5-1977

Consumption, Digestion, and Utilization by Goats of the Dry Matter and Nitrogen in Diets Containing Oak (Quercus gambelii) Foliage

Anastasios Stefanos Nastis

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CONSUMPTION, DIGESTION, AND UTILIZATION BY GOATS OF THE 
DRY MATTER AND NITROGEN IN DIETS CONTAINING OAK 
(QUERCUS GAMBEII) FOLIAGE 
and 

ESTIMATION OF IN VIVO DIGESTIBILITY OF OAK-CONTAINING 
DIETS BY MICRO-DIGESTION TECHNIQUES 
by 
Anastasios Stefanos Nastis 

A thesis submitted in partial fulfillment of the requirements for the degree of 

MASTER OF SCIENCE 
in 
Range Science 

Approved: 

Major Professor 

Committee Member 

Committee Member 

Dean of Graduate School 

Committee Member 

UTAH STATE UNIVERSITY 
Logan, Utah 
1977
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I express appreciation to my parents who taught me in childhood the value of an education and to my wife, Stella, I express my appreciation for her encouragement and patience during all phases of this endeavor.

Anastasios Stefanos Nastis
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ABSTRACT

Consumption, Digestion, and Utilization by Goats of the Dry Matter and Nitrogen in Diets Containing Oak (Quercus gambelii) Foliage and Estimation of In Vivo Digestibility of Oak-Containing Diets by Micro-Digestion Techniques

by

Anastasios Stefanos Nastis, Master of Science
Utah State University, 1977

Major Professor: Dr. John C. Malechek
Department: Range Science

Part I

A study of animal performance was made with goats fed, ad libitum ground and pelleted mixtures of Gambel oak and alfalfa. Intake, digestibility, metabolizable nitrogen, and metabolized energy of oak diets were generally lower than those for a control alfalfa diet. However, no significant differences in live weight gains of experimental animals were found. No apparent toxicity was detected in goats fed diets containing up to 80 percent oak for 72 days.

Part II

Mixtures of oak (Quercus gambelii) and alfalfa (Medicago sativa) were used to evaluate three laboratory techniques, the Tilley and
Terry (1963) two-stage technique, the Van Soest et al. (1966) neutral detergent technique, and the Van Soest (1967) summative equation for their accuracy and precision to estimate in vivo digestibility. Additionally, the effects of inoculum donors' diet, oak phenology, and temperature of drying oak foliage were evaluated in terms of their independent and combined effects upon estimates of in vitro digestibility.

The Tilley and Terry (1963) acid pepsin digestion technique was the best indicator of in vivo digestibility \( (r^2 = 0.97) \), followed by the Van Soest et al. (1966) neutral detergent method \( (r^2 = 0.76) \), while estimates from the Van Soest (1967) summative equation were not significantly correlated \( (P>0.05) \) with in vivo digestion. However, the use of either in vitro method for predicting in vivo digestibility requires development of separate regression equations because both techniques generally over-estimated in vivo digestion. Also, separate regression relationships would be required for plant material varying widely in maturity.

In vitro digestibility of oak-containing rations was inversely related to both percentage of oak in the diets and the amount of oak in the inoculum donors' diet. However, oak content of diets was of much greater importance. Digestibility was also depressed by high drying temperatures and the effect was greater for material collected in early summer when foliage was relatively immature than it was in late summer when foliage reached maturity.
Part I

CONSUMPTION, DIGESTION, AND UTILIZATION BY GOATS OF THE

DRY MATTER AND NITROGEN IN DIETS CONTAINING OAK

(QUERCUS GAMBELII) FOLIAGE
INTRODUCTION

Recently there has been an increased interest in native forage plants and their contribution to the nutrition and production of grazing animals. Shrubby plants are attracting special attention because many such species provide relatively good sources of crude protein to grazing animals during dry or cold seasons when grasses and herbs are generally deficient. Additionally, they are frequently desirable in reclamation of disturbed sites, such as mine spoils (McKell 1976), and they are often the primary food source of big game animals. However, the difficulty and expense of obtaining sufficient quantities of shrubby forage material for feeding trials has been a major obstacle in determining the general nutritional value of most browse species. Additionally, many shrub species contain secondary compounds that may adversely affect their nutritive value (Burns et al. 1972).

Gambel oak, a shrub or small tree, occupies several million hectares of land in the Rocky Mountain and Intermountain areas of the western United States. The plant is consumed in varying degrees during the year by domestic and wild animals grazing these lands, but its nutritional value has been inadequately assessed.

Since the nineteenth century, foliage from various oak species has been classified as potentially toxic to livestock, the toxicity being attributed to the relatively high tannin content. Immature leaves and buds have a reported fatal effect when consumed in large
quantities by cattle, sheep, and goats (Kingsbury 1964). Radeleff (1970) concluded that oak poisoning effects are highly variable depending on the percentage of oak in the diet in excess of 30-40 percent and also on the oak species. The primary lesions are always gastritis and nephritis with hemorrhages, which can be detected by increased levels of blood urea nitrogen (BUN) or serum glutamate oxalacetate transaminase (SGOT), and by the decreased percentage of red blood cell volume (PCV) or hemoglobin concentration. On the other hand, Huston and Shelton (1967) found increased mohair production from Angora goats when oak (Q. virginiana) was incorporated in their basal ration, and oak of several species is a verified major constituent of diets of many range animals (Kufeld 1973, Kufeld et al. 1973, Malechek and Leinweber 1972).

The effects of tannins in ruminant nutrition are complicated. Hatfield (1970) found improved ruminant performance when feed was treated with unspecified quantities of tannic acid. This effect was attributed to the improved efficiency of utilization of dietary nitrogen by "protection" of dietary protein from microbial hydrolysis in the rumen. Nishimuta et al. (1972) found a trend for increased nitrogen retention when tannic acid was added to a basal diet at levels comparable to those which might be expected in oak-containing diets of browsing herbivores, but there were not significant differences from a control treatment. McLeod (1974) concluded that most natural forage tannins offer little value as "built-in" means of protein protection because the majority are of the condensed type as opposed to the hydrolyzable type which are useful in this regard.
However, work by Feeny and Bostock (1968) showed one oak species
(Quercus robur) to contain appreciable quantities of hydrolyzable
tannin.

The purpose of this study was to explore some nutritional
properties of Gambel oak at two phenological stages, with special
reference to its interactions with the processes of digestion
when consumed in varying amounts.
MATERIALS AND METHODS

Hand-harvested oak herbage mixed in graduated proportions with alfalfa was fed to goats in six digestion-balance trials. The oak material included only terminal portions (<15 cm length) of current year’s growth. It was harvested during two periods: in June, during the season of rapid growth, and in August, after twig elongation had ceased and stems had hardened. Alfalfa hay used in these diets was obtained commercially as pellets and was reground before mixing with oak. Oak collected in June was air dried, ground in a commercial forage chopper, and then mixed with alfalfa to form a diet containing 80 percent oak. The diet was then formed into pellets 26 mm diameter in a laboratory pelletizer. The material from the August collection was prepared similarly, but four diets containing 20, 40, 60 and 80 percent oak were formulated. Results from preliminary acceptance trials indicated that roughly 80 percent was the maximum level of oak that could be incorporated in a diet without substantially reducing intake by the goats.

Eight yearling female "Spanish" type goats of mixed breeding and weighing from 25 to 30 kg initially were used in this experiment. They were randomly separated into two groups and placed in metabolism stalls. In the sequence of trials group I was fed the diets containing 0, 40, and 80 percent oak, August collection, and 80 percent oak, June collection. Group II was fed diets containing 20 and 60 percent oak, August collection. Animals that showed signs
of behavioral stress (e.g. nervousness, low intake, weight loss) due to confinement in the metabolism stalls were replaced with a similar animal before proceeding to a subsequent digestion trial in the series.

Each feeding trial was preceded by a preliminary period of 10 days followed by a 7-day collection period (Harris 1970). Ambient temperature of the metabolism barn where trials were conducted was consistently maintained between 16° and 17°C. The animals were weighed before and after each trial, following a 12-hr fast without water. Feed was offered daily at 8:00 a.m. in amounts approximately 10 percent in excess of the maximum predetermined voluntary intake. During the experimental periods all animals had free access to water, salt, and mineral licks.

Feces and urine were collected and measured once daily and then composited by animal over the 7-days of a trial. To prevent nitrogen loss from urine, 75 ml of 25 percent sulfuric acid was added daily to the plastic urine collectors. Feed, feces, and urine samples were stored at -3°C until analyzed.

Subsamples of feed and feces were freeze-dried preparatory to laboratory analysis. Feed, feces, and urine were then analyzed for nitrogen content by a macro-Kjeldahl procedure (A.O.A.C. 1960). Feed and fecal samples were also analyzed for energy content by oxygen bomb calorimetry (A.O.A.C. 1960). Urinary energy was calculated from nitrogen content according to Street et al. (1964). Feed samples were analyzed for cellular constituents (Van Soest 1967),
lignin, and acid detergent fiber content (Van Soest 1963), and for tannin content (expressed as tannic acid equivalent) by the spectrophotometric method of Burns (1963).

On the 9th and 17th days of each trial, samples of jugular blood were drawn for evaluation of possible toxicosis. Tests on serum included blood urea nitrogen determined by the Gentzkow and Masen (1942) technique and serum glutamate oxalacetate transaminase by the method of Reitman and Frankel (1957). Tests on whole blood included packed cell volume by microhematocrit procedures, and hemoglobin was determined as cyanmethemoglobin (Cannan 1955).

Data were subjected to analysis of variance. For evaluating the significance of differences among means, Duncan's New Multiple Range test was used (Steel and Torrie 1960). Differences between means at the $\alpha = 0.05$ level of probability were considered statistically significant.

