to alfalfa since they grow well in southern climates.

Perennial peanut has been found to be nutritionally similar to alfalfa for ruminants and horses (Prine et al., 1981, 1986; Lieb et al., 1993; French and Prine, 1998). For example, both have similar crude protein (CP) content: Gelaye et al. (1990) reported a CP in alfalfa of 18.0% and a CP in perennial peanut of 17.1%. Prine et al. (1981) reported a range of CP values for perennial peanut (12-19%) depending on maturity of the forage. Gelaye et al. (1991) found perennial peanut to have a greater dry matter digestibility (62.5%) than alfalfa (55.7%) in goats. Lieb et al. (1993) found similar digestibilities of perennial peanut and alfalfa hays in mature horses.

'Coastal' bermudagrass is a warm-season grass with a moderate to low nutritional quality (CP ranges from 3.8 to 10.9%, depending on stage of maturity and fertilization). Coastal bermudagrass grows well in Florida and semiarid climates and serves some classes of horses well (maintenance and light work); however, broodmares and other horses that require more energy will have to be supplemented. A more recent bermudagrass cultivar, 'Tifton 85', has been found to be nutritionally superior to Coastal bermudagrass in ruminants (Hill et al., 1993; Mandebvu et al., 1998a, b; Mandebvu et al., 1999). Much research has been conducted with Tifton 85 for cattle and it has been well received by cattle operations in the Southeast. However, limited research has been conducted with horses.

Our objective was to evaluate the nutritive quality and value of perennial peanut, annual peanut, and Tifton 85 as hay for horses. A digestibility trial was conducted with six horses fed

diets of perennial peanut, Coastal bermudagrass, or Tifton 85 hays. An *in vitro* experiment was performed to assess the approximate nutritive value of the same hays used in the digestibility trial, as well as annual peanut and perennial peanut fresh forages and hays, and alfalfa hay. Finally, a selection trial was conducted with six horses offered a choice of perennial peanut, alfalfa, or annual peanut hay to assess preference.

CHAPTER 2 LITERATURE REVIEW

Introduction

In the Southeastern United States, the most common forages fed to horses include alfalfa (*Medicago sativa*) and 'Coastal' bermudagrass (*Cynodon dactylon*). Alfalfa is a cool-season legume and does not grow well in the acidic sandy soils that persist in the deep South. A warm-season forage is therefore needed as an alternative to alfalfa, such as perennial or annual peanut. Coastal bermudagrass grows well in the Southeast. It is a recommended forage for horses because of its high digestibility and palatability (Lieb, 2003). However, it is grass hay, and therefore is nutritionally inferior to a legume. If properly fertilized and harvested, Coastal bermudagrass will generally put weight on young and mature horses on maintenance diets (Lieb, 2003). An improved bermudagrass variety, 'Tifton 85', has been developed that has been shown to be nutritionally superior to Coastal bermudagrass when fed to ruminants (Hill et al., 1993; Mandebvu et al., 1998a, 1998b, 1999; West et al., 1998; Chambliss and Dunavin, 2003). However, horses are not ruminants but hindgut fermenters; whether perennial peanut, annual peanut, and Tifton 85 bermudagrass are suitable for horses has not been thoroughly researched.

Forages and Their Importance to Livestock

Forages, from pastures to silage, are meant to supply the nutrient needs of herbivores. Grazers, such as cattle and horses, and browsers, such as sheep and goats, subsist on plants containing hemicellulose and cellulose. This digestible or partially digestible fiber is what these animals derive energy from. Forages provide nitrogen (N, some more than others) that serves to feed the microbes that digest plant cell walls. These microbes, in turn, may serve as a source of N for the animal (especially for ruminants). It is unlikely that microbial N contributes much to the N requirement of the horse, however (Pagan, 1998). The energy and N provided by forages is

often enough for the animal to live a productive life; only pregnant, lactating, or growing animals need a secondary source of energy and perhaps, N (Ball et al., 2002). Hardworking animals, such as racehorses, also need more energy than even high quality forage can supply. The N requirement for hardworking horses may or may not be greater than that of maintenance, depending on the condition of the animal (NRC, 1989).

Nitrogen requirements are expressed as protein requirements for because particular amino acid requirements have yet to be elucidated for the majority of horses and ruminants. The only individual amino acid requirement known is lysine for growing horses, which need an adequate supply of lysine for optimum growth rates (NRC, 1989). When typical ingredients (mid- to high-quality forage) are used to meet protein requirements, essential amino acids are adequately supplied. Non-protein nitrogen (NPN) sources, such as urea, are not necessary for horses. Including urea in the diet of a horse may or may not show an increase in nitrogen balance. Horses on low-N diets may use some urea N to meet daily N requirements, however, growing horses cannot achieve maximum growth rates and lactating mares cannot produce maximum milk production when urea is the major portion of the N requirement. Urea can be beneficial to cattle on high-grain diets, but it may not be of any benefit to ruminants on a high-forage, low-protein diet (it may not provide sufficient N for the microbes). When natural feedstuffs are used in feeding both horses and cattle, the protein (and hence amino acid and N) requirements are generally satisfied; however, cattle may need another source of rumen degradable protein, aside from NPN (NRC, 1989; NRC, 2000).

Therefore, forage needs to be the basis of every grazing livestock's nutritional program. Wild and pastured horses would normally spend as many as 18 hr a day grazing (Thorne et al., 2005). The chewing process releases saliva to buffer the acidity of the stomach (or the rumen in

ruminants) and also serves to occupy the animal. Ruminants also spend the majority of their time chewing or ruminating (Church, 1988). For example, it is known that increasing the amount of fiber in dairy cattle diets motivates the cow to chew more, thus contributing more saliva to buffer the acidity of the rumen (Mertens, 1997). Without the salivary buffer system working, the rumen becomes more acidic, the acetate to propionate ratio is reduced, and the animal has nothing to occupy itself with; all of which results in a decrease in animal production and health (Jung and Allen, 1995; Mertens, 1997).

Grasses Versus Legumes

There are many differences between grasses and legumes, however, both are important in the diet of ruminants and other herbivores. Grasses and legumes differ in morphology, nutritive value (chemical composition), voluntary intake (palatability), and digestibility.

Morphology

There are many differences and some similarities between the morphology of grasses and legumes. For instance, the leaves of grasses are flat and narrow and attached to a node (a swelling of the stem); whereas legumes have leaves that are generally more rounded, usually in groups of three (such as clovers), and each leaf (or group of leaves) are attached to the stem with a petiole or petiolule (Barnes et al., 2003).

There are two types of stems in grasses: the non-reproducing or vegetative tiller (vertical stem with leaves) that is short with nodes and unelongated internodes; and the flowering or reproductive culm where the shoot apex (growing point) has differentiated into an inflorescence (the reproductive structure) and has elongated internodes. A few other important structures that are a part of the stems include rhizomes, stolons, and axillary buds. Rhizomes are underground stems that are projected laterally from the main tiller. They produce roots at each node, which aid in vegetative propagation. Stolons are creeping, lateral stems that are aboveground, but are

otherwise similar to rhizomes, producing roots at each node and aiding in vegetative propagation. Bermudagrass has both rhizomes and stolons, which aid in their suitability for the Southeastern United States. Axillary buds are formed at the axil of each leaf and contribute new tillers, especially if the shoot apex (the tissue which generates node, leaves, and axillary buds) is removed. The shoot apex is usually found close to the ground and protected by tillers and leaves, so it is unlikely that grazing or harvesting will remove it. Grazing and harvesting is more likely to be detrimental to grasses if it is close and frequently defoliated, thus robbing the stems and roots of the main producers of carbohydrates and N-containing compounds and killing the plant (Barnes et al., 2003).

The stems of legumes do not have different types for reproducing or non-reproducing stems (except for the presence of flowers). The shoot apex of legumes is located at the ends of the stems. The position of the tops of the stems, however, varies between species. Alfalfa and sweetclover, for instance, display the stems at the top of the canopy, leaving them vulnerable to removal by grazing or harvesting. Legumes like alfalfa will then regrow from axillary buds. Other legumes, such as red and alsike clovers, display their leaves at the top of the canopy, protecting their shoot apices. This is one of the reasons why red clover is commonly used for grazing, especially in mixed grass/legume swards. Similar to grasses, legumes may have stolons, such as white clover (which keeps the shoot apices near the ground) to aid in vegetative propagation. The axillary buds are located at each axil of each leaf (or group of leaves) and contribute to branching and flowering or regrowth in the case of a lost shoot apex (Barnes et al., 2003).

The dominant root systems of grasses and legumes are also distinctive. Grasses have adventitious and fibrous roots (several small branches) whereas legumes have taproots (one main

root that grows downward with few to several branches). Stolons and rhizomes (of either grasses or legumes) have adventitious roots at each node. Both root systems vary in depth and (or) branching for each species. Grasses with deeper root systems are more drought tolerant, such as smooth Bromegrass, bahiagrass, and bermudagrass. Kentucky bluegrass, however, has shallow root systems, making it more susceptible to droughts, and therefore unable to grow well in the Southeastern US (Barnes et al., 2003).

The roots of grasses are not generally the sites for storage, and will grow as long as the plant is not frequently and closely defoliated. However, perennial and biennial legumes store carbohydrates and N-containing compounds in their roots, making defoliation less of a problem, making them more suitable for intense grazing (Barnes et al., 2003).

The flowers of grasses and legumes are perhaps the most dissimilar from each other. Grasses have florets, which are found on spikelets, the basic reproductive unit. They usually have three stamens. The inflorescence displays the spikelets with different grass species having different arrangements of spikelets on the inflorescence. Most grasses are cross-pollinated by wind and some are self-pollinated within the floret. Temperate grasses generally flower once a year after a specific sequence of events (fall, winter, spring). Warm-season grasses, such as bermudagrasses, usually flower more than once in a growing season because they do not have the cold temperature (winter) requirement. After the tiller produces an inflorescence and seed, it dies. Therefore, each tiller of perennial grasses lives about 12 to 18 months (Barnes et al., 2003).

Most legumes have flowers displayed in a “butterfly-like” arrangement (papilionaceous). There are five petals: one standard, two wings, and a keel, which is two petals more or less fused together. At the base of the petals is the calyx, comprised of five sepal petals surrounding and protecting the corolla. The corolla is the tube that is formed by the flower petals. At the base of

the corolla, nectar is produced attracting bees and other insects. The corolla varies in length, and longer corollas prevent many bees and insects from reaching the nectar, affecting cross-pollination. The ovary, style, and stamens (usually 10, with one free and the other nine fused at the base) are enclosed by the keel. Some legumes are self-pollinating and almost completely self-compatible. Several legumes are self-sterile or mostly so. After flowering, the plant may or may not die, depending on whether it is an annual, perennial, or biennial (Barnes et al., 2003).

The seed unit of grasses is the caryopsis, which is mostly composed of endosperm (the source of “food” for the growing seed). The caryopsis is protected by the lemma and palea, which may prevent absorption of water (the first step to germination). Legumes have a fruit, which is a pod that contains several seeds. There is no endosperm at maturity; instead, reserve food is stored in two cotyledons. Many legumes have “hard seed”, where the seed is alive, but is protected by an impervious seed coat that restricts water uptake. Only after time underground where microbes soften the seed coats, and cycles of hot, freezing temperatures, wetting, and drying, is the seed allowed to germinate (Barnes et al., 2003).

Once the seed of either grasses or legumes germinates, it begins to grow in different areas to reach the soil surface. Grasses grow by elongating either the coleoptile, which houses the true leaves (festucoid, mainly cool-season grasses) or the subcoleoptile internode (panicoid, mainly warm-season grasses). Grasses have hypogeal emergence, where the coleoptile is pushed upwards through the soil surface by the epicotyl. Once the leaves emerge, photosynthesis begins. Most legumes have epigeal emergence, where the cotyledons (the first structures to emerge and begin photosynthesis for leaf growth) are pushed above the soil level by elongating the hypocotyl. The epicotyl or hypocotyls form an arch to break the surface of the soil and then

straightens once hit by light. Once the cotyledons emerge, they open and begin photosynthesis (Barnes et al., 2003).

Influence of Fertilization

Most fertilization is done to address a soil deficiency in certain minerals or nutrients (such as Ca), unwanted soil characteristics (acid pH), or to boost plant production. However, there is one distinction between grasses and legumes: N. Legumes have a symbiotic relationship with bacteria, collectively called rhizobia that are found in nodules on the roots. These bacteria fix atmospheric N for the plant while receiving carbohydrates from the plant. The N that the legumes receive is vital to the survival and growth of the plant. Therefore, N fertilization is not usually needed for legume fields. However, grasses, which only obtain N from the soil, respond favorably to N fertilization, increasing DM production and CP content (Johnson et al., 2001; Barnes et al., 2003).

Effect of Frost

Frost is a severe weather condition that can kill plants. Grasses are generally more frost tolerant because of their hypogeal emergence, which keeps the coleoptiles protected underground. Legumes are generally more susceptible to frosts because most have epigeal emergence, which does not leave the cotyledons underground (Barnes et al., 2003).

Nutritive Value and Animal Performance

In general, legumes have greater nutritive value than grasses. However, the nutritive value of plants is dependent on species, leaf to stem ratio, maturity, weather, amount of fertilization, soil characteristics (pH and mineral concentration, for example), and pests that may harm or kill the plant before harvest. Furthermore, even if the forage was of high quality (good nutritional value, little antiquality factors such as tannins, and of a reasonable potential intake), the animal performance is still difficult to predict and reproduce (Schneider and Flatt, 1975). The plant-

animal interface is difficult to research and therefore much of the knowledge of what makes forage nutritious for animals remains imprecise. Nonetheless, nutritive value and quality are becoming easier to determine; and with grazing and metabolism trials, significant advances have been made to ascertain what the best forages are for each animal species and situation (Barnes et al., 2003).

Grasses and legumes have different cell wall compositions that affect their nutritive value. For instance, the cell walls of grasses tend to have three to four times as much hemicellulose than legumes, whereas legumes tend to have more of the highly digestible pectins in their cell walls (Norton and Poppi, 1995; Barnes et al., 2003). The leaves of legumes also contain less cell wall material than the leaves of grasses (Jung and Allen, 1995). Cell wall material is generally the most difficult to digest. As a plant ages, the concentration of the cell walls increases; however, legume leaves do not exhibit such increases, rendering them more digestible than grass leaves at similar maturities (Jung and Allen, 1995). Legume cell walls usually have greater concentrations of lignin than grass cell walls, however, this difference is inflated by the commonly used acid-detergent lignin method of analysis (Jung and Allen, 1995; Norton and Poppi, 1995). Even though legumes often contain greater amounts of lignin than grasses, the lignin is generally located in the stem vascular tissue, whereas in grasses it is located in many different cell types (Norton and Poppi, 1995).

Plant anatomy and morphology may also differ between grasses and legumes, which has an impact on nutritive value. Grasses and legumes are typically divided between two general classes: cool-season and warm-season. Cool-season grasses and legumes typically have greater nutritive value than their warm-season counterparts, however, the difference is greatly accentuated when comparing cool-season and warm-season grasses. Most warm-season grasses

have a unique leaf anatomy, the C4 “Kranz” anatomy, which enables the plant to use a more efficient photosynthetic pathway to survive in the harsher conditions of tropical or subtropical regions (often higher temperatures) than those found in temperate areas. A feature of this type of leaf anatomy is the parenchyma bundle sheath, which is thicker and arranged in such a way as to limit bacterial digestion, thus decreasing overall digestibility (Coleman et al., 2004).

Harvesting and storage may also affect nutritive value differently in grasses and legumes. The process of harvesting causes physical losses, such as leaf shatter (a known problem when harvesting annual peanut) and loss, and increased respiration, which decreases the concentration of digestible carbohydrates (Barnes et al., 2003). Rain damage during harvest will compound the physical losses with the leaching of soluble constituents, thereby decreasing the nutritive value of forages. Legumes differ from grasses in that they are more susceptible to rain damage (Barnes et al., 2003). Other problems associated with harvesting and storage that affect both legumes and grasses equally include weathering, aerobic conditions in silage or haylage, and overheating.

Primary Forages in the Southeastern United States

In the Southeastern United States, the prominent grasses and legumes are of low nutritional quality. The grasses that grow well in the soils and weather of this subtropical region are predominantly warm-season grasses, which are generally lower in quality than cool-season grasses. Some warm-season legumes are of higher quality, but there are few choices. Cool-season legumes do not grow well in the acidic sandy soils, and the high rainfall in the spring and potential drought conditions in the summer pose problems for cool-season grasses as well.

In general, the Southern U.S. can be separated into four different zones, but for the purposes of this thesis, the zones will be condensed into two: the upper and lower South. The upper South includes the Southern parts of Kentucky, Tennessee, and the northern regions of

Mississippi, Alabama, and Georgia. The lower South includes the Southern regions of Mississippi, Alabama, and Georgia, along with the Gulf Coast of Florida and peninsular Florida.

In the lower Midwest and upper South, cool-season grasses grow moderately well and provide quality forage for livestock. Breeding programs have produced adapted varieties of timothy and orchardgrasses. These are high quality grasses and can serve as forage for horses, which require higher quality forages because of their decreased capacity to ferment cellulose and hemicellulose compared to ruminants (Ball et al., 2002).

Warm-season perennial grasses are also becoming more popular in the upper South. Big bluestem, Caucasian bluestem, switchgrass, and indiagrass are used as forage in livestock production. These grasses may not be as nutritious as cool-season grasses if made into hay, but as young pasture, they can be used for grazing herds of horses (Ball et al., 2002).

