

EFFECTS OF THREE PRACTICAL DIETS ON FEEDING BEHAVIOR,
NUTRITIONAL STATUS, RUMEN HEALTH, AND GROWTH OF CAPTIVE MULE
DEER (ODOCOILEUS HEMIONUS) FAWNS

By

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Abstract

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Digestive upset and failure to thrive are common among captive exotic ruminants, especially browsers, often resulting from improper nutrition. Zoo nutritionists have begun including more soluble fiber and less starch in pelleted diets in an attempt to correct these problems. Therefore, we examined the effects of feeding 24 juvenile mule deer (*Odocoileus hemionus*) 3 complete pelleted diets with different starch, fiber, and crude protein content on behavior, physiology and growth. At birth (May 2007), mule deer were randomly assigned to one of the 3 diets, which included low-starch, high-fiber (LSHF; 51.6 % NDF, 3.0 % starch, and 14.8 % CP), high-starch, low-fiber (HSLF; 33.3 % NDF, 20.0 % starch, and 19.6 % CP), and moderate-starch, low-fiber (MSLF; 35.7 % NDF, 16.6 % starch, and 19.3 % CP) diets. Deer were fed these diets *ad libitum* with no more than 25 % alfalfa hay from 10 d old until September 2008 (68 wk old). During 5

sampling periods beginning in November 2007 and spaced 6 to 12 weeks apart thereafter, we measured intake, digestion and passage, rumen chemistry and morphology, growth, and body condition. Intake, mean retention time of particles, and DM and apparent energy digestibility did not differ among diets. However, crude protein digestibility was greatest in MSLF, neutral detergent solubles digestibility greatest in HSLF, and NDF digestibility greatest in LSHF. The higher fiber content of LSHF led to longer rumination and feeding times of animals fed this diet than those fed either HSLF or MSLF diets. At an earlier age, animals consuming the higher starch diets tended to have poorer fecal consistency than those consuming the LSHF diet, but all groups improved over time. For animals consuming LSHF, blood acetate and ruminal acetate:propionate tended to be higher and BUN lower than for those animals consuming either HSLF or MSLF. Deer had the same growth and body fat among treatments. Our findings show pelleted diets with low starch and high fiber met the energy and protein requirements of growing mule deer, a medium-sized browsing ruminant, with potentially fewer digestive problems, as well as grain-based diets.

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CHAPTER ONE

FEEDING ECOLOGY AND PLANT DYNAMICS: IMPLICATIONS FOR FEEDING EXOTIC RUMINANTS IN CAPTIVITY

Exotic (i.e., non-domesticated) ruminants are difficult to feed in captivity because they tolerate or require fibrous plant foods yet often consume inadequate amounts of grass or alfalfa hay and few descriptive or experimental data are available on their natural feeding behavior and nutrient requirements. Ruminants that are adapted to consuming the foliage and parts of trees, shrubs, forbs, flowers or fruit (i.e., browsers) present particular challenges when formulating diets for life in captivity. Here, I review what is known about nutrition and feeding ecology of exotic ruminants.

Nutritional Content of Forages

Plants provide the most abundant source of food energy on the planet, but much of the energy is unavailable to vertebrates unless they contain symbiotic microflora in their digestive systems. Cellulose, a structural carbohydrate, makes up a major portion of plant cell walls. Together with hemicellulose and lignin, these structural carbohydrates give the plant its structure and rigidity and are quantitatively referred to as neutral detergent fiber (NDF; Goering and Van Soest, 1970; Van Soest, 1994). These components of the cell are indigestible by mammalian enzymes (Van Soest, 1994). Pectin, another portion of cell wall, is easily fermented but at a more gradual rate than the nonstructural carbohydrates (e.g., starches, sugars) present in the cell solubles that form the rest of the cell (Strobel and Russell, 1986; Van Soest, 1994; NRC, 2007). The cell solubles also contain proteins, lipids, plant defense chemicals, and minerals. Proteins in

plants are variable depending on stage of growth, plant part, and type of plant. Plant foliage is naturally low in fat, ranging from 3 to 10% DM depending on the species and plant part (Byers and Schelling, 1988). Plants also produce a variety of chemicals that serve secondary metabolic functions (i.e., plant secondary metabolites - PSM), such as pollination, protection from the environment, and defense against herbivory (Van Soest, 1994; Metlen et al., 2009). Chemical defenses, such as tannins, flavonoids, and terpenoids, serve to reduce the nutritional quality or are toxic to ruminant herbivores by binding with nutrients to form insoluble, indigestible complexes, inhibiting gut microbial and enzymatic activity, disrupting metabolic pathways, and decreasing intake by decreasing palatability (Robbins, 1983; Van Soest, 1994; Foley et al., 1999; Makkar, 2003; Marsh et al., 2006). Mineral content in plants often reflects underlying soil chemistry, but include Ca, P, Mg, K, Na, S, Cu, Co, Mn, and Mo (Robbins, 1983; Kincaid, 1988; Van Soest, 1994; Metlen et al., 2009).

The nutritional quality of forages varies among plant species, plant parts, stage of development, photosynthetic pathway (C3 v. C4, see Mooney, 1997; Van Soest, 1994), geography, soil type, season, and temperature (Van Soest, 1994; NRC 2007). For example, plant species have been classified taxonomically and physically as monocots (grasses and sedges) and dicots (woody browses and forbs). Because monocots and dicots differ in growth form, placement of apical meristem, structural components of cell wall, and PSM, they offer different nutritional resources for ruminants.

Monocots are generally comprised of a relatively thick cell wall made up of 20 to 40% cellulose and 15 to 40% hemicelluloses (Robbins, 1983). Neutral detergent fiber values for these can range from 50 to 70% dry matter (DM; Duncan and Poppi, 2008;).

PSMs are typically absent from monocotyledonous forage (Robbins, 1983). On the other hand, dicots usually exhibit a relatively thin, lignified cell wall (including a greater proportion of the soluble fiber, pectin) that encompasses a high proportion of cell solubles (NDF = 30 to 50% DM; Robbins, 1983; Duncan and Poppi, 2008). Because a greater proportion of the monocot cell is fiber, monocots generally contain less protein than dicots. However, PSM present in dicots, such as tannins, bind with plant protein and make it unavailable to the animal (Robbins 1983; Van Soest, 1994).

Extracting Nutrients from Forages

Evolution has magnificently engineered the four-chambered stomach and the symbiotic microorganisms (protozoa, bacteria, fungi) of ruminant animals to subsist on plant foods high in cellulose. Through convergent evolution, a number of mammalian taxa, including ruminants in Artiodactyla, have evolved digestive systems that house microorganisms that ferment cellulose into energy-rich byproducts (e.g., volatile fatty acids [VFA]). The two major locations for fermentation are pre-gastric (foregut) or post-gastric (hindgut), each with its own advantage and disadvantage in extracting nutrients from plants.

Hindgut fermenters ferment cellulose in the cecum (e.g., smaller animals like rodents and rabbits), or the colon (e.g., larger animals like horses and elephants). Because food is digested enzymatically in the true stomach first, hindgut fermenters absorb nutrients more efficiently from the non-fiber, digestible cell solubles in the plant than does the foregut fermenter (Demment and Van Soest, 1985; Van Soest, 1996). Although cellulose is fermented in the hindgut where VFAs are absorbed, the flow of particles out of the system is not impeded. Thus fibrous particles pass relatively rapidly

through the digestive system, which means the animal can digest the most nutritious plant parts and excrete the remainder.

