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Five experiments were conducted to evaluate tree forage as a feedstuff for small livestock, using the foliage of black locust (*Robinia pseudoacacia*) and hybrid poplar (*Populus* spp.). In the first trial, rabbits were fed diets containing 40% alfalfa (control), and 10%, 20% and 40% poplar leaves (PL) from untrimmed or uncoppiced growth, and 10% and 20% PL from trimmed or coppiced growth. At the 20% level, crude protein (CP) was better digested for the coppiced PL than for the uncoppiced ($p < .05$). No significant difference was evident between treatments in the average daily gain (ADG) or feed efficiency, but dry matter (DM) intake increased for all the PL treatments compared to the control ($p < .01$). In the second experiment, black locust (BL) leaves were treated by various methods in an attempt to counter the effect of tannins. Rabbits were fed diets containing 50% alfalfa (control), 25% alfalfa and 25% black locust meal

(BLM) (BLM control), BLM + 1% polyethylene glycol (PEG), BLM + 1% phytase, and BLM + 0.3% L-methionine and 0.3% choline chloride. In general, nutrient utilization and ADG were better for the control than for the BLM treatments. The addition of PEG increased CP digestibility ($p < .01$) and ADF ($p < .03$) compared to the BLM control, partially alleviating the reduced nutrient availability. The addition of L-methionine and choline chloride increased ADF digestibility ($p < .02$). In the third trial, black locust (BL) bark and other tree products were used to study the possible toxic effects of lectins in BL bark. Rabbits were fed diets containing 25% BL bark, oak sawdust, red alder bark, or red alder sawdust (all diets also included 25% alfalfa). A 50% alfalfa diet served as a control. In general, nutrient digestibilities and ADG were higher for the control than the treatments and, also, higher for the BL bark diet than the alder bark diet. The ADG with the BL bark diet was lower than for the alder bark diet ($p < .01$), which indicated a possible toxic effect of the BL bark. The fourth trial examined the feeding value of poplar leaves for sheep. Sheep were fed diets containing 50% PL or 50% alfalfa. Nutrient digestibilities were lower for the PL diet ($p < .01$). In the fifth experiment, BL leaves were fed to sheep and goats in order to determine if goats, being browsers instead of grazers like the sheep, are better equipped to

tolerate the anti-nutritive effects from BL forage. Sheep and goats were fed diets composed solely of BL leaves or alfalfa (control). Overall, the nutrient digestibilities were higher for alfalfa than for BL leaves, and there was no difference in terms of digestibility between species. Although the leaves of black locust and poplar contain anti-nutritive factors, it was concluded that the trees have potential as multipurpose trees from which the leaves could be harvested as animal fodder, particularly in temperate areas of the developing world. (Key words: black locust, poplar, agroforestry, tannins)

EVALUATION OF TREE FORAGE AS A NONTRADITIONAL FEEDSTUFF FOR SMALL LIVESTOCK

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INTRODUCTION

The 5 experiments described in this thesis explored, along with the feeding values, some of these areas of interest which arise in feeding tree forage to small livestock, both monogastric and ruminant. In the first trial, rabbits were fed diets containing poplar leaf meal from coppiced and uncoppiced sources, examining the induction of the defense mechanism in coppiced trees. In the second trial, rabbit were fed diets containing black locust leaves which had been treated by various methods in an attempt to alleviate the negative affects of tannins. In the third trial, rabbits were fed diets containing black locust bark and other tree products in an examination of the toxic qualities of the bark, particularly of lectins in the bark. In the fourth trial, sheep were fed diets containing poplar leaf meal in order to determine its feeding value. In the fifth experiment, black locust leaves were fed to sheep and goats in order to determine if goats, being browsers instead of grazers like the sheep, are better equipped to tolerate the antinutritive effects in black locust forage.

REVIEW OF LITERATURE

The Potential of Tree Forage as Feed for Small Livestock

The use of tree forage as a feedstuff for livestock has not only world-wide potential, but also a long history. According to Snook (1986), the Romans fed tree leaves to animals 2000 years ago. In the U.S. and other industrialized countries, the possibility exists to utilize forest industry by-products such as leaves, bark, and sawdust in animal feeding. Trees could also be incorporated into agroforestry systems, integrating trees with crops and animals to better make use of natural resources. In many developing countries, particularly in the tropics, agroforestry has already been put to work in a practical way. Trees have a role in animal feeding as browse. Trees could also be cultivated in forage plantations.

Deforestation in the tropics occurs at a rate of 3000 acres an hour or an area the size of Pennsylvania (27 million acres) per year (World Resources Institute, 1985). Most deforestation of tropical moist forests is due to expansion for agriculture, cattle ranching, and the harvesting of timber (Caufield, 1982). The "slash and burn" or migrating agriculture functioned well in the past, but now with increased population pressure, the land does not lie fallow long enough to recuperate for agriculture (National Geographic Society, 1990). Clearing the rainforest erases rich biodiversity and initiates a downward spiral of environmental degradation. Removing forest cover in the tropics creates a greater danger of erosion than in temperate areas due to intense rainfall, and burning the forest releases stored CO_2 which may contribute to the greenhouse effect (Caufield, 1982). The soil beneath tropical moist forests is virtually sterile because the nutrients are contained in the vegetation (Caufield, 1982). Annual crops can usually not be supported for more than 2-3 years, after which the soil has been leached of nutrients and is unsuitable for cropland. It is often then used for cattle pasture; however, heavy cattle grazing may not be appropriate for sustainable agriculture in the tropics. According to Benge (1987) deriving firewood and fodder from the forest also contributes to deforestation. In a study of two Indian villages, 79% of all fodder consumed came from the forest (Benge, 1987).

Agroforestry systems which incorporate multipurpose trees, particularly fast-growing, nitrogen-fixing trees, can contribute towards sustainable agriculture in the tropics and help alleviate the pressure on the forest.

Uses of trees may include hillside plantations in hedgerows along contour lines to provide a physical barrier to slow soil erosion and aid in watershed protection. Nurse or shade trees are often planted in perennial plantations of coffee, tea or cacao, or in alley-cropping systems where the trees are interdispersed with rows of annual crops and the trees coppiced regularly to incorporate the leaves into the soil as green mulch. Further uses of trees may include "protein banks" or small tree plots which are maintained for supplemental feed, protection against desertification, firewood, windbreaks, timber, bee forage, shelter, medicinal purposes, live fences, and fodder for animals (OTS and CATIE, 1986; Benge, 1987). Leguminous trees fix atmospheric nitrogen and the leaves are often very high in protein, up to 35% protein has been reported (Cheeke and Shull, 1985). Tree leaves could be a valuable supplemental source of fodder in dry seasons because of tree roots being able to reach water below surface. In temperate areas, agroforestry systems incorporating livestock and pine trees have received attention (Anderson et al., 1988).

Trees and shrubs have a role in animal feed as browse. According to McKell (1980), the difference between a tree and a shrub is simply form and size. Browse may be defined as "shoots or sprouts, specially tender twigs and stems of woody plants with their leaves, which are cropped to a varying extent by domestic and wild animals" (Torres, 1983). Shrubs are especially important in areas that experience dry periods and, according to McKell (1980), the cycle of livestock use would be in trouble without browse. Perennial, woody plants have a deep root system and are, therefore, more drought resistant than shallow-rooted herbaceous plants. This quality is of interest for arid and semi-arid zones where protein content is often the most limiting factor affecting liveweight gain in livestock (Torres, 1983). In the U.S. Great Basin desert rangeland, shrubs provide 50-70% of sheep diet and 40% of cattle diet during the wintertime (McKell, 1980).

Trees may be managed in forage plantations for the purpose of harvesting them as a crop to feed to animals (Barrett, 1992).

Small livestock have an important role to play in sustainable agriculture. Species such as rabbits, sheep and goats do not require as much space as cattle.

Rabbits are an example of an efficient way to convert forage to high quality meat and they have great potential in meeting the food needs of the world. According to Cheeke (1980), rabbits fed a high alfalfa diet have a feed conversion of 3-3.5 pounds of feed per pound of gain, while beef cattle fed high roughage diets require 12-15 pounds of feed per pound of gain. From an acre of alfalfa, five times as much meat can be produced by rabbits than by beef cattle (Cheeke and Patton, 1979).

Rabbits can utilize high-forage, low-grain diets that are noncompetitive with human food. They have a high biological potential due to their remarkable capacity for reproduction and their fast growth rate (Cheeke, 1980). A doe which is rebred postpartum under intensive production is capable of being pregnant with one litter, nursing another litter, and having a third litter weaned to a separate cage and growing to market size. A fryer can reach market weight within 9-10 weeks on high-quality diets. The small size of rabbits makes them easy to manage and they act as "biological refrigerators". There is no need to store the small carcass since it can be consumed by a family at one meal (Cheeke et al. 1987).

Rabbits can be raised on a backyard scale in developing countries and fed locally available materials and table scraps; or they may be raised intensively on a commercial scale and fed high quality diets (Cheeke et al., 1987). According to Cheeke (1980), economic return is more important than maximum food production in the United States (U.S.). The rabbit industry is very small in the U.S. In European countries such as France, Italy and Spain, where the consumption of rabbit meat has a long history and the industry is highly developed, rabbit carcass production per capita is between 3.6 and 4.0, compared to only 0.07 in the U.S. (Lukefahr and Cheeke, 1991).

Some Trees with Potential to be Used in Animal Feeding

Some examples of tropical trees which have received attention in animal agroforestry are *Leucaena leucocephala* (National Academy of Sciences, 1984), *Gliricidia sepium* (Smith and van Houtert, 1987), and calliandra (National Research Council, 1983).

In temperate areas, tagasaste (Snook, 1986), honey locust (Le Roux, 1959), black locust and poplar have been of interest. Black locust and poplar are two species were evaluated as a feedstuff for small livestock in this thesis.

Black locust (*Robinia pseudoacacia*) is indigenous to the eastern U.S. Also known as false acacia or yellow acacia, it is in the family Leguminosae (Papilionaceae) (National Academy of Sciences, 1983) and is capable of fixing atmospheric nitrogen. According to Hanover (1992), the species was introduced to Europe sometime after 1601 by Jean Robin, the herbalist of Henry IV of France, or by his son, Vespasien Robin, from whom the genus, *Robinia*, was named. The name, locust, was mistakenly given by early settlers in Jamestown, where houses were built upon this durable wood, who thought the tree resembled the Mediterranean carob due to the production of pods. The species became very popular and widespread in Europe and its native U.S. during the late 1700's. It was a custom for New York farmers to plant plots of these trees when a child was born to provide for its future. However, interest in the tree declined until the mid-1900's when it was revived by the use of black locust in erosion control, stabilizing disturbed areas such as strip mines and for timber production (Hanover, 1992).

It is an aggressive, pioneering species and due to its adaptability to a wide range of conditions and N fixation, it has been planted in disturbed areas world-wide, second only to eucalyptus species, according to Geyer and Bresnan (1992). It is cold hardy and receives about 1000 mm of rainfall in its native area, but can tolerate up to 6 months dry (National Academy of Sciences, 1983). It sprouts readily after coppicing. Black locust can reach 70 feet in height (Dirr, 1990). It has a wide root system which suckers and makes it valuable for stabilizing sand dunes (National Academy of Sciences, 1983). Drawbacks include thorns and crookedness in growing (Hanover, 1992).

Black locust is the major hardwood forestry plantation species cultivated in Hungary, covering 18% of the forested area, where it has undergone extensive selection and improvement work (Hanover, 1992). Rotation may be 25-30 years (National Academy of Sciences, 1983). Since 1980, it is gaining popularity in the U.S. as a hardwood plantation species. It has not enjoyed popularity as a timber species in the U.S. due to the locust borer, the prevention of which is a research priority. This pest is not a problem elsewhere because it does not exist outside the U.S. (Hanover, 1992). The wood is durable, decay-resistant and has been compared to black walnut and teak in terms of fine quality and strength, but products made of black locust are not well-known except for wood fences. Although it has a very rapid growth rate, 2-6 cm per day, the wood is of high density. Black locust is also used for pulp, round timbers and railroad ties (National Academy of Sciences, 1983). It is valuable as a bee forage (George Ayers, 1992).

Black locust can be cultivated in short-rotation, intensive-culture systems for high biomass yields (Geyer and Bresnan, 1992; Baldelli, 1992). It is considered to be the best firewood in North America (National Academy of Sciences, 1983). The wood can be converted to liquid fuels such as ethanol through fermentation or gas by anaerobic digestion in which bacterial degradation converts organic matter to methane and carbon dioxide (Geyer and Bresnan, 1992). The energy crisis has focused on tree biomass as a renewable source of energy as opposed to the nonrenewable fossil fuels petroleum and coal. Using biomass reduces pollution and the increase of CO_2 which contributes to the greenhouse effect (Baldelli, 1992).

Due to its high-protein forage and fast growth, forage plantations have been researched (Barrett, 1992). The leaves have been utilized commercially in animal feeding. In China, the trees are grown to provide leaves as a principle source of rabbit feed and as a source of pigmenting agents for poultry feed which is exported to Japan (Cheeke, 1992).

The forage, which contains about 20-25% crude protein, resembles alfalfa in composition; however, it generally gives low animal performance (Cheeke, 1992). Horton and Christensen (1981) found that when lambs were fed black locust leaf meal (BLM) as a sole feed, the digestibilities of organic matter (OM), crude protein (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF) and phosphorus (P) were less than half those of alfalfa. Although no significant differences were seen between lambs fed BLM and alfalfa in terms of ADG or DM intake, it was concluded that BLM was not a satisfactory replacement for alfalfa. Harris et al. (1984) found ADG for weanling rabbits fed a 40% black locust leaf meal (BLM) diet to be lower than the alfalfa control even though DM intake was higher for the BLM diet. Raharjo et al., (1990) also found ADG to be lower for rabbits fed a 50% BLM diet when compared to an alfalfa control. Singh and Negi (1986) fed supplements of BLM and biul (*Grewia optiva*), along with a concentrate diet, to angora rabbits and found low protein digestibility and low wool production with BLM when compared to biul. Cheeke et al. (1983) fed BLM to chicks at 20% of the diet and found that growth decreased by 30%. This was partially overcome by autoclaving the BLM. The poor animal performance and low digestibilities seen when BLM is fed are probably due to low nutrient availability which will be discussed in more detail.

Some animal feeding trials with BLM have given positive results. Baertsche et al. (1986), using the nylon-bag technique to determine the digestibility of 10 short-rotation, intensive-culture trees, found that black locust had a high feeding value for cattle, particularly because of its high CP content. Liu and Jung (1979) fed BLM or alfalfa at 3% of broiler diets and found that, although the average body weight was slightly higher for the alfalfa treatment, the difference was not significant. It was concluded that the BLM was of a value almost equal to the alfalfa. Takada et al. (1980) fed a 5% BLM diet to broilers and found no difference between BL and alfalfa treatments in terms of body weight gain, feed intake and mortality. Black locust leaf meal was 84% efficient as yellow maize in pigmenting egg yolks.

Black locust leaf meal has been fed to laboratory animals in trials investigating its negative effects. Horigome et al. (1988) found that inclusion of BLM leaves at 1% of the diet reduced the activities of trypsin and α-amylase in the rat intestine.

Hybrid poplar (*Populus* spp.) was the second species of tree forage evaluated in this thesis. The family *Salicaceae,* which includes poplars and willows, has a wide range of temperature tolerance. Poplar is native to the forests of the northern hemisphere from the Arctic circle to latitude 30°. In nature hybrids are common because cross-fertilization often occurs. Poplars are pioneering species and can grow in poor areas; they are highly demanding of water. Since ancient times in the Mediterranean area, country people have raised poplar trees from cuttings close to their homes for fuelwood, timber and forage for animals. Poplars mark areas of human habitation all over the world (FAO, 1980), including abandoned homesteads of the western U.S.

Poplar species, known in the U.S. as cottonwood, are dioecious woody plants. Aspen, *Populus tremuloides*, is the most widespread tree species in North America (Bas et al., 1985). Poplars have a rapid growth rate and may be used in 20-25 years or earlier depending on the use. The wood is valuable for sawn lumber, paper and pulp industry, fibreboards, box making, firewood, and wooden shoes in the Netherlands (FAO, 1980). It is very important as ornamental, landscaping plant. Poplar can be grown in association with farm crops and is useful on lands not suited for agriculture. It has been used as a windbreak in Argentina and the East (FAO, 1980).

According to FAO (1980), poplars are major timber producers for countries without forests and considered "blessed trees" in some countries of the Near East because they may be the only tree that will grow there. Poplars have been cultivated since very early times. In Europe, poplars have been grown since 18th century when North American poplars were hybridized with European ones. Poplar can be grown in short-rotation, intensive-culture systems. Propagation is usually done by vegetative means, cuttings, instead of seeds.

According to FAO (1980), the foliage of poplars is comparable to alfalfa. The literature is scarce for feeding trials involving poplar forage. Forwood and Owensby (1985) found poplar to be of intermediate value for livestock feed. Cattle selected poplar leaves fallen to the ground in September over the understory vegetation. Baertsche et al. (1986), using the nylon bag technique to determine the digestibility of 10 SRIC trees, found poplar to have potential for cattle feed. Some studies have been done with aspen, *Populus tremuloides*. Bas et al. (1985) fed lambs diets containing 0, 25, 50 and 75% aspen leaves with alfalfa as the other dietary ingredient. They found that intake, nutrient digestibilities and weight gain decreased as the level of aspen in the diet increased. Aspen is considered an excellent forage for elk (Canon et al., 1987). Fisher (1980) fed steam-treated aspen chips to lactating cows at 10%, as a partial substitute for corn silage, and found that dry matter (DM) and crude protein digestibilities were lowered. Milk protein content and fat test decreased with the addition of 20% aspen chips. Efficiency of milk yield was not affected.

The Anti-Nutritive Qualities of Tree Forage

The anti-nutritive effects evident in feeding trials with black locust and poplar has generally been attributed to the presence of tannins.

Tannins are considered to be the most widely distributed of secondary plant compounds. They are largely responsible for a plant's chemical defenses against bacteria, fungi and herbivores (Singleton, 1981). Tannins are currently of great interest because of their presence in many potential feed stuffs such as agricultural by-products and forage crops (Kumar and Singh, 1984). Their presence has both beneficial and adverse effects in livestock feed and in human consumption (Mehansho et al., 1987). Their relationships to ecology in terms of plant-herbivore co-evolution are areas under investigation (Freeland et al., 1985).

Mehansho et al. (1987) points out that tannins are found in many foods consumed by humans, thus reflecting our preference for tannins in nutrient-poor foodstuffs. They list some of the common foods containing tannins: red wine, tea, cider, coffee, cocoa, beer, bananas, persimmon, apples, raspberries, betel quid, fava beans, pinto beans, sorghum and barley.

Tannins are highly reactive, water-soluble polyphenolic compounds, well-known for their ability to precipitate proteins (Kumar and Horigome, 1986) (Figure 1). The reactions of phenols and proteins have been extensively researched. The first scientific paper written on this subject was in 1803 by Sir Humphry Davy (Beart et al., 1985). These compounds are also historically renown for their ability to "tan" leather by hydrogen-bonding with the collagen protein (Cheeke and Shull, 1985) and for other uses such as burn treatment and barium enemas (Mehansho et al., 1987). A practical definition of tannins is offered by Mehansho et al. (1987): "tannins are water-soluble phenolic metabolites of plants with a molecular weight of 500 or greater and with the ability to precipitate gelatin and other proteins from aqueous solutions".