Annual ryegrass and cereal grains (oats, wheat, and rye) are also important to livestock operations, especially for winter annual pastures. Breeding work on perennial clovers, such as red clover, has produced legumes that grow well throughout the South, yielding high quality legumes, which have previously not done well in low-fertility soils, subsoil acidity, heat, and drought; all characteristics of the Southern U.S. Varieties of red clover, such as 'Kenland' and others, are more disease resistant and are now produced on a wider scale than before. Ladino white clover varieties, such as 'Regal', and intermediate white clovers, such as 'Louisiana S-1', are also grown all over the South and extend winter grazing periods (Ball et al., 2002).

Other legumes that grow well in the upper South include alfalfa, Common annual lespedeza, and Korean annual lespedeza. Breeding work has also been done on these legumes to improve their suitability to growing in the upper South. Certain varieties of alfalfa may even

grow moderately well in the Gulf Coast; however, the expense of maintenance may override its use in livestock production for the lower South (Ball et al., 2002).

In the lower South and along the Gulf Coast, weather patterns and soils are extreme, necessitating the use of different species or improved cultivars. For instance, common bermudagrass grows well in the lower South; however it is not generally nutritious. The work of G. W. Burton at the University of Georgia has produced many improved cultivars of bermudagrass, notably 'Coastal' and 'Tifton 85' bermudagrasses. These grasses grow well, are productive, and offer potential nutritious forage for livestock (Ball et al., 2002).

Bahiagrass is also popular for livestock operations in the lower South. Bahiagrass is not of sufficient quality for horses, especially as hay or silage, but it is grazing tolerant and best used for grazing horses (Lieb, 2003).

Legumes that grow well in the lower South include varieties of sericea (a perennial lespedeza), Berseem and other rose clovers, and perennial and annual peanuts. Breeding work has proved successful for sericea, where low-tannin, fine-stemmed varieties have improved palatability and nutritive quality. Berseem and other rose clovers are not well known, but they can be used as winter annuals in certain areas of the lower South. Perennial and annual peanuts grow extremely well, and can be as nutritious as alfalfa. Perennial peanut has very few pest problems, however, older varieties of annual peanut are susceptible to many pests. More recent varieties of annual peanut have fewer problems with pests (Gorbet et al., 1994). Perennial peanut also makes good quality hay or pasture (Gelaye et al., 1990; Williams et al., 1991; Williams et al., 2004; Hammond et al., 1992; Lieb et al., 1993; Bennett et al., 1995; Staples et al., 1997; Hernández Garay et al., 2004).

The Southeastern U.S is certainly not lacking in varieties of grasses and legumes. However, it is not as simple to grow high quality forages in the Southeast as it is in more temperate regions because of the acidic, sandy soils, heat, low soil fertility, and the potential drought conditions (Ball et al., 2002). Instead, different strategies must be used to provide the livestock industry of the Southeast with high quality forages.

Digestive System of a Horse

Horses are characterized as hindgut fermenters. They rely on symbiotic bacteria to breakdown fibrous particles, much like a ruminant, however, this occurs predominantly in the large intestine, rather than the forestomach. Horses can only digest around 30 to 50% of the fiber in forages because of the much smaller pre-intestinal capacity and lower microbial activity than ruminants. Therefore, forages provided must be of high quality (immature, low indigestible fiber, high digestible fiber, high protein, and lacking in antinutritive factors) for optimal performance (Pagan, 1998; Ball et al., 2002).

Basic Anatomy of the Horse's Digestive System

Starting with the mouth, the horse chews the food, producing saliva to moisten the food, preventing it from being lodged in the esophagus. The food boluses travel down the esophagus by means of peristaltic waves and are deposited in the stomach. It is generally uncommon for the esophagus to become obstructed and the horse to choke. The stomach of a horse is actually the smallest part of the digestive tract. It is small because the horse evolved to eat small meals continuously throughout the day. Once in the stomach, acid hydrolysis and enzymatic digestion of protein commences. The partially digested food does not remain in the stomach for long, however, and quickly passes into the small intestine (Frape, 1998; Jackson, 1998).

The small intestine averages 21 m long and can hold about 56 L of ingesta in the mature horse. The continuous flow of pancreatic juice buffers and digests the acidic ingesta coming

from the stomach. This is the major zone of absorption of simple sugars, amino acids, free fatty acids, and fat-soluble vitamins (A, D, E) and some minerals in the digestive tract of horses. The cecum and colon come after the small intestine and are analogous to the rumen and reticulum of ruminants. The symbiotic bacteria, fungi, and protozoa in the cecum digest cellulose and other fibrous compounds in the feed. Cecal and colonic microbes also synthesize B vitamins and vitamin K (Frape, 1998; Jackson, 1998).

The major difference in the digestive processes of horses and ruminants is that, in horses, fermentation occurs after the digestion of carbohydrate, protein, and fat in the horse; as opposed to before the small intestine in the ruminant. This leads to differences in efficiency of plant cell wall breakdown and absorption. Horses are not as efficient at fiber breakdown when compared to ruminants. The microbes in the cecum and colon of horses can only digest about 30 to 50% of the plant cell wall, whereas ruminants can digest up to 90% of the cell wall (Pagan, 1998; Ball et al., 2002). Therefore, ruminants can live on lower quality forages than horses since ruminants can digest more of the plant cell walls and fiber. The small intestine is better suited to absorption of compounds than the large intestine. As a result, compounds produced from enzymatic breakdown and fermentation in the rumen and reticulum are absorbed in greater quantities than those of the stomach in horses (Pagan, 1998).

The site of fermentation also has an effect on protein metabolism. Some of the microbes that fermented the food travel into the small intestine of ruminants and can be broken down. As a result, some of the microbial protein is absorbed as well as any protein that wasn't broken down by the microbes in the rumen. Therefore, ruminants do not necessarily need high-protein forages, unlike horses. Microbial protein may or may not be absorbed by the large intestine of horses; if it

is, it is not a major contribution of protein. Horses require high-protein forages, otherwise growth and performance suffers (Pagan, 1998; Ball et al., 2002).

General Horse Nutrition

Feeding programs for horses should be as close to the horse's natural diet as possible. Horses evolved to eat small continuous meals consisting of mostly forage and any departure from this will have the largest effects on gut function and motility (and consequently on the behavior of the horse). The NRC (1989) for horses recommends that all horses should have access to pasture or be fed at least 1% of their body weight/d (DM basis) of good quality forage. If horses have access to pasture and forage *ad libitum*, the horses will voluntarily consume 2 to 2.5% of their BW in a 24 hr period (NRC, 1989). However, some horses require more energy than forage can provide alone, therefore, concentrates are fed, but should be offered in small portions throughout the day and have at least 1 kg of forage DM/100 kg of BW/d.

Other considerations when feeding horses include maintaining feeding programs with no drastic changes, feeding at optimum inclusion rates (e.g. corn should not be more than 23% of a concentrate to avoid digestive disorders and obesity caused by the low fiber and high energy composition of corn), proper storage of feed to prevent mold, rancidity, and loss of feed nutritional quality, feeding by weight and not volume, regular exercise, and proper dentition and deworming (Jackson, 1998).

Importance of Forages in the Diet of Horses

Along with water, forages are required for every horse. Forages provide nutrients and maintain proper gastrointestinal function. Microorganisms in the horse's cecum or large intestine require a source of digestible fiber for energy. Indigestible fiber, which is also high in forages and low in grains, is also needed to maintain gastrointestinal pH, motility, and function of the

digestive system. Too much grain and not enough forage in a horse's diet can lead to harmful and potentially deadly symptoms, such as diarrhea and colic (Lewis, 1996; Pagan, 1998).

The horse has evolved to eat forages and if not given the opportunity to graze or eat forage, it may lead to stable vices such as wood chewing and tail chewing (Lewis, 1996, Pagan, 1998; Thorne et al., 2005). Feeding hay will increase feeding time, chewing activity, and saliva production, thus keeping the horse occupied (Zeyner et al., 2004). Feeding hay also has a soothing effect on microbial fermentation compared to feeding grain (Zeyner et al., 2004).

Zeyner et al. (2004) conducted a study to investigate the influence of the amount of hay fed on intestinal characteristics such as pH and short-chain fatty acid (SCFA) concentration and whether the sequence of feeding hay and a constant amount of oats (1.00 kg crushed oats/100 kg BW \times d) affect these parameters. Horses were fed oats 30 min before orchardgrass hay or in the reverse order in a crossover design. The amount of hay offered was increased gradually in the first period from 0.50, 0.67, and 0.83 to 1.00 kg/100 kg BW \times d and in the second period reduced gradually using the same amounts. Oats was offered three times a day (morning, noon, and evening) and hay was offered twice a day (morning and evening). They found that horses consuming 1.00 kg hay/100 kg BW \times d had a more balanced hindgut environment with high amounts of cellulolytic bacteria even though dietary grains were consistent. Feeding lower amounts of hay was related to adverse behaviors in the horses, including nervousness, restlessness, and aggressive behavior at feeding times. In addition, the horses also tended to eat their own feces. None of the horses exhibited these behaviors when fed 1.00 kg hay /100 kg BW \times d, regardless of whether oats were fed before or after the hay. Increasing hay intake in the range of 0.50 to 1.00 kg hay/100 kg BW/d causes a dose-dependent and significant increase in pH and the molar percentage of acetate in the feces/fecal water mixture. This also causes a

decrease in the concentration of SCFA and the molar percentage of propionate. These results indicate that hay is important in providing the proper buffering capacity. The researchers concluded that horses should be fed at least 1.00 kg hay/100 kg BW × d to sustain proper intestinal and likely metabolic function as well as eliminating behavior abnormalities.

Coastal and Tifton 85 Bermudagrasses

Coastal bermudagrass is a warm-season grass that grows well in the deep South (Ball et al., 2002). Researchers, however, have been experimenting with different hybrids to find one or more cultivars that grow well and have a better nutritive value than Coastal. The 'Tifton 85' cultivar has so far been the most promising of this hybrid research (Ball et al., 2002).

Agronomic Aspects of Tifton 85 Bermudagrass

Visually, Tifton 85 bermudagrass grows taller than other bermudagrasses, has larger stems, broader leaves, and is a darker green in color (Hill et al., 1993). Tifton 85 is rhizomatous, like Coastal, but has fewer rhizomes than Coastal or 'Tifton 44' bermudagrass (Hill et al., 1993). However, like Coastal, Tifton 85 bermudagrass needs to be fertilized with N in order to have adequate protein levels for ruminants (Ball et al., 2002).

Tifton 85 bermudagrass also has a greater dry matter (DM) yield than Coastal bermudagrass. Hill et al. (1993) conducted one of the first experiments to compare Tifton 85 to other bermudagrasses and found 26% greater DM yield and 11% greater in vitro dry matter disappearance (simulates ruminant digestion) than Coastal in two 3-year trials.

Also like Coastal, the forage quality of Tifton 85 decreases dramatically with stage of maturity. The age of plants when harvested usually has the greatest effect on the nutritive quality of plants than any other factor (Ball et al., 2002). Mandebvu et al. (1999) recommends that Tifton 85 bermudagrass be harvested at 3 to 5 wk of regrowth to obtain the best possible nutritional quality.

Nutritional Aspects of Tifton 85 Bermudagrass

As mentioned above, Hill et al. (1993) found Tifton 85 bermudagrass has a higher in vitro dry matter disappearance than Coastal bermudagrass. Mandebvu et al. (1999) conducted a more detailed study of the differences between Coastal and Tifton 85 bermudagrass. They found Tifton 85 to have greater concentrations of neutral detergent fiber (NDF) and acid detergent fiber (ADF) than Coastal; but Tifton 85 was still more digestible than Coastal bermudagrass in DM, NDF, and ADF in growing beef steers. Coastal bermudagrass also had a greater ether-linked ferulic acid lignin concentration than Tifton 85. Ether-linked ferulic acid is thought to be one of the ways lignin restricts cell wall degradation by rumen microbes (Jung and Allen, 1995). They also found that steers fed more mature Tifton 85 bermudagrass had decreased digestion of NDF, ADF, hemicellulose, and cellulose, similar to that found with increased maturity in Coastal.

Three studies have compared the digestibilities of Coastal and Tifton 85 bermudagrasses in horses. McCann et al. (1995) found that both bermudagrasses had similar digestibilities. Dry matter digestibility of Coastal and Tifton 85 bermudagrasses averaged 52.4 and 54.1%, respectively. Crude protein digestibility of Coastal and Tifton 85 bermudagrasses was 65.4 and 63.5%, respectively. Coastal and Tifton 85 bermudagrasses had similar NDF and ADF digestibilities; 54.6 and 57.2%, and 56.5 and 55.0%, respectively. There was no data on maturity of the grasses or intake by the horses, which may have affected these results.

Lieb and Mislevy (2001) compared the intake and digestibility of three subtropical grass forages (Florona stargrass, and Tifton 85 and Florakirk bermudagrasses) to the intake and digestibility of Coastal bermudagrass. All hays were offered *ad libitum* for 14 d and then at 80% of *ad lib* forage intake for 10 d, with a total fecal collection the last 5 d of each period. Tifton 85 (harvested after 35 d of regrowth) was similar to Coastal (harvested at a similar time) in DM, NDF, and energy digestibilities. Dry matter digestibilities of Coastal and Tifton 85

bermudagrasses were 44.7 and 48.1%, respectively. Neutral detergent fiber digestibilities of Coastal and Tifton 85 bermudagrasses were 46.4 and 51.4%, respectively. Energy digestibilities of Coastal and Tifton 85 bermudagrasses were 41.9 and 44.6%, respectively. Crude protein digestibility was greater in Coastal than Tifton 85 (54.4 and 48.4%, respectively; $P < 0.05$). Coastal bermudagrass had a greater CP content than Tifton 85 (13.2 and 9.1%, respectively). Average daily DM intake (as % of BW) was greater ($P < 0.05$) for Coastal than Tifton 85 (2.58 and 2.10%, respectively). Tifton 85 and Coastal bermudagrasses produced average body weight changes of -11.8 and 5.5 kg/28d, respectively. It was concluded that even though Tifton 85 bermudagrass produced similar digestibilities to Coastal bermudagrass, the reluctance of horses to eat Tifton 85 bermudagrass and the inability of Tifton 85 to maintain a positive body weight suggests that Tifton 85 should be offered with other nutrient sources.

Lieb and Mislevy (2003) repeated their first study with the same forages; only they were visually of better quality, having finer leaves, a fresher smell, and less foreign plants. Crude protein content was greater for Tifton 85 and lesser for Coastal (16.6 and 9.3%, respectively). Average daily DM intake (as % of BW) was lower for all hays compared to the previous trial. Tifton 85 and Coastal intakes were 1.71 and 1.81%, respectively. Coastal and Tifton 85 bermudagrasses produced positive average body weight changes (4.54 and 2.27 kg/14 d, respectively). Crude protein digestibility was 44.4 and 57.3% in Coastal and Tifton 85 bermudagrasses, respectively, with Tifton 85 being greater than Coastal bermudagrass ($P < 0.05$). Coastal and Tifton 85 bermudagrasses had different NDF digestibilities (33.5 and 50.2%, respectively; $P < 0.05$). Energy digestibility was 48 and 42.7% for Coastal and Tifton 85, respectively, with Tifton 85 being greater than Coastal ($P < 0.05$). The researchers concluded that Tifton 85 could be equal to or as good as Coastal bermudagrass in digestibility; however,

intake of Tifton 85 is not sufficient to maintain the body weight of mature horses and should not be fed without supplemental nutrient sources.

Perennial and Annual Peanut

One of the most promising warm-season legumes is perennial peanut (*Arachis glabrata*), which differs from annual peanut (*Arachis hypogaea*) in that it is vegetatively propagated with rhizomes, hence the common name of “rhizoma peanut”. The University of Florida produced the well-known cultivar 'Florigraze' in 1981, which grows well in the Southern part of the peanut belt (Gorbet et al., 1994; Hill, 2002).

Annual peanut (*Arachis hypogaea*) forage has not been extensively researched in either ruminant or non-ruminant nutrition, even though hay from residual vines after seed pod harvest has been fed for over 75 yr in the U.S (Hill, 2002). Annual peanut grown specifically as forage and harvested as hay may be as nutritious as alfalfa hay.

Agronomic Aspects of Perennial and Annual Peanut

Perennial peanut is most commonly grown in north Florida, Southern Georgia, and Southern Alabama (Hill, 2002). Approximately 11,000 hectares of perennial peanut was being grown in Florida alone in 2005, and the number is increasing (French et al., 2006).

Perennial peanut is vegetatively propagated from rhizomes and therefore does not need to be replanted every year (Hill, 2002; French et al., 2006). Perennial peanut is drought resistant, once established, and is resistant to many diseases and pests (French et al., 2006). The one drawback to perennial peanut is its comparatively slow establishment period, often taking nearly 2 to 3 yr to entirely cover fields (Hill, 2002). Eventually however, the high yields and lower production costs in later years cover the cost of establishment. In Florida, perennial peanut yields anywhere from 7 to 13 Mg/ha each year and 8 to 10 Mg/ha each year in Texas when cut two to three times annually (Bennett et al., 1995).

Perennial peanut is associated with N-fixing bacteria and therefore does not need to be fertilized with N. Its association with mycorrhizal organisms also increases the availability of P to the plant, reducing the need for P fertilization (French et al., 2006).