---

1/ Sigma Chemical Company, St. Louis, Missouri.

2/ Dade Chemical Company, Miami, Florida.
RESULTS AND DISCUSSION

Table 1-1 describes the chemical composition of the diets fed in this study. Crude protein and acid detergent fiber content declined while cellular constituents, lignin, tannin, and caloric content increased with increasing percentages of oak in the diets. Comparisons of the two 80 percent oak diets indicated lower lignin and caloric contents, but higher crude protein and tannin concentrations, in the diet from the June collection. These differences attributable to chemical changes in oak foliage associated with advancing maturity, are generally in agreement with previous research on other woody species (Short et al. 1972, Urness et al. 1975). However, Feeny (1970) reported peak tannin levels of 5.5 percent in the most mature leaves of Q. robur, in contrast to our findings of seasonally high levels of almost 11 percent in immature oak foliage. The difference may be attributed partly to the different species studied.

Voluntary intake was significantly higher for the animals when they were consuming the alfalfa diet than all but the 40 and 80 percent (August collection) oak diets (Table 1-2). Intake was not significantly different among animal groups consuming oak-containing diets. The inconsistently low intake values for the 20 percent and 60 percent oak diets probably reflect behavioral aberrations of the experimental animals more than intrinsic properties of the diets.
Table 1-1. Chemical composition and caloric content of dry matter of five oak-containing diets in comparison to an alfalfa diet.

<table>
<thead>
<tr>
<th>Constituents</th>
<th>% Oak</th>
<th>% Alfalfa</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80¹/²</th>
</tr>
</thead>
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<tr>
<td>Crude protein (%)</td>
<td>17.4</td>
<td>17.5</td>
<td>16.7</td>
<td>15.9</td>
<td>15.7</td>
<td>16.1</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cellular constituents (%)</td>
<td>56.0</td>
<td>57.9</td>
<td>58.6</td>
<td>60.3</td>
<td>63.1</td>
<td>63.4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lignin (%)</td>
<td>7.8</td>
<td>8.5</td>
<td>9.7</td>
<td>10.1</td>
<td>10.5</td>
<td>9.8</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Acid detergent fiber (%)</td>
<td>39.6</td>
<td>38.0</td>
<td>36.6</td>
<td>35.3</td>
<td>32.8</td>
<td>31.0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tannin (%)</td>
<td>1.0</td>
<td>2.4</td>
<td>3.2</td>
<td>4.8</td>
<td>6.9</td>
<td>8.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gross energy (Kcal/g)</td>
<td>4.1</td>
<td>4.5</td>
<td>4.6</td>
<td>4.7</td>
<td>4.8</td>
<td>4.7</td>
<td></td>
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</tbody>
</table>

¹/² Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table 1-2. Intake, dry matter digestibility, and body weight gain by goats consuming five oak-containing diets and an alfalfa diet.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80 ( \uparrow )</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Intake (g·day(^{-1}·kg(^{-1}))</td>
<td>54.7±9.1a(^2)</td>
<td>38.4±11.9b</td>
<td>46.7±6.9ab</td>
<td>40.1±11.7b</td>
<td>42.0±4.0ab</td>
<td>35.7±2.8b</td>
<td></td>
</tr>
<tr>
<td>Dry matter digestibility (%)</td>
<td>56.9±1.3a</td>
<td>53.9±3.2ab</td>
<td>53.0±1.8bc</td>
<td>50.9±2.6bc</td>
<td>49.8±2.5c</td>
<td>54.0±2.5ab</td>
<td></td>
</tr>
<tr>
<td>Gain (kg·7 days(^{-1}))</td>
<td>2.2±1.1</td>
<td>1.2±1.3</td>
<td>1.5±0.5</td>
<td>1.3±0.7</td>
<td>1.6±1.3</td>
<td>0.6±0.3</td>
<td></td>
</tr>
</tbody>
</table>

\(^1\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.

\(^2\) Means (±95% confidence intervals) followed by a common letter or by no letter are not significantly different (P<0.05).
Three of the original four animals assigned these two diets (Group II) had to be replaced during the course of the experiment because of extreme nervousness and refusal of food, whereas only one of the Group I animals was replaced. Thus all intake-related parameters for the 20 percent oak and 60 percent oak diets were probably negatively biased, and the additional experimental error variation introduced often precluded conclusive statistical interpretations.

A comparison of the 80 percent oak diets indicated that the one from the early summer collection was associated with an apparently lower but not significant different (P<0.05) intake rate. Depressed intake of diets containing either early-summer or late summer foliage might possibly be attributed to tannin content and its effect on palatability (Donnelly 1954) or to interactions of tannins with the activity of various enzymes in the ruminant digestive system (Tagari et al. 1965). However, McLeod (1974) maintains that the significance of tannin as a cause of low intake has not been clearly established.

Dry matter digestibility decreased as the percentage of oak increased in the diets (Table 1-2). Any two diets from the August collection differing 40 percent or more in their oak content differed significantly (P<0.05) in their digestibility. Both lignin and tannin may be implicated in this response, as both possess demonstrated digestion-reducing properties (Van Soest 1963, McLeod 1974) and both, especially tannin, increased in concentration as oak content of the diets rose. However, comparison of the two 80 percent oak diets suggests that the role of tannin in suppression of dry matter digestion is probably minor in comparison to that of lignin.
Lignin content rose slightly from June to August, while tannin declined considerably (Table 1-1). The lower digestibility of the August-based diet (Table 1-2) is more reflective of its lignin content than its tannin content. The other possibilities exist that maturity affects the way both lignin and tannin interact with dry matter digestion or that other unmeasured dry matter constituents such as cutin (Storr 1961) or silica (Van Soest 1967) contributed to variations observed in digestibility.

Weight response of goats (Table 1-2) indicated that all rations supplied levels of nutrients somewhat in excess of maintenance. There were no important differences in weight response among rations.

Nitrogen intake generally followed the same pattern as dry matter intake, hence values for the 20 percent and 60 percent oak diets probably reflect the same bias described above. Animals consuming the alfalfa diet had a significantly higher nitrogen intake rate than for all but the 40 percent and 80 percent (August collection) diets (Table 1-3), but no differences were detected among the oak-containing diets. Variation among animals was generally high in nitrogen intake, digestion, metabolism and excretions.

Observations on nitrogen dynamics of the animals (Table 1-3) were not generally conclusive as to whether net nitrogen retention was influenced by levels of oak in the diets. Exceptions were the relatively small quantity retained by animals consuming the 20 percent oak diet from August collection and the 80 percent oak diet from the June collection. This small retention might be a result of the low...
Table 1-3. Nitrogen balance of goats over a 7-day period.

<table>
<thead>
<tr>
<th>Diets</th>
<th>% Oak</th>
<th>% Alfalfa</th>
<th>N Consumed (g)</th>
<th>Fecal N (g)</th>
<th>Urinary N (g)</th>
<th>N apparently digested (g) (%)</th>
<th>N retained (g) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>00 100</td>
<td>295.4±87.4a</td>
<td>79.5±7.8bc</td>
<td>148.8±67.4a</td>
<td>216.0±65.9a</td>
<td>73.1a</td>
<td>67.1±18.5a 22.7a</td>
<td></td>
</tr>
<tr>
<td>20 80</td>
<td>210.0±81.7b</td>
<td>75.2±43.8c</td>
<td>114.7±25.5b</td>
<td>134.8±44.3bc</td>
<td>64.2b</td>
<td>20.1±22.9cd 9.5b</td>
<td></td>
</tr>
<tr>
<td>40 60</td>
<td>272.4±28.9ab</td>
<td>110.5±23.0ab</td>
<td>114.5±12.6b</td>
<td>161.9±11.9b</td>
<td>59.4c</td>
<td>47.4±19.6abc 17.4a</td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>211.4±79.2b</td>
<td>99.2±32.3bc</td>
<td>74.8±22.1cd</td>
<td>112.4±35.2c</td>
<td>53.1d</td>
<td>37.6±21.5bcd 17.8a</td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>243.4±35.3ab</td>
<td>125.3±11.1a</td>
<td>64.8±3.0d</td>
<td>118.0±10.9c</td>
<td>48.5e</td>
<td>53.3±11.6ab 21.9a</td>
<td></td>
</tr>
<tr>
<td>80°</td>
<td>20</td>
<td>210.7±21.7b</td>
<td>104.1±17.6abc</td>
<td>88.9±8.0bc</td>
<td>106.6±7.5c</td>
<td>50.6de</td>
<td>18.4±6.8d 8.7b</td>
</tr>
</tbody>
</table>

1/ Means (±95% confidence intervals) in the same column followed by a common letter are not significantly (P<0.05) different.

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
level of metabolized energy present in these diets, leading to
catabolism of protein for energy production. The declining trend
in output of urinary nitrogen with higher percentages of oak
indicates that the animals were more efficient in utilizing the
digested nitrogen. However, a corresponding increase in fecal
nitrogen output effectively nullified any increase in nitrogen
retained.

Chalupa (1975) in a recent review, concluded that retention
of plant proteins by protection from bacterial hydrolysis in the
rumen is not always improved. Tannin may decrease protein solubility
in the rumen, but it may also decrease digestion of protein in the
small intestine. The effect of tannin on protein digestion in the
small intestine is unknown (McLeod 1974).

Few important differences among diets were found for energy. Due
to the higher gross energy content of oak in comparison to alfalfa,
the overall energy consumed was not significantly (P<0.05) different
among rations (Table 1-4), although dry matter intake was generally
decreased in the diets that contained greater proportions of oak.