Tannins are phenolic compounds, but not all phenolic compounds are tannins (Salunke et al., 1990). Plant phenolics are classified as phenolic acids, coumarins, flavonoid compounds, and tannins which are broken down into hydrolzable and condensed. Condensed tannins are also referred to as procyanidins (Kumar and Vaithiyanathan, 1990). The changes in taste of ripening fruit is associated with changes in the chemical forms of polyphenols in the fruit (Salunke et al., 1990).

Tannins are capable of several types of bonding in the formation of tannin-protein complexes. As noted by Salunke et al. (1990), these bonds are: hydrogen bonding, hydrophobic interaction, electrostatic attraction, and covalent bonding associated with oxidation. Hydrogen bonds, forming between the phenolic hydroxy groups of the tannins and the carbonyl groups of the peptide linkages of the proteins, and hydrophobic interactions are the principle linkages in tannin-protein complexes (Salunke et al., 1990). Kumar and Horigome (1986) note that the amount of tannin bound up in tannin-protein complexes depends on, along with pH, ionic strength, nature of the protein, and the molecular size of the tannin. The incorporation of tannin increases with molecular size (Kumar and Singh, 1984). For a condensed tannin, a molecular weight larger than 5000 will reduce its ability to form leather and will cause no astringent taste (Kumar and Singh, 1984).

Methods of detecting tannins include the vanillin assay, oxidation-reduction methods which include the Folin-Denis method and the Prussian Blue assay, protein-precipitation methods, and UV spectrophotometry (Salunke et al., 1990).

The dietary effects include unpalatability (astringency) which results in reduced feed intake. Tannins complex with salivary proteins or tie up enzymes, thus making the plant unpalatable to an animal as a major role in deterring herbivores (Kumar and Singh, 1984). According to these authors, the refusal level of plant tissue has been reported to be 20 mg of tannin per g of dry matter for grazing animals.

Tannins precipitate dietary proteins, thus diminishing their digestibilities in the animal and usually with an accompanying decrease in the feed utilization efficiency (Mehansho et al., 1987). The authors reviewed sources that report growth depression in rats, hamsters, mice, swine and chicks, and indicates that tannins increase fecal material and the amount of fecal nitrogen of endogenous sources. A decrease in protein utilization also occurs in humans (Mehansho et al., 1987). According to Kumar and Singh (1984), protein supplementation may counteract the depression in growth, but twice the amount of protein compared to tannin may be required to completely incorporate tannin into insoluble tannin-protein complexes.

Reduced digestibility may be explained by the binding of digestive enzymes, such as trypsin and amylase, by tannins (Kumar and Singh, 1984). Condensed tannins in particular inhibit cellulases, pectinases and lipases in vitro (Salunke et al., 1990). According to Kumar and Singh (1984), these tannin-enzymes reactions may be of two types: a) reducing the solubility of the enzyme protein by forming insoluble protein-phenolic complexes or b) inhibiting the enzyme activity by forming a soluble but inactive enzyme-inhibitor complex.

Controversy exists regarding to what degree the inhibition of digestive enzymes by tannins occurs in vivo and regarding the toxicity of tannins. According to Mehansho et al. (1987), the mechanism of action of the antinutritional effects of tannins is due to the increase in fecal nitrogen due to endogenous sources (most likely from tannin-binding salivary proline-rich proteins). The potential for toxicity is reduced because tannins are probably not absorbed in the digestive tract due to of their large size and due to membrane barriers (Salunke et a., 1990); however, human fatalities have been attributed to tannic acid used in burn and enema treatments (Singleton, 1981). Freeland et al. (1985) fed mice 5% tannic acid and noted a 700% increase in the size of the parotid gland during the first 4 days. This later decreased to 100% after 9 days. After 7 days, the mice given tannic acid weighed less than control mice. The weight loss was associated with a reduced feed intake during the first 2 days which later increased to normal. The authors proposed that the toxicity of ingested tannin is due to erosion of the intestinal mucosa instead of the inhibition of protein digestion. They stated that, although the belief is commonly held that tannins inhibit digestive enzymes, this does not occur in vivo in monogastric animals. They believe that the increased fecal nitrogen is due to endogenous sources from eroded intestinal cells and mucus. They attributed the negative effects of tannins to be from sodium depletion. Other effects on animals due to a tannin-rich diet is the chelating of minerals such as Na, Fe, Ca, Cu, and Mg which results in depletion of Na stores and a general disturbance in balance of the other minerals, and irritation to the intestinal mucosa (Freeland et al., 1985). Freeland et al. (1985) found that mice on a 5% tannic acid diet for seven days showed a lower body sodium concentration than the control, and that after 9 days on this diet, they exhibited severe erosion of the intestinal mucosa.

Further evidence that tannins adversely affect mineral balance is the case of tannin interference with iron absorption in humans. A study by Merhav et al. (1985) describes the effect on tea-drinking on infants in Israelian culture. According to the authors it is a common practice in Jerusalem to give infants tea. The occurrence of iron deficiency is between 26-68% in infants. Of the infants examined in the study, the "percentage of tea drinking infants with microcytic anemia (32.6%) was significantly higher that of the non-tea drinkers (3.5%). This was attributed to the chelating effect of tannin on iron.

Protein precipitation may be viewed as a beneficial effect in the ruminant, preventing bloat, due to the binding of the soluble proteins which form the matrix of stable foam (Kumar and Singh, 1984). A research priority has been the introduction of tannins into common forages such as alfalfa to prevent bloat.

The capacity of tannins to complex with proteins is also of interest in by-pass protein. It may be considered a beneficial effect that tannin complexes with dietary proteins in the rumen where the pH is around 4-6 and later releases these proteins at the lower pH of the abomasum (about 2) upon breakdown of the tannin-protein complexes (Freeland et al., 1985). The protein could then be absorbed in the small intestine, thus delivering the nutrient benefit of the protein to the host animal instead of to the rumen microbes. However, the same problems that occur in monogastrics could still occur in ruminants in the postrumen tract, such as the liberated tannins eroding the intestine wall or forming complexes with endogenous protein (Salunke et al., 1990).

Some animals have been able to adapt to a tannin-rich diet. Atsatt and Ingram (1983) examined the woodrat *Neotoma fuscipes* which prefers a diet with high phenol content. Woodrats chose a diet of *Quercus agriflolia* leaves which contain 40% phenolics and 16% condensed tannin. Following digestion, only one-third of the phenolics and 10% of the condensed tannins remained in the feces. The authors concluded that since the woodrat limits its diet to very specific feeds, its microbes may be more efficient, which allows increased feed-intake necessary here for the maintenance of nitrogen balance. The animals also practice coprophagy, which contributes to their ability to survive on a marginal food (Atsatt and Ingram, 1983). The silk worm requires the phenols present in the mulberry it feeds upon for proper development (Singleton, 1981). It has even been suggested that phytotoxins in angiosperms contributed to the demise of the dinosaurs; angiosperms were replacing gymnosperms at about the same time mammals were replacing reptiles, but the mammals were able to adapt to angiosperms while the reptiles did not (Singleton, 1981).

Kumar and Singh (1984) point out that since tannins are coded for only by one or two genes that it would not be difficult to eliminate the tannin factor; however, researchers must realize that this will greatly decrease the chemical defenses of a plant, needed to combat predation by insects and herbivores, and bacterial and fungal infection. Eighty percent of tannins can be removed from sal seed meal by soaking, washing and boiling the feedstuff; however, water-soluble nutrients may be washed out since 18-23% of the dry matter was lost. Kumar and Singh (1984) mention that work has been done with alkali treatment, removing 74 and 100% of tannins from sal seed meal by treatment with NaOH and Ca(OH)2, respectively. Ammonia treatment is another tannin removal technique which is promising for ruminants because it could mean an increase in non-protein nitrogen. Treatment with formalin converts the tannins to nonreactive resins Kumar and Singh, 1984). Heat treatment is not considered effective because tannins are heat stable (Garrido et al., 1991).

Kumar and Singh mention two adsorbents, polyvinylpyrrolidone (PVP) and polyethylene glycol (PEG), to which the tannins bind more readily than to proteins. Garrido et al. (1991) found that adding 160 mg of PVP per mg of tannin to whole faba beans reduced the tannin index by 97%. Horigome et al. (1984) found that adding 1.6% PEG to rat diets containing BL leaves increased CP digestibility from 49.1% to 70.7%. Pritchard et al. (1988) fed sheep *Acacia aneura* (which contained 6.1% tannins) and found that drenching the sheep every day with PEG increased body weight and wool growth. These findings contrast to those of Nuñez-Hernandez et al. (1991) who determined that adding PEG to diets containing mountain mahogany leaves, which have condensed tannins, did not improve nutrient digestion or affect N balance in goats and lambs. Garrido et al. (1991) found that the addition of PEG to faba bean seed coat, at a level of 200 mg of PEG per g of seed coat, increased CP digestibility in vitro from 52.4 to 75.5%.

Inactivation of tannins may occur through their binding by certain proteins that are rich in proline and have a high affinity for tannins (Austin et al., 1989; Mehansho et al., 1987). Mehansho and his colleagues found that when rats were fed a high-tannin diet, the parotid glands increased fourfold along with an increase in proline-rich proteins (PRP). Rats and mice synthesize PRP (containing up to 44% protein) that are 40% carbohydrate. This indicates that glycoproteins do bind to tannins and the carbohydrate portion is important in the affinity these protein have for tannins (Salunke et al., 1990). According to Mehansho et al., PRPs make up 70% of the protein in human saliva and proposes that this is the "first line defense" against tannins. It has been suggested that humans consume diets which keep the parotid glands in a constantly induced state of producing PRP (Salunke et al., 1990). Mehansho et al. (1987) maintain that PRPs in human saliva, along with binding tannins, may contribute to "calcium binding, hydroxylapatite binding, formation of acquired dental pellicle, and agglutination of oral bacteria". Mehansho et al. (1987) outline the characteristics of tannin-binding proteins as being "large size, open loose structure, high proportion of hydrophobic amino acids, and high proline content", and proteins less likely to bind tannins as being "small, compact and cross-linked with disulfide bonds". Proline's structure makes it less able to fit into an alpha helix, thus contributing to open protein structure (Mehansho et al., 1987). According to Salunke et al. (1990), condensed tannins can differentiate between high or low affinity proteins. Austin et al. (1989) tested the saliva of deer, cattle and sheep for the presence of tannin-binding proteins. Some herbivores in a tannin-rich environment have developed systems for dealing with the tannins. Austin et al. (1989) proposed that deer, being browsers, must overcome the adverse effect of tannins since their natural diet includes a high level of tannins; whereas cattle and sheep, being grazers, do not. Using a unique methodology that is capable of detecting salivary PRP in a small sample of saliva, the authors determined that the saliva of mule deer is richer in proline, contains two to three times more nitrogen, and is more capable of binding tannin than is the saliva of cattle or sheep. By this binding mechanism, deer minimize nitrogen loss and toxicity that could result from a tannin-rich diet (Austin et al.,

1989).

Plant secondary compounds are closely associated with the environment. Their fluctuations and induction influence dietary selection by wildlife. Jakubas et al. (1989) found that ruffed grouse preferred certain aspen trees over others. The nonpreferred trees had higher levels of a simple phenol, coniferyl benzoate. Wounding by simulated herbivory may induce plant defenses. Bradshaw et al. (1989) found that when hybrid poplars were wounded in the lower leaves, the upper leaves code for 'defense' genes. One of the amino acid sequences of these is similar to trypsin inhibitors found in legume seeds. Clausen et al. (1989) found that simulated herbivory of an insect, the large aspen tortrix, caused an increase of two phenolic compounds within 24 hours, and was considered a short-term induced defense mechanism. In a study by Basey et al. (1988), beavers in a newly occupied area, normally select aspen trees of a smaller size for foraging, but in an area previously occupied by beavers, they selected against the smaller, juvenile trees. The authors proposed that this was due to a higher level of a phenolic compound induced in the juvenile trees in response to browsing. Meyer and Montgomery (1987) considered leaf age in a study of the feeding habits of the gypsy moth on poplar leaves. The moth preferred to feed on the old growth of poplar foliage rather than the new leaves. The concentrations of phenols was three times higher in the new leaves than the old leaves, and the moths showed 85% less growth when fed the new leaves. Tannins were not suspected as the antinutritive phenol in this case because the levels did not differ between old and new leaves (the authors suspected phenolic glycosides), but tannin level did change with the seasons.

Lectins are not phenolic compounds, but their presence in black locust has a dietary effect. They are hemagglutins and have been suspected of poisoning horses that strip the bark of black locust trees to which they are tied (Cheeke and Shull, 1985). The lectins in black locust fluctuate during seasons. In the late spring, lectins were equally present in bark and other vegetative tissues such as leaves, roots, stems and flowers, but in the winter, it was found that lectins increased by 50 fold in the bark compared to summer levels.

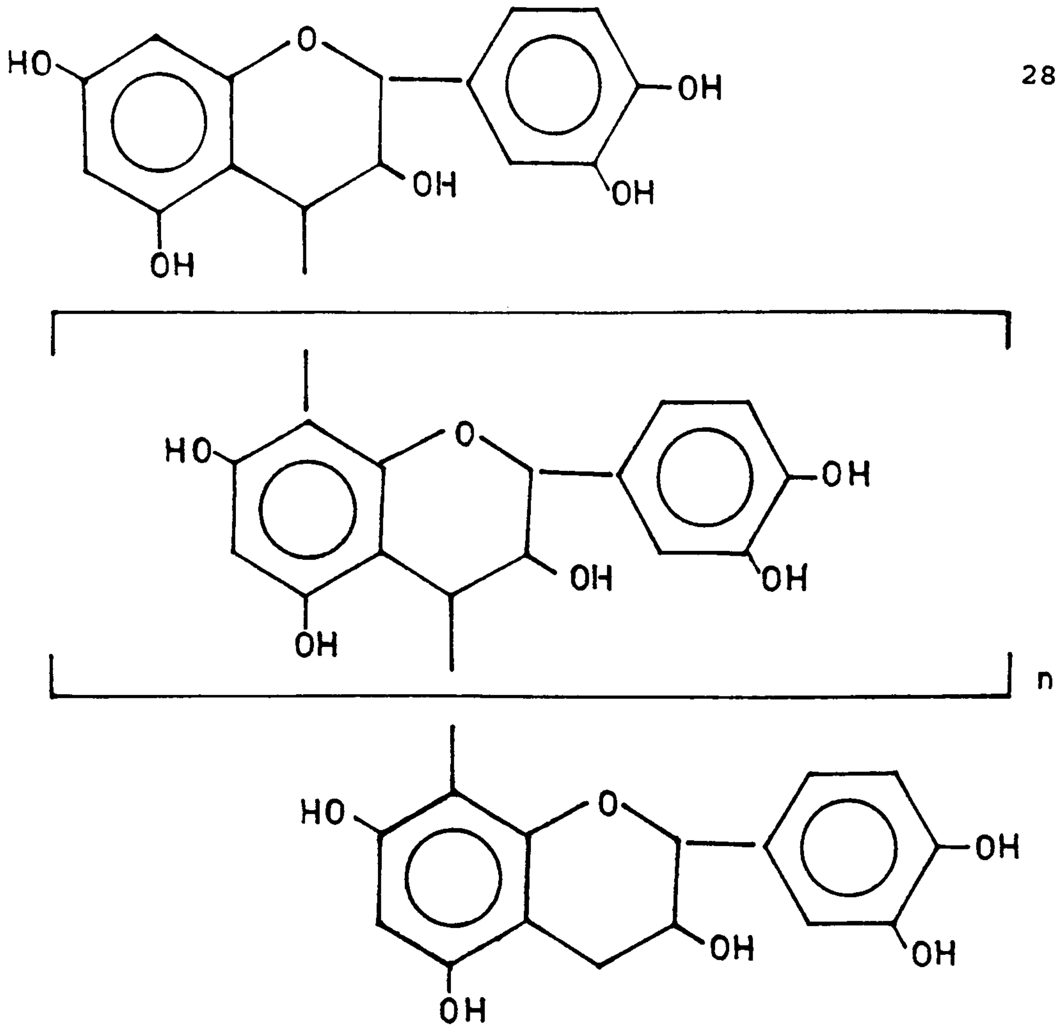

Figure 1. EXAMPLE OF TANNIN STRUCTURE

References

Anderson, G.W., R.W. Moore and P.J. Jenkins. 1988. The integration of pasture, livestock and widely-spaced pine in South Western Australia. Agroforestry Syst. 6:195-211.

Atsatt, P.R. and T. Ingram. 1983. Adaptation to oak and other fibrous, phenolic-rich foliage by a small mammal, *Neotoma fuscipes*. Oecologia 60:135-42.

Austin, P.J., L.A. Suchar, C.T. Robbins, A.E. Hagerman. 1989. Tannin-binding proteins in saliva of deer and their absence in saliva of sheep and cattle. J. Chem. Ecol. 15:1335-1347.

Ayers, G.S. 1992. Characteristics of black locust as bee forage. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.243-251.

Baertsche, S.R., M.T. Yokoyama and J.W. Hanover. 1986. Short rotation, hardwood tree biomass as potential ruminant feed - chemical composition, nylon bag ruminal degradation and ensilement of selected species. J. Anim. Sci. 63:2028-2043.

Beart, J.E., T.H. Lilley, E. Haslam. 1985. Plant polyphenols - secondary metabolism and chemical defence: some observations. Phytochemistry 24:33-38.

Baldelli, C. 1992. Black locust as a source of energy. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.237-243.

Barrett. R.P. 1992. Forage Plantation Management for black locust. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.259-272.

Bas, F.J., F.R. Ehle and R.D. Goodrich. 1985. Evaluation of pelleted aspen foliage as a ruminant feedstuff. J. Anim. Sci. 61:1030-1036.

Basey, J.M., S.H. Jenkins and P.E. Busher. 1988. Optimal central-place foraging by beavers: Tree-size selection in relation to defensive chemicals of quaking aspen. Oecologia 76:278-282.

Benge, M.D. 1987. Multipurpose uses of contour hedgerows in highland regions. World Anim. Rev. 64:31-39.

Bradshaw, H.D., J.B. Hollick, T.J. Parsons, H.R.G. Clark and M.P. Gordon. 1989. Systemically wound-responsive genes in poplar trees encode proteins similar to sweet potato sporamins and legume Kunitz trypsin inhibitors. Plant Molecular Biology 14:51-59.

Canon, S.K., P.J. Urness, N.V. DeByle. 1987. Habitat selection, foraging behavior, and dietary nutrition of elk in burned aspen forest. J. Range Manage. 40:433-438.

Caufield, C. 1982. Tropical Moist Forests: the resource, the people, the threat. Russel Press Ltd., Nottingham, UK. 67p.

Cheeke, P.R. 1980. Editorial: The potential role of the rabbit in meeting world food needs. J. Appl. Rabbit Res. 3:3-4.

Cheeke, P.R. 1992. Black locust forage as an animal feedstuff. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp 32-38.

Cheeke, P.R., M.P. Goeger, G.H. Arscott. 1983. Utilization of black locust (*Robinia pseudoacacia*) leaf meal by chicks. Nitrogen-Fixing Tree Res. Rep. 1:41.

Cheeke, P.R. and N.M. Patton. 1979. Utilization of alfalfa by rabbits. p. 25 in: The Domestic Rabbit: Potentials, Problems and Current Research. Published by OSU Rabbit Research Center.