Annual peanut plants do not need to be fertilized with N because it is a legume, however, most cultivars will need fungicide application. Many cultivars of *Arachis hypogaea* are prone to leaf spot caused by *Cercospora arachidicola* or *Cercosporidium personatum*, but can be controlled with effective fungicide. Gorbet et al. (1994) found that recent cultivars are more resistant to late leaf spot than 'Florunner', a commonly planted cultivar of annual peanut at that time. They also reported similar forage yields of the ten cultivars to that of perennial peanut.

Nutritional Aspects of Perennial Peanut and Annual Peanut

Many studies have been done on the nutritive value of perennial peanut in ruminants, but very few for non-ruminants. Hammond et al. (1992) reported the suitability of perennial peanut hay as a protein source for wintering cows on low protein hay. The perennial peanut hay used averaged an in vitro organic matter disappearance (IVOMD) of 58.7% and a CP (as a percent of DM) of 12.2%. Gelaye et al. (1990) conducted a metabolism trial with goats fed perennial peanut hay and compared it to that of alfalfa hay. Dry matter, OM, NDF, and ADF digestibilities of perennial peanut hay (62.5, 64.0, 49.5, and 48.8%, respectively) were greater ($P < 0.05$) than those fed alfalfa (55.7, 55.6, 33.9, and 33.8%, respectively). Crude protein digestibility was similar in perennial peanut and alfalfa hays (72.7 and 71.4%, respectively). Goats fed perennial peanut had improved feed to gain ratio compared to those fed alfalfa. These researchers also found perennial peanut to have greater gross energy (60.2%), hemicellulose (50.2%), and cellulose (58.9%) digestibilities than alfalfa (55.1, 34.5, and 45.6%, respectively).

Perennial peanut mixed with bermudagrass is a good pasture for use for growing stockers. Williams et al. (1991) performed a 2-year study with steers grazing perennial peanut-mixed

pastures and the steers had a greater ADG (0.79 kg) than those grazing bahiagrass only pastures (0.51 kg).

The only study that researched the digestibility of perennial peanut in horses was done by Lieb et al. (1993). The researchers found that perennial peanut hay was similar to alfalfa in DM (56 and 62%, respectively), CP (70 and 79%, respectively), and NDF (43 and 45%, respectively) digestibilities, as well as digestible energy (55 and 61%, respectively). They also found that the perennial peanut hay was readily accepted by the horses and the voluntary intake was greater for perennial peanut than alfalfa or Coastal bermudagrass.

Annual peanut hay is highly nutritious when fed to ruminants. Ronning et al. (1953) found annual peanut hay that was harvested as a whole plant, dried, and then nuts removed to be nutritionally similar (total feed intake, hay intake, and milk production) to alfalfa in dairy cows. McBrayer et al. (1981) reported similar total digestible nutrients (TDN) in dairy cattle when fed either concentrates plus coarsely ground alfalfa hay, unground annual peanut hay, and ground annual peanut hay.

Gorbet et al. (1994) reported an average IVOMD of 70.4% for the first cutting of ten cultivars of annual peanut. The second cutting produced an average IVOMD of 63.9% of the ten cultivars, however, when cut only once at the end of the growing season, the average IVOMD dropped to 59.6%. The ten cultivars had an average CP content of 16.9 and 16.5% in the first and second cuttings, respectively.

In Nigeria, Africa, annual peanut (called “groundnut”) forage has been recommended by Larbi et al. (1999) as a supplement to cattle during the dry season for smallholder crop-livestock systems. They investigated 38 late-maturing annual peanut cultivars and found that all the cultivars had higher leaf and stem CP concentrations than maize, millet, and rice, which are

typically low in N. Leaf and stem DM losses after 48 hours of incubation were 60.8-77.5% and 47.9 to 59.6%, respectively. The study concluded that breeding programs could be developed to maximize forage quality and yield as well as seed yields to maximize profits.

There has been no research in the digestibility of annual peanut hay in non-ruminants. It may be that annual peanut hay is nutritionally similar to alfalfa hay for horses. Annual peanut hay grows well in Southern climates, like perennial peanut, and may be less expensive for horse owners in the Southern state.

CHAPTER 3
DIGESTIBILITY AND NUTRIENT BALANCE OF PERENNIAL PEANUT AND
BERMUDAGRASS HAYS BY MATURE HORSES

Introduction

Perennial peanut (PP, *Arachis glabrata*) is rapidly becoming popular as a forage legume for horses in the Southeast United States. Perennial peanut grows well in the acidic soil, which is characteristic of the Southeast regions and is not prone to disease and insect pests (French et al., 2006). Perennial peanut has been found to be highly nutritious for ruminants (Gelaye et al., 1990; Williams et al., 1991; Williams et al., 2004; Hammond et al., 1992; Bennett et al., 1995; Staples et al., 1997; Hernández-Garay et al., 2004). Gelaye et al., (1990) reported that goats fed PP had greater DM, OM, NDF, ADF, and hemicellulose digestibilities than alfalfa hay. The goats also voluntarily ate more PP than alfalfa. Hammond et al. (1992) found that PP forage is a good supplement feed for wintering cattle, especially for those on low protein grass hay. Not much is known, however, about the nutritional value of PP in non-ruminants, specifically horses. Lieb et al., (1993) conducted the only study to investigate the digestibility of PP in horses. These researchers found PP to be nutritionally similar to alfalfa in digestible energy, as well as DM, CP, and NDF digestibilities. Perennial peanut was also readily accepted by horses; voluntary intake was greater for PP than for alfalfa.

Coastal bermudagrass (*Cynodon dactylon*, CB) is a popular source of forage fed to horses and ruminants. Researchers with the USDA-ARS and the University of Georgia at the Coastal Plain Experiment Station in Tifton, Georgia have released a hybrid bermudagrass, called Tifton 85 (T85) that has been shown to be nutritionally superior to CB in ruminants (Hill et al., 1993; Mandebvu et al., 1999). Tifton 85 also has improved productivity and is more vigorous than previously released bermudagrass hybrids, including CB (Chambliss and Dunavin, 2003). McCann et al. (1995) found no differences between the digestibilities of DM, CP, NDF, and

ADF between CB and T85 in mature horses. Lieb and Mislevy (2001, 2003) found variability in the digestibility of T85 and CB hays in mature horses. In the first experiment, T85 had similar digestibilities to CB (except for CP digestibility, which was greater in CB). Horses did not consume as much T85 as CB and had trouble maintaining their body weight when fed T85. In the second trial, T85 had greater digestibilities than CB, however, the horses were reluctant to eat T85. Our objectives were to evaluate and compare apparent DM, OM, CP, and NDF digestibilities, as well as Ca, P, and nitrogen balances in mature horses fed PP, CB, or T85 hays.

Materials and Methods

Animals and Experimental Design

In this experiment, six horses were used to estimate various digestibility characteristics of three hays (PP, CB, and T85) that lasted 52 d at the University of Florida, Gainesville, Florida. One horse was unable to complete the trial and was removed from the study after the second collection. Five Thoroughbreds and one Quarter horse of similar weights (Mean \pm SD, 542 \pm 37 kg) were used in a 3x3 Latin Square design with two horses assigned to a group and each group randomly assigned to one of three hays for the initial period (Period 1). Because of the concern for the horse's health when transitioning from the high-quality PP hay to CB, the groups were assigned a treatment for the next two periods. Therefore, after a group was fed PP, they were then fed T85 in the next period before being offered CB.

Feeding and Housing

Horses were transported to the University of Florida main campus from the University of Florida Horse Research Center (Ocala, FL) and were allowed to acclimate to their new surroundings for 11 d before the first adjustment period began (d -11 to d 0). Three periods of a 10 d adjustment period and a 4 d total fecal and urine collection followed (d 1 to d 42). Horses were offered two equally sized meals at 0700 and 1400 daily. Diets were adjusted gradually

when changing to a new diet (%new:%old, 50:50 for two days, 60:40 for one day, 75:25 for two days). The horses were fed diets of 100% CB, T85 hay, or PP hay for at least five days prior to collection. Water and trace-mineralized salt blocks were provided *ad libitum* and, when needed (to stimulate water intake because of the bulkiness of T85), loose salt was provided at both feedings. Horses were weighed before the trial started (d -5 after horses arrived, but before the start of the first period), twice during the trial (d 23 and d 37), and after the trial was completed (d +3, at the University of Florida's Horse Research Center). A body condition score was recorded at the end of every week during the trial (scale 1-9; Henneke et al., 1983).

Horses were fed *ad libitum* before the start of the trial during the 10 d acclimatization period, and then at 2% BW during the adjustment phase of period one. However, because of the substantial amount of orts, the diets were lowered to 1.7% BW. The horses, as a result, lost weight (Mean \pm SD; 30 \pm 15 kg) and body condition score. As a result, for period 2, the amount of hay offered was increased to 2% of BW for both adjustment and collection. The horses did not improve and 3 the diets were brought to 2% BW throughout adjustment and collection periods.

Horses were housed in an 8-stall open barn in individual stalls (3 x 3 m) that were large enough for the horses to turn around and lay down in. Rubber matting covered the floor of each stall. Stalls were bedded with pine shavings during the adjustment phase; all bedding was removed during the collection phase of each period. During the trial, horses were restricted to 3 x 3 m stalls and walked by hand everyday for 15 min.

Diets

Perennial peanut hay ('Florigraze') was grown in Madison County, Florida. The hay was from a delayed first cutting in August 2005 with no rain damage and from a mature field. Both the CB and the T85 hays were grown in Suwannee County, Florida. The maturity for T85 averaged five wk and the CB four wk regrowth. There was no rain damage and both

bermudagrass hays were harvested in August 2005. After each cutting (removed at 4 – 6 wk intervals beginning in May), the bermudagrass fields were irrigated and fertilized with 90 kg of actual N/ha. In the spring, 448 kg/ha of 0-5-28 (N, P, K) were applied; and every year, 7 to 13 Mg of poultry litter/ha were applied.

Sample Collection

A random grab sample of each hay was obtained at each feeding during the collection phase of each period, compiled daily, and weighed. Any feed refusals were also collected and weighed. Total urine and feces were collected using a specialized harness from StableMaid (Melbourne, Australia) with a large plastic bag for the feces and smaller, doubled plastic bags for the urine. Feces and urine were collected at 0800, 1500, and 2300 for period 1 and at 0700, 1500, and 2300 for periods 2 and 3. At each collection, feces were weighed, homogenized, mixed, subsampled, and frozen (-4°C). Feces subsamples were compiled by 24 hr period after the trial. Urine samples were thoroughly mixed and subsampled right after each collection (~100 mL). Five milliliters of 1 N HCL was added to urine subsamples to reduce the evaporative loss of nitrogen. Urine was composited by day.

Sample Preparation and Analysis

The hay samples taken during the 4 d collection period (one of each hay for each day, totaling 12 per period) were dried at 65°C in a forced-air oven for 48 hr and weighed for dry weight. The samples were then ground through a 1mm screen using a Wiley mill (Arthur H. Tomas Co., Philadelphia, PA), mixed, and subsampled. The subsamples were then composited by hay and period, yielding nine hay samples in all (three samples for each hay). The composited hay samples were analyzed for DM, OM, NDF, ADF, CP, Ca, and P.

Hay refusals for each individual horse were collected daily during the adaptation and collection phases, dried at 65°C in a forced-air oven for 48 hr and weighed for dry weight. The refusals collected during the 4 d collection phase were ground through a 1mm screen using a Wiley mill and composited by horse and by period and analyzed for DM to determine net hay intake for digestibility determination.

After the completion of the trial, the feces were thawed and composited by horse by day by period, with day 1 starting at 1500 h the first day of collection and ending at 0800 or 0700 on the second day of collection. A 0.30 kg sample was weighed out, and dried at 60°C in a forced-air oven for 2 d and re-weighed for a dry weight. These dried subsamples were then ground through a 1mm screen in a Wiley mill. Feces were analyzed for DM, OM, NDF, ADF, N, Ca, and P.

Urine was thawed before analyses. After thawing, urine was centrifuged at 2500 rpm for 10 min (5°C) to separate out particulate matter before analysis. Urine was analyzed for N, Ca, and P.

Dry matter concentration of the hay and feces was obtained by drying samples (1.5 g; in duplicate) in an oven dryer at 105°C in an aluminum tin overnight. Afterwards, samples were put in a dessicator, allowed to reach room temperature, and weighed (AOAC, 1990). Ash was determined by placing the samples in a muffle furnace at 600°C overnight. Afterwards, the samples were placed in a dessicator, permitted to cool to room temperature, and weighed (AOAC, 1990).

Neutral detergent fiber was determined on the hay and feces samples using an ANKOM Fiber Analyzer (ANKOM Technology Corp., Fairport, NY). Following the manufacturer's instructions, 1.5 g of sample (in duplicate) was placed into an ash-free, N-free filter bag (F57;

ANKOM, Macedon, NY). For one run, as many as 24 bags (22 samples, 2 blanks) were positioned in the bag suspender, which was then put into the fiber analyzer, and 2000 mL of neutral detergent solution was added to the vessel. The samples were then heated and agitated for 75 min. Afterward, the samples were rinsed three times with hot (70 to 90°C) deionized water. The samples were then soaked in acetone for 3 min and then allowed to air-dry so that all the acetone evaporated before being put into an oven at 105°C overnight. Finally, the samples were placed in a dessicator, permitted to cool to room temperature, and re-weighed. Neutral detergent fiber concentration was determined by the amount lost during analysis.

Acid detergent fiber was determined in a similar manner as NDF using the ANKOM Fiber Analyzer. About 1.5 g of sample (in duplicate) was placed in ash-free, N-free filter bags and placed in the analyzer using the bag suspender (F57; ANKOM, Macedon, NY). Approximately 2000 mL of acid detergent solution was added and the samples were heated and agitated for 60 min. The samples were then rinsed three times with hot deionized water (70 to 90°C) and soaked in acetone for 3 min. The samples were allowed to air-dry and placed in a 105°C oven overnight, and were placed in a dessicator, cooled to room temperature, and re-weighed to determine ADF concentration.

Nitrogen concentration in hay, feces, and urine samples was determined by catalytic tube combustion using the Vario Max CN (Elementar Americas, Inc., Mt. Laurel, NJ). Hay and feces samples (2.5 to 3.5 g; in duplicate) were run after blanks and standards using the 'Aspar125' program. Urine used the same method, but only 1 mL was added, with an additional 100 mg of table sugar to facilitate combustion. Crude protein was calculated as $6.25 \times \%N$.

Calcium, P, (hay, feces, and urine) and other minerals (hay; Mg, K, Cu, Mn, Fe, Zn, and Na) were determined in triplicate by digesting the ash in nitric acid according to Miles et al.

(2001). All digested samples were then analyzed by atomic absorption spectroscopy for all minerals except P (Atomic absorption spectrometer, model 5000; PerkinElmer, Waltham, MA). Phosphorus was determined colorimetrically using a Phosphorus Microplate Reader (KC Junior software; Biotek© Instruments, Inc., Winowski, VT) according to the protocol used by Harris and Propat (1954).

Calculations

Dry matter apparent digestibility (AD) was calculated every 24 h during collection periods by using the following equation:

$$\%AD = (DMI - DME / DMI) \times 100$$

where

DMI = Dry matter intake

DME = Dry matter fecal excretion.

Organic matter, NDF, CP, and energy apparent digestibilities (AD) were calculated every 24 h during collection periods by using the following general equation:

$$\%AD = 100 \times ((DMI \times (\%OM \text{ hay [or NDF, CP] / 100)) - (DME \times (\%OM \text{ feces [or NDF, CP] / 100))) / (DMI \times (\%OM \text{ hay [or NDF, CP] / 100))).$$

Nitrogen balance (NB) was calculated (g/d) by the following equation using averages of daily total hay intake, fecal excretion, and urine excretion:

$$NB = DMI \times (\%N \text{ hay}/100) - DME \times (\%N \text{ feces}/100) - (\text{Urine Output} \times (\%N \text{ urine}/100) \times \text{SGU})$$

where

SGU = specific gravity of average horse urine, 1.028 g/d (Cohen et al., 2002).

Ca (CaBal) and P (PBal) balances were calculated (g/d) by the following equation using averages of daily total hay intake, fecal excretion, and urine excretion:

$$\text{CaBal [PBal]} = 100 \times (DMI \times (\%Ca \text{ hay [%P hay]}/100) - DME (\%Ca \text{ feces [%P feces]}/100) - (\text{Urine Output} \times (\%Ca \text{ urine [%P urine]}/100) \times \text{SGU})) / DMI \times (\%Ca \text{ hay [%P hay]}/100)$$

where

SGU = specific gravity of average horse urine, 1.028 g/d (Cohen et al., 2002).

Statistical Analysis

Data were analyzed using PROC GLM of SAS version 9 (2002). Dry matter digestibility (DMD), OM digestibility (OMD), NDF digestibility (NDFD), CP digestibility (CPD), nitrogen balance (NB), calcium balance (CaBal), and P balance (PBal) were compared between PP and

the mean of both CB and T85; and the mean of CB was compared with that of T85. Treatment means were separated with a PDIFF statement. The model for analysis was TRT = DMD OMD NDFD CPD NB CaBal PBal. Period and horse were not included in the model because they had no effect ($P > 0.05$) on the dependent variables.