In the foregut fermenter, cell solubles are fermented before they reach the true stomach, thus the microbes have first access to plant nutrients. Therefore, cell solubles are less efficiently digested by the foregut fermenter. Cellulose is also fermented and the end products absorbed in the foregut, before moving to the true stomach. In ruminant foregut fermenters, passage of plant fiber is delayed in the foregut (rumen-reticulum) by the reticulo-omasal orifice. Cell wall particles must be re-chewed (ruminated) and fermented to a small enough size to move on through the digestive tract. The mastication and moistening of foods in the mouth are important in increasing surface area, reducing particle size, and increasing particle density. This allows for greater microbial access to food items. As a result, ruminants more efficiently and thoroughly digest plant fiber than do hindgut fermenters. Approximately 95% of fiber consumed is digested in the rumen (Huhtanen, 2006). The extent of fiber digestion in the foregut depends on the characteristics of the cell wall, amount of chewing, composition of the rumen microbial ecosystem, pH of the rumen, and rate of intake (Owens and Goetsch, 1988; Van Soest, 1994). However, delaying passage of plant fiber reduces intake in ruminants, which can be problematic on extremely fibrous diets (Van Soest, 1994; Allen, 1996). A further benefit of foregut fermentation is that the animal can digest and absorb microbial protein as it flushes out of the rumen-reticulum and into the true stomach and lower tract, whereas in hindgut fermenters, microbial protein is lost to all but cecotrophic species.

The major end products of fermentation, VFAs (acetate, propionate, and butyrate), depend on the amount of structural and non-structural carbohydrates in the diet.

Diets high in fibrous forage and low in non-structural carbohydrates (starches, sugars) favor cellulolytic flora, which produce greater proportions of acetate. In contrast, higher levels of starches favor amylolytic flora, which produce more propionate (Van Soest, 1994; Owens and Goetsch, 1988).

Volatile fatty acids are utilized in different biochemical pathways. For example, propionate is a major precursor for gluconeogenesis by the liver, where the majority of propionate is metabolized (van Houtert, 1993). Minimal (1 to 4%) acetate is metabolized by the rumen epithelium, and the bulk enters the portal blood stream, contributing to lipogenesis (van Houtert, 1993; Van Soest, 1994). On the other hand, up to 90% of the butyrate produced is metabolized by the rumen wall for epithelial development (Van Soest, 1994; Hofmann, 1988). In addition, acetate, and butyrate provide oxidative energy sources in the citric acid cycle (Van Soest, 1994). The absorption of VFAs and other fermentative end products is hastened by papillae lining the rumen wall which increase the absorptive surface area of the rumen epithelium (Van Soest, 1994).

After cycling within the rumen ecosystem, particles are either filtered through the reticulo-omasal orifice or re-chewed during rumination. Stimulating salivation, rumination and chewing serve as important buffering sources for the rumen environment (Church, 1988; Mertens, 1997). Rumination also reduces particle size, allowing passage through the omasal orifice for further digestion in the omasum, abomasum (the 'true' acid-secreting stomach) and intestines (Owens and Goetsch, 1988; Van Soest, 1994). Similar to hindgut fermenters, the small intestine of ruminants is the major site for enzymatic digestion and absorption of amino acids, fatty acids, sugars and other nutrients that escape the foregut (Robbins, 1983; Merchen, 1988). The colon, or large intestine, in

ruminants serves chiefly as an absorptive site for electrolytes, minerals, water, nitrogen, and VFAs and has limited fermentation (Van Soest, 1994). This multi-chambered system allows efficient fermentation of high fiber foods, but presents challenges when feeding ruminant animals.

Nutritional Ecology

Because plants differ in morphology and nutrient composition at many spatial and temporal scales, herbivores have differentiated into feeding niches. Hofmann (1973) classified domestic and exotic herbivores as browsers (stem, leaf, and fruit eaters), grazers (grass and roughage eaters) or mixed feeders (intermediate feeders) based on the proportion of grasses (herbaceous monocots) and browses (woody plants and herbaceous dicots) they consumed. He characterized anatomical differences among ruminant herbivores based on this classification. Although more recent studies suggest that body size, rather than feeding strategy, accounts for these anatomical and physiological differences among ruminants (Robbins et al., 1995; Demment and Van Soest, 1985; Gordon and Illius, 1994; Codron et al., 2007), Hofmann's (1973) classification provides a foundation for understanding diet choice and for diet formulation in captivity.

Grazing animals have developed digestive systems adapted to feeding on grasses and sedges. Grasses have relatively thick cell walls consisting mainly of cellulose. Grasses typically grow in a two-dimensional, relatively homogenous sward. Grazers tend to have a large reticulo-rumen, a small reticulo-omasal orifice, and short dense papillae. These adaptations are expected to prolong retention in the rumen for more complete digestion of cellulose (Demment and Van Soest, 1985; Van Soest 1994). To more efficiently harvest grasses, grazers tend to have wide muzzles (Gordon and Illius, 1988;

Janis and Ehrhardt, 1988). Grazers typically exhibit high-crowned (hypsodont) teeth, a trait that is hypothesized to have evolved as a result of high silica content (and thus abrasive quality) of natural forage (Van Soest, 1994). Because animals ruminate in direct proportion with the amount of cell wall ingested, the higher portion of cell wall ingested by grazers leads to longer rumination episodes when compared to browsers consuming dicotyledonous forage (Van Soest, 1994).

Browsers, on the other hand, feed mostly on leaves, twigs, fruits, flowers, vines or other dicotyledonous plants. Because browsers tend to have thinner cell walls, with the cell wall being more lignified and less digestible, these animals tend to have smaller rumens, larger reticulo-omasal orifices, thicker and denser papillae, and larger hindguts. These adaptations allow quick absorption of rapidly fermented cell solubles, and allow very indigestible portions of the plant to escape the rumen rapidly. Because they are less efficient at digesting cellulose, browsers use narrow muzzles and prehensile lips and tongues to efficiently select and harvest the most digestible plants and plant parts. Because selective foraging takes time, browsers tend to be small, requiring less food per day. In addition, browsers with smaller rumens tend to feed in smaller bouts interspersed with frequent rumination periods (Hofmann, 1973; 1988). Unlike grasses, dicots tend to produce many plant secondary metabolites that can be toxic or reduce the nutritional quality of forages (e.g., condensed tannins). Therefore, browsers tend to have larger livers for detoxification and larger parotid salivary glands that secrete salivary binding proteins which bind to condensed tannins, which can form insoluble complexes with the proteins in the plant (Hofmann, 1988; Robbins, 1983)

Captive Feeding of Ruminants

Much of what is known about the nutrient requirements of ruminants has been obtained using controlled experiments on domestic livestock. While domestic animals can provide a baseline for formulating rations in captivity, caution must be exercised in selecting a model ruminant for formulating diets for exotic ruminants. Most domestic livestock are classified as grazers (except for goats which are considered intermediate feeders), whereas grazers represent only ¼ of ruminant species (Hofmann, 1989). Because harvesting and digesting food differs in several ways between browsers and grazers, diets formulated for domestic animals may be inadequate for the majority of exotic herbivores.