Cheeke, P.R., N.M. Patton, S.D. Lukefahr, J.I. McNitt. 1987. Rabbit Production. The Interstate Printers and Publishers, Inc., Danville, IL 472p.

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Westport, CT. 492p.

Clausen, T.P., P.B. Reichardt, J.P. Bryant, R.A. Werner, K. Post and K. Frisby. 1989. Chemical model for short-term induction in quaking aspen (*Populus tremuloides*) foliage against herbivores. J. Chem. Ecol. 15:2335-2346.

Dirr, M.A. 1990. Manual of Woody Landscape Plants: Their Identification, Ornamental Characteristics, Culture, Propagation and Uses. Stipes Publishing Co., Champaign, IL. pp 753-756.

Food and Agriculture Organization (FAO). Poplars and Willows : in wood production and land use. Food and Agriculture Organization of the United Nations 1980. 328p.

Fisher, L.J. 1980. An evaluation of steam-treated aspen as a substitute for corn silage in the rations of lactating cows. Can. J. Anim. Sci. 60:379-384.

Forwood, J.R. and C.E. Owensby 1985. Nutritive value of tree leaves in the Kansas Flint Hills. J. Range. Manage. 38:61- 64.

Freeland, W.J., P.H. Calcott, D.P. Geiss. 1985. Allelochemicals, minerals, and herbivore population size. Biochemical Systematics and Ecology 13:195-206

Garrido, A., A. Gómez-Cabrera, J.E. Guerrero and J.M. van der Meer. 1991. Effects of treatment with polyvinylpyrrolidone and polyethylene glycol on Faba bean tannins. Anim. Feed Sci. Tech. 35:199-203.

Geyer, W.A. and D.F. Bresnan. 1992. Characteristics and uses of black locust for energy. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.221-236.

Hanover, J. 1992. Black locust: an historical and future perspective. Pro. Int. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp.7-28.

Harris, D.J., P.R. Cheeke and N.M. Patton. 1984. Evaluation of black locust leaves for growing rabbits. J. Appl. Rabbit Res. 7:7-9.

Horigome, T., T. Ohkuma and M. Muta. 1984. Effect of condensed tannin of false acacia leaves on protein digestibility, as measured with rats. Jap. J. Zootech. Sci. 55:299-306.

Horigome, T., R. Kumar and K. Okamoto. 1988. Effects of condensed tannins prepared from leaves of fodder plants on digestive enzymes in vitro and in the intestine of rats. Brit. J. Nutr. 60:275-285.

Horton, G.M.J. and D.A. Christensen. 1981. Nutritional value of black locust tree leaf meal (*Robinia pseudoacacia*) and alfalfa meal. Can. J. Anim. Sci. 61:503-506.

Jakubas, W.J., G.W. Gullion and T.P. Clausen. 1989. Ruffed grouse feeding behavior and its relationship to secondary metabolites of quaking aspen flower buds. J. Chem. Ecol. 15:1899-1915.

Kumar, R. and T. Horigome. 1986. Fractionation, characterization, and protein-precipitating capacity of the condensed tannins from *Robinina pseudoacacia L.* leaves. J. Agric. Food Chem. 34:487-489.

Kumar, R. and M. Singh. 1984. Tannins: their adverse role in ruminant nutrition. J. Agric. Food Chem. 32:447-53.

Kumar R. and S. Vaithiyanathan. 1979. Occurrence, nutritional significance and effect on animal productivity of tannins in tree leaves. Anim. Feed Sci. Tech. 30:21-38.

Le Roux, P.L. 1959. Sheep find thin honey-locust pods palatable. Farming in South Africa 18-19.

Liu, C.G. and Y. Jung. 1979. Nutritional value of locust (*Robinia pseudoacacia*) leaf meal. Unpublished report. Chinese Academy of Agricultural Sciences.

Lukefahr, S.D. and P.R. Cheeke. 1991. Rabbit project development strategies in subsistence farming systems. World Anim. Rev. 68:60-70.

McKell, C.M. 1980. Multiple use of fodder trees and shrubs - a world wide perspective. Proc. Int. Sym. on Browse in Africa: The Current State of Knowledge. Le Houérou, H.N. (Ed.). pp. 141-149.

Mehansho, H., L.G. Butler, D.M. Carlson,. 1987. Dietary tannins and salivary proline-rich proteins: interactions, induction, and defense mechanisms. Ann. Rev. Nutr. 7:423-40.

Merhav, M., Y. Amitai, H. Palti, S. Godfrey. 1985. Tea drinking and microcytic anemia in infants. Amer. J. Clin. Nutr. 41:1210-13.

Meyer, G.A. and M.E. Montgomery. 1987. Relationships between leaf age and the food quality of cottonwood foliage for the gypsy moth, *Lymantria dispar*. Oecologia 72: 527-532.

National Academy of Sciences. 1983. Firewood Crops: Shrub and Tree Species for Energy Production, Vol. 2. National Academy Press, Washington, DC. 92p.

National Academy of Sciences. 1984. Leucaena. Promising Forage and Tree Crop for the Tropics. Washington, DC.

National Geographic Society. 1990. The Emerald Realm: Earth's Precious Rain Forests. Published by National Geographic Society. 196p.

National Research Council. 1983. Calliandra: A Versatile Small Tree for the Humid Tropics. National Academy Press. Washington, DC. 48p.

Nuñez-Hernandez, G., J.D. Wallace, J.L. Holechek, M.L. Galyean and M. Cardenas. 1991. Condensed tannins and nutrient utilization by lambs and goats fed low-quality diets. J. Anim. Sci. 69:1167-1177.

Organización para Estudios Tropicales (OTS) y Centro Agronómico Tropical de Investigación y Enseñanza (CATIE). 1986. Sistemas Agroforestales: Principios y Aplicaciones en los Trópicos. Publicado por La Organización para Estudios Tropicales, Costa Rica. 818p.

Pritchard, D.A., D.C. Stocks, B.M. O'Sullivan, P.R. Martins, I.S. Hurwood and P.K. O'Rourke. 1988. The effect of polyethylene glycol (PEG) on wool growth and live weight of sheep consuming a Mulga (*Acacia aneura) diet. Proc. Aust. Soc. Anim. Prod. 17:290-293.*

Raharjo, Y.C., P.R. Cheeke and N.M. Patton. 1990. Effect of cecotrophy on the nutrient digestibility of alfalfa and black locust leaves. J. Appl. Rabbit Res. 13:56-61.

Salunkhe, D.K., J.K. Chavan and S.S. Kadam. 1990. Dietary Tannins: Consequences and Remedies. CRC Press, Boca Raton, FL. 200p.

Singh, B. and S.S. Negi. 1986. Effect on wool production of supplementing black locust (*Robinia pseudoacacia*) and biul (*Grewia optiva*) leaves *ad libitum* to a concentrate diet of angora rabbits. J. Appl. Rabbit Res. 9:159-163.

Singleton, V.L. 1981. Naturally occurring food toxicants: phenolic substances of plant origen common in foods. In Advances in Food Research. Chichester, C.O. (Ed.). Academic Press, New York, NY. pp.149-242.

Smith, O.B. and M.F.J. van Houtert. 1987. The feeding value of *Gliricidia sepium:* a review. World Anim. Rev. 62:57-67.

Snook, L.C. 1986. Tagasaste, Tree Lucerne: High Production Fodder Crop. Night Owl Pub.Shepparton, Australia. p. 93.

Takada, K., T. Nakazato, K. Ono, H. Honda and T. Yamane. 1980. Feeding value of leaf meal of acacia on poultry feed. Jap. Poul. Sci. 17:299-305.

Torres, F. 1983. Role of woody perennials in animal agroforestry. Agroforestry Sys. 1:131-163.

World Resources Institute. 1985. Keep Tropical Forests Alive. Published by World Resources Institute, Washington, DC. 8p.

EVALUATION OF HYBRID POPLAR LEAVES AS A FEEDSTUFF FOR RABBITS

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Abstract

Sixty New Zealand White weanling rabbits were fed diets containing 40% alfalfa (control), and 10%, 20% and 40% hybrid poplar leaves (PL) from the uncoppiced crown of the tree, and 10% and 20% PL from the regrowth after cutting of coppiced trees. Digestibilities of crude protein (CP), acid detergent fiber (ADF), neutral detergent fiber (NDF), dry matter (DM) and ash were measured, along with average daily gain (ADG), DM intake and feed efficiency. Crude protein, NDF and ash were better digested for the control compared to PL treatments (respective p values < .01, .02 and .01). The 40% alfalfa control showed better utilization of CP, NDF and ash, than when the alfalfa was completely replaced in the diet by PL (respective p values < .01, .01 and .01). The only significant difference seen between uncoppiced and coppiced leaves of the 10% and 20% PL levels was at the 20% level where CP was better utilized for the coppiced PL (p < .05). No differences were evident in the ADG and feed efficiency; however, a trend was evident (p < .06)

for the feed/gain ratio to be less efficient for the 40% PL diet than for the alfalfa control. DM intake increased significantly for all the PL treatments when compared to the control ($p < .01$) and when a complete substitution is made ($p < .01$). The literature suggests some anti-nutritive factors due to plant defense have been suspected of reducing digestibilities, such as tannins and other phenols; however, since ADG did not decrease, it was concluded that PL are a suitable replacement for alfalfa in rabbit rations if a slightly higher feed efficiency is acceptable.

Introduction

The use of tree foliage for animal feed has potential in the U.S. as a by-product from the forestry industry. In the U.S. Pacific Northwest, plantations of hybrid poplars for pulp and paper have been established. The foliage could be collected for use as a feedstuff when the trees are harvested. In developing countries which often suffer from severe deforestation, tree forage may have a broader use in agroforestry systems designed to use multiple purpose trees for improving soil conditions, crop interaction and as animal feed (Benge, 1987).

The family *Salicaceae*, poplars and willows, has a wide range of temperature tolerance and grows in cold parts of the northern hemisphere from the Arctic circle to latitude 30° (FAO, 1980). Aspen, *Populus tremuloides*, is the most widespread tree species in North America (Bas et al.,1985). Poplar species, known in the U.S. as cottonwood, are dioecious woody plants.

Tree leaves have been examined for their nutritive value, including the senescent leaves that fall to the ground every year which livestock have been seen to consume (Forwood and Owensby, 1985). Forwood and Owensby (1985) found poplar to be of intermediate feeding value to livestock. Tree forage has been tested for domestic ruminants and shown to have potential (Baertsche et al., 1986), but the literature for nonruminants is scarce.

Many trees may have secondary chemical defense mechanisms to protect against herbivory (Jakubas et al., 1989). It has been noted that livestock, while refraining from eating resprouted growth from coppiced hybrid poplar trees, will readily consume the leaves from the crown of felled trees (personal observation). We hypothesize that the resprouted growth from coppiced trees contains more chemical defenses than the leaves in the crown, since the coppiced plant has been "under attack". "Coppiced" trees are trees which have been cut and from which growth has resprouted. Basey et al. (1988) proposed that heavy cutting by beavers in an area causes increased chemical defenses in the bark of aspen suckers which leads beavers to select forage from mature trees, instead of the usually preferred juvenile trees.

The objectives of this study were to compare hybrid poplar leaves to alfalfa in rabbit diets, using growth rate and nutrient digestibility as indices of performance, and to compare leaves from uncoppiced versus coppiced hybrid poplar trees.

Materials and Methods

Sixty New Zealand White 5-week-old weanling rabbits were randomly assigned to six treatments with ten rabbits in each treatment. The treatments were:

1) 40% alfalfa (control)
2) 10% uncoppiced poplar leaves; 30% alfalfa
3) 20% uncoppiced poplar leaves; 20% alfalfa
4) 40% uncoppiced poplar leaves
5) 10% coppiced poplar leaves; 30% alfalfa
6) 20% coppiced poplar leaves; 20% alfalfa.

The trial consisted of a 14-day adaptation period followed by a 7-day digestibility period with two replicates of five animals per treatment. The second replicate was put on the experiment 40 days after the first replicate. Rabbits were housed in individual cages with automatic dewdrop water valves. Feed intake was measured throughout the trial and during the digestibility period. Initial, weekly and final weights were taken, along with initial and final weights for the digestibility period. Total fecal output for the digestibility period was collected with screens suspended underneath each cage.

The animals were fed the diets *ad libitum* (Table 1). Poplar leaves were hand-harvested from a local farm in September 1990. The leaves considered as "uncoppiced" growth were taken from the crowns of recently felled 10-year-old trees. Leaves considered as "coppiced" growth were collected from growth 5-7 feet high, sprouted from coppiced trunks cut earlier the same year. The leaves were air-dried on screens for 3 weeks and then passed through a shredder until coarsely ground.

After the end of the digestibility period, the total wet feces were weighed and oven-dried at 60° C for 48 hours. Dry total feces were weighed and a subsample was ground in a Wiley mill to pass through a 1 mm screen. Feed and feces samples were analyzed for DM and ash by standard procedures (AOAC, 1984). Samples were analyzed for ADF and NDF as described by Goering and Van Soest (1970), modified by a micro method (Waldern, 1971). Crude protein was determined by the Macro Kjeldahl method (AOAC,

1984).

The data were analyzed following the general linear model procedure (SAS, 1985). Initial F-screening was done at the .05 level. Treatment means were compared using a series of four pre-planned comparisons. Of the 60 animals on the experiment, the records of 57 were included in the analyses. One rabbit from the second replicate died and was not replaced. Two rabbits were removed from all analyses because one spilled its fecal collecting tray and the other was considered an outlier.

Results and Discussion

Composition of the alfalfa and poplar leaves is shown in Table 2. The leaves from uncoppiced trees were lower in crude protein and higher in fiber than alfalfa. The leaves from coppiced trees were similar in composition to alfalfa. Digestibility data are given in Table 3 and growth performance shown in Table 4.

The digestibilities of dry matter, crude protein, ADF, NDF and ash were higher for the 40% alfalfa diet (control) than for the treatments with poplar leaves (Table 3). The values for crude protein, NDF and ash were statistically higher (respective p values $< .01$, .02 and .01) for the control than for the poplar diets (Table 5).

At a level of 20% poplar leaf meal in the diet, the crude protein digestibility was significantly higher ($p < .05$) for coppiced than for uncoppiced leaves (Table 5). This does not suggest a higher content of protein-binding phenolic compounds in the "attacked", resprouted trees.

For the average daily gain (ADG) and feed efficiency (feed/gain), there were no statistical differences between treatments; however, there was a trend ($p < .06$) for the feed efficiency to be higher for the 40% PL diet than for the alfalfa control. There were statistical differences in dry matter intake: less feed was consumed on the control ($p < .01$) than on all the other treatments (Comparison 1) and less feed was consumed on the control ($p < .01$) compared to the 40% PL diet (Comparison 4).

Poplar leaves contain tannins (Meyer and Montgomery, 1987). According to these authors, the leaves of *Populus deltoides* contained 3.6-5.0% condensed tannins. Tannins can bind dietary proteins and enzymatic proteins to reduce CP digestibility (Cheeke and Shull, 1985). Bas et al. (1985) fed pelleted aspen foliage to lambs at levels of 0, 25, 50 and 75%, with alfalfa as the other ingredient, and found that lamb weight and intake decreased as aspen level increased. Van Soest (1982) proposed that an insoluble tannin-protein complex is formed which is recovered in the ADF fraction. In the present study, the poplar leaves had about two times the amount of N bound to ADF fraction as the alfalfa (Table 2), which may have contributed to poorer crude protein digestibility. Bradshaw et al. (1989) found that when hybrid poplars were wounded in the lower leaves, the upper leaves coded for 'defense' genes. The amino acid sequences for two of these were similar to chitinases found in tobacco and barley. A third was similar to trypsin inhibitors found in legume seeds.

The ADF utilization showed no significant difference between treatments whereas the NDF did. Thus the hemicellulose fraction was better utilized for alfalfa than for the PL treatments.

The minerals in the tree forage were not as well utilized as in alfalfa, as evidenced by the lower ash digestibility. Baertsche et al. (1986) found poplar to be lower than alfalfa in Ca and P, but higher in microminerals.

Jakubas et al. (1989) found that ruffed grouse preferred certain aspen trees over others. The nonpreferred trees had higher levels of a simple phenol, coniferyl benzoate. Grouse preferred catkins over buds. Buds were higher in coniferyl benzoate. Since grouse feeding preference was not related to tannins or total phenols, they proposed that the grouse may have adapted to tannins in the diet.

We offer the following explanation for the higher CP digestibility with 20% coppiced leaves than for the uncoppiced leaves in the diet. Meyer and Montgomery (1987) point out that *Populus deltoides* exhibits free shoot growth as opposed to fixed, meaning that the leaves are continuously produced throughout the season instead of in a flush within a few weeks after budbreak. According to Kramer and Kozlowski (1979), the extent that later-season leaves are produced diminishes with tree age. Owing to this, the leaves from uncoppiced growth were generally older than those from the coppiced trees because they came from older mature trees less likely to be producing young leaves in September when we harvested. Young leaves are higher in water and N and contain less fiber (McKey, 1979) which agrees with the leaf composition analyzed in the present study (Table 2). The literature suggests that young leaves would contain more chemical defenses than older leaves. Meyer and Montgomery (1987) found that the gypsy moth prefers to feed on the old growth of poplar (*Populus deltoides*) foliage rather than the new leaves. The moths showed 85% less growth when fed new leaves. They found that the concentration of phenols was three times higher in the new leaves than in the old leaves. The active phenol was not identified. Levels of flavenoids and condensed tannins did not differ between leaf ages. Tannic acid is not present in this type of poplar. The authors suspected phenolic glycosides. In our study, although the younger hybrid poplar leaves from the less mature coppiced trees may have contained more chemical defenses than the older leaves from the uncoppiced mature trees, it did not seem to be enough to cancel out the higher nutritive value. It would be of interest to carry out a similar experiment in which the leaves from uncoppiced and coppiced sources could be harvested early in the season before the effect of old vs young leaves could set in.

Growth rate was not adversely affected by any of the poplar leaf treatments. The slightly lower digestibility of diets containing poplar leaf meal was compensated for by increased feed intake. All the diets seemed to be palatable. The poplar leaf diets had a sweet odor, characteristic of poplar. This odor might be due to low molecular weight, phenolic compounds that are found in poplar.

Poplar can be intensively grown and coppiced. Baertsche et al. (1986) examined 10 short rotation, hardwood trees and found several had potential as livestock feed. *Populus deltoides* contained 22% CP in the initial growth. When cut and regrown in the same season, it had 19% CP.

Hybrid poplar has been planted in some temperate developing countries such as Nepal. The trees could be planted as cuttings in contour hedgerows on hillsides to slow soil erosion from water runoff, coppiced to provide fuelwood and animal forage, in alley-cropping systems, and as a source of material for reforestation (Benge, 1987). The author cites Dula (1982) in reporting that poplar at close spacing can produce 65 tons of dry biomass per hectare per year.

Digestibility of diets containing PL is lower than those containing alfalfa due to some anti-nutritive qualities which the literature has suggested may include tannins or phenolic glycosides; however, since feed efficiency did not increase significantly, it may be concluded that poplar leaves are a satisfactory replacement of alfalfa in the diet when the leaves are economically available and if a slightly less efficient feed/gain ratio is accpetable. Poplar trees could be an integral part of an agroforestry system as animal feed.