Results

Forage Analysis

The nutritive value of PP hay was numerically different than either of the bermudagrass hays. Dry matter concentrations were similar for PP, CB, and T85 (Table 3-1). Ash content was the greatest numerically in PP and both the bermudagrasses were similar. Perennial peanut had a greater CP content than both CB and T85. Neutral detergent fiber was lowest in PP compared with CB and greatest in T85. For ADF, PP and CB were similar but T85 was the greatest. Calcium content was numerically highest in PP with the two bermudagrasses nearly equal. Phosphorus content was similar between PP and T85, but lowest in CB. Magnesium, K, Mn, and Fe were numerically greater in PP than the two bermudagrasses. Copper was numerically highest in CB, with PP and T85 similar. Zinc was very similar between all three hays. Sodium was numerically greatest in CB and T85 compared to PP.

Apparent Digestibility

Dry matter digestibility was greater ($P = 0.0004$) for PP hay than for the mean of the bermudagrass hays (Table 3-2); but apparent DMD of the bermudagrasses did not differ from each other ($P = 0.80$). Apparent OM digestibility had similar results as DMD, with PP having a greater ($P = 0.05$) OMD than the mean of the bermudagrass. Coastal bermudagrass and T85 had a similar OMD ($P = 0.49$). Crude protein digestibility was greater ($P = 0.003$) for PP than for the mean of T85 and CB; both bermudagrasses were similar ($P = 0.32$). There was no difference

between PP and the mean of the bermudagrass hays in apparent NDF digestibility ($P = 0.51$) and no difference between CB and T85 ($P = 0.47$).

Ca and P Balances

Calcium balances for horses fed PP, T85, and CB were 0.56, 0.60, and 0.58 g/d, respectively (Table 3-2). There was no difference between PP hay and the mean of the bermudagrass hays ($P = 0.52$) or between the two bermudagrass hays ($P = 0.65$).

Phosphorus balances for horses fed PP, T85, and CB were -2.62, -3.75 and -4.30 g/d, respectively (Table 3-2). Perennial peanut had a greater PBal than the mean of the bermudagrasses ($P = 0.03$). Coastal bermudagrass and T85 did not differ ($P = 0.40$).

Nitrogen Balance

Nitrogen balance was numerically greatest for PP and least for T85 (Table 3-2). Coastal bermudagrass was intermediate. The mean NB for PP was greater than the mean NB for the two bermudagrasses ($P = 0.013$). The mean NB for CB was greater than that of T85 ($P = 0.012$).

Refusals

There was a noticeable difference for refusals among the three hays. It was observed that the horses did not eat as much T85 as CB or PP when offered at 2% of BW. This data (Figure 3-1) was not statistically analyzed, but the horses' reluctance to eat the hay should be noted as it could have affected digestibility.

Discussion

Forage Analysis

Overall, the nutritive composition of PP was greater than the bermudagrass hays; this is not surprising because legumes are generally more nutritious than grasses. The higher NDF content of Tifton 85 bermudagrass compared to Coastal bermudagrass is consistent with previous

research (Hill et al., 1993; West et al., 1998; Mandebvu et al., 1998a, 1999; Chambliss and Dunavin, 2003). Perennial peanut also had less NDF than either of the bermudagrasses; it is known that legumes contain less NDF than grasses, especially warm-season grasses (Ball et al., 2002; Barnes et al., 2003).

Prine et al. (1981) reported that CP in PP can range from 12 to 19% depending on maturity and season of harvest. Hammond et al. (1992) found a 2-year mean CP (DM basis) of 12.2% while Romero et al. (1987) reports a wide range, from 11.3% CP (DM basis) in commercial PP to 14.4% CP (DM basis) in hay cut at 6 wk regrowth in the summer. Gelaye et al. (1990) reported PP to have 17.1% CP (DM basis). In this trial, the delay in harvest of PP due to weather conditions resulted in a more mature hay. As a result, the CP content of PP may have been depressed slightly but was still close to previously reported data.

A 500 kg adult horse fed PP (90% DM) with 12% CP at 2% of its BW would receive 702 g of CP/d if the CP in the hay were 65% digestible. This is more than 11% above the current NRC requirement (630 g/d) for adult horses at maintenance (NRC, 2007). The horses fed PP during Period 1 (average DM intakes of 1.5% of BW) were receiving an average 3% less than their requirement, but the horses fed PP during Period 2 (average DM intakes of 1.7% of BW) were receiving an average 4% more CP than their requirements. During Period 3, the horse on PP (average DM intakes of 2% of BW) was receiving 40% more CP than its requirement. Perennial peanut can therefore supply average size horses with their required amount of protein at maintenance.

The CP contents of CB and T85 in this trial were typical for bermudagrass grown in Florida. In Georgia, Tifton 85 and Coastal bermudagrasses could have a CP content of 15 or 16% at 5 wk of growth with well fertilized fields, according to previously reported data (Mandebvu et

al., 1998a, 1999; Hill et al., 1993). In Florida, however, CP content of bermudagrass hays is typically lower than 16%. Chambliss et al. (2006) reported a 4 wk regrowth CP average of 13.6% (DM basis) of bermudagrasses grown in Florida. The NRC (1989) reports a CP of 10.9% for 4 to 5 wk maturity CB. The disparities may be due to differences in field maintenance (rain damage), N fertilization, or forage maturity. For instance, bermudagrasses will respond well to N fertilization; increasing CP content as N fertilization increases (Johnson et al., 2001; Chambliss et al., 2006). The fields of CB and T85 used in this trial may not have been fertilized with enough N to increase protein content to optimum levels. Protein content will also decrease with increasing plant maturity in bermudagrasses (Ball et al., 2002; Chambliss et al. 2006). Another reason why the CP of T85 was low could be the later harvest (5 wk regrowth). Lieb and Mislevy (2001, 2003) found different CP concentrations in 5 wk regrowth CB (grown by a commercial producer in north-central FL) and 5 wk regrowth T85 (grown in Ona, FL). In 2001, CP (DM basis) of CB was 13.2%, versus in 2003 where it decreased to 9.3%. Tifton 85 improved in CP (DM basis) content: in 2001 the analyzed CP content was 9.1%, whereas in 2003 CP improved to 16.6%.

Horses in this study only met or exceeded their protein requirement while on CB fed at 2% of BW (of the two bermudagrasses). No horse was able to absorb enough protein from T85 to meet his requirement, even when fed at 2% of BW. A 500 kg adult horse at maintenance would need to eat more than 2.5% of its BW in T85 (90% DM, 9% CP) in order to meet its daily requirement (NRC, 2007). Due to the reluctance of horses to eat T85, this might not be feasible; thereby requiring a supplemental source of protein for horses fed T85. The CP content of T85 would have to be above 12% and voluntary intake at 2% of BW for a 500 kg adult horse at

maintenance to meet its requirement. Tifton 85 produced a negative NB; a greater CP content in T85 or the whole ration would be needed to raise the NB in horses fed T85.

Perennial peanut hay had a Ca:P ratio of 5.6:1, which exceeds the recommended upper end-point ratio of 2.5:1 (NRC, 1989). The horses in this study received enough Ca to meet (CB and T85) or exceed (PP) their daily requirement, even when fed at 1.5% BW. The daily requirement for a 500 kg adult horse at maintenance is estimated to be 20 g of Ca (NRC, 2007). If the same horse were fed PP (1.18% Ca) at 2% of BW (DM 90%) and half of the Ca in the hay was absorbed, the horse would receive over 50 g of Ca/d. Under the same scenario, horses fed alfalfa hay (1.37% Ca, 90% DM) would receive around 60 g of Ca/d (assuming half of the Ca in the hay was absorbed). Under these circumstances, PP and alfalfa hay would be similar in their Ca value for horses.

Phosphorus content is also similar between PP (0.18 – 0.3%; values from this research, Prine et al., 1981) and alfalfa hay (0.19 – 0.3%; NRC, 1989). Horses fed PP in this study were in negative P balance, even when fed at 2% BW. This is typical of feeding more mature forages (Lewis, 1996). Adult horses that are able to graze or are offered younger forage would likely meet their P requirement. A 500 kg adult horse at maintenance would require 14 g of P/d in their diet. If that horse were fed at 2% of BW, perennial peanut (0.21% P, 90% DM) and alfalfa (0.30% P, 90% DM) would not provide enough P (9.45 or 13.5 g/d, respectively) if half of the P in the hays were absorbed by the horse (NRC, 2007). Hay maturity, Ca and P availability, and anti-nutritive factors (such as oxalates or phytates) may affect the actual amounts of Ca and P absorbed by the horse. The NRC takes into account the limited availability of P in forages, but according to the NRC, these horses should have met their P requirement when fed at 1.5% of

BW. However, since the P balances were still negative for horses fed these forages, more investigation is needed to ensure that certain forages really meet horse requirements.

Both CB and T85 had Ca:P ratios within the recommended range (1.9:1 and 1.5:1, respectively). However, the horses fed T85 and CB did not meet their Ca requirements until fed at 2% of BW, where they exceeded their requirement by 8% (for both bermudagrasses). Phosphorus requirements were not met, even when bermudagrass hays were fed at 2% BW. Mature grass hay often does not contain enough P for mature horses to meet their P requirements (Lewis, 1996). Adult horses that have a chance to graze young plants are able to obtain enough P to satisfy their requirement without detrimental effects (Lewis, 1996). Supplemental P is recommended for young horses where maximum growth rate and size is desired, however (Lewis, 1996).

Digestible energies of the three hays were provided by a commercial testing facility (NIRS method, Dairy One, Ithaca, NY). The horses fed PP obtained enough energy from the hay when fed at 1.7% and 2% of BW (9% and 37% more than their requirements, respectively). When horses were fed at 1.5% of BW, horses received 5% less of the energy they required (NRC, 2007). Coastal bermudagrass and T85 only supplied enough energy to the horses when fed at 2% of BW (32 and 11% more than their requirements, respectively). When horses were fed at 1.5% of BW, CB and T85 did not meet the horses' energy requirements (8 and 13% less than requirement, respectively). At 1.7% of BW, the horses' energy intake decreased slightly from the more restricted intake when fed CB and T85 (10 and 14% less than requirement, respectively). One possible reason for this could be that the increase in intake changed the rate of passage, thereby changing the efficiency of digestion.

Apparent Digestibility

It appears that PP is a quality legume that horses will eat and can digest well. Comparisons between these results and other literature data suggest that PP has a similar digestibility to alfalfa (Table 3-3).

Crude protein digestibility of PP in this study (67%; Table 3-3) was lower than the digestibility reported for alfalfa (79%) but similar to PP (70%) by Lieb et al. (1993). LaCasha et al. (1999) found an alfalfa (early-bloom) CPD in horses of 83%, which they recognized as greater than previously reported (Table 3-3). For instance, Crozier et al. (1997) found that mid-bloom alfalfa to have only 73% CPD (Table 3-3). The maturity of the alfalfa in Lieb et al. (1993) is unknown, although it is most likely early-bloom alfalfa. Maturity is known to decrease nutritional value of forages: Darlington and Hershberger (1968) reported a decrease in DM and CPD with increased maturity of alfalfa in horses. It is likely that the differences in maturity are partly responsible for the disparity between the CPD of the PP in this study and that of the alfalfa hays of the other studies (Table 3-3).

Darlington and Hershberger (1968) found that CPD of alfalfa, timothy, and orchardgrass hay was directly proportional to the CP content of the hays. The CP content of the alfalfa hays in the three previously cited studies (19 to 20%) was greater than the perennial peanut in both the current study (13%) and that in Lieb et al. (1993, 16%). Therefore, another reason as to why PP has a lower CPD than typical of alfalfa could be the difference in CP content. This could also explain the greater CPD and N balance in PP versus the two bermudagrasses.

The greater concentration of CP in PP compared to the two bermudagrasses may partly explain the greater CPD found in PP. Lieb and Mislevy (2001, 2003) reported an increase in CPD of T85 when the CP concentration increased from one year to the next.

Maturity may have also affected CPD in the bermudagrasses. Mandebvu et al. (1998b) found a decrease in forage quality for T85 when harvested at 7 wks compared to 3.5 wks. The decrease made the forage quality comparable to that of CB. This may be due to the decreased CP content or increased NDF content of more mature forages. The bermudagrass forages in this study were not of similar maturity; T85 was more mature than CB. Therefore, the maturity of T85 may have affected CPD (and other digestibilities).

Lieb et al. (1993) did not discuss where the PP hay came from and how mature it was, so there could be differences between the PP hay used for this experiment and the PP hay they used. However, despite the lower digestibilities of PP hay compared to alfalfa, Lieb et al. (1993) remarked that there was a higher intake of PP hay, which may have resulted in the greater positive weight gain of those horses fed the PP hay compared to alfalfa or the grass hays. The authors also stated that PP hay was readily accepted by the horses even though they may not have had contact with the hay before.

It is interesting that the apparent digestibility of NDF was numerically greatest for CB, then T85, and finally PP, although the differences were not statistically significant. Tifton 85 bermudagrass has been found to have a greater NDF digestibility than Coastal bermudagrass in ruminants (Mandebvu et al., 1999). However, when T85 was more mature than CB, as is the case in this experiment, the NDF digestibility was similar between the two bermudagrasses (Mandebvu et al., 1999). It may be that T85 would have a greater NDF digestibility than CB if both bermudagrasses were harvested at a similar maturity.

McCann et al. (1995) does not mention the maturity of the bermudagrass hays fed to the horses. They found that T85 had a numerically greater NDF digestibility ($57.2\% \pm 2.1$ SE) than CB ($54.6\% \pm 2.1$ SE), but these results were not significant. McCann et al. (1995) also found that

CB had a numerically greater CP digestibility ($65.4\% \pm 1.1$ SE) than T85 ($63.5\% \pm 1.1$ SE; Table 3-4).

Calcium and Phosphorus Balances

The lack of difference between the CaBal in any of the hays may have been due to individual animal differences, the different amounts of hay offered for each period, or collection problems, such as loss of sample and contamination (results found in Table 3-2). The difference in PBal between PP and the two bermudagrasses could be from a number of factors, such as differences in biological availability, the different amounts of hay offered over the course of the study, and the differing intakes of the three hays.

There have been no reports on the mineral absorption by horses fed PP hay; therefore it is unknown whether the Ca and P balances observed here are typical for PP hay. Sturgeon et al. (1999) reported apparent Ca and P absorptions in alfalfa to be 72% and 24%, respectively, in mature horses. Crozier et al. (1997) reported apparent absorptions of Ca and P of alfalfa in mature horses to be 46% and 8%, respectively. In comparison, PP hay in the present study had a Ca digestibility of 56%. This disparity is likely caused by differences in hay composition, feeding level, contamination, and collection methods. Therefore, it is possible that PP hay has similar apparent absorptions of Ca and P as alfalfa; further studies need to be done, however, to provide more information.

Actual mineral absorptions are difficult to obtain and reproduce because of interactions with other minerals and compounds, and reactions that occur in the digestive system. For instance, Ca may affect P absorption, especially in mature animals, as is the case in pigs and ponies (Jongbloed, 1987; Van Doorn et al., 2004). Phytates and oxalates that are present in some forage also prevent mineral absorption by binding certain minerals, such as P and Ca. Tropical forages (such as bermudagrasses) tend to have more oxalates than Ca, which depresses Ca

absorption (McDowell, 2003). It is not likely that oxalates played a role in depressing the absorption of Ca in PP and CB; however, it should not be ruled out.

Nitrogen Balance

Nitrogen balance for PP and CB were both positive, whereas the NB for T85 was negative (Table 3-2). Horses lost an average of 14 kg when fed T85 and 16 kg when fed CB, whereas horses gained an average 6 kg when fed PP over the course of the study. If Period 1 weight loss is excluded (because of the greater weight loss due to restricted intake; Periods 2 and 3 represent a more normal feeding situation), horses lost more weight on average when fed T85 (-3 kg) than CB (-1 kg). Horses gained an average 22 kg when fed PP (excluding Period 1 weight loss). When the horses were losing weight, catabolism was greater than synthesis, therefore more nitrogen was lost than the forage could provide.

The greater NB of PP compared to the two bermudagrasses could be partly explained by the average weight gain by the horses when fed PP over the course of the study. The higher CP content of PP, and the observation that horses ate more of the PP than CB and T85 when offered at 2% of BW, is also likely to have contributed to the greater NB seen when feeding PP compared to CB and T85. The negative NB of T85 may have been due to the numerically greater weight loss of horses fed T85. The difference between average weight losses of horses fed CB and T85 is not large, but the negative NB of T85 may have also been influenced by the reluctance of horses to eat T85 compared to CB and PP. Although not statistically analyzed, horses appeared to have retained less N (as % of intake, averaged over all periods) when fed T85 (-4.4) versus CB (23.2) or PP (28.4). It may be that N was not as efficiently absorbed from T85 by the horses as it was from CB and PP.

Refusals

The horses in this experiment seemed to prefer PP to the grass hays. Figure 3-1 shows the amount of refusals as a percent of intake for the hays. Perennial peanut hay had the least amount of refusals and consisted mainly of stems and not leaves. The horses did not eat as much of the T85 compared to CB. This could be because T85 has thicker stems than CB. Tifton 85 bermudagrass, because of its thicker stems, may have contributed to a greater fill effect, keeping the horses from eating more. The texture of T85 may also have reduced intake because of the time needed for horses to manipulate (more mastication to reduce to smaller particles).

Table 3-1. Analyzed nutrient composition of perennial peanut (PP), Coastal bermudagrass (CB), and Tifton 85 bermudagrass (T85) hays offered to horses (DM basis).