Furthermore, diets formulated for domestic animals are designed to meet production demands, such as milk, wool, or meat. The diets necessary to provide domestic animals with adequate energy supply to meet short-term production demands use energy-yielding, readily fermentable, cost-effective ingredients like oats, wheat, corn and barley. These ingredients exhibit high fermentation rates, rapidly producing acids within the rumen (Hummel et al., 2006a, b). The resulting drop in ruminal pH makes for a difficult challenge for livestock managers to ensure their animals eat at levels consistent with production goals while balancing the rumen environment against acidosis (see Stone, 2004). Exotic ruminants, however, are unique in that quality and longevity of life are often top priority. Exotic herbivores are not faced with production demands and therefore consuming rations for domestic animals means that they are consuming diets that far surpass their energy requirements or physiological limits.

Surplus energy and inadequate consumption of forage can manifest in several metabolic disorders, including bloat (Cole et al., 1945; Essig et al., 1988; Cheng et al.,

1998), hoof overgrowth and laminitis (Nocek, 1997; Garrett et al., 1998), rumenitis (Thomson, 1967), gastro-intestinal tract obstructions (Wenninger, 1999; Davis et al., 2009), and urolithiosis (Woolf et al., 1973; Wolfe et al., 2000). Of particular concern and frequently reported among captive ruminants is ruminal acidosis. This condition is the result of rapid fermentation of soluble carbohydrates without the presence of adequate fiber leading to an accumulation of unbuffered acids and concurrent drop in pH of the rumen environment (Essig et al., 1988). Ruminal acidosis causes diarrhea by altering motility and absorptive capacity of the GIT and has also been linked to inappetite, laminitis, and liver and lung abscesses (Garry, 2002; Essig et al. 1988; Nocek, 1997; Stone 2004). In addition, the excess absorption of acids from the rumen into the blood stream can cause systemic acidosis, which leads to dehydration, reduced renal function, and death (Essig et al., 1988). Moreover, decreased pH increases susceptibility of rumen wall to hemorrhaging and inflammation, which can lower resistance to systemic invasion by bacteria present in the rumen or externally introduced species. Lower pH also alters the microbial population, often resulting in increased numbers of harmful bacteria species that encourage enterotoxin production (Essig et al., 1988; Ternouth, 1988).

Clearly, formulating diets for exotic ruminants presents many challenges. One goal in the formulation of modern diets for exotic ruminants is to decrease the rapidly digested portion of the diet (starch) and increase the amount of fiber to levels that more closely mimic natural vegetation while still meeting their nutritional requirements. Because many exotic ruminants are reluctant or unable to consume adequate forages like alfalfa or grass hay and because providing large quantities of fresh browse can be logistically and financially difficult, many captive facilities rely on pelleted diets (Claus,

2003; Clauss and Dierenfeld, 2008). Citrus and beet pulp have recently been included as fiber sources in these rations, which have been readily accepted by many species of exotic ruminants and have had positive feedback on GIT health because of their balanced fermentation rates within the rumen (Van Soest, 1991, 1996; Shochat et al., 1997; Kearney, 2005; Hummel et al., 2006b). For some species, especially strict browsers like moose, sawdust has been used as a fiber source, offering higher cellulose without starch. However, few controlled experiments with an adequate sample size of exotic ruminants have been conducted that have allowed an objective test of the benefits and detriments of providing low starch, high fiber pelleted diets to exotic ruminants.

Therefore, in Chapter 2, I examine the responses of mule deer (*Odocoileus hemionus*) consuming 3 pelleted rations resembling commercially-available diets sold specifically for exotic ruminants. Mule deer are classified as browsers, preferentially consuming the foliage of woody plants (Robbins et al., 1995). By using a mule deer as a model, I hoped to gain a better understanding of how wild browsers respond when consuming diets in captivity that vary in starch content and have differing fiber sources and levels.

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CHAPTER TWO

THE EFFECTS OF STARCH AND FIBER IN PELLETTED DIETS ON BEHAVIOR, PHYSIOLOGY, AND GROWTH IN MULE DEER

INTRODUCTION

Formulating affordable and practical diets for exotic (i.e., non-domesticated) ruminants that provide adequate growth and reproduction while maintaining digestive health has proved challenging for zoos and wildlife agencies (Baker and Hobbs, 1985; Baker et al., 1998). Traditionally, exotic ruminants have been fed diets designed for domestic ruminants (e.g., cows, sheep, and goats). Pelleted and mixed rations made for domestic ruminants are typically formulated with ingredients such as cereal grains that are high in starches and other rapidly fermenting, non-structural carbohydrates to provide energy to meet short-term production demands (e.g., milk, meat, wool) at low cost to the producer (Van Soest, 1994; Kearney, 2005; Hummel et al., 2006b). However, these diets may be unsuitable for maintaining exotic ruminants that are not used for production because these animals may be unable to absorb and metabolize the end products of fermentation (volatile fatty acids, VFA) at a rate equal to their production. Excess acid in the blood (systemic acidosis) or rumen (rumen acidosis) compromises long-term health of ruminants by increasing the risk of laminitis, rumenitis, inappetite, weight loss, liver abscesses, or creates a general failure to thrive (Thomson, 1967; Essig et al., 1988; Nocek, 1997; Garry, 2002).

To offset the nutritional problems caused by consuming rapidly fermentable diets, both domestic and exotic ruminants are supplemented with long forages (e.g., grass or alfalfa hay). Because forages require more chewing, they stimulate saliva production,

which buffers acid production in the rumen (Mertens, 1997; Beauchemin et al., 2003). However, many exotic ruminants such as moose (*Alces alces*), blue duikers (*Cephalophus monticola*), okapis (*Okapia johnstoni*), and giraffes (*Giraffa camelopardalis*) that are strictly browsers (i.e., stem and leaf-eaters) are often reluctant to ingest or unable to digest adequate amounts of grass or alfalfa hay. Therefore, they may consume too much soluble carbohydrate and become at greater risk of acidosis than grazing ruminants (i.e., grass eaters) that readily ingest hay forage (Van Soest, 1994; Shochat et al., 1997; Dierenfeld et al., 2002; Clauss et al., 2003; Kearney, 2005; Clauss and Dierenfeld, 2008).

As a solution to this problem, ingredients like beet and citrus pulp have been introduced in pelleted and mixed rations to provide fibrous, energy-yielding substitutes for ingredients that are high in sugars or starches (Fegeros et al., 1995; Clark and Armentano, 1997; Kearney 2005; Clauss and Dierenfeld, 2008). Beet and citrus pulp have higher pectin and hemicellulose content and lower levels of non-structural carbohydrates, which slow ruminal fermentation and reduce lactate production, thereby reducing the potential for acidosis (Van Soest et al., 1991; Van Soest, 1994). Similar to long forage supplements, the higher fiber content of pelleted diets containing beet and citrus pulp are expected to increase the time spent chewing and ruminating, helping to stimulate saliva production and balancing rumen pH against the production of acids (Clark and Armentano, 1997).