EXPERIMENT 1

TABLE 1. COMPOSITION (%) OF EXPERIMENTAL DIETS FED TO RABBITS (DRY MATTER BASIS)

Diet	1	2	3	4	5	6
Alfalfa meal, sun-cured	40	30	20	-	30	20
Corn oil	1	1	1	1	1	1
Dicalcium phosphate	0.25	0.5	0.5	0.5	0.5	0.5
Molasses	3	3	3	3	3	3
Limestone	-	0.25	0.5	0.5	0.25	0.5
Poplar uncoppiced	-	10	20	40	-	-
Poplar coppiced	-	-	-	-	10	20
Soybean meal	10	10	10	10	10	10
Trace mineral salt*	0.5	0.5	0.5	0.5	0.5	0.5
Wheat mill run	49	48.5	48.3	48.3	48.5	48.3
Vitamin mix**	0.25	0.25	0.25	0.25	0.25	0.25
Chemical Analysis:						
Crude protein	18.9	18.0	18.1	18.7	19.1	19.2
ADF	20.0	19.7	19.8	18.4	18.9	18.4
NDF	38.8	37.3	34.7	33.3	37.1	36.2
Ash	7.4	7.6	7.9	7.7	7.9	8.3

*Ingredients: Sodium chloride, zinc oxide, manganous oxide, iron carbonate, copper oxide

**Vitamin A 3,000,000 IU
D-3 1,000,000 IU
E 1000 IU
K 0.5 gm
B-12 5 mg
Riboflavin 3 gm
Pantothenic acid 5 gm
Niacin 20 gm
Choline chloride 200 gm
Folic acid 200 mg
Ethoxyquin 56.75 gm

* and ** manufactured by Inman & Co., Inc.; 12530 SE Jennifer St., Clackamas, OR 97015.

EXPERIMENT 1

TABLE 2. NUTRIENT COMPOSITION (%) OF POPLAR LEAVES AND ALFALFA.

	Crude Protein	ADF	NDF	Ash	N-ADF*
Poplar leaves Uncoppiced	14.77	29.26	42.09	8.98	0.94
Poplar leaves Coppiced	21.63	21.03	31.53	9.49	0.79
Alfalfa	20.29	29.45	38.65	10.12	0.41

*Nitrogen contained in the ADF fraction

EXPERIMENT 1

TABLE 3. PERCENT NUTRIENT DIGESTIBILITIES (MEANS ±SE) IN WEANLING RABBITS OF DIETS CONTAINING VARIOUS PROPORTIONS OF ALFALFA AND POPLAR LEAVES (PL).

Treatment	Crude Protein	ADF	NDF	Dry Matter	Ash
1. 40% Alfalfa Control	78.75 ± 2.11	26.28 ± 4.01	36.87 ± 3.82	64.35 ± 2.76	60.62 ± 2.02
2. 10% PL Uncoppiced	68.71 ± 2.11	19.04 ± 4.01	29.53 ± 3.82	59.02 ± 2.76	51.93 ± 2.02
3. 20% PL Uncoppiced	60.05 ± 2.11	18.40 ± 4.01	20.88 ± 3.82	55.72 ± 2.76	48.95 ± 2.02
4. 40% PL Uncoppiced	54.76 ± 2.36	10.15 ± 4.48	18.79 ± 4.27	57.63 ± 3.08	44.73 ± 2.26
5. 10% PL Coppiced	69.92 ± 2.22	21.07 ± 4.22	31.21 ± 4.03	59.88 ± 2.91	52.40 ± 2.13
6. 20% PL Coppiced	66.11 ± 2.11	21.97 ± 4.01	31.01 ± 3.82	59.34 ± 2.76	50.81 ± 2.02
Average of 2 3 4 5 6	63.91 ± 2.18	18.13 ± 4.15	26.28 ± 3.95	58.32 ± 2.85	49.76 ± 2.09

EXPERIMENT 1

TABLE 4. MEANS (±SE) OF GROWTH PERFORMANCE INDICATORS AND FEED INTAKES OF WEANLING RABBITS FED DIETS CONTAINING VARIOUS PROPORTIONS OF ALFALFA AND POPLAR LEAVES (PL).

Treatment	Ave. Daily Gain (g)	Daily DM Intake (g)	Feed/ Gain
1. 40% Alfalfa Control	35.5 ±2.7	105.6 ±4.9	3.4 ±0.2
2. 10% PL Uncoppiced	39.7 ±2.7	117.3 ±4.9	3.3 ±0.2
3. 20% PL Uncoppiced	36.7 ±2.7	122.3 ±4.9	3.8 ±0.2
4. 40% PL Uncoppiced	37.9 ±3.1	133.9 ±5.5	4.0 ±0.2
5. 10% PL Coppiced	37.3 ±2.9	119.4 ±5.2	3.7 ±0.2
6. 20% PL Coppiced	37.1 ±2.7	114.3 ±4.9	3.5 ±0.2
Average of 2 3 4 5 6	37.7 ±2.8	121.4 ±5.1	3.7 ±0.2

EXPERIMENT 1

TABLE 5. STATISTICAL COMPARISONS FOR DIGESTIBILITY (%) AND GROWTH PERFORMANCE DATA OF WEANLING RABBITS FED DIETS CONTAINING VARIOUS PROPORTIONS OF ALFALFA AND POPLAR LEAVES (PL) FROM UNCOPPICED (U) AND COPPICED (C) SOURCES.

Contrasts	Crude Protein	ADF	NDF	Dry Matter	Ash	ADG (g)	Dry Matter Intake (g)	Feed/ Gain
1. Control vs PL treatments (Groups 1 vs 2 3 4 5 6)	78.75 vs 63.91*	26.28 vs 18.13	36.87 vs 26.28*	64.35 vs 58.32	60.62 vs 49.76*	35.5 vs 37.7	105.6 vs 121.4*	3.4 vs 3.7
2. U vs C PL at 10% (Groups 2 vs 5)	68.71 vs 69.92	19.04 vs 21.07	29.53 vs 31.21	59.02 vs 59.88	51.93 vs 52.40	39.7 vs 37.3	117.3 vs 37.3	3.3 vs 3.7
3. U vs C PL at 20% (Groups 3 vs 6)	60.05 vs 66.11*	18.40 vs 21.97	20.88 vs 31.01	55.72 vs 59.34	48.95 vs 50.81	36.7 vs 37.1	122.3 vs 114.3	3.8 vs 3.5
4. Complete Substitution (Groups 1 vs 4)	78.75 vs 54.76*	26.28 vs 10.15	36.87 vs 18.79*	64.35 vs 57.63	60.62 vs 44.73*	35.3 vs 37.9	105.6 vs 133.9*	3.4 vs 4.0

* indicates statistical significance at .05 level

References

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Baertsche, S.R., M.T. Yokoyama and J.W. Hanover. 1986. Short rotation, hardwood tree biomass as potential ruminant feed - chemical composition, nylon bag ruminal degradation and ensilement of selected species. J. Anim. Sci. 63:2028-2043.

Bas, F.J., F.R. Ehle and R.D. Goodrich. 1985. Evaluation of pelleted aspen foliage as a ruminant feedstuff. J. Anim. Sci. 61:1030-1036.

Benge, M.D. 1987. Multipurpose uses of contour hedgerows in highland regions. World Anim. Rev. 64:31-39.

Basey, J.M., S.H. Jenkins and P.E. Busher. 1988. Optimal central-place foraging by beavers: Tree-size selection in relation to defensive chemicals of quaking aspen. Oecologia 76:278-282.

Bradshaw, H.D., J.B. Hollick, T.J. Parsons, H.R.G. Clark and M.P. Gordon. 1989. Systemically wound-responsive genes in poplar trees encode proteins similar to sweet potato sporamins and legume Kunitz trypsin inhibitors. Plant Molecular Biology 14:51-59.

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Inc. Westport, CT. p 333.

Dula, J.C. 1982. *Newsletter*. No. 2-82. Canby, OR. Dula's Nurseries.

FAO 1980. Poplars and Willows : in wood production and land use. Food and Agriculture Organization of the United Nations 1980. pp.12-13.

Forwood, J.R. and C.E. Owensby. 1985. Nutritive value of tree leaves in the Kansas Flint Hills. J. Range. Manage. 38:61-64.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Jakubas, W.J., G.W. Gullion and T.P. Clausen. 1989. Ruffed grouse feeding behavior and its relationship to secondary metabolites of quaking aspen flower buds. J. Chem. Ecol. 15:1899-1915.

Kramer, P.J. and T.T. Kozlowski. 1979. Physiology of woody plants. Academic Press, New York, p 811.

McKey, D. 1979. The distribution of secondary compounds within plants. In: Rosenthal, G.A., Janzen, D.H. (eds) Herbivores - Their interactions with secondary plant metabolites. Academic Press, New York, pp 56-133.

Meyer, G.A. and M.E. Montgomery. 1987. Relationships between leaf age and the food quality of cottonwood foliage for the gypsy moth, *Lymantria dispar*. Oecologia 72:527-532.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc., Cary, NC.

Van Soest, P.J. 1982. Nutritional Ecology of the Ruminant. O & B Books, Inc. Corvallis, OR.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67-69.

EFFECT ON WEANLING RABBITS OF THE ADDITION OF POLYETHYLENE GLYCOL, PHYTASE, METHIONINE AND CHOLINE CHLORIDE TO DIETS CONTAINING BLACK LOCUST (*ROBINIA PSEUDOACACIA)* LEAF MEAL

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Abstract

Forty New Zealand White weanling rabbits were fed diets containing 50% alfalfa (control), 25% alfalfa and 25% black locust leaf meal (BLM) (BLM control), BLM + 1% polyethylene glycol (PEG), BLM + 1% phytase, and BLM + 0.3% L-methionine and 0.3% choline chloride. The digestibilities of dry matter (DM), crude protein (CP), ADF, NDF, ash, calcium and phosphorous were significantly higher for the 50% alfalfa control than for the BLM treatments (respective p values < .01, .01, .01, .01, .01, .01, .01). The average daily gain was higher for the control (p < .01) and feed/gain was lower (p < .01) than for the BLM treatments. The lower utilization of nutrients in the BLM diets is probably due to the presence of condensed tannins. The addition of PEG increased CP (p < .01) and ADF (p < .03) digestibilities compared to the BLM control. The addition of L-methionine and choline chloride increased ADF digestibility (p < .02) compared to the BLM control. It was concluded that PEG is partially

effective in alleviating the reduced nutrient availability due to tannins.

Introduction

Black locust (*Robinia pseudoacacia*) is a fast-growing, leguminous tree; its leaves can be harvested for animal feed. In China, the trees are grown to provide leaves as a principle source of rabbit feed (Cheeke and Patton, 1987) and as a source of pigmenting agents for poultry feed (Takada et al., 1980). Black locust (BL) is intensively cultivated in Hungary for saw logs (particularly a variety called "shipmast locust"), poles and posts, beekeeping and decorative planting (Keresztesi, 1983).

Black locust is indigenous to the eastern U.S. It has a rapid growth rate which can average 2 feet per year, and the tree can reach a height of 70 feet. It is very adaptable to soils and climate, having the ability to grow in any type of soil except permanently wet soil and to fix atmospheric nitrogen (Dirr, 1990).

Although BL leaves are similar to alfalfa meal in nutrient composition (Cheeke, 1992), feeding trials with BL leaf meal have generally given poor animal performance (Horton and Christensen, 1981, Harris et al., 1984, Raharjo et al., 1990, Takada et al., 1980).

Black locust leaves contain the anti-nutritive factors robin and tannins. Robin is a lectin that may cause adverse effects on animals by binding to the intestinal wall and is suspected of poisoning horses when they eat the bark (Cheeke and Shull, 1985). Tannins are phenolic substances which bind dietary proteins and digestive enzymes and reduce nutrient digestibility (Cheeke and Shull, 1985). Tannins are responsible for the low protein digestibility of BL leaves (Horigome et al., 1984).

Various methods of detoxifying tannins have been demonstrated. Dietary supplementation with polyethylene glycol (PEG) reduces adverse effects because tannins bind more strongly to PEG than to proteins (Oh et al., 1980). Methionine and choline chloride may act as methyl donors to facilitate the excretion of absorbed phenolic acids (Cheeke and Shull, 1985). Sulfur-containing amino acids counteract adverse effects of tannic acid in chickens (Fuller et al., 1967).

Tree leaves are often low in phosphorous (Kumar and Vaithiyanathan, 1990). Phosphorous (P) was found to be poorly utilized by lambs fed BL leaves as a sole feed (Horton and Christensen, 1981). A phytase-rich diet including rye bran has been shown to increase P availability in swine (Pointillart, 1991).

The objectives of this study were to determine the effects of adding PEG, phytase, and methionine and choline chloride to diets containing BL leaves on nutrient digestibility and growth performance in weanling rabbits. A diet with an equal amount of alfalfa meal in place of BL served as the control.

Materials and Methods

Forty New Zealand White weanling rabbits aged 5 to 6 weeks old were randomly assigned to five treatment groups:

1) 50% alfalfa (control)

2) 25% alfalfa and 25% black locust leaf meal (BLM) (BLM control)

3) 25% alfalfa and 25% BLM + 1% PEG[1]

4) 25% alfalfa and 25% BLM + 1% phytase[2]

5) 25% alfalfa and 25% BLM + 0.3% L-methionine[3] and 0.3% choline chloride[4].

The trial consisted of a 10-day adaptation period followed by a 10-day digestibility period. Rabbits were

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[1]Sigma Chemical Co., St. Louis, MO. Average molecular weight:3350.

[2]Alltech Biotechnology, Inc., Nicholasville, KY

[3]U.S. Biochem. Corp., Cleveland, OH

[4]U.S. Biochem. Corp., Cleveland, OH

housed in individual cages with automatic water valves. Feed intake was measured throughout the trial and during the digestibility period. Feed intake was adjusted by collecting and measuring wasted feed which fell from the feeder. Initial, weekly and final weights were taken, along with initial and final weights for the digestibility period. Total fecal output for the digestibility period was collected with screens suspended underneath each cage.

Animals were fed the diets *ad libitum* (Table 1). The BL leaves were hand-harvested from mature trees felled on a local farm in July 1990. The branches and leaves were air-dried on screens. The leaves were stripped from branches and put through a shredder. The leaves were dried in an oven at 60° C for 48 hours and then ground through a 2 mm screen.

After the end of the digestibility period, the total wet feces were weighed and a subsample taken which was dried in an oven at 60° c for 72 hours. The dry subsample was weighed and ground through a 1 mm screen. Feed and feces samples were analyzed for dry matter and ash by standard procedures (AOAC, 1984). Samples were analyzed for ADF and NDF as described by Goering and Van Soest (1970), modified by a micro method (Waldern, 1971). Crude protein was determined by the Macro Kjeldahl method (AOAC 1984). Calcium (Ca) was measured by atomic absorption spectrophotometry (AOAC 1984) and P was measured with a spectrophotometer by the vanadomolybdate method (AOAC 1984).

The data were analyzed following the general linear model procedure (SAS, 1985). Initial F-screening was done at the .05 level. Treatment means were compared using a series of contrasts from 4 pre-planned comparisons.

Results and Discussion

The digestibility and growth performance data are given in Table 2 and statistical comparisons in Table 3.

In Comparison 1, the digestibilities of dry matter (DM), crude protein (CP), ADF, NDF, ash, Ca and P were significantly higher for the alfalfa control than for the treatments with the BLM (respective p values $< .01$, .01, .01, .01, .01, .01, .01). The overall model for average daily gain (ADG) approaches significance with the F-screening ($p < .09$), but when examining Comparison 1, it is evident that the ADG was significantly higher ($p < .01$) for the alfalfa control than for the BLM treatments. Feed efficiency was lower for the alfalfa control than for the BLM treatments ($p < .01$).

Comparison 2 (BLM control vs. BLM + PEG) showed significantly higher crude protein ($p < .01$) and ADF digestibilities when PEG was added to the diet ($p < .03$).

Comparison 3 (BLM control vs. BLM + phytase) showed a slight, although not significant, increase in digestibility of dry matter when phytase was added to the diet ($p < .07$).

In Comparison 4 (BLM control vs. BLM + L-methionine and choline chloride), the addition of L-methionine and choline chloride statistically increased ADF digestibility ($p < .02$) and gave a slight, although not significant, increase of P digestibility ($p < .07$).

There was no mortality on any of the treatments.

The diets containing BLM were more poorly digested than the alfalfa control. This is most likely due to the presence of tannins in the BL leaves. Tannins may be classified as condensed or hydrolyzable tannins (Salunkhe et al., 1990). Black locust leaves contain primarily condensed tannins (Kumar and Horigome, 1986). The leaves contain about 5-7% condensed tannins (B. Luick, 1992 personal communication, University of Alaska). Tannins precipitate proteins by hydrogen-bonding and hydrophobic interactions (Haslam, 1974). Kumar and Horigome (1986) examined the nature of BL tannins. The proanthocyanidins (condensed tannins) in BL may exist in five molecular sizes. Their protein-precipitating capacity increases with an increasing degree of polymerization.

Horton and Christensen (1981) found that when lambs were fed BL leaves as a sole feed, the digestibilities of organic matter, CP, ADF, NDF and P were less than half those of alfalfa. Although no significant differences were seen between lambs fed BL leaves and alfalfa in terms of ADG or DM intake, it was concluded that BLM was not a satisfactory replacement for alfalfa. Harris et al. (1984) found ADG for weanling rabbits fed a 40% BL diet to be lower than the alfalfa control even though DM intake was higher for the BL diet. Raharjo et al. (1990) also found ADG to be lower for rabbits fed a 50% BL diet when compared to an alfalfa control, and reported twice as much N bound to the ADF fraction in the diets containing BL leaves as compared to the alfalfa control diet.

In our experiment, the addition of 1% PEG improved crude protein digestibility from 58.8% to 66.3%. This is in agreement with the findings of Horigome et al. (1984), who found that adding 1.6% PEG to rat diets containing BL leaves increased CP digestibility from 49.1% to 70.7%. Only 46% of the PEG was recovered in the feces compared to 91% from a diet containing brewers' grains, thus showing that a water-insoluble complex was formed in the gut. Pritchard et al. (1988) fed sheep *Acacia aneura* (which contained 6.1% tannins) and found that drenching the sheep every day with PEG increased body weight and wool growth.

These findings contrast with those of Nuñez-Hernandez et al. (1991), who determined that adding PEG to diets containing mountain mahogany leaves, which have condensed tannins, did not improve nutrient digestion or affect N balance in goats and lambs. Garrido et al., (1991) found that the addition of PEG to faba bean seed coat, at a level of 200 mg of PEG per gram of seed coat, increased CP digestibility in vitro from 52.4% to 75.5%. A reduction of 97% of the tannins was observed by the addition of increasing levels of polyvinylpyrrolidine.

Tannins may affect not only CP digestibility but the digestibilities of other nutrients as well. A lower digestibility of DM and energy was observed in lambs fed BLM as compared to alfalfa (Horton and Christensen, 1981). In our study, ADF digestibility was also improved by addition of PEG to the BLM diet. The ADF was poorly digested for the control BLM diet (2.63%), but increased slightly to 8.55% with addition of PEG. Insoluble tannin-fiber complexes may be measured as ADF in the feces to give very low ADF digestibilities (Cheeke, 1992). It is evident from the NDF values in this study that the hemicellulose portion is better utilized for the alfalfa control than the BLM treatments.

The effect that tannins may have on mineral availability is not entirely clear, but tannins appear to form insoluble complexes with divalent metal ions which make them less absorbable (Salunke et al., 1990). Horton and Christensen (1981) found P to be poorly digested by lambs fed BLM. In our study, Ca and P were less available in the BLM diets as compared to the alfalfa control and the addition of phytase did not improve P or Ca utilization; however, there was a trend towards improved DM digestibility with addition of phytase. Pointillart (1991) found that feeding a 20% rye bran diet high in phytase (1200 IU/kg) to pigs increased P absorption (55 vs 36%) and retention (50 vs 36%) when compared to a control without rye bran.