Nutrient	PP	CB	T85
DM, %	85	88	84
Ash, %	7	5	5
CP, %	12	10	9
DE, Mcal/kg ¹	2.14	1.98	1.87
NDF, %	46	73	77
ADF, %	34	36	42
Ca, %	1.18	0.3	0.29
P, %	0.21	0.16	0.20
Mg, %	0.44	0.13	0.15
K, %	1.52	1.28	1.38
Cu, ppm	4.30	6.12	4.85
Mn, ppm	101.18	35.49	32.49
Fe, ppm	100.63	85.29	94.66
Zn, ppm	30.85	24.11	29.15
Na, ppm	153.48	174.45	173.19

¹Energy values provided by a commercial testing facility (NIRS method, Dairy One, Ithaca, NY) and were averaged by period.

Table 3-2. Apparent digestibilities (%) of dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF); and balances (g/d) of calcium (CaBal), phosphorus (PBal), and nitrogen (NB) of perennial peanut (PP), Coastal bermudagrass (CB), and Tifton 85 bermudagrass (T85) hays when fed to mature horses.

Item	PP	CB	T85	SEM ¹	P-value	
					PP vs. CB and T85	CB vs. T85
DM	65	53	52	2.2	0.0004	0.80
OM	66	53	57	2.5	0.0006	0.68
CP	67	60	57	1.9	0.0035	0.47
NDF	45	50	46	3.2	0.51	0.47
CaBal	0.56	0.60	0.58	0.28	0.52	0.65
CaD ²	56	60	58	3.83	0.48	0.70
PBal	-2.62	-4.30	-3.75	0.46	0.03	0.40
NB	53.6	36.8	-3.4	10.2	0.013	0.012

¹SEM = Standard error of the mean, n=6 (T85 and CB); n = 5 (PP).

²CaD = Ca digestibility (%).

Table 3-3. Comparison of the apparent digestibility of perennial peanut (PP) from our study (Eckert) with PP and alfalfa (A) from Lieb et al. (1993), and A from Crozier et al. (1997) and LaCasha et al. (1999).

Digestibility, %	Eckert PP	Lieb PP	Lieb A	Crozier A	LaCasha A
DM	65	56	62	58	63
OM	66	ND ¹	ND	ND	74
CP	67	70	79	73	83
NDF	45	43	45	47	24

¹"ND" = not determined.

Table 3-4. Comparison of apparent digestibility of Coastal bermudagrass (CB) and Tifton 85 (T85) bermudagrass in our study (Eckert), with that of McCann et al. (1995), Lieb et al. (1993), and LaCasha et al. (1999).

Digestibility, %	Eckert CB	Eckert T85	McCann CB	McCann T85	Lieb CB	LaCasha CB
DM	53	52	52	54	41	46
OM	53	57	ND ¹	ND	ND	60
CP	60	57	65	64	63	64
NDF	50	46	55	57	42	52

¹"ND" = not determined.

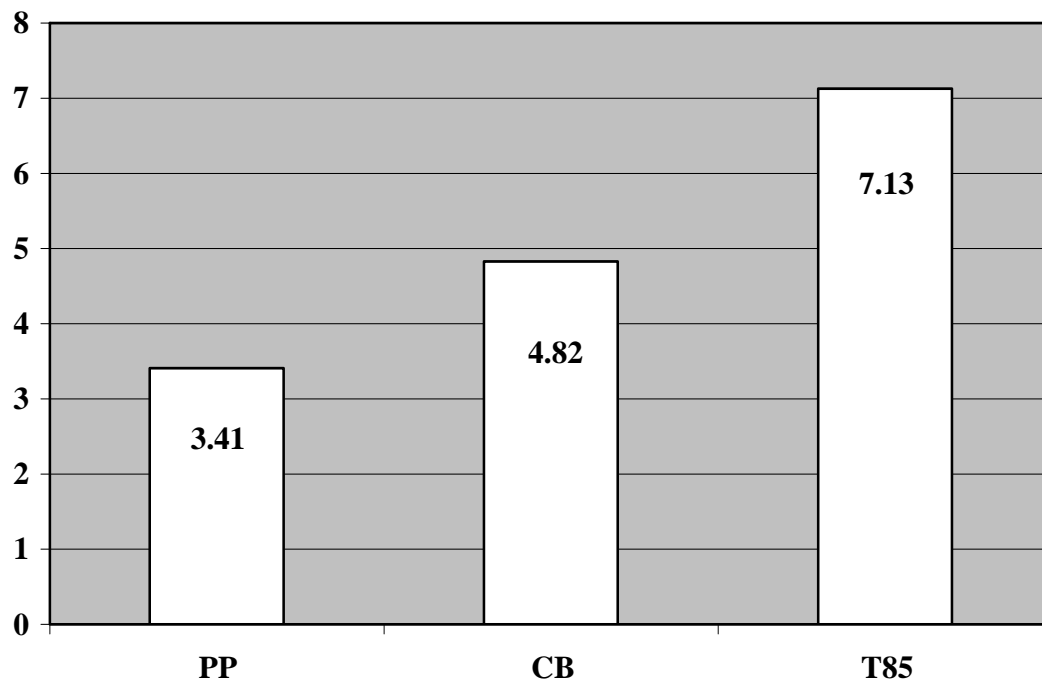


Figure 3-1. Hay refusals (as % of intake) of perennial peanut (PP), Coastal bermudagrass (CB), and Tifton 85 bermudagrass (T85; standard deviation = 1.88).

CHAPTER 4
IN VITRO ESTIMATION OF NUTRITIVE VALUE OF PERENNIAL PEANUT, ANNUAL
PEANUT, ALFALFA, AND BERMUDAGRASS FORAGES FOR HORSES

Introduction

Traditional digestion trials that use the actual animal are becoming increasingly difficult to perform due to time and cost constraints. Therefore, methods have been developed to estimate the digestibility of feedstuffs that only need the animal as a representative source of microbes, such as rumen or cecal fluids (Tilley and Terry, 1963; Applegate and Hershberger, 1969). The use of horse feces as a source of microbes in estimating digestibility is non-invasive and is comparable to cecal fluid and *in vivo* digestibility estimates in predicting forage digestibility in horses (Lowman et al., 1999; Ringler et al., 2005; Warren and Kivipelto, 2005; Lattimer et al., 2007).

In vitro digestion studies were traditionally done using the gas production method (Tilley and Terry, 1963). This is time consuming, labor-intensive, and not as precise as a closed system fermentation apparatus (Holden, 1999; Lattimer et al., 2007). Recently, technology has made it easier to analyze different types of samples simultaneously by using the incubators (DAISY^{II}; ANKOM Technology Corp., Fairport, NY). Holden (1999) compared the method using the incubators to the traditional gas production method and found no difference between the *in vitro* dry matter digestibility (IVDMD) of the methods, even when forages and grains were incubated in the same vessel. The authors used rumen fluid from cows as the source of microbial inoculum. Ringler et al. (2005) used horse feces as a source of inoculum and found no difference between *in vivo* and *in vitro* dry matter digestibility when testing alfalfa hay, timothy hay, alfalfa-oat mix, and a timothy-oat mix. Lattimer et al. (2007) also found *in vitro* and *in vivo* digestibility

estimates to be similar when testing the effectiveness of yeast culture on horses fed high-concentrate or high-forage diets.

Perennial (*Arachis glabrata*) and annual peanuts (AP; *Arachis hypogea*) are warm-season legumes that grow well in the Southeastern US. Very little is known of the nutritional value of these forages for horses. Lieb et al. (1993) found perennial peanut (PP) was similar to alfalfa in DM, CP, and NDF digestibilities as well as digestible energy. In addition, the horses voluntarily consumed more of the PP than alfalfa. There have been no studies done with horses on the suitability of AP. Researchers have found that AP is highly nutritious for ruminants (Ronning et al., 1953; McBrayer et al., 1981; Gorbet et al., 1994; Larbi et al., 1999); thus AP may be a good forage for horses. The 'Florigraze' cultivar of PP was released by the University of Florida in 1981. Other varieties that have been released include 'Arbrook' and 'Ecoturf'. The University of Florida is continuing to research different varieties of PP to maximize the use of the forage for all types of livestock. *In vitro* digestibility procedures would be valuable for this type of research in helping to identify potentially high quality varieties.

Coastal bermudagrass is commonly fed to horses in the Southeastern US as a source of forage. It is highly digestible and palatable to horses if fertilized and harvested properly. Recently, Tifton 85 bermudagrass has eclipsed CB in popularity in the feeding of ruminants. This particular bermudagrass cultivar is more digestible than CB in DM, NDF, and ADF in ruminants (Mandebvu et al., 1999). McCann et al. (1995) compared the digestibility of CB and T85 bermudagrass hays in mature horses. These researchers found similar digestibility between the bermudagrass hays. Lieb and Mislevy (2001, 2003) found that Tifton 85 bermudagrass hay could be as nutritious as or better than Coastal bermudagrass hay in horses. In the study

mentioned in Chapter 3, Coastal and Tifton 85 bermudagrasses were found to have similar digestibility estimates.

Our objectives of this experiment were to compare the *in vitro* true digestibility (IVTD) of a number of samples incubated together. Comparisons were made between samples of a similar type or from the same trial

- Dried fresh forage samples of annual peanut from two fields (AP–Fresh1 and –Fresh2) and one field of perennial peanut (PP–Fresh),
- Dried fresh forage samples of annual peanut (AP–Fresh1 and –Fresh2) and perennial peanut (PP–Fresh) compared to their hay counterparts (PP06–hay and AP– hay),
- Hay samples of perennial peanut (PP06–hay), annual peanut (AP–hay, from the AP–Fresh2 field), and commercial alfalfa (A–hay),
- Hay samples from three periods each of perennial peanut (PP05–hay), Tifton 85 bermudagrass (T85–hay), and Coastal bermudagrass (CB–hay) from a previous *in vivo* digestion trial (Chapter 3),
- Dried fresh forage samples from six varieties of PP (Arbrook, AR; Floragrazer, FG; Tom Burton Selection, TB; Ecoturf, EC; Elite Line, EL; and Good Line, GL).

Materials and Methods

Samples

The forage or hay samples included: three pasture cuttings per field from two different fields of annual peanut (AP–Fresh1 and –Fresh2) and one field of perennial peanut (PP–Fresh, Marianna, FL; collected in August 2006 from mature fields); perennial peanut hay (PP06–hay, cut in 2006 from a mature field), annual peanut hay (AP–hay, cut in 2006 from the same mature field as AP–Fresh2), alfalfa hay (A–hay, purchased locally and was grown in Iowa, 2006); three periods (P1, P2, and P3) each of perennial peanut hay (PP05–hay, Madison County, FL, delayed first cutting August 2005 from a mature field with no rain damage), Coastal bermudagrass hay (CB–hay), and Tifton 85 bermudagrass hay (T85–hay; both bermudagrass hays from Suwannee County, FL, cut 2005 from irrigated and fertilized fields with no rain damage), all hays from a

previous *in vivo* digestibility trial (Chapter 3); and dried fresh forage samples from six mature perennial peanut varieties ('Arbrook', AR; 'Floragrazer', FG; 'Tom Burton Selection', TB; 'Ecoturf', EC; 'Elite Line', EL; and 'Good Line', GL; variety trial, Marianna, FL, 2005).

Dried fresh forage samples (AP–Fresh1 and –Fresh2, and PP –Fresh) were obtained by using a meter square. The square was tossed eight times in different parts of the field (eight samples total per field; which were later combined by field). Plant material inside the square was hand clipped to a stubble height of 10 to 12 cm.

Hay samples (PP06–hay, AP–hay, A–hay, PP05–hay, CB–hay, and T85–hay) were obtained using a hay corer (3 cm diameter and 30 cm length). At least 20 random samples were collected from small bales (enough sample was obtained for chemical analysis and the *in vitro* fermentation procedure).

All samples were dried in a forced-air oven at 65°C. After drying, the field cuttings of AP–Fresh1 and –Fresh2, PP–Fresh, and PP varieties (AR, FG, TB, EC, EL, and GL) were stored for approximately eight months before the trial began. Samples of hays from 2006 (PP06, AP, and A) were stored for approximately two months. All samples were stored in sealed plastic bags at room temperature (23°C).

Animals, Feeding, and Housing

This *in vitro* trial used yearling horses as a source of feces for inoculation of the *in vitro* system. Horses only acted as a source of feces and did not need different housing or feeding from what they received at the UF Horse Teaching Unit. Horses were not fed any of the forages tested, as there has been no difference found between type of forage fed and subsequent digestion coefficients (Applegate and Hershberger, 1969). The horses were fed their normal diet of concentrate, CB hay, and pasture.

***In Vitro* Digestion Procedure**

Freshly voided feces were collected from the ground of the stall (no shavings collected) from seven horses into plastic bags and put into a warmed thermos. The feces were processed within 20 min of collection.

Samples of hays or dried fresh forages were ground through a 1-mm screen in a Wiley mill (Arthur H. Tomas Co., Philadelphia, PA). About 0.5 g of sample was put into ash-free, N-free incubation bags (F57; ANKOM, Macedon, NY) and heat-sealed.

In vitro fermentation was conducted using an ANKOM© DAISY^{II} incubator (ANKOM Technology Corp., Fairport, NY). Each of the four jars in the incubator contained the 21 individual forage samples and 2 blanks (thus, each forage sample was analyzed in quadruplicate).

Two buffers were made according to ANKOM© *In Vitro* directions. Buffer solution A contained 10 g of KH_2PO_4 , 0.5 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 0.5 g of NaCl , 0.1 g of $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, and 0.5 g of reagent grade urea in 1 L of deionized water. Buffer solution B was made with 15.0 g of Na_2CO_3 and 1.0 g of $\text{Na}_2\text{S} \cdot 9\text{H}_2\text{O}$ in 1 L of deionized water. After an overnight incubation period at 39°C for both buffers, the buffers were thoroughly mixed. The feces collected from the horses were pooled and 200 g of the resulting mixture were weighed out and combined with approximately 400 mL of buffer in a Wiley electric mixer (Arthur H. Tomas Co., Philadelphia, PA). The blender was then purged with CO_2 . Once thoroughly macerated, the fecal/buffer mixture was poured into a graduated cylinder, the blender rinsed with remaining buffer, and poured into a graduated cylinder to reach a total of 1800 mL. This resulting mixture was immediately poured into a digestion jar and purged with CO_2 . To each jar, 21 samples and 2 blank bags were added (12 on one side of the divider, 11 on the other) and then the fecal

inoculum. The jars were purged with CO₂ for 30 sec and placed in the incubators. Four jars were heated at 39°C and rotated at the same time for 48 h (Jennifer Ringler, personal communication; Warren and Kivipelto, 2005).

After the incubation period, samples were rinsed in deionized water until the water ran clear and were then subjected to NDF analysis using the ANKOM Fiber Analyzer (ANKOM Technology Corp., Fairport, NY). After NDF analysis, samples were soaked in acetone for 3 to 5 min, placed in a fume hood for acetone to evaporate, dried overnight in a forced-air oven (105°C), and re-weighed.

Sample Analysis

Chemical composition of AP–Fresh1 and –Fresh2, PP–Fresh, PP06–hay, AP–hay, A–hay, and the PP varieties (AR, FG, TB, EC, EL, and GL) were determined by a commercial forage analysis laboratory (NIRS method; Dairy One, Ithaca, NY). These data are summarized in Table 4-1.

Calculations

In vitro true digestibility (DM basis) was calculated with the following equation:

$$\text{IVTD (DM basis)} = 100 - ((W_3 - (W_1 \times C_1)) \times 100) / (W_2 \times (\text{DM}/100))$$

where

W₁ = Bag tare weight

W₂ = Sample weight

W₃ = Final bag weight after *in vitro* fermentation and sequential NDF determination

C₁ = Blank bag correction (Final oven dried weight / original blank bag weight)

DM = % Dry matter.

Statistical Analysis

Data were analyzed using PROC MIXED of SAS version 9 (2002). Each sample type had four replications. The LSMEANS were calculated and separated using a PDIFF statement. Comparisons were made between samples of a similar type, e.g. the hays from the digestion trial to each other, the field samples to each other, and the six PP varieties to each other. Also fresh

dried forage samples of annual and perennial peanut were compared with their respective hays. Therefore, each sample type served as the experimental unit (n=4 per forage).

Results

Dried Fresh Forage and 2006 Hays

In vitro total digestibility of fresh dried annual and perennial peanut forages harvested in 2006 are presented in Table 4-2. *In vitro* total digestibility was similar between AP–Fresh1 and AP–Fresh2 (P = 0.48). PP–Fresh had a lower IVTD than AP–Fresh1 (P = 0.008) and AP–Fresh2 (P = 0.0011). For the hays harvested in 2006, A–hay had greater IVTD than AP–hay (P = 0.011) but similar IVTD as PP06–hay (P = 0.203). The IVTD of PP06–hay did not differ from AP–hay (P = 0.183; Table 4-2). Annual peanut–Fresh1 and AP–Fresh2 were greater in IVTD than AP–hay (P = 0.0001) but PP–Fresh did not differ from PP06–Hay (P = 0.32).

***In Vivo* Digestion Trial Hays**

In vitro total digestibility of the three hays used in a previous *in vivo* digestibility trial (Chapter 3) are presented in Table 4-3. Across all three periods, PP05–hay had greater IVTD than CB–hay and T85–hay (P = 0.0001). The IVTD of CB–hay did not differ from T85–hay in P1 (P = 0.24) or P2 (P = 0.71); however IVTD of CB–hay tended to be greater than T85–hay in P3 (P = 0.06). An effect of period was only observed for CB–hay; IVTD of CB–hay was greater in P3 than P1 CB–hay (P = 0.05) and P2 CB–hay (P = 0.001).