However, the benefits of these new diets to animal behavior, physiology and growth have yet to be rigorously tested in browsing ruminants, especially during somatic growth. Therefore, we compared behavior (intake, feeding and rumination time),

physiology (digestibility, fecal scores, rate of passage, VFAs, blood urea nitrogen [BUN], serum Ca, P, and glucose, ruminal papillae, and pH of ruminal fluid) and growth and morphology (body mass, body fat, bone density, and frame size) of a growing browsing ruminant (mule deer, *Odocoileus hemionus*) fed 3 pelleted diets that differed in the content of starch, fiber, and crude protein.

We specifically hypothesized that mule deer fed diets higher in starch would have poorer fecal consistency than those fed lower starch diets as a result of a higher digestible dietary energy content, higher blood glucose concentrations and lower acetate:propionate ratios in the ruminal fluid caused by increased propionate production. Increased VFA production is expected to result in increased ruminal absorptive surface area via longer and denser ruminal papillae. On the other hand, we expected that growing mule deer fed diets with higher fiber and lower crude protein would consume more and digest less of the diet, ruminate longer, have a slower rate of passage, have lower BUN, less body fat, and slower growth than those on lower fiber diets.

METHODS

Study Animals and Diets

All research was conducted according to Washington State University's (WSU) Institutional Animal Care and Use Committee protocol #3705. We selected 24 mule deer fawns born at WSU's Wild Ungulate Facility (WUF) from captive females in May of 2007. Each fawn was removed from its dam within 24 h of birth for hand-rearing. Hand-rearing procedures followed those outlined in Parker and Wong (1985). The milk formula consisted of whole cow's milk mixed with powdered lamb milk replacer (10 %

milk weight) supplemented with 0.65 g powdered casein (sodium caseinate, Prestige Proteins, Boca Raton, FL), 0.35 g ground pediatric multi-vitamin with iron, and 0.70 g CaCO₃.

To compare digestion, physiology, and growth, we assigned fawns to one of three completely-balanced herbivore diets (Table 1, 2) at birth stratified by birth weight, sex, and date. Siblings were not assigned to the same diet. Three males and 5 females made up a treatment group. Animals were offered assigned diets *ad libitum* beginning at 10 d of age and throughout the experiment, except during the 5- to 7-d intake rate trials described in the following section. Alfalfa hay was restricted to 25% of total intake on an as fed basis.

Diets differed in ingredients and ranged from 15 to 19 % CP, 30 to 50 % NDF, and 3 to 20 % starch (Table 1, 2). The diets also differed in sources of carbohydrate. LSHF diet (low starch, high fiber) contained ground soybean hulls, dehulled soybean meal, dried beet pulp, ground aspen, and ground oat hulls. HSLF diet (high starch, low fiber) contained alfalfa meal, wheat middlings, ground corn and soybean meal. Finally, MSLF (moderate starch, low fiber) diet contained wheat middlings, alfalfa, oat hulls, canola meal, soy hulls and meal, and soft wheat. Sodium sesquicarbonate (NaSC) was added to LSHF (0.4 %) and rumen-undegradable protein was added to MSLF (soybean meal-based, 0.5 %). These diets were designed as practical diets for exotic ruminants, and attempts were made to standardize as many nutritional parameters as possible (e.g., minerals) in order to minimize variation.

Fawns were housed in 2 m x 4 m chain-link pens in groups of 2 or 3 with limited access (4 to 6 h/d) to grass pasture until they were 16 wk old, and then were housed by

treatment groups in 0.5-ha pastures composed of grass species (Table 3) that was mowed to the ground at regular intervals to minimize grass intake. We collected routine fecal samples from each group that were sent to the Washington Disease Diagnostic Laboratory (WADDL, Pullman, WA) for bacteriological and parasitological analyses. Additionally, all animals were subject to hematological screening during each sampling period to determine if blood parameters were within the normal range documented for ruminants. In the event treatment was necessary, animals were subject to an appropriate course of antibiotic therapy under the direction of WSU veterinary staff and allowed to stabilize before we collected samples from them.

We removed animals from the experiment if two of three criteria established *a priori* were experienced and did not improve with nutritional or antibiotic therapy. These criteria included 1) an animal lost over 15% body mass (BM; from peak value), 2) loose feces (fecal score < 50) observed for ≥ 4 wk with no identified pathogen, or 3) a continuous decline in intake (consuming < 2% body mass for over 1 wk) of the treatment diet. Nutritional support included the incorporation of one or more of the following in addition to the treatment diet: unmolassed beet pulp, FIBRevive (Oxbow Animal Health, Murdock, NE), Nutri-Cal (EVSCO Pharmaceutical, Buena, NJ), plain yogurt, Probios (Vets Plus, Knapp, WI), or electrolyte solution.

Intake and Digestion

Intake

During 5 sample periods beginning on 29 November 2007 (Period 1 = P1) when fawns were 26 wk old and every 6 to 12 wk thereafter (January 2008, P2; April 2008, P3;

June 2008, P4, and September 2008, P5), we measured dry matter intake (DMI, g/d) of the assigned diet and alfalfa cubes (restricted to $\leq 25\%$ of intake) by each animal daily for 7 d. During P1 to P3, we offered fawns four, 1-h meals per day in individual feeders, and in P4 and P5, we reduced them to three, 1-h meals per day. All meals were offered *ad libitum* until 1 h passed.

Feeding Activity

To compare rumination time of animals on each of the three diets, we observed feeding activity for one 24-h period for each diet using scan sampling in August 2008. Animals were denied pasture access for a period of 24 h before and during rumination observations. Every 5 min during the 24-h observation period, observers classified behavior of all animals as 1) ruminating 2) feeding or 3) other activities.

Digestion

To determine the digestibility of dry matter (DMD), apparent energy (AED), protein (CPD), neutral detergent fiber (NDFD), and NDF solubles (NDS) of the diets, we conducted total collection digestion trials with each animal between 5 May and 8 August in 2008. Animals were housed in 1.5 m x 1.5 m digestion crates for a 2-d acclimation period followed by a 5-d digestion trial. Feces fell to collection screens and urine was funneled into a pan underneath the crate. Approximately 60 mL of acetic acid was added to the collection pan each day to maintain a pH below 7 to reduce loss of nitrogen via ammonia.

During the trial period, we weighed the daily ration of pelleted diet and fed it *ad libitum*, along with water *ad libitum* to each deer. A fresh sample of food was weighed and dried at 100° C to correct for dry matter composition. Residual food and feces were

collected each morning and weighed. A subsample of feces was dried at 100° C for 24 h to correct for DM content. A second subsample was frozen. Urine was collected, the volume measured and a subsample (2 % total volume) frozen for subsequent analysis.

To determine the nutrient content of food, feces and urine, we first ground fecal and food samples in a Wiley Mill to pass a 1-mm screen. Fecal and food samples were analyzed for gross energy (GE, bomb calorimetry), nitrogen (N, carbon-nitrogen analyzer – TruSpec®, LECO Corporation, St. Joseph, MI), and fiber composition (neutral detergent fiber, NDF; acid detergent fiber, ADF; acid detergent lignin, ADL; Goering and Van Soest, 1970) using Ankom filter bag technology (Ankom, Fairport, NY). Urine samples were analyzed for GE and N content as described previously. Apparent DMD, AED, CPD, and digestibility of NDF and NDF solubles were calculated as in Robbins (1983). Digestible energy intake (DEI, MJ/d) was estimated as the product of DMI, GE (KJ/g) and AED, and digestible protein intake (DPI, g protein/d) was the product of DMI, dietary crude protein content (g protein/g DM) and CPD.