Percent energy digested for diets containing oak from the late
summer collection demonstrated a declining pattern similar to that
of nitrogen digestibility but with smaller deviations among diets.
Our results do not indicate whether high percentages of oak in the
diets had any effect on the percent of energy metabolized (Table 1-4).
The only conclusion that can be derived is that oak collected in
early summer yielded a smaller percentage of metabolized energy than
that collected in late summer.
Table 1-4. Energy balance of goats over a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>% Alfalfa</th>
<th>Energy Consumed (Mcal)</th>
<th>Energy In Feces (Mcal)</th>
<th>Energy In Urine (Mcal)</th>
<th>Digestibility (Mcal) (%)</th>
<th>Metabolized 2/ (Mcal) (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>00</td>
<td>100</td>
<td>48.7±14.4†</td>
<td>20.8±5.6</td>
<td>3.3±1.3b</td>
<td>27.9±8.8a</td>
<td>57.3a</td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td>35.3±13.7</td>
<td>16.1±6.5</td>
<td>2.6±0.5c</td>
<td>19.2±7.4c</td>
<td>54.4ab</td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td>49.3±5.2</td>
<td>23.3±3.4</td>
<td>2.5±0.2c</td>
<td>26.1±1.9ab</td>
<td>52.9bc</td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td>40.9±15.3</td>
<td>20.0±7.6</td>
<td>1.7±0.5d</td>
<td>20.9±7.9bc</td>
<td>51.1c</td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>48.6±4.5</td>
<td>24.3±2.7</td>
<td>1.5±0.1d</td>
<td>24.3±1.8abc</td>
<td>50.0c</td>
</tr>
<tr>
<td>80 2/</td>
<td>20</td>
<td>39.7±4.0</td>
<td>18.9±2.4</td>
<td>4.6±0.5a</td>
<td>20.8±1.6bc</td>
<td>52.4b</td>
</tr>
</tbody>
</table>

† Means (±95% confidence intervals) in the same column followed by a common or no letter are not significantly (P<0.05) different).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.

3/ Uncorrected for methane production.
Fecal energy did not vary significantly among diets, but urinary energy decreased with increased percentages of oak in the diets. Urinary energy probably reflects urinary nitrogen content.

Comparing the two 80 percent oak diets, the one containing material from the early summer collection revealed approximately four times more urinary energy. This is probably a result of tannin content, the only chemical fraction different between these two diets.

None of the four tests used for monitoring possible toxicosis revealed any effect. Means over all animals and oak diets are presented in Table 1-5 in comparison to averages over all animals for a control alfalfa diet. Kingsbury (1964) has suggested supplementing oak-containing rations with up to 10 percent alfalfa as a means of preventing oak poisoning. All diets tested in this study contained 20 percent or more alfalfa, which may have acted as an effective preventative agent.

Table 1-5. Average blood and serum parameters for goats fed an alfalfa diet and five oak-containing diets.

<table>
<thead>
<tr>
<th>Diet</th>
<th>Hemoglobin g·100 ml⁻¹</th>
<th>SGOT units·ml⁻¹</th>
<th>BUN mg·100 ml⁻¹</th>
<th>PCV %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alfalfa</td>
<td>13.3</td>
<td>93.1</td>
<td>25.9</td>
<td>36.7</td>
</tr>
<tr>
<td>Oak-alfalfa, mean of all levels</td>
<td>13.6</td>
<td>95.8</td>
<td>19.6</td>
<td>38.3</td>
</tr>
</tbody>
</table>
SUMMARY AND CONCLUSIONS

Intake rates of most oak-containing diets were lower than those of an alfalfa diet, with a trend for lower intake with increased dietary percentages of oak. Digestibility was significantly lower for oak diets than for the control and was inversely related to the percentage of oak in the diets.

Oak foliage from an early summer collection resulted in lower amounts of metabolizable energy than did an equivalent diet formulated from material collected in late summer. This was not expected since crude protein was not significantly different for the two diets, while dry matter digestibility was higher for the early summer diet. Further research involving the possible implications of tannin in this relationship is needed.

Nutrients available from diets containing up to 80 percent oak were in excess of maintenance requirements for goats. Gains resulting from late summer oak diets were not significantly different from those of an alfalfa diet. The relatively high digestibility of gross energy and crude protein of oak during the growing season suggest that it may serve an important role in nutrition of domestic as well as for wild animals under certain rangeland conditions.
LITERATURE CITED


Part II

ESTIMATION OF IN VIVO DIGESTIBILITY OF OAK-CONTAINING DIETS BY MICRO-DIGESTION TECHNIQUES
INTRODUCTION

Conventional digestion-balance trials are of limited feasibility for estimation of the digestibility of range forage plants. The expense involved in collection and preparation of sufficient quantities of material for such feeding trials is beyond the budget of many research projects. Alternative micro-techniques that have been used extensively for digestibility prediction include various in vitro techniques (Tilley and Terry 1963, Barnes 1967) and chemical partitioning of dry matter into fractions of known digestibility (Van Soest et al. 1966, Van Soest 1967 and 1968, Goering and Van Soest 1970, Fonnesbeck and Harris 1970). Such techniques, when applied within narrow and specifically defined boundaries of forage variation, have proven to be both accurate and inexpensive. They are suitable for analyzing large numbers of samples at a time, thus have had particular appeal to plant breeders interested in determining preliminary nutritional information on large numbers of plant cultivars.

The use of indirect methods for estimating the in vivo digestibility of range forage in general and shrub species in particular has been problematical (Short et al. 1974). Fiber content of shrubs is relatively high and is known to increase with tissue maturity more rapidly than in conventional forage grasses and legumes (Dietz 1972). Digestibility of dry matter in shrubs declines drastically after the period of rapid growth which
frequently occurs during a relatively brief portion of the growing season (Short et al. 1972, Urness et al. 1975). Maturation affects not only the cell constituents or "cell solubles" to "cell wall" ratio, but also the concentration of lignins, cellulose, hemicellulose and other fibrous components of the cell wall, resulting in reduction of digestibility (Short et al. 1974).

Digestibility of immature browse twigs is adequately predicted by the two-stage in vitro digestion technique of Tilley and Terry (1963) and the summative equation of Van Soest (1967), but estimates of digestibility for mature foliage from browse plants by either technique are generally lower than in vivo digestion (Urness et al. 1977). Van Soest et al. (1966) suggested that in the two-stage Tilley and Terry (1963) procedure, some bacterial matter remained occluded in the fiber of the final residue, thus, yielding lower estimates of true digestibility. They concluded also that if the second stage of acid pepsin digestion of the Tilley and Terry (1963) method was substituted by the neutral detergent extraction procedure of Van Soest et al. (1966) all bacterial matter was dissolved. This conclusion arose from their findings of no difference in the quantity of intact plant material dissolved by either acid pepsin and neutral detergent. Thus, any differences found in treating samples of microbial fermentation residues by the two methods were attributed to undigested bacterial cells. Wilson et al. (1971) suggested that the retention of bacterial cells in the fiber is greater in forages of low digestibility. Secondary chemical compounds such as tannins and terpenes found in many shrub species may also
erratically affect estimation of digestion. Burns et al. (1972) comparing in vitro digestibility of serica lespedeza with alfalfa, found alfalfa more digestible by 14.2 digestibility units. These two forages are similar in structural constituents but lespedeza contains approximately three times more tannins and phenolic compounds than does alfalfa.

Method of sample preparation has been another important source of variation in laboratory estimates of dry matter digestibility. Tilley and Terry (1963) found no differences in digestibility of conventional forages dried at various temperatures below 100°C. However, Noller et al. (1966) reported higher in vitro digestibility for lyophilized alfalfa and corn foliage in comparison to material dried at 60° or 80°C. This can be explained by the Van Soest (1965) findings of increased fiber and "apparent lignin" content for conventional forages dried at 100°C in comparison to those dried at 20°C or lyophilized. Validations of the above findings are needed for shrub species having chemical composition significantly different than conventional grass and legume forages. They are especially needed for species that contain significant proportions of tannins that may act either as microbial inhibitors (Tagari et al. 1965) or as complexing agents that form insoluble polymers with plant proteins (McLeod 1974).

Knipfel and Troelsen (1966) and Calder (1970), studying microdigestion of diets composed of alfalfa, wheat straw, and barley grain in varying proportions, found that inoculum used for in vitro studies had significant effects on digestibility
estimates. Significant inoculum-related effects would be expected when donor animals consume diets high in shrub species that contain large quantities of digestion reducing compounds such as tannins.

The research reported in this paper was conducted to compare in vivo digestibility of oak-containing diets with estimates derived from three microdigestion techniques, and to explore the effects of inoculum donors' diet and drying temperatures upon digestion of oak foliage.
MATERIALS AND METHODS

Oak was collected at two discrete periods, in June during the season of rapid growth and in August after elongation had ceased and stems had hardened. Oak foliage included only terminal portions (up to 15 cm length) of the current year's growth. From the early collection a diet was formulated containing 80 percent oak and 20 percent alfalfa. From the later collection four additional diets were formulated containing 20, 40, 60, and 80 percent oak with alfalfa making up the complement. A pure alfalfa diet was used as the experimental control.

The six diets were tested for in vitro dry matter digestibility by the Tilley and Terry (1963) two-stage acid-pepsin digestion technique (IVAP) modified by terminating fermentation with HCl instead of HgCl and by the method proposed by Van Soest et al. (1966) where the second stage consists of extracting in vitro fermentation residues with neutral detergent (IVND). Additionally, the dry matter of the diets was partitioned into the various components specified by Van Soest (1967) and true dry matter digestibility was calculated by his summative equation, omitting the SiO$_2$ term.