The addition of methionine and choline chloride to the BLM diet did not improve CP digestibility, but it did improve ADF digestibility, giving an increase from 2.63% to 8.81%. This may perhaps be explained by the tannins binding more strongly to protein than to fiber, allowing a better use of ADF. There was also a trend towards improved P utilization with addition of methionine and choline chloride. In a study by Myer et al. (1986), 0.1% DL-methionine was added to diets containing bird-resistant (high tannin) and non-bird-resistant (low tannin) grain sorghum. Dietary methionine was not effective in alleviating the detrimental effects that tannins have on swine. No improvement was seen in ADG or feed efficiency.

Fuller et al. (1967) found that adding MHA (methionine hydroxy analogue) or choline to diets containing 0, 0.5, 1% tannic acid partially alleviated the growth depression due to the tannic acid in chicks. They proposed that the methionine probably provides labile methyl groups for the production of 4-O-methyl gallate, found by Booth et al. (1959) to be the major metabolite in the urine of rats and rabbits fed tannic acid and gallic acid.

Black locust leaf meal is not a satisfactory replacement of alfalfa in the diets of weanling rabbits. The nutrient digestibilities and ADG were lower for the BLM diets than for the alfalfa control. Since the DM intake did not increase to compensate for this, palatability may be a problem. The addition of PEG to diets containing BLM leaves improved nutrient utilization and warrants further investigation into the use of black locust as animal feed when treated with PEG. At the levels used in this study, addition of phytase and L-methionine and choline chloride had little effect on nutrient utilization. Further trials could be conducted using higher levels.

EXPERIMENT 2

TABLE 1. COMPOSITION AND CHEMICAL ANALYSIS (%) OF DIETS FED TO RABBITS (DRY MATTER BASIS).

Diet #	1	2	3	4	5
Alfalfa meal, sun-cured	50	25	25	25	25
Black locust leaves	-	25	25	25	25
Ground barley	42.5	42.5	41.5	41.5	41.5
Molasses	5	5	5	5	5
Trace mineral salt*	0.25	0.25	0.25	0.25	0.25
Vegetable oil	2	2	2	2	2
Vitamin mix**	0.25	0.25	0.25	0.25	0.25
Choline chloride	-	-	-	-	0.3
L-Methionine	-	-	-	-	0.3
Phytase	-	-	-	1	-
Polyethylene glycol	-	-	1	-	-
Chemical Analysis:					
Crude protein	15.53	15.7	15.6	15.9	16.31
ADF	18.53	18.34	18.85	18.49	18.86
NDF	30.32	30.27	30.11	29.55	29.80
Ash	7.02	7.44	7.68	7.84	7.67
Ca	0.98	1.49	1.61	1.43	1.64
P	0.33	0.27	0.25	0.28	0.30

*Ingredients: sodium chloride, zinc oxide, manganous oxide, iron carbonate, copper oxide

**Contains: Vitamin A 3,000,000 IU

D-3 1,000,000 IU
E 1000 IU
K 0.5 gm
B-12 5 mg
Riboflavin 3 gm
Pantothenic acid 5 gm
Niacin 20 gm
Choline chloride 200 gm
Folic acid 200 gm
Ethoxyquin 56.75 gm

* and ** manufactured by Inman & Co., Inc.; 12530 SE Jennifer St., Clackamas, OR 97015

EXPERIMENT 2

TABLE 2. MEANS (±SE) OF DIGESTIBILITIES (%) OF NUTRIENTS AND GROWTH INDICATORS OF WEANLING RABBITS FED DIETS CONTAINING ALFALFA AND BLACK LOCUST LEAF MEAL (BLM) MODIFIED BY VARIOUS TREATMENTS.

Trt	CP	ADF	NDF	DM	Ash	Ca	P	ADG	DM Intake	Feed/ Gain
1. 50% alfalfa (Control)	77.07 ±1.34	15.81 ±1.79	25.78 ±1.20	68.80 ±0.85	75.26 ±1.34	84.51 ±1.58	70.33 ±2.56	37.0 ±1.7	95.9 ±3.2	2.9 ±0.1
2. 25% alfalfa & 25% BLM (BLM Control)	58.80 ±1.34	2.63 ±1.79	19.79 ±1.20	62.32 ±0.85	66.32 ±1.34	75.62 ±1.58	58.48 ±2.39	30.4 ±1.7	93.0 ±3.2	3.4 ±0.1
3. BLM Control + PEG	66.30 ±1.34	8.55 ±1.79	18.38 ±1.20	63.64 ±0.85	68.13 ±1.34	75.33 ±1.58	61.60 ±2.39	32.4 ±1.7	100.1 ±3.2	3.5 ±0.1
4. BLM Control + phytase	60.06 ±1.34	6.91 ±1.79	18.94 ±1.20	64.50 ±0.85	68.67 ±1.34	73.72 ±1.58	63.65 ±2.39	31.4 ±1.7	91.2 ±3.2	3.2 ±0.1
5. BLM Control + L-methionine & choline chloride	57.77 ±1.34	8.81 ±1.79	17.14 ±1.20	62.43 ±0.85	67.90 ±1.34	74.58 ±1.58	64.83 ±2.39	33.3 ±1.7	94.4 ±3.2	3.2 ±0.1
Average of 2, 3, 4 and 5	60.73 ±1.34	6.73 ±1.79	18.56 ±1.20	63.22 ±0.85	67.76 ±1.34	74.81 ±1.58	62.14 ±2.39	31.9 ±1.7	94.7 ±3.2	3.3 ±0.1

EXPERIMENT 2

TABLE 3. STATISTICAL COMPARISONS BETWEEN NUTRIENT DIGESTIBILITY MEANS (%) AND GROWTH INDICATOR MEANS OF WEANLING RABBITS FED DIETS CONTAINING ALFALFA AND BLACK LOCUST LEAF MEAL (BLM) MODIFIED BY VARIOUS TREATMENTS.

Contrast	Crude Protein	ADF	NDF	Dry Matter	Ash	Ca	P	ADG (g)	DM Intake g/day	Feed / Gain
1	77.07 vs 60.73*	15.81 vs 6.73*	25.78 vs 18.56*	68.80 vs 63.22*	75.26 vs 67.76*	84.51 vs 74.81*	70.33 vs 62.14*	37.0 vs 31.9*	95.9 vs 94.7	2.9 vs 3.3*
2	58.80 vs 66.30*	2.63 vs 8.55*	19.79 vs 18.38	62.32 vs 63.64	66.32 vs 68.13	75.62 vs 75.33	58.48 vs 61.60	30.4 vs 32.4	93.0 vs 100.1	3.4 vs 3.5
3	58.80 vs 60.06	2.63 vs 6.91	19.79 vs 18.94	62.32 vs 64.50	66.32 vs 68.67	75.62 vs 73.72	58.48 vs 63.65	30.4 vs 31.4	93.0 vs 91.2	3.4 vs 3.2
4	58.80 vs 57.77	2.63 vs 8.81*	19.79 vs 17.14	62.32 vs 62.43	66.32 vs 67.90	75.62 vs 74.59	58.48 vs 64.83	30.4 vs 33.3	93.0 vs 94.4	3.4 vs 3.2

Contrasts are:
1. Control vs. BLM Treatments (Groups 1 vs. 2, 3, 4 and 5)
2. BL Control vs. BLM + PEG (Groups 2 vs. 3)
3. BL Control vs. BLM + phytase (Groups 2 vs. 4)
4. BL Control vs. BLM + L-methionine and choline chloride (Groups 2 vs. 5)

* indicates statistical significance at .05 level

References

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Booth, A.N., M.S. Morse, D.J. Robbins, O.H. Emerson, F.T. Jones and F. Deeds. 1959. The metabolic fate of gallic acid and related compounds. J. Biol. Chem. 234:3014-3016.

Cheeke, P.R. 1992. Black locust forage as an animal feedstuff. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp 32-38.

Cheeke, P.R. and N.M. Patton. 1987. Rabbit production and research in China. J. Appl. Rabbit Res. 10:68-75.

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Inc. Westport, CT p. 333

Dirr, M.A. 1990. Manual of Woody Landscape Plants: Their Identification, Ornamental Characteristics, Culture, Propagation and Uses. Stipes Publishing Co., Champaign, IL. pp 753-756.

Fuller, H.L., S.I. Chang and D.K. Potter. 1967. Detoxification of dietary tannic acid by chicks. J. Nutr. 91:477-481.

Garrido, A., A. Gómez-Cabrera, J.E. Guerrero and J.M. van der Meer. 1991. Effects of treatment with polyvinylpyrrolidone and polyethylene glycol on Faba bean tannins. Anim. Feed Sci. Tech. 35:199-203.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Harris, D.J., P.R. Cheeke and N.M. Patton. 1984. Evaluation of black locust leaves for growing rabbits. J. Appl. Rabbit Res. 7:7-9.

Haslam, E. 1974. Polyphenol-Protein Interaction. Biochem. J. 139:285-288.

Horigome, T., T. Ohkuma and M. Muta. 1984. Effect of condensed tannin of false acacia leaves on protein digestibility, as measured with rats. Jap. J. Zootech. Sci. 55:299-306.

Horton, G.M.J. and D.A. Christensen. 1981. Nutritional value of black locust tree leaf meal (*Robinia pseudoacacia*) and alfalfa meal. Can. J. Anim. Sci. 61:503-506.

Keresztesi, B. 1983. Breeding and cultivation of black locust, *Robinia pseudoacacia*, in Hungary. Forest Ecol. and Manage. 6:217-244.

Kumar, R. and T. Horigome. 1986. Fractionation, characterization and protein-precipitating capacity of the condensed tannins from *Robinia pseudoacacia* L. leaves. J. Agric. Food Chem. 34:487-489.

Kumar, R. and S. Vaithiyanathan. 1979. Occurrence, nutritional significance and effect on animal productivity of tannins in tree leaves. Anim. Feed Sci. Tech. 30:21-38.

Myer, R.O., G.E. Combs and D.W. Gorbet. 1986. Effect of methionine addition on the feeding value of diets containing bird-resistant grain sorghum for growing-finishing swine. Nutr. Reports Int. 33:843-850.

Nuñez-Hernandez, G., J.D. Wallace, J.L. Holechek, M.L. Galyean and M. Cardenas. 1991. Condensed tannins and nutrient utilization by lambs and goats fed low-quality diets. J. Anim. Sci. 69:1167-1177.

Oh, H.I., J.E. Hoff, G.S. Armstrong, and L.A. Haff. 1980. Hydrophobic interaction in tannin-protein complexes. J. Agric. Food Chem. 28:394-398.

Pointillart A. 1991. Enhancement of phosphorous utilization in growing pigs fed phytase-rich diets by using rye bran. J. Anim. Sci. 69:1109-1115.

Pritchard, D.A., D.C. Stocks, B.M. O'Sullivan, P.R. Martins, I.S. Hurwood and P.K. O'Rourke. 1988. The effect of polyethylene glycol (PEG) on wool growth and live weight of sheep consuming a Mulga (*Acacia aneura) diet. Proc. Aust. Soc. Anim. Prod. 17:290-293.*

Raharjo, Y.C., P.R. Cheeke and N.M. Patton. 1990. Effect of cecotrophy on the nutrient digestibility of alfalfa and black locust leaves. J. Appl. Rabbit Res. 13:56-61.

Salunkhe, D.K., J.K. Chavan and S.S. Kadam. 1990. Dietary Tannins: Consequences and Remedies. CRC Press, Boca Raton, FL. p.135.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc., Cary, NC.

Takada, K., T. Nakazato, K. Ono, H. Honda and T. Yamane. 1980. Feeding value of leaf meal of acacia on poultry feed. Jap. Poul. Sci. 17:299-305.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67- 69.

EFFECT ON WEANLING RABBITS OF BLACK LOCUST (*ROBINA PSEUDOACACIA*) BARK, OAK SAWDUST, RED ALDER (*ALNUS RUBRA*) BARK AND RED ALDER SAWDUST IN THE DIET

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Abstract

Forty-eight New Zealand White weanling rabbits were fed diets containing 25% black locust (*Robinia pseudoacacia*) bark, oak sawdust, red alder (*Alnus rubra*) bark, or red alder sawdust (all diets also included 25% alfalfa). A 50% alfalfa diet served as a control, along with a standard OSU fryer diet which provided a performance reference. Average daily weight gain (ADG) was higher for the alfalfa control as compared to the treatments, as were the nutrient digestibilities, with the exception of the ADF and NDF fractions. Nutrient digestibilities of black locust bark were higher than those of alder bark with the exception of NDF; however, the ADG was much lower for the black locust bark diet. According to the literature, black locust bark contains lectins that disrupt gut function. The bark should be avoided in the use of black locust forage for animal feeding. On the basis of ADG, alder bark appears to have a slightly higher feeding value than alder sawdust, and oak sawdust appears to have a higher feeding value than

alder sawdust.

Introduction

The use of tree forage has potential for animal feeding as a by-product from the forestry industry or from multipurpose trees incorporated into agroforestry systems. Although the leaves of black locust (*Robinia pseudoacacia)* have a nutrient composition similar to alfalfa, feeding trials have generally given poor results (Cheeke, 1992). The lower availability of nutrients is probably due to tannins and lectins. Robin is a lectin which is reputed to be present in black locust (BL) (Cheeke, 1992).

Lectins (hemagglutins) are glycoproteins that cause clumping of red blood cells in vitro (Cheeke and Shull, 1985). They are present in beans and a number of other plants including BL. Lectins have a high affinity for sugars, binding to carbohydrate moieties in the intestinal epithelium and interfering with digestion. Some symptoms of lectin poisoning include anorexia, weakness, diarrhea and death. Horses have been poisoned by stripping the bark of black locust trees to which they have been tied (Cheeke and Shull, 1985).

It is of interest to determine the suitability of including BL bark in the diet when the leaves are harvested for use as leaf meal and to examine feeding value of bark in general. It is hypothesized that most of the lectins that exert a toxic effect on animals are contained in the bark. Consumption of bark is common among wildlife, but is not usually a major part of a mammal's diet (Whitten and Whitten, 1987). Aspen twigs, buds, leaves and bark are adequate forage sources for deer and elk and could also be utilized by domestic ruminants (Tengerdy and Nagy, 1988).

Other forestry by-products such as sawdust have been tested for animal feeding with some success, especially when chemically or physically treated to improve digestibility (Vidal and Molinier, 1988; Tengerdy and Nagy, 1988). In Australia, eucalyptus sawdust was treated with high temperature, pressure and a catalyst to break up lignin-cellulose association, after which it was cooked, adding nonprotein nitrogen and minerals to create a product used as 34% of sheep diets. (Anonymous, 1979).

The objectives of this study were to evaluate the effect of adding BL bark to the diet of growing rabbits as compared to alder bark (another nitrogen-fixing tree) and alfalfa control in order to determine whether BL bark has a toxic effect on rabbits; and, secondarily, to examine the feeding value of bark compared to sawdust (alder bark vs. alder sawdust) and oak sawdust compared to alder sawdust.

Materials and Methods

Forty-eight New Zealand White weanling rabbits approximately 5 weeks old were randomly assigned to six treatments with eight rabbits being allocated to each treatment. The treatments were:

1) 25% alfalfa and 25% black locust bark
2) 25% alfalfa and 25% oak sawdust
3) 25% alfalfa and 25% alder bark
4) 25% alfalfa and 25% alder sawdust
5) 50% alfalfa control
6) standard OSU fryer diet.

The trial consisted of a 10-day adaptation period followed by a 10-day digestibility period. Rabbits were housed in individual cages with automatic dewdrop water valves. Feed intake was measured throughout the trial and during the digestibility period. Feed intake was adjusted by collecting and measuring wasted feed. Initial and weekly weights were taken, along with initial and final weights for the digestibility period. Total fecal output for the digestibility period was collected with screens suspended underneath each cage.

Throughout the trial, animals were fed the diets *ad* *libitum* (Table 1). The black locust bark was harvested from recently felled trees on a local farm in June 1991. It was stripped from the trunks and branches and air-dried for 3 weeks. Bark was put through a shredder and oven-dried at 50° C for 5 days. It was then ground through a 2 mm screen. The oak sawdust came from a commercial source. The alder bark was collected from recently felled trees at a local sawmill. Bark was sent through a chipper. These pieces were oven-dried at 50° c for 5 days and then ground through a 2 mm screen. The alder sawdust came from accumulated piles at the same sawmill. It was oven-dried at 50° C for 72 hours.

After the end of the digestibility period, the total wet feces were weighed and a subsample taken which was oven-dried at 60° C for 72 hours. The dry subsample was weighed and then ground through a 1mm screen. Feed and feces samples were analyzed for dry matter and ash by standard procedures (AOAC, 1984). Samples were analyzed for ADF and NDF as described by Goering and Van Soest (1970), modified by a micro method (Waldern, 1971). Crude protein was determined by the Macro Kjeldahl method (AOAC, 1984).

The data were analyzed following the general linear model procedure (SAS, 1985). Initial F-screening was done at the .05 level. Treatment means were compared using a series of contrasts from four pre-planned comparisons. Three rabbits with very poor performance were removed from the trial.

Results and Discussion

The compositions of the experimental feed ingredients are given in Table 2. The digestibility and growth performance data are given in Table 3 and the statistical comparisons in Table 4.

In Comparison 1, the digestibilities of crude protein (CP), dry matter (DM), and ash were higher for the alfalfa control than for all the treatments (respective p values $<$ .01, .01, .01, .01). The NDF digestibility was lower ($p <$ .01) for the alfalfa control than for the treatments. The ADF digestibility did not differ significantly in Comparison 1; however, this is due to the means of 1, 2, 3 and 4 canceling each other out. If all possible pairs are compared, they were all significantly different except for the means of 1 and 4 at the .05 level. Therefore, it is evident that the ADF fraction was better digested for black locust bark (BLB) and alder sawdust (AS) treatments than for the alfalfa control. Average daily weight gain and DM intake were significantly lower for all treatments than for the alfalfa control ($p <$ .01 and .01) and feed/gain was significantly higher for the treatments compared to the alfalfa control ($p < .01$).

In Comparison 2, the CP, ADF and DM were better digested in the BLB diet than the alder bark (AB) diet ($p < .01, .01, .01$), whereas NDF was better digested in the AB diet than in the BLB diet ($p < .01$); however, average daily weight gain and DM intake were significantly lower for the BLB diet than for the AB diet ($p < .01$ and $.01$) and feed/gain was higher for the BLB than the AB diet ($p < .01$). This may indicate a toxic effect of the BLB diet.

In Comparison 3, the CP and ash digestibilities were higher for the AB diet than for the AS diet ($p < .04$ and $.05$), whereas the ADF digestibility was higher for the AS ($p < .01$). Average daily weight gain was slightly, although not significantly higher for the AB than the AS diet ($p < .06$).

In Comparison 4, CP and ash were better digested in the oak sawdust (OS) treatment ($p < .01, .01$), whereas ADF and NDF digestibility were higher for the AS treatment ($p < .01, .01$). Dry matter intake and ADG were higher for the OS treatment ($p < .04, .01$).

There was no mortality on any of the treatments, but three rabbits had to be removed from the BLB treatmnet because they were losing condition to such an extent that death was likely.