Mean Retention Time

To determine the mean retention time of diet particles within the digestive tract of our mule deer, we first marked NDF of a sample of each pelleted diet with YbNO₃. Samples were digested in neutral detergent fiber solution, rinsed with distilled water and dried in an oven at 60° C for 48 h. Digested diet samples were then soaked in YbNO₃ for 12 h, rinsed to remove unbound marker and dried for 24 h at 60° C (Moore et al., 1992).

After acclimating 3 deer selected from each treatment (1 male and 2 females) to the digestion crates for 24 h, we gave each animal a 0.5-g oral pulse-dose of the labeled NDF from their assigned diet, along with their assigned diet and water *ad libitum*. After

dosing, feces were collected at 2-h intervals through hour 40 post-dosing, after which feces were collected every 4 h through the end of the trial (hour 72). Total feces produced was measured at each collection interval, from which a subsample of 30 to 100 g was collected and dried at 60° C for a minimum of 48 h and ground in a Wiley Mill to pass a 1-mm screen.

To determine the Yb concentration of fecal samples, we weighed approximately 1 g of dried, ground feces into a porcelain crucible and ashed this sample at 550° C for 5 h. Ashed samples were transferred into 100-mL tubes and 10 mL of 3N HCl and 2 drops of HNO₃ were added. The solution was then gently boiled for approximately 10 min. Upon cooling, the liquid was passed through #4 filter paper into a 100-mL volumetric flask. Filter paper and remaining residue were rinsed 3 times with distilled water, allowing the rinse to filter into the flask. The solution was then brought to volume with distilled water. The concentration of Yb (ppm) in the fecal samples was determined with inductively coupled plasma mass spectrometry (ICP-MS) using model HP 4500 (Agilent Technologies, Palo Alto, CA) at WSU's GeoAnalytical Laboratory.

We estimated mean retention times for each animal by fitting the following equation (Brandt and Thacker, 1958; Sakaguchi et al., 1987) to the downward portion of the marker concentration curve:

$$Y = Y_0 e^{-kt},$$

where Y is the marker concentration in feces at time t, Y₀ is constant, k is the rate-constant, and t is the time interval after feeding of the markers. MRT equaled the sum of the reciprocal of k and the transit time, which is the initial appearance of the marker after dosing (Sakaguchi et al., 1987).

Fecal Scores

Each week from 10 October 2007 (just before P1) to 9 September 2008 (P5), we visually scored fecal consistency of each animal from 0 (watery diarrhea with no consistency) to 100 (firm, individual pellets, Appendix A).

Diet Preference

To determine whether starch and fiber content of the diet influenced the deer's preference for consuming a diet, we conducted preference trials with 12 of the female mule deer (4 from each treatment) after P5. Each deer was allowed *ad libitum* access to both HSLF and LSHF diets simultaneously for at least one month preceding the preference trial. During February and May 2009, animals were housed in 2 m x 4 m isolation pens and given a 24-h adjustment period to housing prior to trial. For 5 d, both HSLF and LSHF diets were offered *ad libitum* in 4-L buckets fastened adjacently to the isolation pen wall. Intake (as fed basis) of the pelleted diets was calculated daily by weighing the residual food and subtracting it from the amount fed.

Rumen and Blood Chemistry

To obtain samples of blood and ruminal fluid at the end of each of the 5 sampling periods, animals were sedated using 0.3 mg/kg BM xylazine hydrochloride administered intra-muscularly (IM). As a reversal, yohimbine hydrochloride at 0.25 mg/kg BW or tolazoline hydrochloride at 4 mg/kg BM was administered intra-venously (IV).

To compare rumen chemistry among pelleted diets, we collected ruminal fluid during the 5 sampling periods using a tygon gastric tube attached to a fluid evacuator and volumetric flask. We also collected ruminal fluid at time of necropsy. The pH was

measured immediately upon collection using an electronic pH meter. We strained the ruminal fluid through 4 layers of cheese cloth and the resulting fluid was acidified with sulfuric acid to a pH of ≤ 2 in order to stop microbial fermentation. The fluid was then refrigerated up to 2 d until it could be centrifuged at 1300 x g for 20 min at 10° C. Decanted fluid was frozen for later analysis. Ruminal fluid was analyzed for acetic, propionic, and butyric acid (mol/100 mol; millimolar concentration [mM]) profiles using a gas chromatograph at Dairy One Forage Laboratory (Ithaca, NY; Autosystem XL, PerkinElmer, Waltham, MA). Following fluid collection at each of the 5 sampling periods, animals were given a prophylactic dose of either enrofloxacin (Baytril, 7.5 mg/kg) or florfenicol (Nuflor, 40 mg/kg) subcutaneously.

To compare plasma VFA (mM); BUN; and serum glucose, Ca, P, and Ca:P among diets, we collected 20 mL blood from sedated animals via jugular venipuncture into 10-mL serum tubes and 10-mL ethylenediaminetetraacetic acid (EDTA) tubes during the 5 sampling periods. Blood samples were immediately spun into serum or plasma using a centrifuge at 1300 x g at 20 °C. Serum was analyzed for serum chemistry at WSU Clinical Pathology Laboratory (Dimension Xpand plus Chemistry Analyzer, Siemen's Diagnostics, Deerfield, IL) and trace minerals at Michigan State University's Diagnostic Center for Population and Animal Health (DCPAH, Lansing, MI). As a qualitative measure of animal health, whole blood samples were analyzed for hematology at WSU Clinical Pathology Laboratory (Advia 120 Hematology Analyzer, Siemen's Diagnostics, Deerfield, IL). Plasma samples were submitted to Dairy One Forage Laboratory and analyzed for VFA concentration using methods previously described for ruminal fluid samples.

Growth and Body Condition

To monitor growth of mule deer during the experiment, we measured BM of deer to the nearest 0.1 kg using an electronic platform scale weekly to once every 2 wk from birth in May 2007 to September 2008, the completion of the study. During each of the 5 sampling periods, we measured to the nearest 1 mm the length of the hind leg from the tip of the hoof to the end of the calcaneus. We also measured maximum subcutaneous rump fat thickness (MAXFAT; Stevenson et al., 1998; Cook et al., 2007) and thickness of the *longissimus dorsi* muscle (Herring et al., 1995; Cook et al., 2001) between the 12th and 13th rib, adjacent to the spine (hereafter called loin depth), using a portable ultrasonograph (Sonovet 600, Medison Corp. Universal Medical, Newbedford Hills, NY).

Tissue and Bone Sampling

After P5 in September 2008, we sedated 8 males and 1 female deer (3/diet) with xylazine hydrochloride. Animals were transported to WSU Veterinary Teaching Hospital for Dual X-ray Absorptiometry (DEXA) scan (Hologic QDR 4500 Acclaim Series Elite, Bedford, MA), which measured: bone mineral composition (BMC), bone mineral density (BMD), fat mass, lean muscle mass, total mass, and percent fat. Animals were then euthanized using pentobarbital (Beuthanasia-D Special) at 10 mg/kg BM.