Perennial Peanut Varieties

In vitro total digestibility of six different perennial peanut hay varieties is presented in Table 4-4. The IVTD of EL was similar to EC (P = 0.38) but greater than GL (P = 0.02), FG (P = 0.002), TB (P = 0.002), and AR (P = 0.001). The IVTD of EC was similar to GL (P = 0.14) but greater than FG (P = 0.03), TB (P = 0.02), and AR (P = 0.008). The IVTD of GL was similar to TB (P = 0.35), FG (P = 0.43), and AR (P = 0.22). The IVTD of AR was similar to FG (P = 0.65)

and TB ($P = 0.75$). The IVTD of FG was no different from TB ($P = 0.89$). Numerically, the ranking of IVTD within the PP varieties is $EL > EC > GL > FG > TB > AR$.

Discussion

Field Cuttings and 2006 Hays

Annual peanut appears to retain less digestible nutrients than PP through the haying process and storage. Annual peanut hay had similar DM compared to the fresh forage samples (Table 4-1). Neutral detergent fiber and ADF were both numerically greater in the hay than the fresh field cuttings (Table 4-1). Perennial peanut showed only a nominal increase in NDF but did not greatly increase in ADF due to the haying process and storage time (Table 4-1). Harvested and stored forages generally increase in fiber content (measured as NDF, ADF, and lignin) as storage time progresses. This is thought to occur because of increased oxidation of nonstructural carbohydrates (Rotz and Muck, 1994). It appears that the increase in NDF and ADF in AP is greater than in PP due to haying and subsequent storage.

Collins et al. (1987) found that alfalfa increased in NDF and decreased in CP content after the haying process (ADF was not measured). Over 90% of the losses from the harvest were from leaf shatter, decreasing the nutritional value of alfalfa. Mechanical harvesting differs from harvesting by grazing in that it collects as much of the stem as the machine is programmed to harvest. Leaves are also gathered, but much is lost due to leaf shatter. This produces bales that are predominantly stems. Animals, however, preferentially graze leaves and few stems. This results in a different nutritive profile of grazed forages from those baled into hay. Alfalfa is still a good hay crop, however, with only moderate increases in NDF and moderate decreases in CP (41% NDF when grazed versus 53% as hay and 18% CP versus 15%; Collins et al., 1987). Crude protein decreased dramatically in AP due to haying and storage, but only moderately in PP. It is likely that there is substantial leaf shatter during harvesting of AP. Annual peanut may then be

better suited as a grazed crop for horses instead of being baled into hay, since the crop's nutritive value is dramatically reduced. Perennial peanut, however, did not dramatically change in NDF, ADF, or CP concentration as a result of haying and storage; making it excellent as pasture or hay.

The lack of differences in IVTD of PP06-hay and A-hay would indicate that PP is intrinsically as nutritious as A; however an *in vivo* digestion trial will need to be conducted to as IVTD only measures the total potential digestion, but does not take into account intake and individual animal differences. Lieb et al. (1997) found PP to be nutritionally similar to alfalfa in digestible energy, as well as DM, CP, and NDF digestibilities. Perennial peanut was also readily accepted by horses; voluntary intake was greater for PP than for alfalfa. This IVTD data and the study by Lieb et al. (1997) suggest that PP hay could replace alfalfa hay in a horse's diet.

As noted above, the IVTD of AP-hay and PP06-hay did not differ from each other. However, because of the thicker stems of AP hay, it may not be preferred by horses. Given the dramatic changes of AP when harvested as hay, AP may be better suited for horses as a grazed legume.

***In Vivo* Digestion Trial Hays**

The most interesting difference between the digestibilities found *in vivo* and *in vitro* is the lower IVTD in the two grasses than the *in vivo* digestion trial. *In vitro* true digestibilities of the bermudagrasses were lower than those determined *in vivo* (CB-hay and T85-hay mean *in vivo* DM digestibility: 53 and 52%, respectively versus mean IVTD: 42 and 40%, respectively) whereas PP05 did not greatly differ (mean *in vivo* DM digestibility: 65% versus mean IVTD: 69), although it was numerically greater (Table 4-3). Applegate and Hershberger (1969) also observed this in a study involving alfalfa and two grasses. They postulated that alfalfa supplied nutrients to the fermentation medium, which increased the rate of fermentation compared to the

two grasses they evaluated (timothy and orchardgrass). They used the Tilley and Terry method and horse cecal fluid (Tilley and Terry, 1963); however, a similar occurrence could have happened during the present study using the Daisy^{II} incubator and fecal inocula. Even though grasses tend to have lower *in vitro* digestibilities than legumes, the procedure may still be used to separate good quality from poor quality grasses. Lattimer et al. (2007) found that including a lesser amount of sample in the filter bags more closely resembles *in vivo* data. They found that using 0.25 g of sample was more accurate and precise than using 0.50 g of sample, which was the amount used in the present study. Therefore, more accurate data may be obtained if *in vitro* analyses in the present study were repeated using 0.25 g of sample. Another possible method change would be to incubate the samples for 72 hr instead of 48 hr. Ringler et al. (2005) found that even though the IVDMD of samples incubated for 48 hr and 72 hr did not differ, the samples incubated for 72 hr were the ones that did not differ from *in vivo* digestibility estimates. It typically wouldn't take 72 hr to completely digest a forage meal in a horse, but a 72 hr *in vitro* fermentation may be the most beneficial when estimating *in vivo* digestibility. There were four diets tested, including alfalfa hay, alfalfa hay plus oats, timothy hay, and timothy hay plus oats.

Coastal bermudagrass hay demonstrated the most variation in IVTD between periods, whereas T85-hay or PP05-hay did not. The *in vivo* digestibility estimates mirrored the ones from the *in vitro* experiment (Table 4-3). This emphasizes the need of horse owners to periodically check the nutritive value of the hays they are feeding to their horses. Even though the IVTD and *in vivo* DM digestibility values may have been different, the trend between the forages was the same; PP was greater in digestibility than both CB and T85, and CB and T85 were similar.

Perennial Peanut Varieties

The three commercially available varieties of perennial peanut are 'Arbrook', 'Ecoturf', and 'Florigraze'. At this time, 'Florigraze' is the most popular variety used as forage for horses and other livestock (French et al., 1998). From the results of the present study, AR does not appear to be the most digestible PP variety for horses. 'Elite Line' and EC were the most digestible and could potentially be produced and fed to horses. This is likely due to the greater concentration of CP and smaller concentrations of NDF and ADF than the other varieties. All the varieties of PP had high IVTD and could potentially be alternatives to alfalfa or 'Florigraze' PP. The *in vitro* procedure utilizing fecal inoculum would be of benefit to agronomists to help identify potential new varieties of PP for horses. Even so, accurate assessments of digestibility require *in vivo* trials to see if the animal will eat the food and how it performs in the animal.

Table 4-1. Analyzed nutrient composition of dried fresh forage samples, 2006 hay samples, *in vivo* digestion trial hays, and perennial peanut variety samples, % (DM basis).

Forage ¹	DM	CP	NDF	ADF
AP–Fresh1	90.1	23.4	32.2	28.0
AP–Fresh2	90.8	22.7	36.9	30.2
PP–Fresh	89.6	18.4	43.8	34.1
AP–hay	89.7	16.6	49.8	40.4
PP06–hay	89.8	15.5	44.8	33.8
A–hay	90.4	20.8	39.7	31.7
PP05–hay ²	90.7	13.5	43.8	36.1
CB–hay ²	91.7	10.5	72.7	38.9
T85–hay ²	92.7	9.1	76.5	41.6
AR	87.4	19.2	49.3	39.8
FG	87.4	19.3	46.3	35.1
TB	87.8	18.1	51.4	40.9
EC	86.7	20.3	40.5	34.6
EL	86.9	20.5	40.5	34.6
GL	87.8	19.6	44.5	35.9

¹Analyses done by a commercial forage analysis laboratory (Dairy One, Ithaca, NY). AP–Fresh1 and –Fresh2 = dried fresh annual peanut from two fields; PP–Fresh = dried fresh perennial peanut; AP–hay = annual peanut hay, 2006 (from field AP–Fresh2); PP06–hay = perennial peanut hay, 2006, from same field as PP–Fresh; A–hay = alfalfa hay; PP05–hay = perennial peanut hay, 2005; CB–hay = Coastal bermudagrass hay; T85–hay = Tifton 85 bermudagrass hay; AR = 'Arbrook'; FG = 'Floragrazer'; TB = 'Tom Burton Selection'; EC = 'Ecoturf'; EL = 'Elite Line'; GL = 'Good Line'.

²Mean of three periods.

Table 4-2. In vitro true digestibility (IVTD) of fresh dried annual and perennial peanut forages and hays, % (DM basis).

Forage	IVTD ¹	P-value				
		AP-Fresh1	AP-Fresh2	PP-Fresh	PP06-hay	AP-hay
AP-Fresh1 ²	79.6	-	0.4818	0.0084	-	0.0001
AP-Fresh2 ²	77.8	0.4818	-	0.0011	-	0.0001
PP-Fresh ²	70.7	0.0084	0.0011	-	0.32	-
PP06-hay ³	68.1	-	-	0.32	-	-
AP-hay ³	64.6	0.0001	0.0001	-	0.1826	-
A-hay ⁴	71.4	-	-	-	0.2026	0.0106

¹SEM for all samples = 1.83.

²Hand harvested and dried in the laboratory (eight compiled samples). AP-Fresh1 and – Fresh2 = fresh dried annual peanut from two fields; PP-Fresh = dried fresh perennial peanut.

³PP06-hay = perennial peanut hay, 2006, from same field as PP-Fresh; AP-hay = annual peanut hay, 2006, from same field as AP-Fresh2.

⁴A-hay= alfalfa hay. Horse quality hay purchased locally.

Table 4-3. Comparison of IVTD (%) of *in vivo* digestion trial hays and *in vivo* dry matter digestibility estimates: periods 1, 2, and 3 of perennial peanut (PP05–hay), Coastal bermudagrass (CB–hay), and Tifton 85 bermudagrass (T85–hay).

Period	Hay ¹			Comparisons	
	PP05–hay	CB–hay	T85–hay	PP05–hay vs. CB–hay, T85–hay	CB–hay vs. T85–hay
1	68 ^a	42 ^b	39 ^a	**	NS
2	68 ^a	38 ^b	39 ^a	**	NS
3	69 ^a	47 ^a	42 ^a	**	NS
Mean ²	68.5 (65) ³	42.1 (53)	39.8 (52)	**	NS

¹Different letters within a column differ significantly, P <0.05.

²SEM = 1.83.

³Values in parentheses are *in vivo* dry matter digestibility estimates.

**Values differ (P <0.05).

Table 4-4. IVTD of perennial peanut varieties: Arbrook (AR), Floragrazer (FG), Tom Burton Selection (TB), Ecoturf, (EC), Elite Line (EL), and Good Line (GL).

Variety	IVTD ¹ , %	Grouping		P-value					
				EL	EC	GL	FG	TB	AR
EL	75.7	A		-	0.38	0.02	0.002	0.002	0.006
EC	73.5	A	B	0.38	-	0.14	0.03	0.02	0.008
GL	69.6		B C	0.002	0.14	-	0.43	0.35	0.22
FG	67.5		C	0.002	0.03	0.43	-	0.89	0.65
TB	67.2		C	0.002	0.02	0.35	0.89	-	0.75
AR	66.4		C	0.006	0.008	0.22	0.65	0.75	-

¹SEM for all samples = 1.83.

CHAPTER 5 SELECTION OF PERENNIAL PEANUT, ANNUAL PEANUT, OR ALFALFA HAYS IN MATURE HORSES

Introduction

The digestibility and nutritional composition of forages are important factors to consider when deciding what to feed a horse, but only if the forage is not rejected by the animal. Horses may prefer a certain forage to another, even if they are equally digestible. It is important, therefore, to assess the acceptance of forages as well as their nutritional content and digestibility.

Perennial peanut (PP, *Arachis glabrata*) and annual peanut (AP, *Arachis hypogea*) are warm-season legumes that grow well in the Southeastern US. Both have been found to be nutritionally similar to alfalfa in ruminants (Ronning et al., 1953; McBrayer et al., 1982; Gelaye et al., 1990), although preference trials have not been conducted. There is a lack of information regarding the suitability of perennial and annual peanut hays in horses. Lieb et al. (1993) found PP was similar to alfalfa (A) in DM, CP, and NDF digestibilities as well as DE in horses. In addition, horses voluntarily consumed more of the PP than A, indicating preference for PP. By comparison, the suitability of AP for horses has not been investigated. Therefore, the aim of this experiment was to determine the preference of A, PP, or AP hay by horses.

Materials and Methods

Horses

Six Thoroughbred or Quarter horses maintained at the University of Florida Horse Teaching Unit participated in a preference trial in July 2007. These horses were maintained on bahiagrass pasture and free-choice Coastal bermudagrass hay with an evening feeding of grain daily. All horses had been fed A and PP at some point previously; however, they had no prior exposure to AP hay. The study protocol was approved by the University of Florida Institutional Animal Care and Use Committee.

Trial Design

Six horses, acting as their own controls, participated in a 12-d split-plot randomized complete block design with two periods consisting of 6 d ($n = 12$) in which they received six different treatments. The experimental unit was the horse. The comparisons (PP vs. AP, PP vs. A, and A vs. AP) served as the main plot with period serving as the subplot. Period also served as the block.

Perennial peanut hay was harvested from a mature field in 2006 (Marianna, FL). Annual peanut hay was also harvested from a mature field in 2006 (Marianna, FL). Alfalfa hay was purchased locally (grown in Iowa, 2006).

There were six treatments consisting of two hays in all possible side combinations. For example, treatment 1 consisted of PP on the left and AP on the right; treatment 2 consisted of PP on the right and AP on the left (Table 5-1). This was done to minimize any side preference. For each period, each horse was offered one treatment per day; all six treatments were given to each horse in each period (one preference test per day). Therefore, horses were offered each treatment twice (one time per period). Treatments were assigned to the horses randomly for each period.

The amount of each hay offered during each preference test was determined pre-trial (0.40 kg) and presented in a similar fashion as in the trial. Sufficient forage was provided for horses to eat continuously during 10 min; however, the preferred hay may have been entirely consumed towards the end of each observation period.

Feeding Slips and Forage Presentation

Forage preference trials were conducted in 3 m x 3 m feeding slips that permitted individual feeding. The feeding slips were attached to the pasture horses were housed on and were outside, not under the cover of a roof. Horses were accustomed to being in the feeding slips for daily feeding. For the forage preference trials, each slip was equipped with two similar

buckets (19 L; in a range of colors) containing either of two hays placed approximately 0.5 m apart on the opposite side of the horse's entrance (Figure 5-1). Hays were weighed individually in bags prior to each day's observation. Each bucket was assigned a hay to eliminate odor contamination and was removed after every observation period; therefore one horse did not always receive the same bucket each day. Horses were assigned the same feeding slip throughout the two periods.

Horses were brought in from the pasture in the early evening into individual feeding slips. Each horse was observed by an individual person who recorded when the horse started and stopped eating either hay. Total time spent eating either hay was calculated after the trial. After the 10 min observation time was up, both buckets and any distinguishable hay dropped on the ground were taken out of the slip and returned to the bag it came from. Bags with remaining hay were re-weighed and any remaining hay was calculated.

Forage Analysis

Samples of PP, AP, and A hays used were analyzed for DM, CP, NDF, and ADF (Table 5.2) by a commercial forage testing laboratory using near-infrared spectroscopy (NIRS; Dairy One, Ithaca, NY).

Statistical Analysis

Data were analyzed using PROC MIXED of SAS version 9 (2002). Amounts of hay eaten were calculated for each observation on each horse. The amount consumed (kg) for a hay was averaged by horse by period. Comparisons were made between the amounts of PP, A, and AP consumed. The total time (min) spent eating a hay was averaged for each horse in each period. Comparisons were made between the mean times spent on PP vs. AP, PP vs. A, and A vs. AP. There was no period \times time interaction and period was not significant.

Results and Discussion

Forage Analysis

The DM of the three hays was numerically similar, however, AP had a greater CP than PP; and A had the greatest CP content of the three hays (Table 5-2). Annual peanut hay had the highest NDF and ADF concentrations. Perennial peanut and A had similar NDF and ADF. Perennial peanut and A hays were observed to be very leafy with thinner stems, whereas the AP hay contained thicker stems and a lower leaf:stem ratio than the other two legume hays. Even though A had numerically greater CP than PP, the CP content of PP was still enough to provide for horses on maintenance diets comprising solely of forage.

Amount of Hay Consumed and Time Spent on Hay

Horses spent more time eating PP than AP ($P = 0.004$) and more time eating A than AP ($P = 0.0001$; Table 5-3). Horses tended to spend more time on A than PP ($P = 0.06$). Horses consumed more PP than AP ($P < 0.0001$) and more A was eaten than AP ($P < 0.001$), as presented in Table 5-4. Horses tended to eat more A than PP ($P = 0.09$).

It is unlikely that nutritional composition (CP and NDF contents) played a role in the preferences of the horses in this study. In studies measuring voluntary intake, horses may choose to eat more or less of a forage based on CP or NDF content, though these relationships are relatively weak or nonexistent for legume forages (Dulphy et al., 1997; Edouard et al., 2008). Instead, it is more likely that the palatability (odor, taste, and texture) of the forages influenced the preferences of the horses. The AP hay had a smaller amount of leaves compared to stems, had thicker stems, contained more weeds, and was more dusty than A or PP, which may have deterred ingestion by the horses (Prache et al. 1998).