Inoculum for the in vitro digestion trials was obtained by vacuum aspiration of rumen fluid from two ruminally-fistulated goats fed the oak diets. The six diets were fed in sequence beginning with the pure alfalfa control and proceeding in order of increasing oak content. Donor animals were allowed at least 5 days
to adjust to each new diet before rumen fluid was collected for a series of in vitro trials. Rumen inoculum obtained from donor animals consuming a specific test diet was termed a source of inoculum. For each source of inoculum all six diets were fermented in duplicate for each of the two in vitro techniques. The rumen fluid obtained from the two donors at any particular collection was aggregated and handled according to the Tilley and Terry (1963) IVAP procedure.

Results achieved from the three small-sample techniques were compared by regression procedures to in vivo digestibility values determined in a related experiment (Nastis 1977). Only those in vitro values were used that resulted from digestion of a particular test diet by inculum from donors consuming the corresponding diet.

To further isolate sources of variation in estimates of digestibility by the two in vitro techniques, a comparison was conducted on the ability of acid pepsin and neutral detergent to dissolve bacterial cells. For this purpose bacterial samples were separated from rumen liquor as follows: large solids were separated from the liquid phase of the rumen fluid by straining through cheesecloth. This filtrate was then subjected to a modification of Williams and William (1973) method for separation of bacterial cells. A 2000 ml aliquot was first centrifuged at 500 g for 10 minutes to remove sediment after straining through cheesecloth, and then the filtrate was centrifuged at 1400 g for 20 minutes to isolate microbial cells. The residue, composed mainly of bacteria cells (Hungate 1965), was then re-suspended in distilled water, frozen, and freeze-dried.
These samples of bacterial cells were then tested for solubility by acid pepsin and neutral detergent.

Additionally, a test was conducted to determine the effect of drying temperature upon in vitro digestion. Samples of oak foliage were hand harvested at monthly intervals throughout the growing season and sub-samples of these materials were freeze-dried at -2°C, air-dried at 0°C and 25°C, and oven-dried at 55°C, 65°C, and 100°C. These subsamples were then subjected to in vitro (Tilley and Terry 1963) two-stage digestion.

All results were evaluated using analysis of variance procedures and Duncan's New Multiple Range test (Steel and Torrie 1960). In vivo and in vitro digestibility values were regressed on diet composition. Additionally, a multiple regression (Snedecor and Cochran 1967) for predicting in vivo digestibility was investigated, including as independent variables 1) digestibility by the Tilley and Terry (1963) IVAP method, 2) digestibility by the Van Soest et al. (1966) IVND method, 3) digestibility by the Van Soest (1967) summative equation, and 4) percent tannin content (Nastis 1977).
RESULTS AND DISCUSSION

Diet Composition and Digestibility

Average in vivo and in vitro digestibility of experimental diets decreased as the percentage of oak increased (Figure 2-1). Results from the summative equation are not included since they were not significantly correlated with diet composition. Tilley and Terry (1963) IVAP digestion showed a significantly greater decline in digestibility with increasing oak content of diets than the Van Soest et al. (1966) IVND digestion. The rate of decrease over diets (slope of the regression line) for in vivo digestion was not statistically (P<0.05) different from either of the in vitro methods. However, differences in the intercepts were significant among all methods except between the Tilley and Terry (1963) IVAP and in vivo digestion. Both the Tilley and Terry (1963) IVAP method and the Van Soest et al. (1966) IVND in vitro methods over-estimated in vivo digestibility of all diets containing oak foliage from the late summer collection. These results suggest that in vitro digestibility data are of little value for direct estimation of in vivo values.

Comparison of In Vitro Techniques

Regression analysis for comparison of the two in vitro micro-digestion techniques to in vivo digestion (Figure 2-2) indicated that the Tilley and Terry (1963) IVAP method was the most precise
IVND (Δ) y = 75.0 - 0.1X ± 1.2  \( r^2 = 0.83 \)
IVAP (o) y = 66.6 - 0.2X ± 0.2  \( r^2 = 0.99 \)
IVD (□) y = 56.3 - 0.1X ± 1.7  \( r^2 = 0.61 \)

Fig. 2-1. Dry matter digestibility of oak-alfalfa diets in relation to diet composition. Digestion by in vitro with acid pepsin method (IVAP) and in vitro with neutral detergent method (IVND) is compared to in vivo digestion (IVD).
IVAP(o) = 29.1 + 0.4X ± 0.6  \( r^2 = 0.97 \)
IVND(Δ) = -18.6 + 1.0X ± 1.6  \( r^2 = 0.76 \)

**Fig. 2-2.** Relation of in vitro dry matter digestibility, determined by the in vitro with acid pepsin method (IVAP) and the in vitro with neutral detergent method (IVND), to in vivo digestibility.
(r^2 = 0.97). The coefficient determination for the Van Soest et al. (1966) IVND method (r^2 = 0.76) suggested that this procedure accounted for considerably less variation. Neither method, however, was sufficiently accurate to allow estimation of in vivo digestibility from in vitro data without developing a regression relationship.

Although limited, the data suggested that separate regression relationships would be required for testing plant material harvested at distinctly different phenological stages. If digestibility data from the diet containing 80 percent oak collected in early summer were included in the regression analysis, the standard error for both in vitro methods would be increased drastically. The coefficient of determination (r^2) would also drop for the IVAP method from 0.97 to 0.44 and for the IVND method from 0.76 to 0.16. This data point was tested as an outlier (Snedecor and Cochran 1967) and was found to deviate significantly from the remaining points. Sufficient data were not available to establish a separate relationship for June-collected oak in this study.

Digestibility results from the summative equation are not included in Figure 2-2 because they were not significantly correlated (P<0.05) with in vivo digestibility. The summative equation apparently provides unsatisfactory estimates of in vivo digestibility for oak-containing diets. This is because in vivo digestibility for some fractions of the dry matter is different from that assumed by the Van Soest (1967) equation. For example, in a related study (Nastis 1977) the maximum digestibility of cell constituents of oak was 74.8±2.0 percent as compared to 98.0±2.5 percent assumed by the Van
Soest (1967) equation. The reasons for this discrepancy are unclear, but the digestion-reducing properties of tannins are probably implicated.

Average digestibility of dry matter for all diets, considering all inoculum sources, was significantly higher for the Van Soest et al. (1966) IVND method (71.1±0.3) than the Tilley and Terry (1963) IVAP method (56.5±0.3). This difference must be attributed to differences in solubility of the undigested plant residues remaining after microbial fermentation, of bacterial cells, or to some combination of the two. Solubility of bacterial cells was subsequently tested in acid pepsin and neutral detergent. On the basis of nine replications, a small but significantly higher solubility of bacterial cells was found for the neutral detergent method (99.7 percent) than the acid pepsin method (98.1 percent). Therefore these results suggest that the difference in estimates of dry matter digestibility must be attributed to variation in the ability of acid pepsin and neutral detergent to dissolve material of plant origin.

**Inoculum Donors' Diet**

Inoculum source had a significant effect on in vitro digestibility with a trend for lower values as the composition of the donors' diet increased in oak content (Table 2-1). When donors were fed the diet composed of oak collected in June, even lower in vitro digestibility resulted. Moreover, composition of donors' diet and composition of test diets interacted significantly (P<0.05). Test
Table 2-1. Percent digestibility of an alfalfa diet and five oak-containing diets in relation to composition of the inoculum donors' diets. Values are means of estimates from the Tilley and Terry (1963) method and the Van Soest et al. (1966) method.

<table>
<thead>
<tr>
<th>Composition of inoculum donors' diet</th>
<th>Diets</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>00</td>
<td>20</td>
<td>40</td>
<td>60</td>
<td>80</td>
<td>801/</td>
<td>Average2/</td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak</td>
<td>% Alfalfa</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>100</td>
<td>67.1</td>
<td>68.5</td>
<td>65.7</td>
<td>64.5</td>
<td>62.1</td>
<td>64.8</td>
<td>65.5±0.5a</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td>69.3</td>
<td>66.9</td>
<td>64.8</td>
<td>63.6</td>
<td>62.7</td>
<td>65.1</td>
<td>65.4±0.5a</td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td>68.2</td>
<td>67.4</td>
<td>65.4</td>
<td>62.9</td>
<td>60.8</td>
<td>64.0</td>
<td>64.8±0.5ab</td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td>68.2</td>
<td>67.2</td>
<td>64.9</td>
<td>62.9</td>
<td>62.0</td>
<td>61.9</td>
<td>64.5±0.5b</td>
<td></td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>67.7</td>
<td>65.8</td>
<td>63.6</td>
<td>61.4</td>
<td>54.7</td>
<td>61.9</td>
<td>63.3±0.5c</td>
<td></td>
</tr>
<tr>
<td>801/</td>
<td>20</td>
<td>64.6</td>
<td>60.9</td>
<td>61.6</td>
<td>57.1</td>
<td>55.7</td>
<td>55.7</td>
<td>59.3±0.5d</td>
<td></td>
</tr>
<tr>
<td><strong>Average2/</strong></td>
<td></td>
<td>65.5±0.5a</td>
<td>66.1±0.5b</td>
<td>64.3±0.5c</td>
<td>62.1±0.5d</td>
<td>60.5±0.5e</td>
<td>62.2±0.5d</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.

2/ Digestibility averages (±95% confidence intervals) in the same column or row followed by a common letter are not significantly (P<0.05 different).
diets containing high percentages of oak were lower in in vitro digestibility when inoculum donors' diet contained high levels of oak than when it contained only alfalfa or low percentages of oak. Table 2-1 indicates that digestibility of diets containing less than 60 percent oak is slightly affected by the inoculum donors' diet, while digestibility of diets containing 80 percent oak is highly influenced by the inoculum donors' diet.