To examine the health of digestive tissues qualitatively, we collected samples from the reticulum, rumen, omasum, abomasum, duodenum, jejunum, ileum, cecum, colon, kidney, and liver and preserved in 10% formalin. Histological samples of each tissue were embedded in paraffin wax and cut into sections 4 μ (microns) thick and

mounted on glass microscope slides. They were subsequently stained with haematoxylin and eosin and examined microscopically for abnormalities (e.g., swelling, hemorrhaging) by pathologists at WADDL.

To examine length and density of rumen papillae, a 6 cm x 10 cm section of the cranioventral rumen wall was preserved separately in 10% formalin from each of the 9 animals necropsied. From this section, we measured the length (mm) of the longest 10 papillae from five, 1-cm² subsamples using a caliper. We also measured density of papillae in the same five, 1-cm² subsamples using a dissecting microscope.

To quantify Ca and P content and to measure length, antlers were cleaned of tissue and cut below the pedicel using a band-saw. A separate DEXA was performed on each of 8 sets of antlers to determine BMC, BMD, total area, and total mass of antlers. We measured the diameter, circumference, and length of the main beam from both antlers and averaged them for a final value. We also recorded the total number of points for both antlers.

Samples of antlers and metacarpal bones were analyzed for Ca and P at DCPAH. Samples were first de-fatted with ether in a soxlet extractor, dried, then ashed at 500 ° C for 8 h. This ash was solubilized in HNO₃ and the resulting solution was analyzed using inductively coupled plasma-mass spectroscopy (Agilent 7500 CE, Agilent Technologies, Palo Alto, CA)

Statistical Analysis

We compared intake, body condition, blood and rumen chemistry measurements collected during the 5 sampling periods among diets, sampling periods, and diet x period

interactions (d x p) using mixed linear models (PROC MIXED SAS ver. 9.1.3, SAS Inst., Cary, NC, USA) with a repeated measures design for animal and sampling period. If global effects were significant ($\alpha = 0.05$), we assessed comparisons among least squares means. Likewise, we compared weekly fecal score measurements among diets, weeks, and diet x week interactions using mixed linear models. For measurements collected only once (e.g., feeding and rumination times, digestibility, and body condition during necropsies), we used 1-way analysis of variance (PROC ANOVA) to compare among diets. As a measure of preference, we compared the amount of HSLF and LSHF consumed by each deer using a paired t-test (PROC TTEST). All means reported as mean \pm standard error.

RESULTS

Four of 8 animals on HSLF, 5 of 8 animals on MSLF, and 3 of 8 animals on LSHF received injectable antibiotics before the end of the 18-mon trial period. Three of 8 animals on HSLF exhibited severe, prolonged diarrhea with no parasitic or bacterial origins and declining body condition, thus were removed from the experiment in November and December 2007 and January 2008. Despite nutritional and antibiotic therapy, 2 of the animals eventually died. Likewise, one female on MSLF was removed from the experiment in January 2008 because of prolonged diarrhea and failed response to nutritional and antibiotic support, and one female on LSHF was removed from the experiment in January 2008 due to loss of appetite and body condition. From these, hematology results showed elevated white blood cells in 1 HSLF and 1 LSHF individual, indicating an immune response to infection. This left 5 animals on HSLF, 7 in MSLF,

and 7 in LSHF by P3. We also noticed severe hair plucking, particularly elevated in the winter months, in a few individuals in each treatment group. For all other individuals, clinical hematology and serum chemistry results were within normal reference range for sheep and goats determined by WSU Clinical Pathology Laboratory. Glucose was higher than average across all treatments (124.65 – 153.01 mg/dL) in our mule deer when compared to sheep and goats at 46 – 96 mg/dL (Table 4, Appendix B).

Intake and Digestion

Intake, Rumination and Feeding Time

Across the 5 sampling periods, animals fed 3 pelleted diets with $\leq 25\%$ supplemental alfalfa cubes *ad libitum* during meal periods had different daily DMI ($F_{2,19} = 4.93$, $P = 0.02$, Table 5) and DMI relative to body mass (DMI_{BM} , $F_{2,19} = 7.31$, $P = 0.004$, Table 5). As the animals grew, DMI increased with sampling period ($F_{4,60} = 23.70$, $P < 0.0001$), with a similar intake level during P1 to P3 ($779.17 \text{ g} \pm 55.55$ to $797.46 \text{ g} \pm 55.45$), but greater DMI during P4 and P5 ($1175.83 \text{ g} \pm 63.34$ to $1418.44 \text{ g} \pm 92.94$). However, there was no $d \times p$ interaction ($F_{8,60} = 1.39$, $P = 0.22$) for DMI. Similarly, DMI_{BM} also differed between periods ($F_{4,60} = 4.26$, $P = 0.004$) with a similar level during P1 to P3 ($19.93 \text{ g/kg BM} \pm 1.15$ to $24.59 \text{ g/kg BM} \pm 1.49$), and similar DMI_{BM} during P4 and P5 ($21.05 \text{ g/kg BM} \pm 1.25$ to $23.01 \text{ g/kg BM} \pm 0.94$). There were no $d \times p$ interaction ($F_{8,60} = 1.54$, $P = 0.16$) for DMI_{BM} .

During meal feeding, animals fed HSLF consumed 27 % less total DM and DM_{BM} than those fed LSHF (DMI: $t = 2.81$, $P = 0.01$, DMI_{BW} : $t = 3.15$, $P = 0.005$) and 29 % less than those fed MSLF (DMI: $t = 2.74$, $P = 0.01$, DMI_{BM} : $t = 3.57$, $P = 0.002$, Table

5). Animals in LSHF and MSLF did not differ in DMI ($t = 0.03$, $P = 0.98$) or DMI_{BM} ($t = 0.53$, $P = 0.60$, Table 5). However, during digestion trials when animals had 24-h access to pellets, DMI did not differ among diets ($F_{2,16} = 1.41$, $P = 0.27$, Table 5). Additionally, there was a $d \times p$ interaction for the intake (DM) of alfalfa ($F_{8,19} = 2.54$, $P = 0.05$). Intake of alfalfa cubes across all diets differed among periods ($F_{4,19} = 17.85$, $P < 0.0001$) and decreased over time, with higher intake during P1 (Nov) through P3 (April; DMI: 94 – 173 g/d) and lower intake during P4 (June) and P5 (Sept.; DMI: 61 – 76 g/d). However, over the length of the trial, DMI of alfalfa did not differ among diets ($F_{2,19} = 0.62$, $P = 0.55$, Table 5), nor was the alfalfa:pellet intake ratio different ($F_{2,19} = 0.07$, $P = 0.94$, Table 5).

Animals fed LSHF spent more time feeding ($F_{2,16} = 5.63$; $P = 0.01$) than those consuming HSLF ($t = 2.59$; $P = 0.02$) and MSLF ($t = 3.08$; $P = 0.007$), but feeding times of MSLF and HSLF were not different ($t = 0.22$; $P = 0.83$, Table 5). Likewise, animals fed LSHF spent twice as much time ruminating ($F_{2,16} = 14.41$; $P = 0.0003$) than those consuming HSLF ($t = 3.63$; $P = 0.002$) and 2.8 times that of MSLF ($t = 5.18$; $P = < 0.0001$), but rumination times of MSLF and HSLF were not different ($t = 1.10$, $P = 0.29$, Table 5).