Neophobia may have been a factor in limiting the intake of AP. The horses in this study had never been fed AP hay before and the feeding method was relatively new. Herskin et al.

(2003) observed changes in the behaviors of cattle (increased sniffing, decreased feeding duration, and increased self-grooming) when food was presented in a novel way or when novel ingredients (fish oil, eucalyptus oil, or carrots) were added to the cattle's accustomed diet. In the few days immediately prior to this study, the horses were presented with all of the forages in the same arrangement as the trial in an attempt to mitigate the effects of neophobia and to provide data on how much the horses could eat of one forage in the allotted time (10 min). Nevertheless, some individuals may be better at coping with novelty than others.

Perennial peanut hay was accepted by the horses but perhaps not as much as A hay. This may be due to quality of the PP hay. During a portion of the trial, the PP was less leafy and had the same spiny weeds that AP had. It is likely that this "bad" PP affected the results.

Horses evolved a patch-foraging strategy that has allowed them to obtain the most nutrition from their environment; even when sources of food were scant (Prache et al., 1998). Studies on foraging behavior in horses have shown that horses prefer variety, and will eat less preferable forages or flavors along with more preferred forages or flavors (Archer, 1971, 1973; Goodwin et al., 2002; Goodwin et al., 2005a, b; Thorne et al., 2005). This natural instinct of horses may have influenced the results.

These results suggest that horses prefer PP almost as much as A and that AP was less preferable than either PP or A. Further investigation should be done, however, such as increasing the sample size and duration of the study, before definitive conclusions be made.

Table 5-1. Description of treatments.

Treatment	Left Hay ¹	Right Hay
1	PP	AP
2	AP	PP
3	PP	A
4	A	PP
5	A	AP
6	AP	A

¹PP = perennial peanut hay; AP = annual peanut hay; A = alfalfa hay.

Table 5-2. Analyzed composition of annual peanut (AP), perennial peanut (PP), and alfalfa (A) hays, % (DM basis).

Item ¹	DM	CP	NDF	ADF
AP	89.7	16.6	49.8	40.4
PP	89.8	15.5	44.8	33.8
A	90.4	20.8	39.7	31.7

¹Analyses done by a commercial forage analysis laboratory by near-infrared spectroscopy (Dairy One, Ithaca, NY). AP = annual peanut hay, 2006; PP = perennial peanut hay, 2006; A = alfalfa hay, 2006.

Table 5-3. Time spent consuming perennial peanut (PP), annual peanut (AP), or alfalfa (A) hays.

Hay Comparison	Time (min) ¹	P-value	SE ²
PP	5.88	0.004	0.74
AP	2.01		
PP	2.58	0.06	0.72
A	4.80		
A	6.32	0.0001	0.53
AP	1.34		

¹Time spent consuming hay.

²SE = standard error; n = 12.

Table 5-4. Amounts consumed of perennial peanut (PP), annual peanut (AP), and alfalfa (A) hays.

Hay Comparison	Amount consumed (kg)	P-value	SE ¹
PP	0.296 (74%) ²	<0.0001	0.022
AP	0.104 (26%)		
PP	0.160 (40%)	0.09	0.028
A	0.240 (60%)		
AP	0.122 (30%)	<0.001	0.024
A	0.298 (70%)		

¹SE = standard error; n = 12.

²Numbers in parentheses are amounts consumed as a percentage of amount offered.

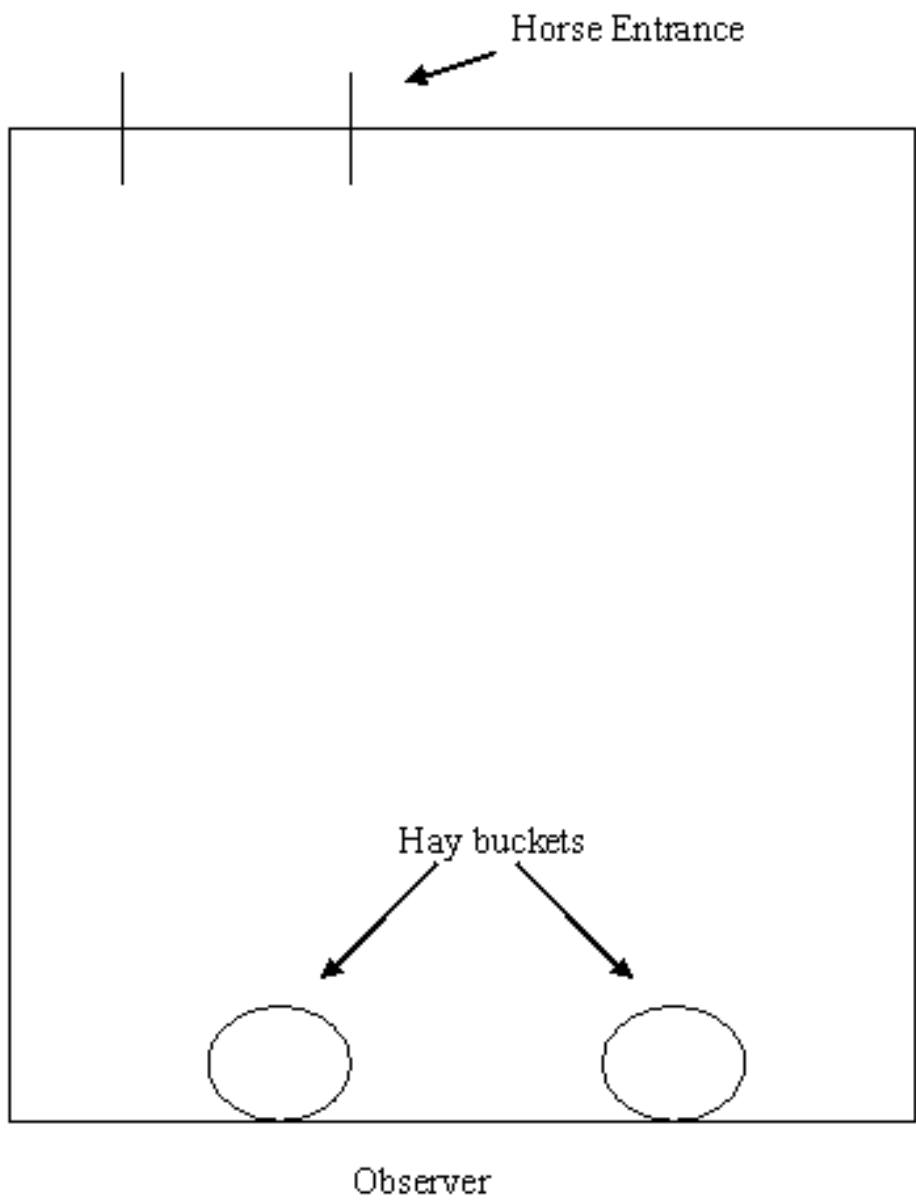


Figure 5-1. Individual feeding slip and hay offering

CHAPTER 6 GENERAL SUMMARY

A series of experiments were conducted to determine the digestibility and suitability of perennial peanut (PP), annual peanut (AP), and Tifton 85 (T85) bermudagrass hays for horses. Perennial peanut (*Arachis glabrata*) is rapidly gaining popularity as legume hay for horses in the Southeastern United States, yet not much is known about its suitability as an equine feed. It grows well in the particular climate and soil types of the coastal plain region and peninsular Florida of the Southeastern U.S., where alfalfa (*Medicago sativa*), the most well-known and popular legume hay for horses, does not grow well. Many studies have found PP hay to be a highly digestible legume for ruminants (Romero et al., 1987; Ocumpaugh, 1990; Terrill et al., 1996; Staples et al., 1997; West et al., 1998; Hill, 2002; Chambliss and Dunavin, 2003; Hernandez Garay et al., 2004; Williams et al., 2004). Lieb et al. (1993) found PP hay to have similar dry matter (DM), crude protein (CP), neutral detergent fiber (NDF), and energy digestibility as alfalfa hay. Perennial peanut hay was also readily accepted by horses and even exceeded alfalfa in voluntary intake.

Annual peanut (*Arachis hypogea*) is similar to PP in that it grows well in the Southeastern U.S., however, even less information is available about its uses in the horse industry. Annual peanut residue hay and peanut by-products have been fed to ruminants for over 75 yr in the United States (Hill, 2002). Annual peanut hay has been found to be nutritious forage for ruminants. Crude protein is generally high in AP dried forage, similar to that of alfalfa. Gorbet et al. (1994) reported an average CP content of ten cultivars of AP grown specifically as hay to be 16.9% or 16.5% in the first or second cuttings, respectively. Annual peanut hay is also utilized in West Africa to supplement cattle during the dry season (Larbi et al., 1999). There have been no studies done with horses to determine the suitability and digestibility of AP hay in horses.

Tifton 85 bermudagrass (T85) has been a popular forage for ruminants since its release in 1992 (Chambliss and Dunavin, 2003). It is more productive and nutritious than its popular counterpart, Coastal bermudagrass (CB). Hill et al. (1993) conducted one of the first experiments to compare T85 to other bermudagrasses and found a 26% greater DM yield and 11% greater in vitro dry matter disappearance (simulates ruminant digestion) than CB in two 3-year trials. Mandebvu et al. (1999) found T85 was more digestible than CB in DM, NDF, and ADF in growing beef steers. Tifton 85 also has different lignin chemistry than CB, making the cell wall of T85 more digestible than CB. However, like CB, the quality of T85 decreases rapidly with maturity. Mandebvu et al. (1999) cautions that after 5 wks of regrowth, the digestibility of NDF, ADF, cellulose, and hemicellulose decreases in both bermudagrasses.

McCann et al. (1995) compared the digestibility of CB and T85 hays in horses. Both bermudagrasses were found to be similar in DM, CP, NDF, and acid detergent fiber (ADF) digestibilities. There was, however, no data on intake or the maturity of the two grasses.

Lieb and Mislevy (2001, 2003) found T85 could be variable in quality. In 2001, they found that T85 was equal to CB in DM, NDF, and energy digestibilities. Coastal bermudagrass was greater in crude protein digestibility (CPD) than T85. Intake of T85 was less than CB and T85 could not maintain a positive body weight on any horse. In 2003, they used T85 of better quality, with a higher CP content and visually looked better. In this trial, T85 was greater than CB in CP, NDF, and energy digestibilities. Intakes of T85 and CB were lower in this trial than the previous one and T85 and CB both produced positive weight gains in the horses. The researchers observed that the horses were still reluctant to eat T85, therefore they cautioned horse owners to provide supplemental sources of nutrition if feeding T85, even if the T85 is of good quality.

The objectives of the present horse *in vivo* digestibility experiment were to evaluate and compare apparent DM, OM, CP, and NDF digestibilities, as well as balances of Ca (CaBal), P (PBal), and N (NB) in mature horses fed PP, CB, or T85 hays. The trial was conducted between April and May of 2006. Six horses were used in a 3X3 Latin square design; with a 10-d adjustment phase and a 4-d total fecal and urine collection phase. Horses were given water *ad libitum* and fed 1.5% to 2% of their body weight in hay (DM basis). Hay was subsampled every day during collection and analyzed for DM, Ash, CP, N, NDF, ADF, Ca, P, Mg, K, Cu, Mn, Fe, Zn, and Na. Hay refusals were gathered during the collection period and analyzed for DM. After collection, feces and urine were mixed separately and then subsampled. Subsamples were later compiled and analyzed for DM, Ash, CP, N, NDF, ADF, Ca, and P.

Dry matter, OM, and CP digestibilities were greater for PP than the mean of the two bermudagrasses ($P = 0.0004$; $P = 0.0002$; $P = 0.004$, respectively). There were no differences between PP and the mean of the two bermudagrasses in NDFD ($P = 0.40$). Coastal bermudagrass and T85 did not differ in DM, OM, CP, and NDF ($P = 0.80$; $P = 0.80$; $P = 0.30$; $P = 0.50$, respectively). Calcium balance was not different between PP and the mean of the bermudagrasses ($P = 0.50$) nor was it different between CB and T85 ($P = 0.70$). Phosphorus balance was greater in PP than the mean of the two bermudagrasses ($P = 0.03$) and was not different between CB and T85 ($P = 0.40$). Nitrogen balance was greater in PP than the mean of the bermudagrasses ($P = 0.01$) and the NB of CB was greater than T85 ($P = 0.01$).

The CB used in this study was younger than the T85 (4 wk versus 5 wk). It is likely the maturity of T85 affected the results. Coastal bermudagrass had a greater NB than T85, probably due to low CP content, overall weight loss of horses fed T85 (even when fed at 2% BW) compared to CB and PP, and decreased intake of T85. Refusals (% of intake) were greater for

T85 (7.13%) than CB (4.82%) and PP (3.41%). Another explanation for the negative NB of T85 could be the less N retained (as % of intake) from T85 compared to CB and PP. Nitrogen may not be as efficiently absorbed from T85 as from CB and PP. From these results and observations made during the trial, 5wk T85 would be able to take the place of 4 wk CB in the diets of horses, however intake will be reduced. It would be interesting to see if the same holds true for less mature T85 (e.g. > 4 wk).

Perennial peanut hay, as expected, was more digestible than CB and T85 (DM, OM, and CP) and has a greater NB. When compared to digestibility data on alfalfa, DM, OM, and NDF digestibilities have been similar to PP (Lieb et al., 1993; Crozier et al.1997; LaCasha et al. 1999). Digestibility of CP of PP in the present experiment was slightly less than previously reported; however, the PP hay used in the present experiment was more mature than normal due to a delayed harvest.

The objectives of the horse *in vitro* digestibility experiment were to compare the IVTD of dried fresh forage samples of annual peanut (AP –Fresh1 and –Fresh2) and perennial peanut (PP –Fresh); hay samples of perennial peanut (PP06 –hay; taken from the PP –Fresh field), annual peanut (AP –hay; taken from AP –Fresh2 field), and commercial alfalfa (A–hay) from the selection trial; hay samples of perennial peanut (PP05–hay), T85–hay, and CB–hay from the *in vivo* digestion trial (Chapter 3); and dried fresh forage samples from six varieties of perennial peanut (Arbrook, AR; Floragrazer, FG; Tom Burton Selection, TB; Ecoturf, EC; Elite Line, EL; and Good Line, GL). An enclosed fermentation incubator (Daisy^{II}; ANKOM Technology Corp., Fairport, NY) was used to ascertain the *in vitro* true digestibility (IVTD) of the samples. Freshly voided horse feces were used as a source of microbial inoculum. About 0.5 g of sample was put into ash-free, N-free bags (F57; ANKOM, Macedon, NY) and heat-sealed. Twenty-one samples

and two blanks were added to one jar, and four jars were used (replicated four times). Jars were heated and rotated simultaneously for 48 h. After the incubation period, samples were put through NDF analysis using the ANKOM Fiber Analyzer (ANKOM Technology Corp., Fairport, NY).

Annual peanut–Fresh1 and AP–Fresh2 were greater in IVTD than PP–Fresh ($P < 0.01$), however, PP06–hay was similar in IVTD to AP–hay ($P = 0.20$), suggesting AP loses a greater amount of nutrients during haying and storage than PP. Comparisons of the IVTD and nutrient composition between the fresh forage and the hay lend weight to this assumption. Perennial peanut06–hay and PP–Fresh IVTD did not differ ($P = 0.32$), whereas the IVTD of both AP–Fresh1 and AP–Fresh2 were greater than AP–hay ($P < 0.0001$ for both). Neutral detergent fiber and ADF both increased in AP as a result of haying and storage, however, only NDF increased in PP. Crude protein content dramatically reduced in AP as a result of hay harvest and storage, suggesting that AP is better suited as silage/haylage or for grazing.

The IVTD of PP06–hay and A–hay were no different from each other. Therefore, PP can be as nutritious as A, if intake is similar. Another *in vivo* digestion trial comparing PP and A in horses is recommended to measure intake and its effect on digestibility of these two hays. Annual peanut–hay and PP06–hay did not differ in IVTD as well. However, this may be the perfect example of the effect of intake on digestibility. During the selection trial (Chapter 5), it was observed that horses did not eat as much of or spend as much time on AP than PP or A.

The IVTD of PP05–hay was numerically greater than the digestibility from the *in vivo* trial (68.5% *in vitro* versus 65% *in vivo*). *In vitro* true digestibility measures the total potential a forage (or other sample) can be digested, therefore, it overestimates actual digestibility. The most

intriguing result of running the same hays from the *in vivo* experiment through an *in vitro* system was the dramatically lower IVTD of the two grass hay than what was found *in vivo*.

The only variation seen between the three adaptation and collection periods of the *in vivo* experiment was in CB. Period 3 CB had a significantly greater IVTD than Periods 1 and 2. The IVTD mirrors that of the *in vivo* data, insofar as the two bermudagrasses were similar in digestibility and less digestible than PP.

The *in vitro* procedure is valuable to agronomists when considering varieties and cultivars of plants to propagate. All PP evaluated in the PP variety trial had high digestibility, but there were some differences. Arbrook, another commercially available variety of PP (the other two are Florigraze and Ecoturf), is not as digestible as the other varieties; it is in fact, the least digestible ($P < 0.05$). The most digestible were EL and EC, which were also numerically greater than PP05 and PP06 (both Florigraze). All varieties had comparable IVTDs to PP and A. It is recommended to further study EL and EC digestibilities in horses.