The interactions of in vitro methods with inoculum sources was significant. As seen in Table 2-2, digestion by IVAP was affected to a slightly greater degree by inoculum source than was IVND digestion. If tannin is indeed responsible for reductions in the enzymatic activity of pepsin, apparently the relatively small amount introduced into the fermentation medium from the inoculum source is not nearly so important as the relatively greater amount present in diets containing large portions of oak.

**Phenological Stage and Drying Temperature**

Digestibility of oak foliage was significantly affected by drying temperatures (Table 2-3). Lower in vitro digestibilities were associated with higher drying temperatures. Van Soest (1965) demonstrated that heating alters carbohydrate structure and diminishes nitrogen solubility by forming insoluble polymers that are measured in the lignin fraction.

Phenological stage of plant material also imparted significant variation upon in vitro digestibility. Averages of in vitro digestion
Table 2-2. In vitro digestibility (%) of dry matter, with inoculum from donors' consuming oak-alfalfa diets in varying proportions, by Tilley and Terry (1963) and Van Soest et al. (1966) methods averaged over all diets.

<table>
<thead>
<tr>
<th>Inoculum donors' diet</th>
<th>% Oak</th>
<th>% Alfalfa</th>
<th>Tilley and Terry apparent</th>
<th>Van Soest true</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>100</td>
<td></td>
<td>58.8±3.0a(^1/)</td>
<td>72.1±4.2ab</td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td></td>
<td>57.5±3.0b</td>
<td>73.3±1.6a</td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td></td>
<td>57.8±3.0b</td>
<td>71.8±3.6b</td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td></td>
<td>57.3±2.6b</td>
<td>71.8±1.3b</td>
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<tr>
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<td>20</td>
<td></td>
<td>55.8±3.1c</td>
<td>70.9±0.9c</td>
</tr>
<tr>
<td>80(^2/)</td>
<td>20</td>
<td></td>
<td>51.6±3.3d</td>
<td>67.0±1.5d</td>
</tr>
<tr>
<td>Average</td>
<td></td>
<td></td>
<td>56.5±0.3</td>
<td>71.1±0.3</td>
</tr>
</tbody>
</table>

\(^1/\) Digestibility averages (±95% confidence intervals) in a particular column followed by common letters are not significant (P<0.05) different.

\(^2/\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table 2-3. Effects of drying temperature and stage of maturity upon Tilley and Terry (1963) in vitro digestion of oak foliage.

<table>
<thead>
<tr>
<th>Collection date</th>
<th>Drying Temperatures (°C)</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th>Average&lt;sup&gt;1/&lt;/sup&gt;</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>-2°</td>
<td>0°</td>
<td>25°</td>
<td>55°</td>
<td>65°</td>
<td>100°</td>
<td></td>
</tr>
<tr>
<td>June&lt;sup&gt;2/&lt;/sup&gt;</td>
<td>55.4</td>
<td>50.6</td>
<td>50.6</td>
<td>42.6</td>
<td>43.7</td>
<td>41.2</td>
<td>47.4±0.9a</td>
</tr>
<tr>
<td>July</td>
<td>46.2</td>
<td>41.3</td>
<td>44.0</td>
<td>38.4</td>
<td>40.1</td>
<td>37.5</td>
<td>41.2±0.9b</td>
</tr>
<tr>
<td>August</td>
<td>43.0</td>
<td>37.6</td>
<td>38.4</td>
<td>36.0</td>
<td>34.0</td>
<td>33.7</td>
<td>37.1±0.9c</td>
</tr>
<tr>
<td>September</td>
<td>38.7</td>
<td>38.5</td>
<td>36.7</td>
<td>36.4</td>
<td>37.0</td>
<td>37.0</td>
<td>37.3±0.9c</td>
</tr>
<tr>
<td>Average&lt;sup&gt;1/&lt;/sup&gt;</td>
<td>45.8±1.1a</td>
<td>42.0±1.1b</td>
<td>42.4±1.2b</td>
<td>38.4±1.2c</td>
<td>38.7±1.2c</td>
<td>37.4±1.1c</td>
<td></td>
</tr>
</tbody>
</table>

<sup>1/</sup> Digestibility averages (±95% confidence intervals) in the same column or row followed by the same letter are not significantly different (P<0.05).

<sup>2/</sup> Within a week from bud emergence.
across all drying temperatures decreased through the growing season from 47.4 percent in June to 37.3 percent in September. Oak collected in June had a significantly higher digestibility (P<0.05), than oak collected in July. These were both different from oak collected in the following months. However, there were no significant differences for oak collected in August or September. Chemical composition of oak through the growing season (unpublished data) showed a significant decrease in crude protein from 20 percent to 13 percent and an increase in acid detergent fiber from 29 percent to 36 percent and also a slight increase in lignin content from 10 percent to 11 percent. These chemical changes probably explain the decrease in digestibility of oak over the range of phenological stages.

Interactions between phenological stage and drying temperature (Table 2-3) were also significant (P<0.05). High temperature depressed IVAP digestibility to a greater extent in early-harvested material than late-harvested material. This is probably related to the effect of elevated temperatures in altering plant carbohydrate composition and formation of insoluble, dark-colored polymers which are intensified by the high moisture content of early-harvested material (Van Soest 1965, Raguse and Smith 1963).

Estimation of In Vivo Digestibility from Laboratory Procedures

Although the Tilley and Terry (1963) IVAP digestibility is the best estimator of in vivo digestibility of mature oak foliage ($r^2 = 0.97$), its value for prediction over a wide range of plant
phenological conditions is doubtful. As discussed previously, when the early summer diet was included in regression relationships, unaccounted variation increased dramatically. In order to estimate the in vivo digestibility of oak foliage at any phenological stage a multiple regression was developed. Independent variables included digestibility by the Tilley and Terry (1963) IVAP method, digestibility by the Van Soest et al. (1966) IVND method, digestibility by the Van Soest (1967) summative equation, and percent tannin content (Nastis 1977). The regression, including only factors with significant F-values (P<0.05), is \( Y = 9.1 + 1.4 \text{ (IVAP)} - 0.7 \text{ (IVND)} + 2.4 \text{ (Tannin)} \) with \( r^2 = 0.99 \).
SUMMARY AND CONCLUSIONS

Digestibility of oak-alfalfa diets decreased as the percentage of oak increased in the diets due to the lower oak digestibility.

The Tilley and Terry (1963) IVAP technique was the best predictor for in vivo digestibility of oak diets followed by the Van Soest et al. (1966) IVND method while the Van Soest (1967) summative equation yielded results not significantly correlated (P<0.05) with in vivo digestibility. Chemical methods are apparently not appropriate for a broad array of species having unusual chemical and structural composition since digestibility of standard fractions apparently differs between species. Digestibility of cell constituents for oak was 74.8 percent in contrast to 98.0 percent for alfalfa.

Differences in dry matter digestibility, as estimated by the Tilley and Terry (1963) IVAP technique and the Van Soest et al. (1966) IVND method, must be attributed mainly to differences in the extent that acid pepsin and neutral detergent dissolve fermentation residues of plant origin. Neither in vitro technique was sufficiently precise for yielding direct estimates of in vivo digestibility. There were small, but statistically significant differences in the extent of bacterial solubility by the two chemicals.

Oak content of inoculum donors' diet was inversely related to in vitro digestibility. Inoculum from donors fed diets with high percentages of oak yielded lower in vitro digestibilities for diets
with high oak content than for diets with little or no oak content. In vitro digestibility of oak foliage decreased with both advanced maturity and increased drying temperatures. The effect of high drying temperatures on digestibility was more pronounced in immature oak foliage than on mature foliage.

A multiple regression equation for estimating in vivo digestibility of oak containing rations over a range of phenological conditions for oak was developed as: \[ \hat{Y} = 9.1 + 1.4 \text{ (IVAP)} - 0.7 \text{ (IVND)} + 2.4 \text{ (Tannin)} \]
LITERATURE CITED


Appendix A

Measurements and Statistical Tests Related to

Determination of In Vivo Digestibility
Table A-1. Average live weight (kg) of goats during the digestion trial of each diet.

| Goat % Alfalfa | % Oak  | 00  | 20  | 40  | 60  | 80  | 80 1/
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<tr>
<th></th>
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<th></th>
<th></th>
<th></th>
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</tr>
</thead>
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<td>32.6</td>
<td>--</td>
<td>34.5</td>
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<td>34.8</td>
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<td>36.8</td>
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</tr>
<tr>
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</tr>
<tr>
<td>6</td>
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<tr>
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<td>31.1</td>
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</tr>
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<td>9</td>
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<td>10</td>
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<td>30.7</td>
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<td>32.8</td>
<td>20.8</td>
<td>34.4</td>
<td>35.4</td>
<td></td>
</tr>
</tbody>
</table>

1/ Oak collected during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-2. Intake of experimental diets by goats (g dry matter/kg body weight·day).

<table>
<thead>
<tr>
<th>% Oak</th>
<th>Diets</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80½</th>
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</thead>
<tbody>
<tr>
<td>Goat % Alfalfa</td>
<td></td>
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<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
<td>20</td>
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<td></td>
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</tr>
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<td></td>
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<td>46.7</td>
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</table>

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-3. Dry matter digestibility (%) by goats of five oak-containing diets in comparison to an alfalfa diet.

<table>
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<th>60</th>
<th>80</th>
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<th>Goat % Alfalfa</th>
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<td>50.9</td>
<td>49.8</td>
<td>54.0</td>
<td></td>
</tr>
</tbody>
</table>

<sup>1/</sup> Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-4. Weight gain (kg/head) by goats over a 7-day period while consuming diets containing oak and alfalfa.