The bark of trees includes dead cells of phloem, the cortex where food is stored and the cork which protects the cambium from temperature changes and fungal attack and prevents dehydration (Feininger, 1968). Whitten and Whitten (1987) found that for a tropical squirrel which feeds mostly on bark (49% of stomach contents), selection of feed trees was not influenced by concentrations of CP, crude fiber, fat, calorific value or condensed tannin levels in bark. Most barks are low in N and carbohydrate calories. The authors theorized that the squirrels were selecting for something such as a combination of trace elements in the bark.

The BLB had a high level of CP (16.9%) whereas AB had a much lower level (1.9%) (Table 2). Whitten and Whitten (1987) found that the CP level of the barks of 13 tropical trees ranged from 0.48 to 3.79%. The high CP level in BLB may be due to the presence of lectins. Nsimba-Lubaki and Peumans (1986), in a study examining the seasonal fluctuations of lectins in BL bark, found that bark lectins made up 5% of the total protein content. BL lectins were not exclusively found in bark. According to the literature, most of the work done with lectins has been with seeds, but lectins are present in other tissues such as roots and leaves as well. Nsimba-Lubaki and Peumans (1986) considered the bark of trees to be a vegetative storage tissue and bark lectins to behave as typical storage proteins. In the late spring, lectins were equally present in bark and other vegetative tissues such as leaves, roots, stems and flowers, but in the winter, they found that lectins increased by 50 fold in the bark compared to summer levels. Nitrogen is normally stored in the form of protein in the bark at the end of the growing season until the following spring. This decreases with age: older trees showed less disappearance of lectins in the summer.

In a diet identical to the experimental BLB diet in which BLB was replaced by BL leaves (Ayers et al., 1992), rabbits showed weight gains 8 times higher than the gains obtained by the rabbits consuming BLB diet. This indicates a lower feeding value and a probable toxic effect of the bark, most likely due to lectins. The animals on the BLB performed so poorly that three had to be removed from the trial so they would not die. The bark used in our study was harvested in the summer, when the lectin levels would be relatively low, but the lectins still appeared to be of a such a higher level than the leaves to show a toxic effect. The involvement of toxins other than lectins cannot be excluded.

Sawdust is about 60% cellulose and 40% lignin and tannins (Anonymous, 1979). The ligno-cellulose structure and cellulose crystallinity prevents enzymes from degrading the cellulose. This may not be a problem for rabbit rations because rabbits do not normally digest fiber to a great extent. Fiber does seem to be necessary to keep intestinal lining of the gut healthy (Cheeke et al., 1987). Fayek et al. (1989) found no difference in weanling rabbit performance between urea-treated and untreated sawdust at levels of 5, 7.5 and 10% of the diet. They concluded that sawdust can be incorporated into the diet at levels up to 10% without deleterious effects on growth. El-Sabban et al. (1970) found that up to 15% sawdust in finishing beef cattle rations did not affect carcass characteristics.

Sawdust has been treated by chemical or physical means to improve digestibility by disrupting the ligno-cellulose structure (Tengerdy and Nagy, 1988). Vidal and Molinier (1988) treated poplar sawdust (an abundant industry by-product in southern Europe) with ozone to increase its value as cattle feed. Ozonolysis of lignan improved in vitro digestibility. Tengerdy and Nagy (1988) used an ammonia freeze explosion treatment to decrease lignin of aspen twigs and chips by 30% and aspen bark by 20%. This improved the in vitro digestibility of twigs and chips. Bark, already high in IVD, did not increase further. According to the authors, most of the digestibility in bark is due to about 40% soluble materials.

In our study, none of the treatments, including the alfalfa control, gave satisfactory ADG when compared to the standard Oregon State University fryer diet. This is due to the deliberate omission of a protein supplement in the experimental diets, in order that the digestibility values would reflect the test substances as much as possible.

It can be concluded that BL bark is very toxic to rabbits, possibly because of its lectins. This would suggest that when BL forage is harvested for animal feeding, the harvesting procedure should be conducted so as to maximize the leaf component and minimize the inclusion of bark from stems. On the basis of ADG, bark appears to have a slightly higher feeding value than sawdust, and in comparing two types of sawdust, OS appears to have higher feeding value than AS.

EXPERIMENT 3

TABLE 1. COMPOSITION (%) OF EXPERIMENTAL DIETS FED TO RABBITS (DRY MATTER BASIS).

Ingredients	1	2	3	4	5
Alder bark	-	-	25	-	-
Alder sawdust	-	-	-	25	-
Alfalfa meal	25	25	25	25	50
Black locust bark	25	-	-	-	-
Ground barley	42.5	42.5	42.5	42.5	42.5
Molasses	5	5	5	5	5
Oak sawdust	-	25	-	-	-
Trace mineral salt*	0.25	0.25	0.25	0.25	0.25
Vegetable oil	2	2	2	2	2
Vitamin mix**	0.25	0.25	0.25	0.25	0.25
Chemical Analysis					
Crude Protein	13.2	11.0	10.3	6.4	11.4
ADF	22.5	23.9	19.5	31.7	17.6
NDF	36.8	81.2	80.9	88.4	31.7
Ash	6.6	5.3	6.1	4.6	8.2

*Ingredients: Sodium chloride, zinc oxide, manganous oxide, iron carbonate, copper oxide

**Vitamin A 3,000,000 IU
- D-3 1,000,000 IU
- E 1000 IU
- K 0.5 gm
- B-12 5 mg
- Riboflavin 3 gm
- Pantothenic acid 5 gm
- Niacin 20 gm
- Choline chloride 200 gm
- Folic acid 200 mg
- Ethoxyquin 56.75 gm

* and ** manufactured by Inman & Co., Inc; 12530 SE Jennifer St., Clackamas, OR 97015.

EXPERIMENT 3

TABLE 2. NUTRIENT COMPOSITION (%) OF EXPERIMENTAL TREE PRODUCTS

Ingredient	Crude Protein	ADF	NDF	Ash
Black locust bark	16.91	42.34	56.08	7.40
Oak sawdust	<.01	62.92	91.60	0.53
Alder bark	1.90	47.36	66.09	4.15
Alder sawdust	<.01	69.25	95.02	0.56
Alfalfa	20.29	29.45	38.65	10.12

EXPERIMENT 3

TABLE 3. MEANS (±SE) OF NUTRIENT DIGESTIBILITIES (%) AND GROWTH PERFORMANCE DATA OF WEANLING RABBITS FED DIETS CONTAINING 25% ALFALFA AND 25% BLACK LOCUST BARK, OAK SAWDUST, ALDER BARK AND ALDER SAWDUST OR 50% ALFALFA CONTROL OR STANDARD FRYER DIET.

Treatments	Crude Protein	ADF	NDF	Dry Matter	Ash	DM Intake (g)	ADG (g)	Feed/ Gain
1. Black Locust Bark	66.94 ±1.95	21.91 ±1.78	29.47 ±1.05	63.73 ±0.85	54.07 ±1.91	53.0 ±3.6	4.4 ±1.4	20.26 ±2.10
2. Oak Sawdust	75.88* ±1.54	2.24 ±1.41	54.37 ±0.83	55.92 ±0.68	62.66 ±1.51	88.9 ±2.9	18.1 ±1.1	5.51 ±1.66
3. Alder Bark	58.81* ±1.54	-9.46 ±1.41	58.29 ±0.83	57.22 ±0.68	57.98 ±1.51	80.1 ±2.9	13.3 ±1.1	6.70 ±1.66
4. Alder Sawdust	54.15* ±1.54	22.10 ±1.41	58.05 ±0.83	56.08 ±0.68	53.59 ±1.51	80.2 ±2.9	10.3 ±1.1	8.72 ±1.66
5. Alfalfa Control	69.15 ±1.54	9.37 ±1.41	21.08 ±0.83	66.29 ±0.68	64.72 ±1.51	91.1 ±2.9	26.9 ±1.1	3.77 ±1.66
6. Standard Fryer Diet	NA	NA	NA	NA	NA	117.6 ±2.9	40.3 ±1.1	3.18 ±1.66
Average of 1, 2, 3, and 4	63.95 ±1.64	9.20 ±1.50	50.05 ±0.89	58.24 ±0.72	57.08 ±1.61	75.6 ±3.1	11.5 ±1.2	10.30 ±1.77

NA: Not Applicable (Treatment 6 served only as a reference for overall growth performance and was not included in nutrient digestibility analyses.)

* The protein digestibility values for these diets reflect the digestibilities of the protein in alfalfa and barley due to the very low values of protein in oak sawdust, alder bark and alder sawdust.

EXPERIMENT 3

TABLE 4. STATISTICAL COMPARISONS OF DIGESTIBILITIES (%) AND GROWTH PERFORMANCE DATA OF WEANLING RABBITS FED ALFALFA CONTROL OR DIETS CONTAINING BLACK LOCUST BARK, OAK SAWDUST, ALDER BARK AND ALDER SAWDUST.

Contrasts	Crude Protein	ADF	NDF	Dry Matter	Ash	DM Intake (g)	ADG (g)	Feed/ Gain
1. Treatments vs Alfalfa Control (Groups 1 2 3 4 vs 5)	63.95 vs 69.15*	9.20 vs 9.37	50.05 vs 21.08*	58.24 vs 66.29*	57.08 vs 64.72*	75.6 vs 91.1*	11.5 vs 26.9*	10.30 vs 3.77*
2. BLB Bark vs Alder Bark (Groups 1 vs 3)	66.94 vs 58.81*	21.91 vs -9.46*	29.47 vs 58.29*	63.73 vs 57.22*	54.07 vs 57.98	53.0 vs 80.1*	4.4 vs 13.3*	20.26 vs 6.70*
3. Alder Bark vs Alder Sawdust (Groups 3 vs 4)	58.81 vs 54.15*	-9.46 vs 22.10*	58.29 vs 58.05	57.22 vs 56.08	57.98 vs 53.59*	80.1 vs 80.2	13.3 vs 10.3*	6.70 vs 8.72
4. Oak Sawdust vs Alder Sawdust (Groups 2 vs 4)	75.88 vs 54.15*	2.24 vs 22.10*	54.37 vs 58.05*	55.92 vs 56.08	62.66 vs 53.59*	88.9 vs 80.2*	18.1 vs 10.3*	5.51 vs 8.72

* indicates statistical significance at .05 level

References

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Anonymous, 1979. Hardwood sawdust into milk and meat. Aust. For. Ind. J. Aust. Log. 45:68-70.

Ayers, A.C., P.R. Cheeke and N.M. Patton. 1992. Effect on weanling rabbits of the addition of polyethylene glycol, phytase, methionine and choline chloride to diets containing black locust (*Robinia pseudoacacia*) leaf meal. J. Appl. Rabbit Res. (in press).

Cheeke, P.R. 1992. Black locust forage as an animal feedstuff. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp.252-258.

Cheeke, P.R., N.M. Patton, S.D. Lukefahr and J.I. McNitt. 1987. Rabbit Production. The Interstate Printers & Publishers, Inc. Danville, IL p. 156.

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Inc. Westport, CT. pp 240-244.

El-Sabban, F.F., T.A. Long and B.R. Baumgardt. 1970. Utilization of oak sawdust as a roughage substitute in beef cattle finishing rations. J. Anim. Sci. 32:749-755.

Fayek, H.M., Y.A. Mady, A.A. Abdel-Aziz and H.M. El-Hosseny. 1989. Urea treated sawdust in rabbit diets. J. Appl. Rabbit Res. 12:185-187.

Feininger, A. 1968. Trees. The Viking Press. New York, NY. pp 65-66.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Nsimba-Lubaki, M. and W.J. Peumans. 1986. Seasonal fluctuations of lectins in barks of elderberry (*Sambucus nigra*) and black locust (*Robinia* *pseudoacacia*). Plant. Physiol. 80:747-751.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc. Car, NC.

Tengerdy, R.P. and J.G. Nagy. 1988. Increasing the feed value of forestry waste by ammonia freeze explosion treatment. Biological Wastes 25:149-153.

Vidal, P.F. and J. Molinier. 1988. Ozonolysis of lignin - improvement of *in vitro* digestibility of poplar sawdust. Biomass 16:1-17.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67- 69.

Whitten, J.E. and A.J. Whitten. 1987. Analysis of bark eating in a tropical squirrel. Biotropica 19:107-115.

EVALUATION OF HYBRID POPLAR LEAVES AS A FEEDSTUFF FOR SHEEP

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Abstract

Ten crossbred mature ewes were fed diets containing 50% hybrid poplar leaves or 50% alfalfa for an 8-day adaptation and a 6-day digestibility period. Nutrient digestibilties, crude protein, ADF, NDF, dry matter and ash were lower ($p < .01$) for the poplar leaf meal (PLM) diet than for the alfalfa diet. There was no difference in nitrogen (N) retention as a percent of N digested between the treatments. The lower digestibility of the PLM diet is probably due to the presence of tannins. These results indicate that PLM is considerably lower in feeding value for sheep than alfalfa, primarily due to low protein digestibility.

Introduction

In the Pacific Northwest of the United States, plantations of hybrid poplars for pulp and paper manufacturing have been established. The foliage could be collected for use as a feedstuff when the trees are harvested. In developing countries which often suffer from severe deforestation, tree forage may have a broader use in agroforestry systems designed to use multiple purpose trees for improving soil conditions, crop interaction and as animal feed (Benge, 1987).

The family *Salicaceae*, poplars and willows, has a wide range of temperature tolerance and grows in cold parts of the northern hemisphere from the Arctic circle to latitude 30° (FAO, 1980). Aspen, *Populus tremuloides*, is the most widespread tree species in North America (Bas et al., 1985). Poplar species, known in the U.S. as cottonwood, are dioecious woody plants. The genus *Populus* is characterized by an ability to resprout vigorously after browsing and spreads quickly by root suckers (Palo, 1984). It can also be vegetatively propagated by cuttings (Benge, 1987).

Poplar leaves have been found to be of intermediate feeding value for cattle (Forwood and Owensby, 1985; Baertsche et al., 1986). Hybrid poplar leaves were found to be a satisfactory replacement for alfalfa in the diets of growing rabbits (Ayers, 1992).

The objective of this study was to evaluate the feeding value of hybrid poplar leaf meal compared to alfalfa in a diet for mature sheep.

Materials and Methods

Ten Suffolk x Hampshire ewes were randomly assigned to each of two treatments: poplar leaf meal (PLM) diet or alfalfa diet (control). The diets contained 50% either alfalfa or PLM (Table 1). The trial consisted of a 8-day adaptation period followed by a 6-day digestibility period. The sheep were housed in wooden metabolism crates in a barn with constant lighting.

During the adaptation period, free consumption of hay was allowed while the amount of diet offered was gradually increased to the full amount two days before the beginning of the digestibility period. In the two days before the start of the digestibility period, no hay was given. During the adaptation period, two sheep on the PLM diet would not consume the diet in adequate amounts and were switched to the alfalfa diet. During the digestibility period, the sheep were offered 2 kg of feed a day of the experimental diets or approximately 3% of body weight. Free access to water was allowed. The amount of feed offered each day was measured and spillage from the feeders was taken into account. The sheep left no orts, but it was not feasible to give them more because of a limited amount of poplar leaf meal. The poplar leaves were harvested at a local tree farm [5], from 1 and 2-year-old trees. Branches were cut from the trees and air-dried on screens, and the leaves stripped off and ground to pass through a 2 mm screen.

The total output of urine was collected each day and placed in airtight containers. 1N solution of sulfuric acid was added to urine collecting vessels to prevent volatilization of ammonia N. The total output of feces was collected each day, placed in plastic bags and frozen.

After the end of the digestibility period, total feces were weighed and a subsample was oven dried at 60° C for 48 hours. The sample was ground to pass through a 1 mm screen. Total urine was weighed and a subsample frozen until further analysis. Feed and feces samples were analyzed for DM and ash by standard procedures (AOAC, 1984). Samples were analyzed for ADF and NDF as described by Goering and Van Soest (1970), modified by a micro method (Waldern, 1971). Crude protein was determined by the Macro Kjeldahl method (AOAC, 1984). The same method was used for determining nitrogen content in the urine for calculating nitrogen retention.

The data were analyzed following the general linear model procedure (SAS, 1985). One sheep on the poplar diet was removed from the trial due to illness.

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Results and Discussion

The nutrient compositions of the alfalfa and poplar leaf meal are compared in Table 2. A statistical comparison of the digestibility and nitrogen retention data is given in Table 3.

All of the nutrient digestibilities, consisting of crude protein (CP), ADF, NDF, dry matter (DM) and ash, were higher for the alfalfa than the poplar leaf meal (PLM) diet (all p values $< .01$). The N retention as a percentage of digested N did not differ for the two treatments, indicating that the poorer utilization of PLM diet as compared to alfalfa was not due to a metabolic problem, but rather to digestive or absorptive interference. When N retention was considered as a percentage of N consumed, it was significantly lower for the PLM diet than for the alfalfa diet, indicating that the N consumed was less efficiently digested ($p < .01$).

The lower nutrient digestibilities of the PLM diet compared to the alfalfa diet were probably due to the presence of anti-nutritive phenols in the poplar leaves, such as tannins. Meyer and Montgomery (1987) found that the leaves of *Populus deltoides* contained 3.6 to 5.0% condensed tannins. According to Palo (1984), phenolics are the only chemical class of secondary compounds that are found to occur in the family *Salicaceae*. Poplar leaves are known to contain tannins (Meyer and Montgomery, 1987). Tannins bind to dietary proteins and enzymatic proteins to reduce protein digestibility (Cheeke and Shull, 1985).

Bas et al. (1985) fed pelleted foliage of aspen, a *Populus* species, to lambs at levels of 0, 25, 50 and 75% with alfalfa as the other ingredient, and found lamb weight and feed intake to decrease as the level of aspen increased. Aspen, however, is considered to an excellent forage of wildlife such as elk (Canon et al., 1987).

In a comparison of the utilization of PLM by sheep and rabbits, the CP digestibility of diets containing approximately one-half PLM was similar for rabbits and sheep (Table 4). In a prior study (Ayers et al., 1992), the CP digestibility of a 40% PLM diet was 54.76% for growing rabbits, or 70% that of the CP digestibility for an alfalfa control diet. In the present study, the CP digestibility of a 50% PLM diet was 55.44% for mature sheep or 66% of the CP digestibility for the alfalfa control diet. While the NDF fraction of the PLM diet was poorly digested in the rabbit (18.79%), it was better digested in the sheep (41.12%), which was 76% of the digestibility of alfalfa diet. The ADF digestibility of PLM diet in the rabbit was only 10.15% but was twice that for the sheep (24.84%). This probably reflects the sheep's ability, as a ruminant, to digest the fiber portion of the feed to a much greater degree than the rabbit. The rabbit selectively excretes fiber and retains the nonfiber portion of feed in the cecum for fermentation (Cheeke, 1991).

An inconsistency is represented in the fact that all of the measured digestibilities are lower for the PLM diets in both studies, yet the DM digestibilities of the PLM diets reach 90% and 89% of the alfalfa diet in the rabbit and sheep studies, respectively. Such high DM digestibilities of the PLM diets would be explained by high digestibilities of the unmeasured ether extract and nitrogen-free extract containing readily available carbohydrates, but there is no reason to assume that these unmeasured digestibilities would be higher for the PLM diets than for the alfalfa diets.

In the rabbit study the PLM was found to be a satisfactory replacement of alfalfa in terms of growth rate. Even though the nutrient digestibilities were lower for the PLM diet, the DM intake of the PLM diet increased sufficiently to allow the average daily weight gain at a level equal to the alfalfa diet. The efficiency of feed utilization was somewhat reduced, although not significantly, because of the higher intake of PLM diet needed to support growth equal to that with alfalfa.