The objective of the selection experiment was to determine the preference of mature horses offered either A, PP (from 2006), or AP hay (from AP–Fresh2 field). Six yearling horses living at the University of Florida Horse Research Center (Gainesville, FL) participated in this trial during July 2007. Horses remained on pasture with Coastal bermudagrass *ad libitum* and an evening feeding of grain. Horses were brought into individual feeding slips before their grain feeding and observed for 10 min. Horses were given a choice of two hays (in buckets) at a time. There were six treatments consisting of hays in all possible combinations to eliminate side preference. The amount of time a horse spent eating either hay was recorded and refusals were collected and weighed.

Results showed horses spent more time on and ate more of both A ($P = 0.0001$ and $P < 0.001$, respectively) and PP ($P = 0.0004$ and $P < 0.0001$, respectively) than AP. Horses tended to spend more time on and eat more of A than PP ($P = 0.09$ and $P = 0.06$, respectively). The texture of AP was probably the reason horses did not spend time eating it, as the AP hay had thicker stems, was less leafy, was very dusty, and had spiny weeds mixed in. Another complication could have been the relative newness of AP to the horses. They were never exposed to AP until shortly before data collection. The PP hay also had the same spiny weeds and less leaves during a portion of the experiment and probably affected the results.

These experiments demonstrate the highly digestible PP ('Florigraze' variety) is suitable for horses. Other varieties of PP were found to have potential as hay for horses. Annual peanut is best recommended as silage/haylage or as a grazed legume because of the losses in nutritive quality during harvest and storage. Also, the hay has thicker stems and is very dusty, both contributing to the reluctance of horses to eat the hay. Tifton 85 hay may be fed to horses, especially if CP content is similar to a legume, but decreased intake should be expected. It is recommended that horses fed T85 hay also be offered other sources of nutrients.

There is still more work that can be done to further the information available for horse owners on the uses of PP, AP, and T85. Suggested studies include

- *In vivo* experiments comparing PP, A, and AP
- Grazing trial comparing PP, A, and AP
- *In vivo/in vitro* experiments comparing T85 and CB of similar maturity (younger than 5 wk)
- *In vivo* experiments evaluating the newer PP varieties that show promise from the *in vitro* experiment
- Repeating the selection trial with more horses and for longer periods of time.

APPENDIX
 SUPPLEMENTAL DATA FOR *IN VIVO* DIGESTIBILITY TRIAL

Table A.1. Body weights of horses throughout trial.

Horse	Pre-Trial (d -5)	Before P2 Collection (d 23)	Before P3 Collection (d 37)	After-Trial (d +3)
A	520	505	505	503.5
B	518	499.5	507.5	500
C	516	475.5	ND ¹	ND
D	578	548.5	555.5	558
E	522	499	511	503
F	599	545.5	596.5	585

¹ND: no data collected

Table A.2. Period 1: Amount of hay offered, amount of hay refused, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of perennial peanut (PP), Tifton 85 bermudagrass (T85), or Coastal bermudagrass (CB) hays by horse.

Horse	Hay	Offered, kg ¹	Refused ² , kg	DM, %	OM, %	CP, %	NDF, %
A	PP	8.85		88.6	91.8	12.08	47.7
B	PP	8.81		88.6	91.8	12.08	47.7
C	T85	8.65		91.0	93.9	8.61	77.8
D	T85	9.68	0.42	91.0	93.9	8.61	77.8
E	CB	9.13		93.9	94.7	9.63	74.2
F	CB	10.48		93.9	94.7	9.63	74.2

¹As-fed basis.

²Total amount of refusals for all days of collection.

Table A.3. Period 2: Amount of hay offered, amount of hay refused, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of perennial peanut (PP), Tifton 85 bermudagrass (T85), or Coastal bermudagrass (CB) hays by horse.

Horse	Hay	Offered, kg ¹	Refused ² , kg	DM, %	OM, %	CP, %	NDF, %
A	T85	8.86	0.43	92.4	94.1	8.39	77.9
B	T85	8.83	0.80	92.4	94.1	8.39	77.9
C	CB	8.63		92.1	94.6	9.62	75.7
D	CB	9.67	0.48	92.1	94.6	9.62	75.7
E	PP	9.14		95.0	91.7	12.11	44.4
F	PP	10.48		95.0	91.7	12.11	44.4

¹As-fed basis.

²Total amount of refusals for all days of collection.

Table A.4. Period 3: Amount of hay offered, amount of hay refused, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of perennial peanut (PP), Tifton 85 bermudagrass (T85), or Coastal bermudagrass (CB) hays by horse for Period 3.

Horse	Hay	Offered, kg ¹	Refused ² , kg	DM, %	OM, %	CP, %	NDF, %
A	CB	12.44	5.05	96.7	93.7	11.2	70.6
B	CB	13.39	5.07	96.7	93.7	11.2	70.6
D	PP	13.69	3.11	94.2	92.2	11.98	46.1
E	T85	12.21	5.66	96.4	94.1	8.86	74.2
F	T85	14.06	8.54	96.4	94.1	8.86	74.2

¹As-fed basis.

²Total amount of refusals for all days of collection.

Table A.5. Period 1: Total fecal excretion, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of feces by horse fed perennial peanut (PP), Tifton 85 bermudagrass (T85) or Coastal bermudagrass (CB) hays.

Horse	Ha	Total Fecal Excretion, kg ¹	DM, %	OM, %	CP, %	NDF, %
A PP						
	d 1	15.90	21.82	86.15	11.48	67.27
	d 2	10.15	21.78	84.58	11.24	64.74
	d 3	10.92	25.10	86.43	10.96	63.83
	d 4	11.50	21.71	83.66	11.04	62.34
	Mean	12.12	22.60	85.21	11.19	64.54
B PP						
	d 1	13.80	25.04	86.04	11.41	66.75
	d 2	10.01	24.68	84.38	11.26	63.76
	d 3	10.59	22.01	85.90	11.58	65.23
	d 4	11.10	21.65	85.69	12.45	63.16
	Mean	11.37	23.35	85.50	11.67	58.83
C T85						
	d 1	19.08	21.63	91.52	10.12	77.55
	d 2	17.06	21.67	92.76	8.53	80.55
	d 3	13.667	21.67	93.33	9.39	81.11
	d 4	20.58	18.67	93.76	8.05	80.71
	Mean	17.60	20.93	92.84	9.02	79.98
D T85						
	d 1	21.88	21.83	93.91	8.32	79.85
	d 2	17.90	18.61	93.38	7.75	80.03
	d 3	22.24	18.67	92.54	7.37	78.65
	d 4	21.34	18.62	92.59	9.14	81.64
	Mean	20.84	19.43	93.11	8.15	80.04
E CB						
	d 1	12.27	18.75	91.96	9.18	77.87
	d 2	24.87	21.64	93.14	8.98	80.29
	d 3	12.44	21.77	92.69	8.56	80.09
	d 4	19.17	21.56	93.84	8.75	80.40
	Mean	17.17	20.93	92.91	8.87	79.66
F CB						
	d 1	18.75	18.73	93.61	8.90	78.92
	d 2	21.64	18.74	93.76	9.57	80.19
	d 3	21.77	18.73	93.61	8.74	80.01
	d 4	21.56	18.66	93.76	8.75	81.10
	Mean	20.93	18.72	93.68	8.99	80.30

¹Wet weight.

Table A.6. Period 2: Total fecal excretion, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of feces by horse fed perennial peanut (PP), Tifton 85 bermudagrass (T85) or Coastal bermudagrass (CB) hays.

Horse	Hay	Total Fecal Excretion, kg ¹	DM, %	OM, %	CP, %	NDF, %
A	T85					
	d1	24.81	18.46	93.32	5.96	0.83
	d2	23.71	15.33	92.93	6.58	0.83
	d3	24.40	18.44	93.81	6.26	0.83
	d4	22.87	21.78	94.12	6.34	0.82
	Mean	23.95	18.50	93.55	6.28	0.83
B	T85					
	d1	20.98	21.66	94.39	6.53	0.81
	d2	22.54	21.67	94.62	6.52	0.82
	d3	19.77	21.67	95.01	6.21	0.84
	d4	20.08	20.23	94.10	6.40	0.83
	Mean	20.84	21.31	94.53	6.41	0.83
C	CB					
	d1	24.55	18.52	93.07	8.27	0.82
	d2	23.78	18.45	94.03	7.94	0.82
	d3	23.42	21.63	93.08	7.85	0.68
	d4	23.72	19.88	93.39	8.19	0.81
	Mean	23.07	19.62	93.73	8.06	0.78
D	CB					
	d1	26.21	15.29	93.38	7.64	0.83
	d2	28.68	18.51	94.08	7.35	0.66
	d3	26.19	15.47	93.76	10.78	0.77
	d4	28.16	15.44	93.76	7.78	0.80
	Mean	27.31	16.18	93.74	8.39	0.78
E	PP					
	d1	15.98	21.41	86.61	11.28	0.66
	d2	14.98	21.51	86.44	10.74	0.69
	d3	14.54	24.54	85.96	11.62	0.67
	d4	13.17	24.59	85.61	11.27	0.67
	Mean	14.67	23.01	85.16	11.23	0.75
F	PP					
	d1	14.12	24.39	86.67	13.22	0.67
	d2	15.32	24.37	85.22	10.54	0.66
	d3	15.82	24.47	85.85	10.42	0.66
	d4	17.32	31.55	85.92	10.72	0.66
	Mean	15.64	23.70	85.91	11.22	0.67

¹Wet weight.

Table A.7. Period 3: Total fecal excretion, dry matter (DM), organic matter (OM), crude protein (CP), and neutral detergent fiber (NDF) content of feces by horse fed perennial peanut (PP), Tifton 85 bermudagrass (T85) or Coastal bermudagrass (CB) hays.

Horse	Hay	Total Fecal Excretion, kg ¹	DM, %	OM, %	CP, %	NDF, %
A						
	CB					
	d1	36.53	18.40	92.09	8.12	78.69
	d2	27.49	18.34	92.65	8.21	80.54
	d3	31.73	18.35	90.93	8.74	79.95
	d4	33.46	15.15	92.32	8.37	81.82
	Mean	32.30	17.56	92.00	8.34	80.25
B						
	CB					
	d1	26.78	18.36	93.40	8.21	78.95
	d2	25.73	18.44	92.96	8.50	79.71
	d3	25.05	18.31	93.63	8.39	80.81
	d4	27.07	18.31	93.28	8.14	80.71
	Mean	26.16	18.36	93.31	8.31	80.04
D						
	PP					
	d1	21.29	18.08	86.35	10.83	63.05
	d2	20.35	21.04	85.61	10.67	66.15
	d3	27.07	21.22	86.42	10.92	66.00
	d4	21.96	18.15	84.84	11.62	61.70
	Mean	20.45	19.62	85.80	11.01	64.23
E						
	T85					
	d1	28.55	18.27	90.63	7.74	78.69
	d2	25.03	18.38	91.21	8.07	78.16
	d3	25.35	18.22	90.29	8.51	75.09
	d4	26.88	18.25	91.91	7.60	78.42
	Mean	26.45	18.18	91.01	7.68	77.79
F						
	T85					
	d1	30.57	18.20	92.34	7.51	80.05
	d2	28.47	15.25	91.98	8.50	75.85
	d3	28.40	18.37	91.79	7.92	75.41
	d4	27.34	18.33	92.04	7.43	76.51
	Mean	28.70	17.54	92.03	7.84	76.95

¹Wet weight.

Table A-8. Daily urine production for horses on perennial peanut hay (PP), Coastal bermudagrass hay (CB), and Tifton 85 bermudagrass hay (T85).

Urine Production, mL			
Horse	Period 1	Period 2	Period 3
A	PP	T85	CB
d1	9020	4280	6720
d2	6950	4920	5720
d3	5500	4420	5900
d4	6590	5960	5840
Mean	7015	4895	6045
B	PP	T85	CB
d1	7342	5460	7680
d2	7400	5320	6620
d3	6780	5580	6285
d4	8200	5480	5100
Mean	7430.5	5460	6421.25
C	T85	CB	
d1	3130	4560	ND ¹
d2	7100	5280	ND
d3	7120	5860	ND
d4	5765	4780	ND
Mean	5778.75	5120	ND
D	T85	CB	PP
d1	6603	6000	9060
d2	4520	3324	9360
d3	4480	5140	11320
d4	6690	4080	9600
Mean	5573.25	4636	9835
E	CB	PP	T85
d1	2000	8830	6080
d2	0	8900	4920
d3	4220	7690	3640
d4	2020	8720	4460
Mean	2060	8535	4775
F	CB	PP	T85
d1	7510	9220	9460
d2	7130	11840	8540
d3	5320	11170	6720
d4	5260	133870	6500
Mean	6305	11525	7805

¹ND = No data.

Table A-9. Mineral concentrations for all periods of perennial peanut hay (PP), Coastal bermudagrass hay (CB), and Tifton 85 bermudagrass hay (T85).

Period	Hay	Ca, %	P, %	Mg, %	K, %	Cu, ppm	Mn, ppm	Fe, ppm	Zn, ppm	Na, ppm
1	PP	1.16	0.20	0.46	1.50	4.72	86.12	80.66	29.05	235.96
2	PP	1.18	0.18	0.46	1.51	4.09	81.02	137.92	29.79	88.28
3	PP	1.20	0.25	0.39	1.54	4.10	136.40	83.30	33.70	136.19
1	CB	0.28	0.14	0.11	1.06	6.85	35.62	70.66	20.80	128.85
2	CB	0.31	0.14	0.13	1.31	5.45	36.18	87.55	20.76	144.77
3	CB	0.29	0.19	0.13	1.46	6.06	34.67	97.66	30.77	249.74
1	T85	0.28	0.18	0.14	1.43	4.07	30.43	80.55	21.64	142.39
2	T85	0.28	0.23	0.17	1.25	5.41	27.32	94.15	36.23	190.26
3	T85	0.32	0.19	0.15	1.47	5.08	39.73	109.30	26.57	186.91

Table A-10. Calcium (Ca) and phosphorus (P) content of feces for all horses and periods (P) for perennial peanut hay (PP), Coastal bermudagrass hay (CB), and T85 bermudagrass hay (T85).

Horse	Hay	Ca, %	P, %
A			
P1	PP	1.55	0.48
P2	T85	0.19	0.33
P3	CB	0.19	0.28
B			
P1	PP	1.58	0.61
P2	T85	0.22	0.36
P3	CB	0.20	0.34
C			
P1	T85	0.26	0.47
P2	CB	0.27	0.30
D			
P1	T85	0.38	0.46
P2	CB	0.28	0.32
P3	PP	1.38	0.48
E			
P1	CB	0.29	0.38
P2	PP	1.35	0.44
P3	T85	0.24	0.37
F			
P1	CB	0.24	0.43
P2	PP	1.48	0.43
P3	T85	0.20	0.44

Table A-11. Calcium (Ca) and phosphorus (P) concentrations of urine for all horses and periods (P) for perennial peanut hay (PP), Coastal bermudagrass hay (CB), and Tifton 85 bermudagrass hay (T85).

Horse	Hay	Ca, % ¹	P, %/mL ¹
A			
P1	PP	0.02	0.00030
P2	T85	0.08	0.00048
P3	CB	0.03	0.00062
B			
P1	PP	0.03	0.00039
P2	T85	0.03	0.00046
P3	CB	0.01	0.00059
C			
P1	T85	0.11	0.00066
P2	CB	0.08	0.00045
D			
P1	T85	0.03	0.00031
P2	CB	0.05	0.00056
P3	PP	0.02	0.00059
E			
P1	CB	0.09	0.00012
P2	PP	0.002	0.00042
P3	T85	0.07	0.00103
F			
P1	CB	0.06	0.00050
P2	PP	0.03	0.00042
P3	T85	0.05	0.00056

¹Values are not corrected for specific gravity.

Table A-12. Nitrogen (N) concentrations of feces for all horses and periods (P) for perennial peanut hay (PP), Coastal bermudagrass hay (CB), and Tifton 85 bermudagrass hay (T85).

Horse	Hay	N, %
A		
P1	PP	1.79
P2	T85	1.56
P3	CB	1.31
B		
P1	PP	1.87
P2	T85	1.32
P3	CB	1.52
C		
P1	T85	1.44
P2	CB	1.27
D		
P1	T85	1.30
P2	CB	1.32
P3	PP	1.42
E		
P1	CB	1.42
P2	PP	1.52
P3	T85	1.37
F		
P1	CB	1.44
P2	PP	1.26
P3	T85	1.45

Table A-13. Nitrogen (N) concentrations of urine for all horses and periods (P) for perennial peanut hay (PP), Coastal bermudagrass hay (CB), and Tifton 85 bermudagrass hay (T85).

Horse	Hay	N, % ¹
A		
P1	PP	1.02
P2	T85	1.40
P3	CB	1.52
B		
P1	PP	1.08
P2	T85	1.17
P3	CB	1.26
C		
P1	T85	1.51
P2	CB	1.41
D		
P1	T85	1.43
P2	CB	1.63
P3	PP	0.89
E		
P1	CB	1.44
P2	PP	0.92
P3	T85	1.37
F		
P1	CB	1.00
P2	PP	0.77
P3	T85	1.12

¹Values are not corrected for specific gravity.

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BIOGRAPHICAL SKETCH

Juliet Eckert was born in Indianapolis, Indiana in 1982. Her family shortly moved to Minnetrista, Minnesota, where she spent most of her formative years. After finishing high school in Palm Harbor, Florida, Juliet moved back to Indiana to complete her BS in biological sciences at Saint Mary-of-the-Woods College in Saint Mary-of-the-Woods, Indiana.

Upon completion of her masters degree, Juliet hopes to enter the field of comparative nutrition. She now lives in Atlanta, GA, and works at the Georgia Aquarium.