<table>
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<th>Goat % Alfalfa</th>
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<th></th>
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<th></th>
<th></th>
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<tbody>
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<td>80</td>
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<td>60</td>
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<td>0.4</td>
<td>0.9</td>
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<td>2.5</td>
<td>--</td>
<td>2.0</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>1.1</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>1.1</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>0.9</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>2.2</td>
<td>1.2</td>
<td>1.5</td>
<td>1.3</td>
<td>1.6</td>
<td>0.6</td>
</tr>
</tbody>
</table>

1/ Oak collected during the season of rapid growth. Oak in other diets was collected in August after elongation had ceased and stems had hardened.
Table A-5. Analysis of variance for dry matter intake, dry matter digestibility, and weight gain by goats.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Dry matter intake</th>
<th>Dry matter digestibility</th>
<th>Weight gain</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td>5</td>
<td>186.1(^{1/})</td>
<td>25.6(^{*})</td>
<td>1.0</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>63.5</td>
<td>4.3</td>
<td>0.5</td>
</tr>
</tbody>
</table>

\(^{1/}\) *Statistically significant (P<0.05).

Table A-6. Comparison of individual treatment means for dry matter intake by goats (g/kg body wt.) using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa Mean intake Mean difference</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100 54.7 +(^{1/}) + + - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>40 60 46.7 - - - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20 42.0 - - - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40 40.0 - - - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 80 38.4 - - - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80(^{2/}) 20 35.7 - - - -</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\(^{1/}\) (+) = significant differences (P<0.05). (-) = no significant difference (P\(\geq\)0.05).

\(^{2/}\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-7. Mean comparison of in vivo dry matter digestibility by goats using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diets</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak</td>
<td>% Alfalfa</td>
<td>Mean</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>100</td>
<td>56.9</td>
<td>+1/</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td>54.0</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>80/2</td>
<td>20</td>
<td>54.0</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td>53.0</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td>50.9</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>49.8</td>
<td>-</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P≥0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-8. Nitrogen intake by goats (g/head) over a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80(^1/)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goat</td>
<td>% Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>353.6</td>
<td>--</td>
<td>272.4</td>
<td>--</td>
<td>233.2</td>
<td>--</td>
</tr>
<tr>
<td>2</td>
<td>336.9</td>
<td>--</td>
<td>294.2</td>
<td>--</td>
<td>262.0</td>
<td>--</td>
</tr>
<tr>
<td>3</td>
<td>275.8</td>
<td>--</td>
<td>278.8</td>
<td>--</td>
<td>238.9</td>
<td>--</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>244.4</td>
<td>--</td>
<td>239.4</td>
<td>--</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>161.4</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>215.4</td>
<td>183.3</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>200.3</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>295.1</td>
<td>--</td>
<td>294.4</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>177.0</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
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<td>--</td>
<td>--</td>
<td>170.8</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>204.2</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>295.4</td>
<td>210.0</td>
<td>272.4</td>
<td>211.6</td>
<td>243.4</td>
<td>210.7</td>
</tr>
</tbody>
</table>

\(^1/\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-9. Nitrogen retained by goats (g/head) over a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80(^1/)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goat</td>
<td>% Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>86.5</td>
<td>--</td>
<td>29.2</td>
<td>--</td>
<td>44.5</td>
<td>12.1</td>
</tr>
<tr>
<td>2</td>
<td>60.9</td>
<td>--</td>
<td>63.4</td>
<td>--</td>
<td>64.0</td>
<td>17.6</td>
</tr>
<tr>
<td>3</td>
<td>56.4</td>
<td>--</td>
<td>50.5</td>
<td>--</td>
<td>49.2</td>
<td>23.6</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>46.4</td>
<td>--</td>
<td>55.0</td>
<td>20.3</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>10.4</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>64.7</td>
<td>5.5</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>21.6</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>42.7</td>
<td>--</td>
<td>52.1</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>17.7</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>33.4</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>47.4</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>67.1</td>
<td>20.0</td>
<td>47.4</td>
<td>37.6</td>
<td>53.2</td>
<td>18.4</td>
</tr>
</tbody>
</table>

\(^1/\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-10. Apparent digestibility of nitrogen (%) by goats.

<table>
<thead>
<tr>
<th>Diet</th>
<th>Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80&lt;sup&gt;1/&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>%</td>
<td>%</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>73.4</td>
<td>56.9</td>
<td>47.6</td>
<td>53.3</td>
<td>49.3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>73.9</td>
<td>58.7</td>
<td>49.0</td>
<td>50.7</td>
<td>53.1</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>3</td>
<td>71.9</td>
<td>59.1</td>
<td>46.9</td>
<td>50.7</td>
<td>53.1</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>4</td>
<td>63.5</td>
<td>63.5</td>
<td>50.7</td>
<td>53.3</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>5</td>
<td>68.8</td>
<td>68.8</td>
<td>50.7</td>
<td>53.3</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>6</td>
<td>72.9</td>
<td>62.6</td>
<td>49.0</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>7</td>
<td>66.3</td>
<td>66.3</td>
<td>51.2</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>8</td>
<td>61.2</td>
<td>61.2</td>
<td>56.9</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>9</td>
<td>56.9</td>
<td>56.9</td>
<td>56.9</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>10</td>
<td>56.9</td>
<td>56.9</td>
<td>56.9</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>11</td>
<td>56.9</td>
<td>56.9</td>
<td>56.9</td>
<td>50.7</td>
<td>53.3</td>
<td>49.0</td>
<td>53.3</td>
</tr>
<tr>
<td>Average</td>
<td>73.0</td>
<td>64.7</td>
<td>59.5</td>
<td>53.5</td>
<td>48.5</td>
<td>50.7</td>
<td></td>
</tr>
</tbody>
</table>

<sup>1/</sup> Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-11. Analysis of variance for nitrogen intake, retained nitrogen, and apparent digestibility of nitrogen by goats.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Nitrogen intake</th>
<th>Retained nitrogen</th>
<th>Digestibility of nitrogen</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td>5</td>
<td>5358.2*1/</td>
<td>1465.1*</td>
<td>349.5*</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>1921.3</td>
<td>164.2</td>
<td>6.7</td>
</tr>
</tbody>
</table>

1/ *Statistically significant (P<0.05).

Table A-12. Individual mean comparison for nitrogen intake (g/head) over a 7-day period using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td>Mean intake</td>
<td>Mean differences</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>-----------------</td>
<td>-------------</td>
<td>------------------</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>295.4</td>
<td>+1/</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>40 60</td>
<td>272.4</td>
<td>+</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>243.4</td>
<td>-</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>211.6</td>
<td>-</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>802/ 20</td>
<td>210.7</td>
<td>-</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 80</td>
<td>210.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P<0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-13. Mean comparison for nitrogen retained, over a 7-day period using Duncan's New Multiple Range test.

<table>
<thead>
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<th>Range (Number of means within a comparison)</th>
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<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>67.1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>53.2</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>40 60</td>
<td>47.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>37.6</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>20 80</td>
<td>20.0</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>18.4</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P<0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-14. Mean comparison for nitrogen digestibility using Duncan's New Multiple Range test.

<table>
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<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td>Digestibility mean</td>
<td>Mean differences</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>-----------------</td>
<td>-------------------</td>
<td>-----------------</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>73.0</td>
<td>4.3 4.2 4.1 4.0 3.8</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 80</td>
<td>64.7</td>
<td>+ + + +</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>40 60</td>
<td>59.5</td>
<td>+ + -</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>53.5</td>
<td>+ -</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>50.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>48.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P ≤ 0.05). (-) = no significant difference (P ≥ 0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-15. Fecal nitrogen excreted by goats (g/head) over a 7-day period.

<table>
<thead>
<tr>
<th>Goat % Alfalfa</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80(^1/)</th>
</tr>
</thead>
<tbody>
<tr>
<td>% Oak</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>94.1</td>
<td>--</td>
<td>117.4</td>
<td>--</td>
<td>122.1</td>
<td>104.9</td>
</tr>
<tr>
<td>2</td>
<td>87.8</td>
<td>--</td>
<td>121.5</td>
<td>--</td>
<td>134.4</td>
<td>115.8</td>
</tr>
<tr>
<td>3</td>
<td>77.5</td>
<td>--</td>
<td>113.9</td>
<td>--</td>
<td>126.9</td>
<td>106.6</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>89.3</td>
<td>--</td>
<td>118.1</td>
<td>89.1</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>50.4</td>
<td>--</td>
<td>--</td>
<td>--</td>
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<tr>
<td>6</td>
<td>58.4</td>
<td>68.5</td>
<td>--</td>
<td>--</td>
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<tr>
<td>7</td>
<td>--</td>
<td>67.5</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>114.6</td>
<td>--</td>
<td>147.7</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>86.3</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>74.6</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>88.1</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>79.5</td>
<td>75.2</td>
<td>110.5</td>
<td>99.2</td>
<td>125.3</td>
<td>104.1</td>
</tr>
</tbody>
</table>

\(^1/\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-16. Urinary nitrogen excreted by goats (g/head) over a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goat</td>
<td>% Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>173.0</td>
<td>125.8</td>
<td>66.6</td>
<td>89.9</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>188.2</td>
<td>109.3</td>
<td>63.6</td>
<td>93.6</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>141.9</td>
<td>114.4</td>
<td>62.9</td>
<td>87.7</td>
<td></td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>108.6</td>
<td>66.3</td>
<td>81.6</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>100.6</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>92.3</td>
<td>109.3</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>111.2</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>137.7</td>
<td>94.6</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>73.0</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>62.8</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>68.7</td>
<td>--</td>
<td>--</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Average</td>
<td>148.8</td>
<td>114.7</td>
<td>114.5</td>
<td>74.8</td>
<td>64.8</td>
<td>88.2</td>
</tr>
</tbody>
</table>

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-17. Analysis of variance for fecal nitrogen and urinary nitrogen excreted by goats.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Mean Squares</th>
<th>Fecal nitrogen</th>
<th>Urinary nitrogen</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td>5</td>
<td>1440.6*¹/</td>
<td></td>
<td>3853.3*</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>410.7</td>
<td>388.6</td>
<td></td>
</tr>
</tbody>
</table>

¹/ *Statistically significant (P<0.05).