It is concluded that PLM is not an equal replacement of alfalfa in sheep diets. In this study, weight gain was not measured and there was not enough PLM material to allow ad libitum feeding which could have allowed the possibility of an increase in DM intake. Further studies are recommended, measuring changes in weight and offering feed ad libitum, in order to determine if PLM could be a satisfactory replacement of alfalfa in sheep diets.

EXPERIMENT 4

TABLE 1. COMPOSITION AND CHEMICAL ANALYSIS (%) OF DIETS FED TO SHEEP (DRY MATTER BASIS)

	Alfalfa Diet	Poplar Diet
Alfalfa, sun-cured	50	-
Ground barley	21.5	21.5
Molasses	5	5
Poplar leaves	-	50
Trace mineralized salt	0.5	0.5
Vegetable oil	3.0	3.0
Wheat mill run	20	20
Chemical Analysis		
Crude protein	16.8	11.9
ADF	20.4	19.8
NDF	34.7	32.0
Ash	8.7	8.7

EXPERIMENT 4

TABLE 2. NUTRIENT COMPOSITION (%) OF EXPERIMENTAL FEED INGREDIENTS (DRY MATTER BASIS)

	Crude Protein	ADF	NDF	Ash
Poplar leaves	11.4	29.45	36.39	9.88
Alfalfa	20.3	26.02	38.65	10.12

EXPERIMENT 4

TABLE 3. MEANS (±SE) OF NUTRIENT DIGESTIBILITIES OF DIETS CONTAINING 50% ALFALFA OR 50% POPLAR LEAF MEAL FED TO SHEEP

	Crude Protein	ADF	NDF	Dry Matter	Ash	N retained as % of N digested	N retained as % of N consumed
Alfalfa Diet	84.25[a] ±1.39	51.74[a] ±3.07	54.12[a] ±2.63	75.48[a] ±0.97	62.64[a] ±1.77	51.97[a] ±2.86	43.74[a] ±2.36
Poplar Diet	55.44[b] ±1.56	24.84[b] ±3.43	41.12[b] ±2.95	67.02[b] ±1.09	51.00[b] ±1.97	52.99[a] ±3.20	29.56[b] ±2.64

Superscripts with different letters in the same column differ at the .05 level.

EXPERIMENT 4

TABLE 4. NUTRIENT DIGESTIBILITIES (%) AND THE COMPARATIVE EFFECTIVENESS OF REPLACING ALFALFA WITH POPLAR LEAF MEAL (PLM) IN THE DIETS OF GROWING RABBITS AND MATURE SHEEP.

	Crude Protein	ADF	NDF	Dry Matter	Ash
Rabbit*					
40% PLM	54.76	10.15	18.79	57.63	44.73
40% Alfalfa	78.75	26.28	36.87	64.35	60.62
Replacement Efficacy	70%	39%	51%	90%	74%
Sheep					
50% PLM	55.44	24.84	41.12	67.02	51.00
50% Alfalfa	84.25	51.74	54.12	75.48	62.64
Replacement Efficacy	66%	48%	76%	89%	81%

*Rabbit data adapted from Ayers, 1992.

References

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Ayers, A.C., P.R. Cheeke and N.M. Patton. 1992. Evaluation of hybrid poplar leaves as a feedstuff for rabbits. J. Appl. Rabbit Res. (in press).

Baertsche, S.R., M.T. Yokoyama and J.W. Hanover. 1986. Short rotation, hardwood tree biomass as potential ruminant feed - chemical composition, nylon bag ruminal degradation and ensilement of selected species. J. Anim. Sci. 63:2028-2043.

Bas, F.J., F.R. Ehle and R.D. Goodrich. 1985. Evaluation of pelleted aspen foliage as a ruminant feedstuff. J. Anim. Sci. 61:1030-1036.

Benge, M.D. 1987. Multipurpose uses of contour hedgerows in highland regions. World Anim. Rev. 64:31-39.

Canon, S.K., P.J. Urness and N.V. Debyle. 1987. Habitat selection, foraging behavior, and dietary nutrition of elk in burned aspen forest. J. Range Manage. 40:433-438.

Cheeke, P.R. 1991. Applied Animal Nutrition: Feeds and Feeding. Macmillan Pub. Co. New York. p. 16

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Inc. Westport, CT. p. 333.

FAO. 1980. Poplars and Willows: in wood production and land use. Food and Agriculture Organization of the United Nations. pp. 12-13.

Forwood, J.R. and C.E. Owensby. 1985. Nutritive value of tree leaves in the Kansas Flint Hills. J. Range. Manage. 38:61- 64.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Meyer, G.A. and M.E. Montgomery. 1987. Relationships between leaf age and the food quality of cottonwood foliage for the gypsy moth, *Lymantria dispar*. Oecologia 72:527-532.

Palo, R.T. 1984. Distribution of birch (*Betula* spp.), willow (*Salix* spp.), and poplar (*Populus* spp) secondary metabolites and their potential role as chemical defense against herbivores. J. Chem. Ecology 10:499-520.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc. Cary, NC.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67- 69.

A COMPARISON OF THE DIGESTIBILITY OF BLACK LOCUST (*ROBINIA PSEUDOACACIA*) LEAVES BY SHEEP AND GOATS

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Abstract

Four crossbred mature sheep and four mature Angora goats were fed diets composed solely of black locust (*Robinia pseudoacacia*) leaves or alfalfa (control). It was hypothesized that goats might be better able to digest black locust (BL) leaves, by binding tannins with proline-rich salivary proteins. A 2 x 2 Latin square design was used in which the treatment assigned to an animal in the first replicate was switched in the second. Following a 7-day digestibility period, the digestibilities of crude protein (CP), ADF, NDF, dry matter (DM) and ash were measured. The nutrient digestibilities were higher for alfalfa than for black locust (BL) leaves with both species (all p values $< .01$). This was probably due to tannins in the BL leaves. There were no differences between sheep or goats in terms of digestibility of alfalfa or BL leaves.

Introduction

Black locust (*Robinina pseudoacacia*) is a fast-growing, leguminous tree; its leaves can be harvested for animal feed. Black locust is intensively cultivated in Eastern Europe for sawlogs (particularly a variety called "shipmast locust"), poles and posts, beekeeping and decorative planting (Keresztesi, 1983). Since 1980, increased attention has been given to black locust in the U.S. for reasons including the focusing on trees capable of rapid growth and high-biomass production due to the energy crisis, success in other countries and biological research potential (Hanover, 1992).

Black locust (BL) is indigenous to the eastern U.S. It can grow as fast as 2 feet per year and may reach a height of 70 feet. It is very adaptable to soils and climate and has the ability to fix atmospheric nitrogen (Dirr, 1990).

Although BL leaves are similar to alfalfa meal in nutrient composition (Cheeke, 1992), feeding trials with BL leaf meal have generally given poor animal performance (Horton and Christensen, 1981; Harris et al., 1984; Raharjo et al., 1990). The low nutrient digestibilties are probably due to tannins, phenolic compounds that bind dietary protein and digestive enzymes (Kumar and Singh,

1984).

The saliva of mule deer, a browser, has more proline rich protein (PRP) and more tannin-binding ability than the saliva of sheep and cattle, both grazers (Austin et al., 1989). It has been proposed that PRPs may provide the first line of defense against tannins (Mehansho et al., 1987). We theorize that goats, being browsers, may also have PRPs in their saliva and be more equipped to tolerate the tannins in BL leaves.

The objectives of this study are to evaluate the feeding value of BL leaves for mature sheep and goats as compared to alfalfa, and to determine if the goats are able to better digest the nutrients in BL leaves than the sheep.

Materials and Methods

Four crossbred sheep and 4 Angora goats were randomly assigned to diets composed solely of black locust leaves or alfalfa (control). The experimental design was a 2 x 2 Latin Square and was carried out in 2 replicates in which the treatments to which the animals were assigned in the first replicate were switched in the second.

In the first replicate, during the 6-day adaptation period, free consumption of hay was allowed with increasing amounts of experimental forage. A 7-day digestibility period followed during which the animals were housed in a barn in wooden metabolism crates with free access to water. During the three weeks between replicates, the animals were housed in common pens and fed alfalfa, barley and hay. Trace mineralized salt was offered. In the 3 days before the start of the second replicate, only alfalfa was given. In the second set, there was no adaptation period since the animals were already accustomed to 100% forage diet. The second replicate consisted of a 7-day digestibility period during which the animals were returned to the metabolism crates.

During the digestibility period, sheep were offered 2200 g of alfalfa per day (2.6% of body weight) or 2000 g of BL leaves per day (2.3 % of body weight). Goats were offered 1100 g of alfalfa per day (2.8% of body weight) or 1000 g of BL leaves per day (2.6% of body weight). The BL leaves were from a forage plantation at Michigan State University. The leaves were air-dried and offered to the animals in a coarsely chopped form. Alfalfa was finely chopped. Orts were collected daily and pooled in plastic bags. During the digestibility period, total feces were collected and frozen.

After the end of the digestibility period, total feces were weighed and a subsample was oven dried at 60 ° C for

48 hours. The sample was ground to pass through a 1 mm screen. Feed and feces samples were analyzed for DM and ash by standard procedures (AOAC, 1984). Samples were analyzed for ADF and NDF as described by Goering and Van Soest (1970), modified by a micro method (Waldern, 1971). Crude protein was determined by the Macro Kjeldahl method (AOAC, 1984).

The data were analyzed following the general linear model procedure (SAS, 1985). Treatment means were compared using a series of orthogonal contrasts from three pre-planned comparisons.

Results and Discussion

The nutrient composition of the alfalfa and the black locust (BL) leaves are presented in Table 1. The nutrient digestibility and DM intake are given in Table 2 and statistical comparisons in Table 3. The comparisons are 1) alfalfa vs. BL leaves (across species) 2) sheep vs. goats on BL leaf diet and 3) sheep vs. goats on alfalfa diet.

All of the nutrient digestibilities crude protein (CP), ADF, NDF, dry matter (DM) and ash were lower ($p < .01$) for the BL when compared to the alfalfa across both species (Comparison 1). This is probably due to the presence of tannins in the BL leaves. Across the species, more alfalfa was consumed than BL leaves ($p < .01$).

There was no difference in the utilization of alfalfa by sheep or goats (Comparison 3). Based on kilogram of approximate body weight, the goats consumed more alfalfa than the sheep ($p < .01$).

There was no difference in the utilization of BL leaves by sheep or goat (Comparison 2). The goats consumed more BL leaves than the sheep ($p < .01$) per kilogram of body weight.

The lower nutrient digestibilities of the BL leaves compared to the alfalfa was probably due to the presence of tannins. Tannins are phenolic compounds which precipitate dietary proteins and digestive enzymes by hydrogen-bonding and hydrophobic interactions, thus lowering nutrient digestibilities (Kumar and Singh, 1984). The leaves of BL contain primarily tannins of the condensed type (Kumar and Horigome, 1986). The leaves contain about 5-7% condensed tannins (B. Luick, 1992, personal communication, University of Alaska).

Horton and Christensen (1981) found that when lambs were fed a diet of 100% BL leaf meal, digestibilities of OM, CP, ADF, NDF and P were less than half those of alfalfa. Although no significant difference were evident in ADF or DM intake between lambs fed BL leaves of alfalfa in the 28-day trial, it was concluded that BL leaves were not a satisfactory replacement of alfalfa.

On a high-quality diet, subterraneum clover, Doyle et al. (1984), found that sheep and goats did not differ in intake or digestion of organic matter (OM), but the sheep showed better N retention. On a low-quality, high-fiber diet, goats digested more OM due to a more extensive digestion of the fiber fraction. The goat rumen had a higher concentration of cellulolytic bacteria and a longer digesta retention time than the sheep rumen. The goats were more efficient at conserving N on poor diets.

In a review, Brown and Johnson (1984) concluded that there is no difference between sheep and goats in terms of forage digestibility.

Nuñez-Hernandez et al. (1991) found that sheep consumed more OM (per kg body weight) than goats when mountain mahogany leaves comprised 25% or 50% of the diet, but OM digestibility did not differ between species. Sheep digested NDF to a greater extent than goats.

It was not possible to calculate N retention in the present study and the literature is not consistent in terms of N retention between sheep and goat species. Nuñez-Hernandez et al. (1991) found that goats had greater N digestibility, but N balance and urinary N did not differ between the species. As mentioned above, Doyle et al. (1984) found that goats were more efficient than sheep at conserving N of poor quality diets. It has been proposed by Nuñez-Hernandez et al. (1991) that condensed tannins in a low-quality diet could increase efficiency of the use of absorbed N in sheep and goats. Some of the protein-tannin complexes prevent N absorption, and less N is excreted in the urine because of low ruminal ammonia concentration.

It is interesting to note that goats have been reported to have a tannase in the rumen mucosa which lowered the toxicity level of tannic acid to 8-10% when added directly to the rumen as compared to 3-5% for cattle (Kumar and Singh, 1984).

Proline-rich proteins (PRP) in the saliva may provide the first line of defense against tannins in the diet (Mehansho et al., 1987). Mehansho et al. (1987) found that feeding a tannin-rich diet to rats enlarged the parotid glands four-fold and increased the production of PRPs. The rats gained weight at the normal rate. According to Austin et al. (1989) the saliva of mule deer, a browser, has more PRP than the saliva of cows or sheep, grazers, and also has more tannin-binding ability. We theorized that goats, being browsers, would be more equipped to tolerate the tannins in BL leaves than the sheep; however, in our study, no differences were found in the nutrient digestibilities between the two species. Perhaps the experimental period was too short for induction of increased PRP production.

In a prior study with weanling rabbits (Ayers et al., 1992), the replacement of alfalfa with only 25% BL leaves of the diet gave CP digestibilities 76% of those attained with alfalfa. When 100% of the alfalfa was replaced with BL leaves for sheep and goats, the efficacy in terms of CP digestibility was 48% and 54%, respectively. This reflects the fact that the effects of tannins may be less severe in ruminants than nonruminants (M). Beneficial effects may include by-pass protein and bloat suppression (Kumar and Singh, 1984). Condensed tannins can inhibit rumen microbes (Nuñez-Hernandez et al., 1991).

It is concluded than BL leaves do not appear to be a satisfactory replacement of alfalfa for sheep or goats due to low nutrient digestibilties. Goat saliva should be examined for the tannin-binding PRP activity. Goats, as browsers, are likely to have PRP in their saliva and thus be better equipped than sheep to tolerate tannins in the diet; however, our study showed no difference between these species in terms of digestibility.

EXPERIMENT 5

TABLE 1. NUTRIENT COMPOSITION (%) OF EXPERIMENTAL DIETS FED TO SHEEP AND GOATS (DRY MATTER BASIS)

	Crude Protein	ADF	NDF	Ash
Alfalfa	18.7	25.0	35.9	10.0
Black locust	17.8	29.8	48.1	6.5

EXPERIMENT 5

TABLE 2. NUTRIENT DIGESTIBILITIES (MEANS ±SE) AND AVERAGE DM INTAKE/ KG OF BODY WEIGHT OF ALFALFA (A) OR BLACK LOCUST LEAF (BL) DIETS FED TO SHEEP AND GOATS.

	Crude Protein	ADF	NDF	Dry Matter	Ash	Daily Intake/kg BW	Daily Intake (g)
Sheep A	78.44	29.68	28.94	62.14	49.89	.0214	1838
Sheep BL	37.56	-15.05	5.81	45.50	33.04	.0159	1371
Goats A	79.22	31.15	28.83	61.57	50.84	.0253	985
Goats BL	42.60	-28.62	-0.11	43.47	33.05	.0201	783
(SE)	(±3.45)	(±7.91)	(±6.57)	(±3.28)	(±4.76)	(±.0067)	
Ave A (Sheep and Goats)	78.83	30.42	28.89	61.86	50.37	.0341	
Ave BL (Sheep and Goats)	40.08	-21.84	2.85	44.49	33.05	.0180	

EXPERIMENT 5

TABLE 3. STATISTICAL SIGNIFICANCE OF CONTRASTS BETWEEN NUTRIENT DIGESTIBILITY MEANS AND AVERAGE DM INTAKE/ KG OF BODY WEIGHT OF ALFALFA OR BLACK LOCUST LEAF DIETS FED TO SHEEP AND GOATS.

Contrast	Crude Protein	ADF	NDF	Dry Matter	Ash	Daily Intake/ kg BW
1. Alfalfa vs Black Locust (across species)	78.83 vs 40.08*	30.42 vs -21.84*	28.89 vs 2.85*	61.86 vs 44.49*	50.37 vs 33.05*	.0341 vs .0180*
2. Sheep vs Goats on Black Locust Leaf diet	37.56 vs 42.60	-15.05 vs -28.62	5.81 vs -0.11	45.50 vs 43.47	33.04 vs 33.05	.0159 vs .0201*
3. Sheep vs Goat on Alfalfa diet	78.44 vs 79.22	29.68 vs 31.15	28.94 vs 28.83	62.14 vs 61.57	49.89 vs 50.84	.0214 vs .0253*

* indicates statistical significance at the .05 level.

References

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Austin, P.J., L.A. Suchar, C.T. Robbins and A.E. Hagerman. 1989. Tannin-binding proteins in saliva of deer and their absence in saliva of sheep and cattle. J. Chem. Ecol. 15:1335-1347.

Ayers, A.C., P.R. Cheeke and N.M. Patton. 1992. Effect on weanling rabbits of the addition of polyethylene glycol, phytase, methionine and choline chloride to diets containing black locust (*Robinina pseudoacacia*) leaf meal. J. Appl. Rabbit Res. (in press).

Brown, L.E. and W.L. Johnson. 1984. Comparative intake and digestibility of forages and byproducts by goats and sheep. A review. Int. Sheep Goat Res. 2:212.

Cheeke, P.R. 1992. Black locust forage as an animal feedstuff. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp.252-258.

Doyle, P.T., Egan, J.K. and A.J. Thalen. 1984. Intake, digestion, and nitrogen and sulfur retention in angora goats and merino sheep fed herbage diets. Aust. J. Exp. Agric. Anim. Husb. 24:165-169.

Dirr, M.A. 1990. Manual of Woody Landscape Plants: Their Identification, Ornamental Characteristics, Culture, Propagation and Uses. Stipes Pub. Co., Champaign, IL. pp. 753-756.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Hanover, J.W. 1992. Black locust: an historical and future perspective. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp.7-18.

Harris, D.J. P.R. Cheeke and N.M. Patton. 1984. Evaluation of black locust leaves for growing rabbits. J. Appl. Rabbit Res. 7:7-9.

Horton, G.M.J. and D.A. Christensen 1981. Nutritional value of black locust leaf meal (*Robinina pseudoacacia*) and alfalfa meal. Can. J. Anim. Sci. 61:503-506.

Keresztesi, B. 1983. Breeding and cultivation of black locust, *Robinia pseudoacacia*, in Hungary. Forest Ecol. and Manage. 6:217-244.

Kumar, R. and T. Horigome. 1986. Fractionation, characterization and protein-precipitating capacity of the condensed tannins from *Robinia pseudoacacia L.* leaves. J. Agric. Food Chem. 34:487-489.

Kumar, R. and M. Singh. 1984. Tannins: their adverse role in ruminant nutrition. J. Agric. Food Chem. 32:447-453.