Digestibility and Mean Retention Time

Despite differences in amount and sources of fiber and nonstructural carbohydrates, DMD ($F_{2,16} = 2.74$, $P = 0.09$) and AED ($F_{2,16} = 0.94$, $P = 0.41$, Table 5) did not differ among diets. However, NDS ($F_{2,16} = 10.22$, $P = 0.001$) and CPD ($F_{2,16} = 8.05$, $P = 0.004$), and NDFD ($F_{2,16} = 37.86$, $P < 0.0001$, Table 5) varied with diet. LSHF had lower NDS than HSLF ($t = 4.51$, $P = 0.0004$) but was similar to MSLF ($t = 1.81$, P

= 0.09). HSLF had higher NDS than MSLF ($t = 2.86$, $P = 0.01$). Likewise, MSLF had higher CPD than HSLF ($t = 2.26$, $P = 0.04$) and LSHF ($t = 3.98$, $P = 0.001$). HSLF and LSHF were not different in CPD ($t = 1.37$, $P = 0.19$, Table 5). HSLF had lower NDFD than both LSHF ($t = 8.7$, $P < 0.0001$) and MSLF ($t = 4.83$, $P = 0.0002$, Table 5). LSHF NDFD was higher than MSLF ($t = 4.24$, $P = 0.0006$).

As a consequence, DEI and DEI with respect to body mass (DEI_{BM}) differed among treatments during the intake trials conducted each sampling period (DEI: $F_{2,19} = 5.4$, $P = 0.01$; DEI_{BM} : $F_{2,19} = 8.38$, $P = 0.003$, Table 5), but were similar during the digestion trials because of similar DMI and digestibility among treatments (DEI: $F_{2,16} = 1.08$, $P = 0.36$; DEI_{BM} : $F_{2,16} = 0.46$, $P = 0.64$, Table 5). During P1 to P5, HSLF had lower DEI and DEI_{BM} than did LSHF (DEI: $t = 2.86$, $P = 0.01$; DEI_{BM} : $t = 3.21$, $P = 0.005$) and MSLF (DEI: $t = 2.96$, $P = 0.008$; DEI_{BM} : $t = 3.91$, $P = 0.0009$). MSLF and LSHF did not differ in DEI ($t = 0.12$, $P = 0.90$) nor DEI_{BM} ($t = 0.79$, $P = 0.44$).

The DPI and DPI with respect to body mass (DPI_{BM}) of pellets also differed among treatments during the intake trials conducted each sampling period (DPI: $F_{2,19} = 14.38$, $P = 0.0002$; DPI_{BM} : $F_{2,19} = 33.18$, $P < 0.0001$, Table 5), and during the digestion trial ($F_{2,16} = 7.36$, $P = 0.005$; DPI_{BM} : $F_{2,16} = 16.26$, $P = 0.0001$). For P1 to P5, animals fed MSLF had 1.5 times higher DPI and DPI_{BM} than those fed HSLF ($t = 4.79$, $P = 0.0001$; DPI_{BM} : $t = 6.82$, $P < 0.0001$) and LSHF (DPI: $t = 4.37$, $P = 0.0003$; DPI_{BM} : $t = 7.19$, $P < 0.0001$). Animals fed HSLF and LSHF had similar DPI ($t = 0.77$, $P = 0.45$) and DPI_{BM} ($t = 0.12$, $P = 0.91$). During the digestion trial, animals fed MSLF had higher DPI and DPI_{BM} than both HSLF (DPI: $t = 2.63$, $P = 0.02$; DPI_{BM} : $t = 3.11$, $P = 0.007$) and LSHF (DPI: $t = 3.68$, $P = 0.002$; DPI_{BM} : $t = 5.67$, $P < 0.0001$). However, there was no

difference in DPI between HSLF and LSHF ($t = 0.73$; $P = 0.48$). Mean retention time of particles was also similar among animals on each treatment ($19.9 \text{ h} \pm 1.7$, $F_{2,6} = 1.95$, $P = 0.22$, Table 5, Appendix C).

Preference

When diets were offered *ad libitum* and simultaneously without supplemental forage, mule deer ate similar amounts of HSLF (DMI: $749.2 \text{ g/d} \pm 159.8$) and LSHF (DMI: $703.3 \text{ g/d} \pm 128.4$, $t = -0.19$, $P = 0.86$).

Rumen and Blood Chemistry

Rumen Fluid Volatile Fatty Acid Profiles and Microbial Populations

Acetate within the ruminal fluid collected via gastric tubing during the 5 sampling periods differed among diets ($F_{2,20} = 7.22$, $P = 0.004$, Table 4) and periods ($F_{4,57} = 5.77$, $P = 0.0006$) with a significant $d \times p$ interaction ($F_{8,57} = 3.96$; $P = 0.0009$). Overall, ruminal fluid from animals consuming LSHF had higher acetate concentration than those consuming HSLF ($t = 2.19$, $P = 0.04$) and MSLF ($t = 3.77$, $P = 0.001$), but acetate did not differ between animals fed HSLF and MSLF ($t = 1.36$, $P = 0.19$). Acetate was higher in ruminal fluid from LSHF animals during P1 ($F_{2,16} = 8.13$; $P = 0.004$), P3 ($F_{2,16} = 8.60$; $P = 0.003$), and P5 ($F_{2,14} = 7.44$; $P = 0.006$) but did not differ among treatment groups during P2 ($73.5 \% \pm 1.1$; $F_{2,15} = 0.72$; $P = 0.50$) or P4 ($69.7 \% \pm 0.90$; $F_{2,16} = 0.40$; $P = 0.68$). However, acetate concentration in the ruminal fluid collected from 9 animals during necropsy did not differ among diets ($F_{2,6} = 4.18$, $P = 0.07$, Table 4).

Propionate within ruminal fluid collected via gastric tubing was similar among animals across treatments ($F_{2,20} = 1.56$, $P = 0.24$, Table 4), but differed among periods

($F_{4,57} = 6.67$, $P = 0.0002$) and there was a $d \times p$ interaction ($F_{8,57} = 2.52$; $P = 0.02$).

Ruminal propionate did not differ among treatment groups during P1 ($20.2 \% \pm 0.8$; $F_{2,16} = 1.61$; $P = 0.23$), P2 ($18.3 \% \pm 0.8$; $F_{2,15} = 0.84$; $P = 0.45$), P3 ($21.2 \% \pm 0.89$; $F_{2,16} = 2.83$; $P = 0.089$), and P5 ($16.0 \% \pm 0.58$; $F_{2,14} = 3.01$, $P = 0.0815$). However, during P4, propionate was higher in animals consuming LSHF ($20.4 \% \pm 1.25$, $F_{2,16} = 3.44$, $P = 0.057$) than in those consuming HSLF ($17.2 \% \pm 0.9$, $t = 2.27$, $P = 0.04$) and MSLF ($17.6 \% \pm 0.57$; $t = 2.20$, $P = 0.04$). Animals consuming HSLF and MSLF did not differ in ruminal propionate concentration ($t = 0.26$, $P = 0.80$). Propionate in ruminal fluid collected from 9 animals during necropsy was similar among diets ($F_{2,6} = 0.70$, $P = 0.53$, Table 4).