Table A-18. Mean comparison for nitrogen in feces using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>125.3</td>
<td>34.0</td>
<td>33.6</td>
<td>33.2</td>
<td>31.5</td>
</tr>
<tr>
<td>40 60</td>
<td>110.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80²/ 20</td>
<td>104.1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>99.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>79.5</td>
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<td></td>
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</tr>
<tr>
<td>20 80</td>
<td>75.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

¹/ (+) = significant differences (P<0.05). (-) = no significant difference (P<0.05).

²/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-19. Mean comparison for nitrogen in urine using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak</td>
<td>% Alfalfa</td>
<td>Nitrogen in urine</td>
<td>Mean differences</td>
<td></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>100</td>
<td>148.8</td>
<td>+1/</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td>114.7</td>
<td></td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td>114.5</td>
<td></td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>802/</td>
<td>20</td>
<td>88.2</td>
<td></td>
<td></td>
<td>+</td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td>74.8</td>
<td></td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>64.8</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P>0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-20. Gross energy consumed by goats (Mcal/head) over a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80¹/²</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goat % Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>58.3</td>
<td>--</td>
<td>49.3</td>
<td>--</td>
<td>46.6</td>
<td>38.9</td>
</tr>
<tr>
<td>2</td>
<td>55.6</td>
<td>--</td>
<td>53.2</td>
<td>--</td>
<td>52.3</td>
<td>42.7</td>
</tr>
<tr>
<td>3</td>
<td>45.5</td>
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<td>50.5</td>
<td>--</td>
<td>47.7</td>
<td>41.0</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>44.2</td>
<td>--</td>
<td>47.8</td>
<td>35.9</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>27.1</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>35.5</td>
<td>30.8</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>33.6</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>49.5</td>
<td>--</td>
<td>56.9</td>
<td>--</td>
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<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>34.2</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>33.0</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>39.5</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>48.7</td>
<td>35.2</td>
<td>49.3</td>
<td>40.9</td>
<td>48.6</td>
<td>39.6</td>
</tr>
</tbody>
</table>

¹/² Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-21. Digestibility of energy (%) by goats.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>Goat % Alfalfa</th>
<th>Diets</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80¹/₁</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
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<td></td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>57.6</td>
<td>--</td>
<td>51.9</td>
<td>--</td>
<td>49.4</td>
<td>--</td>
<td>52.2</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>58.4</td>
<td>--</td>
<td>51.9</td>
<td>--</td>
<td>48.6</td>
<td>--</td>
<td>51.5</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>57.1</td>
<td>--</td>
<td>53.3</td>
<td>--</td>
<td>49.3</td>
<td>--</td>
<td>52.7</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td></td>
<td>55.2</td>
<td>--</td>
<td>53.3</td>
<td>--</td>
<td>54.0</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td></td>
<td>57.9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>55.8</td>
<td>51.9</td>
<td>--</td>
<td>--</td>
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<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>53.6</td>
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</tr>
<tr>
<td>8</td>
<td>--</td>
<td>54.3</td>
<td>--</td>
<td>50.8</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>47.4</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>53.0</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>52.9</td>
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<td></td>
</tr>
<tr>
<td>Average</td>
<td>57.2</td>
<td>54.4</td>
<td>53.1</td>
<td>51.0</td>
<td>50.1</td>
<td>52.6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

¹/₁ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-22. Energy metabolized by goats (Mcal/head) over a 7-day period.

| % Oak | 00 | 20 | 40 | 60 | 80 | 80 1/ 
|-------|----|----|----|----|----|------
| Goat  | % Alfalfa | 100 | 80 | 60 | 40 | 20 | 20 | 20 |
| 1     | 29.7 | -- | 22.8 | -- | 21.5 | 15.9 |
| 2     | 28.3 | -- | 25.2 | -- | 23.9 | 17.0 |
| 3     | 22.9 | -- | 24.4 | -- | 22.1 | 16.9 |
| 4     | --   | -- | 22.0 | -- | 24.0 | 15.3 |
| 5     | --   | 13.5 | -- | -- | -- | -- |
| 6     | 17.8 | 13.6 | -- | -- | -- | -- |
| 7     | --   | 15.5 | -- | -- | -- | -- |
| 8     | --   | 23.8 | -- | 26.7 | -- | -- |
| 9     | --   | -- | -- | 14.6 | -- | -- |
| 10    | --   | -- | -- | 16.1 | -- | -- |
| 11    | --   | -- | -- | 19.2 | -- | -- |
| Average | 24.7 | 16.6 | 23.6 | 19.1 | 22.9 | 16.3 |

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-23. Analysis of variance for gross energy consumed, digestible energy, and metabolized energy by goats.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Mean Squares</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Gross energy consumed</td>
</tr>
<tr>
<td>Diets</td>
<td>5</td>
<td>141.2</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>59.4</td>
</tr>
</tbody>
</table>

1/ *Statistically significant (P<0.05).

Table A-24. Mean comparison of digestibility of gross energy using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>57.2</td>
<td>+1/</td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>20 80</td>
<td>54.4</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>40 60</td>
<td>53.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>802/2</td>
<td>52.6</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>60 40</td>
<td>51.0</td>
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<td>-</td>
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<tr>
<td>80 20</td>
<td>50.1</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P>0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-25. Mean comparison for metabolized energy using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak % Alfalfa</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mean metabolized energy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00 100</td>
<td>24.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>40 60</td>
<td>23.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>22.9</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>60 40</td>
<td>19.1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20 80</td>
<td>16.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>80 20</td>
<td>16.3</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = significant differences (P<0.05). (-) = no significant difference (P<0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-26. Gross energy content (Mcal) of feces excreted during a 7-day period.

<table>
<thead>
<tr>
<th>% Oak</th>
<th>00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80(^1/)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Goat</td>
<td>% Alfalfa</td>
<td>100</td>
<td>80</td>
<td>60</td>
<td>40</td>
<td>20</td>
</tr>
<tr>
<td>1</td>
<td>24.7</td>
<td>--</td>
<td>23.7</td>
<td>--</td>
<td>--</td>
<td>23.6</td>
</tr>
<tr>
<td>2</td>
<td>23.1</td>
<td>--</td>
<td>25.6</td>
<td>--</td>
<td>--</td>
<td>26.9</td>
</tr>
<tr>
<td>3</td>
<td>19.5</td>
<td>--</td>
<td>23.6</td>
<td>--</td>
<td>--</td>
<td>24.2</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>19.8</td>
<td>--</td>
<td>--</td>
<td>22.3</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>11.4</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>15.7</td>
<td>14.8</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>15.6</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>22.6</td>
<td>--</td>
<td>28.0</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>18.0</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>15.5</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>18.6</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>20.8</td>
<td>16.1</td>
<td>23.2</td>
<td>20.0</td>
<td>24.3</td>
<td>18.9</td>
</tr>
</tbody>
</table>

\(^1/\) Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-27. Energy (Mcal) excreted in urine by goats during a 7-day period.

<table>
<thead>
<tr>
<th>Goat % Alfalfa</th>
<th>% Oak 00</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>80 1/</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>3.9</td>
<td>--</td>
<td>2.8</td>
<td>--</td>
<td>1.5</td>
<td>4.5</td>
</tr>
<tr>
<td>2</td>
<td>4.2</td>
<td>--</td>
<td>2.4</td>
<td>--</td>
<td>1.4</td>
<td>4.9</td>
</tr>
<tr>
<td>3</td>
<td>3.1</td>
<td>--</td>
<td>2.5</td>
<td>--</td>
<td>1.3</td>
<td>4.7</td>
</tr>
<tr>
<td>4</td>
<td>--</td>
<td>--</td>
<td>2.4</td>
<td>--</td>
<td>1.5</td>
<td>4.1</td>
</tr>
<tr>
<td>5</td>
<td>--</td>
<td>2.2</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>6</td>
<td>2.0</td>
<td>2.3</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>7</td>
<td>--</td>
<td>2.5</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>8</td>
<td>--</td>
<td>3.1</td>
<td>--</td>
<td>2.2</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>9</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>1.6</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>10</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>1.3</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>11</td>
<td>--</td>
<td>--</td>
<td>--</td>
<td>1.6</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td>Average</td>
<td>3.3</td>
<td>2.6</td>
<td>2.5</td>
<td>1.7</td>
<td>1.4</td>
<td>4.6</td>
</tr>
</tbody>
</table>

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-28. Analysis of variance for fecal energy and urinary energy excreted by goats.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>Degrees of freedom</th>
<th>Fecal energy</th>
<th>Urinary energy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets</td>
<td>5</td>
<td>35.1</td>
<td>5.2*1/</td>
</tr>
<tr>
<td>Error</td>
<td>18</td>
<td>13.5</td>
<td>0.2</td>
</tr>
</tbody>
</table>

*Statistically significant (P<0.05).

Table A-29. Mean comparison for energy content in urine using the Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Number of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diets % Oak % Alfalfa Mean energy content</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.8</td>
<td>0.8</td>
<td>0.7</td>
<td>0.7</td>
<td>0.7</td>
</tr>
<tr>
<td>801/</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>4.6</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>00</td>
<td>3.3</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>20</td>
<td>2.6</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>2.5</td>
<td>+</td>
<td>+</td>
<td></td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>1.7</td>
<td></td>
<td></td>
<td></td>
<td>-</td>
</tr>
<tr>
<td>80</td>
<td>1.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.