Mehansho, H., L.G. Butler and D.M. Carlson. 1987. Dietary tannins and salivary proline-rich proteins: interactions, induction, and defense mechanisms. Ann. Rev. Nutr. 7:423- 40.

Nuñez-Hernandez, G., J.D. Wallace, J.L. Holechek, M.L. Galyean and M. Cardenas. 1991. Condensed tannins and nutrient utilization by lambs and goats fed low-quality diets. J. Anim. Sci. 69:1167-1177.

Raharjo, Y.C., P.R. Cheeke and N.M. Patton. 1990. Effect of cecotrophy on the nutrient digestibility of alfalfa and black locust leaves. J. Appl. Rabbit Res. 13:56-61.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc., Cary, NC.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67- 69.

CONCLUSIONS

Hybrid poplar leaves have potential as a feedstuff for rabbits at a level of at least 40% of the diet, if the leaves are economically available and if a slightly less efficient feed/gain ratio is acceptable compared to alfalfa. The literature suggests that leaves from coppiced sources are more likely to contain higher chemical defenses and to be of less nutritional value, but our study indicated that the leaves from the coppiced source were more digestible than the uncoppiced with no indication of higher chemical defenses. It is concluded that hybrid poplar leaves may be harvested from any source without risk.

Hybrid poplar leaves at 50% of the diet had poor digestibilities compared to alfalfa for sheep. The use of additives such as polyethylene glycol (PEG) may be a means of improving the utilization of poplar leaves. Further work is necessary in order to conclusively identify tannins as the factor causing the poor animal performance.

Hybrid poplar leaves do have potential as multipurpose trees in agroforestry systems in temperate areas of the developing world where they could provide animal fodder as well as firewood, aid in erosion control, etc.

Untreated black locust leaves are of low feeding value for both sheep and goats. The goats, as browsers, may be able to better digest the leaves, but it was not evident in our study. Perhaps a longer experimental trial is needed to allow sufficient time for induction of adaptation mechanisms such as salivary tannin-binding proteins.

Black locust leaves have potential in animal feeding if the leaves are pre-treated to counteract the adverse affects of the tannins. Polyethylene glycol (PEG) appears to be an effective treatment.

In general, sawdust and bark are of little feeding value even for a hind-gut fermenter such as the rabbit. Black locust bark is very toxic to rabbits, probably due to lectins, and harvesting of the leaves should be done to minimize the inclusion of bark from stems.

Black locust, a fast-growing, nitrogen-fixing tree, has potential, as evidenced by the research which has been focused on black locust plantations for timber, forage and energy, as a multipurpose tree from which the leaves could be harvested for animal fodder. Processing methods which include the addition of feed additives, such as PEG, will probably be necessary.

RECOMMENDATIONS FOR FURTHER RESEARCH

A feeding trial should be carried out with coppiced and uncoppiced sources of hybrid poplar leaves early in the season to avoid the confounding effects of young vs old leaves.

Feeding trials should be conducted with hybrid poplar leaves with leaves that have been treated to counteract tannins.

Feeding trials should be done with black locust leaves treated with varying amounts of PEG to determine the appropriate amount needed to reach a feeding value closer to alfalfa.

Feeding trials should be performed with black locust treated with polyvinylpyrrolidine (PVP) to aid in confirmation that tannins are the major anti-nutritive factor.

Goat saliva should be examined for PRP activity and a longer feeding trial carried out in which black locust leaves are fed to goats.

A feeding trial should be done with autoclaved black locust bark to inactivate the lectins by heat and determine if this reduces the toxicity of black locust bark.

BIBLIOGRAPHY

Anderson, G.W., R.W. Moore and P.J. Jenkins. 1988. The integration of pasture, livestock and widely-spaced pine in South Western Australia. Agroforestry Syst. 6:195-211.

Anonymous, 1979. Hardwood sawdust into milk and meat. Aust. For. Ind. J. Aust. Log. 45:68-70.

AOAC. 1984. Official Methods of Analysis (14th Ed.). Association of Official Analytical Chemists, Washington, DC.

Atsatt, P.R. and T. Ingram. 1983. Adaptation to oak and other fibrous, phenolic-rich foliage by a small mammal, *Neotoma fuscipes*. Oecologia 60:135-42.

Austin, P.J., L.A. Suchar, C.T. Robbins, A.E. Hagerman. 1989. Tannin-binding proteins in saliva of deer and their absence in saliva of sheep and cattle. J. Chem. Ecol. 15:1335-1347.

Ayers, A.C., P.R. Cheeke and N.M. Patton. 1992. Evaluation of hybrid poplar leaves as a feedstuff for rabbits. J. Appl. Rabbit Res. (in press).

Ayers, A.C., P.R. Cheeke and N.M. Patton. 1992. Effect on weanling rabbits of the addition of polyethylene glycol, phytase, methionine and choline chloride to diets containing black locust (*Robinia pseudoacacia*) leaf meal. J. Appl. Rabbit Res. (in press)

Ayers, G.S. 1992. Characteristics of black locust as bee forage. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.243-251.

Baertsche, S.R., M.T. Yokoyama and J.W. Hanover. 1986. Short rotation, hardwood tree biomass as potential ruminant feed - chemical composition, nylon bag ruminal degradation and ensilement of selected species. J. Anim. Sci. 63:2028-2043.

Baldelli, C. 1992. Black locust as a source of energy. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.237-243.

Barrett. R.P. 1992. Forage Plantation Management for black locust. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.259-272.

Bas, F.J., F.R. Ehle and R.D. Goodrich. 1985. Evaluation of pelleted aspen foliage as a ruminant feedstuff. J. Anim. Sci. Vol 61, No 5:1030-1036.

Basey, J.M., S.H. Jenkins and P.E. Busher. 1988. Optimal central-place foraging by beavers: Tree-size selection in relation to defensive chemicals of quaking aspen. Oecologia 76:278-282.

Beart, J.E., T.H. Lilley, E. Haslam. 1985. Plant polyphenols - secondary metabolism and chemical defence: some observations. Phytochemistry 24:33-38.

Benge, M.D. 1987. Multipurpose uses of contour hedgerows in highland regions. World Anim. Rev. 64:31-39.

Booth, A.N., M.S. Morse, D.J. Robbins, O.H. Emerson, F.T. Jones and F. Deeds. 1959. The metabolic fate of gallic acid and related compounds. J. Biol. Chem. 234:3014.

Bradshaw, H.D., J.B. Hollick, T.J. Parsons, H.R.G. Clark and M.P. Gordon. 1989. Systemically wound-responsive genes in poplar trees encode proteins similar to sweet potato sporamins and legume Kunitz trypsin inhibitors. Plant Molecular Biology 14:51-59.

Brown, L.E. and W.L. Johnson. 1984. Comparative intake and digestibility of forages and byproducts by goats and sheep. A review. Int. Sheep Goat Res. 2:212.

Canon, S.K., P.J. Urness, N.V. DeByle. 1987. Habitat selection, foraging behavior, and dietary nutrition of elk in burned aspen forest. J. Range Manage. 40:433-438.

Caufield, C. 1982. Tropical Moist Forests: the resource, the people, the threat. Russel Press Ltd., Nottingham, UK. 67p.

Cheeke, P.R. 1980. Editorial: The potential role of the rabbit in meeting world food needs. J. Appl. Rabbit Res. 3:3-4.

Cheeke, P.R. 1992. Black locust forage as an animal feedstuff. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.). pp 32-38.

Cheeke, P.R., M.P. Goeger, G.H. Arscott. 1983. Utilization of black locust (*Robinia pseudoacacia*) leaf meal by chicks. Nitrogen-Fixing Tree Res. Rep. 1:41.

Cheeke, P.R. and N.M. Patton. 1979. Utilization of alfalfa by rabbits. p. 25 in: The Domestic Rabbit: Potentials, Problems and Current Research. Published by OSU Rabbit Research Center.

Cheeke, P.R. and N.M. Patton. 1987. Rabbit production and research in China. J. Appl. Rabbit Res. 10:68-75.

Cheeke, P.R., N.M. Patton, S.D. Lukefahr, J.I. McNitt. 1987. Rabbit Production. The Interstate Printers and Publishers, Inc., Danville, IL 472p.

Cheeke, P.R. and L.R. Shull. 1985. Natural Toxicants in Feeds and Poisonous Plants. Avi Pub. Co., Westport, CT. 492p.

Clausen, T.P., P.B. Reichardt, J.P. Bryant, R.A. Werner, K. Post and K. Frisby. 1989. Chemical model for short-term induction in quaking aspen (*Populus tremuloides*) foliage against herbivores. J. Chem. Ecol. 15:2335-2346.

Dirr, M.A. 1990. Manual of Woody Landscape Plants: Their Identification, Ornamental Characteristics, Culture, Propagation and Uses. Stipes Publishing Co., Champaign, IL. pp 753-756.

Doyle, P.T., J.K. Egan and A.J. Thalen. 1984. Intake, digestion, and nitrogen and sulfur retention in angora goats and merino sheep fed herbage diets. Aust. J. Exp. Agric. Anim. Husb. 24:165-169.

Dula, J.C. 1982. *Newsletter*. No. 2-82. Canby. OR. Dula's Nurseries.

El-Sabban, F.F., T.A. Long and B.R. Baumgardt. 1970. Utilization of oak sawdust as a roughage substitute in beef cattle finishing rations. J. Anim. Sci. 32:749-755.

FAO. 1980. Poplars and Willows : in wood production and land use. Food and Agriculture Organization of the United Nations 1980. 328p.

Fayek, H.M., Y.A. Mady, A.A. Abdel-Aziz and H.M. El-Hosseny. 1989. Urea treated sawdust in rabbit diets. J. Appl. Rabbit Res. 12:185-187.

Feininger, A. 1968. Trees. The Viking Press. New York, NY. pp 65-66.

Fisher, L.J. 1980. An evaluation of steam-treated aspen as a substitute for corn silage in the rations of lactating cows. Can. J. Anim. Sci. 60:379-384.

Forwood, J.R. and C.E. Owensby 1985. Nutritive value of tree leaves in the Kansas Flint Hills. J. Range. Manage. 38:61- 64.

Freeland, W.J., P.H. Calcott, D.P. Geiss. 1985. Allelochemicals, minerals, and herbivore population size. Biochemical Systematics and Ecology 13:195-206

Fuller, H.L., S.I. Chang and D.K. Potter. 1967. Detoxification of dietary tannic acid by chicks. J. Nutr. 91:477-481.

Garrido, A., A. Gómez-Cabrera, J.E. Guerrero and J.M. van der Meer. 1991. Effects of treatment with polyvinylpyrrolidone and polyethylene glycol on Faba bean tannins. Anim. Feed Sci. Tech. 35:199-203.

Geyer, W.A. and D.F. Bresnan. 1992. Characteristics and uses of black locust for energy. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State Univ. Hanover, J.W. (Ed.). pp.221-236.

Goering, H.K. and P.J. Van Soest. 1970. Forage Fiber Analyses (apparatus, reagents, procedures, and some applications). Agric. Handbook 379. USDA, ARS, Washington, DC.

Hanover, J.W. 1992. Black locust: an historical and future perspective. Proc. Int. Conf. on Black Locust: Biology, Culture and Utilization. Dept. of Forestry, Michigan State University. Hanover, J.W. (Ed.) pp.7-28.

Harris, D.J., P.R. Cheeke and N.M. Patton. 1984. Evaluation of black locust leaves for growing rabbits. J. Appl. Rabbit Res. 7:7-9.

Haslam, E. 1974. Polyphenol-Protein Interaction. Biochem. J. 139:285-288.

Horigome, T., T. Ohkuma and M. Muta. 1984. Effect of condensed tannin of false acacia leaves on protein digestibility, as measured with rats. Jap. J. Zootech. Sci. 55:299-306.

Horigome, T., R. Kumar and K. Okamoto. 1988. Effects of condensed tannins prepared from leaves of fodder plants on digestive enzymes in vitro and in the intestine of rats. Brit. J. Nutr. 60:275-285.

Horton, G.M.J. and D.A. Christensen. 1981. Nutritional value of black locust tree leaf meal (*Robinia pseudoacacia*) and alfalfa meal. Can. J. Anim. Sci. 61:503-506.

Jakubas, W.J., G.W. Gullion and T.P. Clausen. 1989. Ruffed grouse feeding behavior and its relationship to secondary metabolites of quaking aspen flower buds. J. Chem. Ecol. 15:1899-1915.

Keresztesi, B. 1983. Breeding and cultivation of black locust, *Robinia pseudoacacia*, in Hungary. Forest Ecol. and Manage. 6:217-244.

Kramer, P.J. and T.T. Kozlowski. 1979. Physiology of woody plants. Academic Press, New York, p 811.

Kumar, R. and T. Horigome. 1986. Fractionation, characterization, and protein-precipitating capacity of the condensed tannins from *Robinina pseudoacacia L.* leaves. J. Agric. Food Chem. 34:487-489.

Kumar, R. and M. Singh. 1984. Tannins: their adverse role in ruminant nutrition. J. Agric. Food Chem. 32:447-53.

Kumar R. and S. Vaithiyanathan. 1979. Occurrence, nutritional significance and effect on animal productivity of tannins in tree leaves. Anim. Feed Sci. Tech. 30:21-38.

Le Roux, P.L. 1959. Sheep find thin honey-locust pods palatable. Farming in South Africa 18-19.

Liu, C.G. and Y. Jung. 1979. Nutritional value of locust (*Robinia pseudoacacia*) leaf meal. Unpublished report. Chinese Academy of Agricultural Sciences.

Lukefahr, S.D. and P.R. Cheeke. 1991. Rabbit project development strategies in subsistence farming systems. World Anim. Rev. 68:60-70.

McKell, C.M. 1980. Multiple use of fodder trees and shrubs - a world wide perspective. Proc. Int. Sym. on Browse in Africa: The Current State of Knowledge. Le Houérou, H.N. (Ed.). pp. 141-149.

McKey, D. 1979. The distribution of secondary compounds within plants. In: Rosenthal, G.A., Janzen, D.H. (eds) Herbivores - Their interactions with secondary plant metabolites. Academic Press, New York, pp 56-133.

Mehansho, H., L.G. Butler, D.M. Carlson,. 1987. Dietary tannins and salivary proline-rich proteins: interactions, induction, and defense mechanisms. Ann. Rev. Nutr. 7:423-40.

Merhav, M., Y. Amitai, H. Palti, S. Godfrey. 1985. Tea drinking and microcytic anemia in infants. Amer. J. Clin. Nutr. 41:1210-13.

Meyer, G.A. and M.E. Montgomery. 1987. Relationships between leaf age and the food quality of cottonwood foliage for the gypsy moth, *Lymantria dispar*. Oecologia 72: 527-532.

Myer, R.O., G.E. Combs and D.W. Gorbet. 1986. Effect of methionine addition on the feeding value of diets containing bird-resistant grain sorghum for growing-finishing swine. Nutr. Reports Int. 33:843-850.

National Academy of Sciences. 1983. Firewood Crops: Shrub and Tree Species for Energy Production, Vol. 2. National Academy Press, Washington, DC. 92p.

National Academy of Sciences. 1984. Leucaena. Promising Forage and Tree Crop for the Tropics. Washington, DC.

National Geographic Society. 1990. The Emerald Realm: Earth's Precious Rain Forests. Published by National Geographic Society. 196p.

National Research Council. 1983. Calliandra: A Versatile Small Tree for the Humid Tropics. National Academy Press. Washington, DC. 48p.

Nsimba-Lubaki, M. and W.J. Peumans. 1986. Seasonal fluctuations of lectins in barks of elderberry (*Sambucus nigra*) and black locust (*Robinia pseudoacacia*). Plant. Physiol. 80:747-751.

Nuñez-Hernandez, G., J.D. Wallace, J.L. Holechek, M.L. Galyean and M. Cardenas. 1991. Condensed tannins and nutrient utilization by lambs and goats fed low-quality diets. J. Anim. Sci. 69:1167-1177.

Oh, H.I., J.E. Hoff, G.S. Armstrong, and L.A. Haff. 1980. Hydrophobic interaction in tannin-protein complexes. J. Agric. Food Chem. 28:394-398.

Organización para Estudios Tropicales (OTS) y Centro Agronómico Tropical de Investigación y Enseñanza (CATIE). 1986. Sistemas Agroforestales: Principios y Aplicaciones en los Trópicos. Publicado por La Organización para Estudios Tropicales, Costa Rica. 818p.

Palo, R.T. 1984. Distribution of birch (*Betula* spp.), willow (*Salix* spp.), and poplar (*Populus* spp.) secondary metabolites and their role as chemical defense against herbivores. J. Chem. Ecol. 10:499-520.

Pointillart A. 1991. Enhancement of phosphorous utilization in growing pigs fed phytate-rich diets by using rye bran. J. Anim. Sci. 69:1109-1115.

Pritchard, D.A., D.C. Stocks, B.M. O'Sullivan, P.R. Martins, I.S. Hurwood and P.K. O'Rourke. 1988. The effect of polyethylene glycol (PEG) on wool growth and live weight of sheep consuming a Mulga (*Acacia aneura) diet. Proc. Aust. Soc. Anim. Prod. 17:290-293.*

Raharjo, Y.C., P.R. Cheeke and N.M. Patton. 1990. Effect of cecotrophy on the nutrient digestibility of alfalfa and black locust leaves. J. Appl. Rabbit Res. 13:56-61.

Salunkhe, D.K., J.K. Chavan and S.S. Kadam. 1990. Dietary Tannins: Consequences and Remedies. CRC Press, Boca Raton, FL. 200p.

SAS. 1985. SAS User's Guide: Statistics. SAS Inst., Inc., Cary, NC.

Singh, B. and S.S. Negi. 1986. Effect on wool production of supplementing black locust (*Robinia pseudoacacia*) and biul (*Grewia optiva*) leaves *ad libitum* to a concentrate diet of angora rabbits. J. Appl. Rabbit Res. 9:159-163.

Singleton, V.L. 1981. Naturally occurring food toxicants: phenolic substances of plant origen common in foods. In Advances in Food Research. Chichester, C.O. (Ed.). Academic Press, New York, NY. pp.149-242.

Smith, O.B. and M.F.J. van Houtert. 1987. The feeding value of *Gliricidia sepium*: a review. World Anim. Rev. 62:57-67.

Snook, L.C. 1986. Tagasaste, Tree Lucerne: High Production Fodder Crop. Night Owl Pub.Shepparton, Australia. p. 93.

Takada, K., T. Nakazato, K. Ono, H. Honda and T. Yamane. 1980. Feeding value of leaf meal of acacia on poultry feed. Jap. Poul. Sci. 17:299-305.

Tengerdy, R.P. and J.G. Nagy. 1988. Increasing the feed value of forestry waste by ammonia freeze explosion treatment. Biological Wastes 25:149-153.

Torres, F. 1983. Role of woody perennials in animal agroforestry. Agroforestry Sys. 1:131-163.

Van Soest, P.J. 1982. Nutritional Ecology of the Ruminant. O & B Books, Inc. Corvallis, OR.

Vidal, P.F. and J. Molinier. 1988. Ozonolysis of lignin - improvement of *in vitro* digestibility of poplar sawdust. Biomass 16:1-17.

Waldern, D.E. 1971. A rapid micro-digestion procedure for neutral and acid detergent fiber. Can. J. Anim. Sci. 51:67.

Whitten, J.E. and A.J. Whitten. 1987. Analysis of bark eating in a tropical squirrel. Biotropica 19:107-115.

World Resources Institute. 1985. Keep Tropical Forests Alive. Published by World Resources Institute, Washington, DC. 8p.