2/ (+) = significant differences (P<0.05). (-) = no significant difference (P<0.05).
Table A-30. Blood and serum parameters for goats fed an alfalfa and five oak-containing diets.

<table>
<thead>
<tr>
<th>Oak (%)</th>
<th>Alfalfa (%)</th>
<th>Hemoglobin g·100 ml⁻¹</th>
<th>SGOT units·ml⁻¹</th>
<th>BUN mg·100ml⁻¹</th>
<th>PCV %</th>
</tr>
</thead>
<tbody>
<tr>
<td>00</td>
<td>100</td>
<td>13.3±0.4⁴¹/</td>
<td>93.1± 2.2</td>
<td>25.9±1.6</td>
<td>36.7±2.8</td>
</tr>
<tr>
<td>20</td>
<td>80</td>
<td>13.5±1.8</td>
<td>102.0±10.9</td>
<td>18.8±2.6</td>
<td>35.9±5.9</td>
</tr>
<tr>
<td>40</td>
<td>60</td>
<td>14.4±0.7</td>
<td>100.0± 6.6</td>
<td>19.9±1.7</td>
<td>39.3±2.0</td>
</tr>
<tr>
<td>60</td>
<td>40</td>
<td>13.3±1.6</td>
<td>93.6±11.8</td>
<td>21.1±6.2</td>
<td>38.6±4.5</td>
</tr>
<tr>
<td>80</td>
<td>20</td>
<td>13.5±0.8</td>
<td>88.9± 8.9</td>
<td>18.7±5.6</td>
<td>39.3±2.3</td>
</tr>
<tr>
<td>80²²/</td>
<td>20</td>
<td>13.5±0.8</td>
<td>95.1±12.6</td>
<td>19.4±2.8</td>
<td>38.6±3.3</td>
</tr>
<tr>
<td>Average</td>
<td></td>
<td>13.6±0.3</td>
<td>95.4± 4.2</td>
<td>20.6±1.4</td>
<td>38.1±1.3</td>
</tr>
</tbody>
</table>

¹/ Means ±95% confidence intervals.

²/ Oak collected in June during the season of rapid growth. Oak of the other diets was collected in August after elongation had ceased and stems had hardened.
Appendix B

Measurements and Statistical Tests Related to Determination of In Vitro Digestibility
Table A-31. Analysis of variance for diets, methods, and inoculum effect on in vitro dry matter digestibility.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>df</th>
<th>Sum of squares</th>
<th>Mean squares</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inoculum</td>
<td>5</td>
<td>593.2</td>
<td>118.6*</td>
</tr>
<tr>
<td>Methods IVAP-IVND</td>
<td>1</td>
<td>6744.7</td>
<td>6744.6*</td>
</tr>
<tr>
<td>Diets</td>
<td>5</td>
<td>773.4</td>
<td>154.7*</td>
</tr>
<tr>
<td>Inoculum X Methods</td>
<td>5</td>
<td>22.5</td>
<td>4.5*</td>
</tr>
<tr>
<td>Methods X Diets</td>
<td>5</td>
<td>449.0</td>
<td>89.8*</td>
</tr>
<tr>
<td>Inoculum X Diets</td>
<td>25</td>
<td>79.6</td>
<td>3.2*</td>
</tr>
<tr>
<td>Inoculum X Methods X Diets</td>
<td>25</td>
<td>82.6</td>
<td>3.3*</td>
</tr>
<tr>
<td>Error</td>
<td>61</td>
<td>92.1</td>
<td>1.5</td>
</tr>
<tr>
<td>Total</td>
<td>132</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Statistically significant (P<0.05)
Table A-32. Mean comparison for effect of inoculum upon in vitro dry matter digestibility using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Range (Numbers of means within a comparison)</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample size</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>% Oak</td>
<td>% Alfalfa</td>
<td>Mean</td>
<td>Mean differences</td>
<td></td>
<td></td>
</tr>
<tr>
<td>23</td>
<td>00</td>
<td>100</td>
<td>65.5</td>
<td>+1/</td>
<td>+</td>
</tr>
<tr>
<td>22</td>
<td>20</td>
<td>80</td>
<td>65.4</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>21</td>
<td>60</td>
<td>40</td>
<td>64.8</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>23</td>
<td>40</td>
<td>60</td>
<td>64.5</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>24</td>
<td>80</td>
<td>20</td>
<td>63.3</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>22</td>
<td>802/</td>
<td>20</td>
<td>59.3</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = statistically significant (P<0.05). (-) = no significant difference (P>0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-33. Mean comparison for effect of oak content of diet upon in vitro dry matter digestibility of oak rations using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Sample size</th>
<th>Diets</th>
<th>Mean differences</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% Oak</td>
<td>% Alfalfa</td>
</tr>
<tr>
<td>26</td>
<td>00</td>
<td>100</td>
</tr>
<tr>
<td>27</td>
<td>20</td>
<td>80</td>
</tr>
<tr>
<td>23</td>
<td>40</td>
<td>60</td>
</tr>
<tr>
<td>26</td>
<td>60</td>
<td>40</td>
</tr>
<tr>
<td>27</td>
<td>80</td>
<td>20</td>
</tr>
</tbody>
</table>

1/ (+) = statistically significant (P<0.05).

Table A-34. Mean comparisons for dry matter digestibility between acid pepsin-neutral detergent methods using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Sample size</th>
<th>Methods</th>
<th>Mean digestibility</th>
</tr>
</thead>
<tbody>
<tr>
<td>63</td>
<td>IVND</td>
<td>71.1</td>
</tr>
<tr>
<td>67</td>
<td>IVAP</td>
<td>56.4</td>
</tr>
</tbody>
</table>

1/ (+) = statistical significant (P<0.05).
Table. A-35. Analysis of variance for inoculum, months, and drying temperatures affects on in vitro dry matter digestibility of oak forage.

<table>
<thead>
<tr>
<th>Source of variation</th>
<th>df</th>
<th>Sum of square</th>
<th>Mean squares</th>
</tr>
</thead>
<tbody>
<tr>
<td>Inoculum</td>
<td>5</td>
<td>282.3</td>
<td>56.5*</td>
</tr>
<tr>
<td>Months</td>
<td>3</td>
<td>2029.7</td>
<td>676.6*</td>
</tr>
<tr>
<td>Temperatures</td>
<td>5</td>
<td>1023.8</td>
<td>204.8*</td>
</tr>
<tr>
<td>Inoculum X Months</td>
<td>15</td>
<td>149.2</td>
<td>9.9</td>
</tr>
<tr>
<td>Inoculum X Temperatures</td>
<td>25</td>
<td>84.8</td>
<td>3.4</td>
</tr>
<tr>
<td>Months X Temperatures</td>
<td>15</td>
<td>336.5</td>
<td>24.2*</td>
</tr>
<tr>
<td>Error</td>
<td>59</td>
<td>376.3</td>
<td>6.4</td>
</tr>
<tr>
<td>Total</td>
<td>127</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ Statistically significant (P<0.05).
Table A-36. Mean comparison for inoculum effect on in vitro dry matter digestibility of oak forage using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Sample size</th>
<th>Diets</th>
<th>Mean digestibility</th>
<th>6</th>
<th>5</th>
<th>4</th>
<th>3</th>
<th>2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>% Oak</td>
<td>% Alfalfa</td>
<td>1.8</td>
<td>1.8</td>
<td>1.7</td>
<td>1.7-1.6</td>
<td>1.6-1.5</td>
</tr>
<tr>
<td>22</td>
<td>60</td>
<td>40</td>
<td>42.4</td>
<td></td>
<td>+1/</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>00</td>
<td>100</td>
<td>41.9</td>
<td></td>
<td>+</td>
<td>+</td>
<td>-</td>
</tr>
<tr>
<td>24</td>
<td>40</td>
<td>80</td>
<td>41.6</td>
<td></td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>24</td>
<td>20</td>
<td>60</td>
<td>41.4</td>
<td></td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>24</td>
<td>80</td>
<td>20</td>
<td>39.6</td>
<td></td>
<td></td>
<td></td>
<td>+</td>
</tr>
<tr>
<td>23</td>
<td>802/</td>
<td>20</td>
<td>37.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

1/ (+) = statistically significant differences (P<0.05). (-) = no significant difference (P>0.05).

2/ Oak collected in June during the season of rapid growth. Oak in the other diets was collected in August after elongation had ceased and stems had hardened.
Table A-37. Mean comparison for digestibility of oak forage collected in different months using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Sample size</th>
<th>Months</th>
<th>Mean digestibility</th>
<th>Mean differences</th>
</tr>
</thead>
<tbody>
<tr>
<td>41</td>
<td>June</td>
<td>47.4</td>
<td>±1/</td>
</tr>
<tr>
<td>40</td>
<td>July</td>
<td>41.2</td>
<td>+</td>
</tr>
<tr>
<td>40</td>
<td>September</td>
<td>37.4</td>
<td>-</td>
</tr>
<tr>
<td>40</td>
<td>August</td>
<td>37.1</td>
<td></td>
</tr>
</tbody>
</table>

1/ (±) = statistically significant (P<0.05). (-) = no significant difference (P>0.05).

Table A-38. Mean comparison for drying temperature affects on in vitro dry matter digestibility using Duncan's New Multiple Range test.

<table>
<thead>
<tr>
<th>Sample size</th>
<th>Temperatures °C</th>
<th>Mean digestibility</th>
<th>Mean differences</th>
</tr>
</thead>
<tbody>
<tr>
<td>26</td>
<td>-2</td>
<td>45.8</td>
<td>±1/</td>
</tr>
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<tr>
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1/ (+) = statistically significant (P<0.05). (-) = no significant difference (P>0.05).
Table A-39. Solubility (%) of microbia cells by acid pepsin and neutral detergent.

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