Across the trial, the acetate to propionate ratio (A:P) for ruminal fluid collected via gastric tubing was similar in animals on regardless of diet ($F_{2,20} = 3.25$, $P = 0.06$, Table 4), but differed among periods ($F_{4,57} = 5.86$, $P = 0.0005$) and had $d \times p$ interactions ($F_{8,57} = 2.80$; $P = 0.01$). Animals consuming LSHF had higher A:P during P1 (4.34 ± 0.45 , $F_{2,16} = 3.56$, $P = 0.05$) and P3 (4.10 ± 0.27 , $F_{2,16} = 3.93$, $P = 0.04$) than those consuming MSLF (P1: 3.11 ± 0.21 , $t = 2.67$, $P = 0.02$; P3: 3.17 ± 0.30 , $t = 2.39$, $P = 0.03$) and HSLF (P3: 3.08 ± 0.27 , $t = 2.40$, $P = 0.03$). However, A:P was similar among animals on each treatment during P2 ($F_{2,15} = 0.99$, $P = 0.39$), P4 ($F_{2,16} = 1.98$, $P = 0.17$), and P5 ($F_{2,14} = 3.04$, $P = 0.08$). Likewise across diets, A:P was similar in ruminal fluid collected from animals during necropsy ($F_{2,6} = 1.14$, $P = 0.38$, Table 4).

Butyrate in the ruminal fluid collected from animals via gastric tubing varied among treatments overall ($F_{2,20} = 12.29$, $P = 0.0003$, Table 4) and among periods ($F_{4,20} = 4.04$, $P = 0.01$), but had no $d \times p$ interaction ($F_{8,57} = 1.20$, $P = 0.31$). Butyrate was lower

in animals consuming LSHF than those consuming HSLF ($t = 3.42$, $P = 0.003$) and MSLF ($t = 4.78$, $P = 0.0001$), but butyrate was similar in animals consuming MSLF and HSLF ($t = 1.10$, $P = 0.28$).

Butyrate in the ruminal fluid of 9 necropsied animals varied among diets ($F_{2,6} = 5.17$, $P = 0.05$, Table 4). Animals consuming LSHF had a lower proportion of butyrate in the ruminal fluid at the time of necropsy than animals consuming HSLF ($t = 3.11$, $P = 0.02$). However, animals fed LSHF and MSLF had a similar proportion of butyrate in the ruminal fluid ($t = 2.28$, $P = 0.06$), as did those fed HSLF and MSLF ($t = 0.83$, $P = 0.44$).

Blood Chemistry

Blood acetate concentrations of animals varied among treatments ($F_{2,20} = 13.25$, $P = 0.0002$, Table 4) and periods ($F_{4,20} = 42.05$, $P < 0.0001$) with a $d \times p$ interaction ($F_{8,20} = 2.77$, $P = 0.03$). Across periods, blood acetate was 34% higher in animals consuming LSHF than those consuming HSLF ($t = 4.18$, $P = 0.0005$) and MSLF ($t = 4.59$, $P = 0.0002$), but was not different between those consuming HSLF and MSLF ($t = 0.12$, $P = 0.91$). When analyzed by period, blood acetate was higher for animals consuming LSHF in P2 ($0.58 \text{ mM} \pm 0.05$, $F_{2,20} = 4.06$, $P = 0.03$) and P3 ($0.69 \text{ mM} \pm 0.04$, $F_{2,16} = 9.77$, $P = 0.002$) than those consuming HSLF (P2: $0.47 \text{ mM} \pm 0.03$, $t = 2.12$, $P = 0.05$; P3: $0.45 \text{ mM} \pm 0.03$, $t = 4.00$, $P = 0.001$) and MSLF (P2: $0.44 \text{ mM} \pm 0.03$, $t = 2.69$, $P = 0.01$; P3: $0.50 \text{ mM} \pm 0.04$, $t = 3.50$, $P = 0.003$). Blood acetate of animals consuming HSLF and MSLF did not differ (P2: $t = 0.48$, $P = 0.64$; P3: $t = 0.80$, $P = 0.44$), and blood acetate did not differ among diets during P1 ($0.29 \text{ mM} \pm 0.05$, $F_{2,20} = 0.73$, $P = 0.49$), P4 ($0.35 \text{ mM} \pm 0.02$, $F_{2,16} = 2.36$, $P = 0.13$) and P5 ($0.48 \text{ mM} \pm 0.03$, $F_{2,16} = 2.72$, $P = 0.10$). On the other

hand, blood propionate was similar among diets ($F_{2,20} = 0.16$, $P = 0.85$, Table 4), averaging 0.04 ± 0.04 mM.

Blood urea nitrogen (BUN) varied across diets ($F_{2,20} = 9.32$, $P = 0.001$, Table 4), periods ($F_{4,20} = 14.88$, $P < 0.0001$), with a d x p interaction ($F_{8,20} = 4.57$, $P = 0.003$). Across periods, animals fed LSHF had lower BUN than those fed HSLF ($t = 3.36$, $P = 0.003$) and MSLF ($t = 3.97$, $P = 0.0008$), but HSLF and MSLF animals did not differ ($t = 0.48$, $P = 0.64$). When analyzed by period, animals fed LSHF had a lower BUN during P1 (22.37 mg/dL ± 1.19 , $F_{2,19} = 4.97$, $P = 0.02$) and P5 (22.29 mg/dL ± 1.06 , $F_{2,16} = 19.99$, $P < 0.0001$) than both HSLF (P1: 31.57 mg/dL ± 3.44 , $t = 2.82$, $P = 0.01$; P5: 34.00 mg/dL ± 1.76 , $t = 4.36$, $P = 0.0005$) and MSLF (P1: 30.71 mg/dL ± 2.06 ; $t = 2.56$, $P = 0.02$; P5: 37.14 mg/dL ± 2.32 ; $t = 6.06$, $P < 0.0001$). However, the BUN of animals fed HSLF and MSLF was similar (P1: $t = 0.25$, $P = 0.80$; P5: $t = 1.17$, $P = 0.26$). BUN did not vary among dietary treatments during P2 ($F_{2,17} = 2.29$, $P = 0.13$), P3 ($F_{2,16} = 3.05$, $p = 0.08$) and P4 ($F_{2,16} = 2.96$, $P = 0.08$).

Serum glucose concentration was similar among treatments ($F_{2,20} = 1.33$, $P = 0.29$, Table 4). However, serum glucose differed among periods ($F_{3,48} = 3.68$, $P = 0.02$), but did not have a d x p interaction ($F_{6,48} = 0.53$, $P = 0.78$). Serum glucose was highest during P4 (155.05 mg/dL ± 6.85) and P5 (147.58 mg/dL ± 5.54) but lower during P2 (131.57 mg/dL ± 7.14) and P3 (130.84 mg/dL ± 4.68). We did not examine serum glucose during P1.

Serum Ca levels ($F_{2,20} = 14.52$, $P = 0.0001$) and Ca:P ($F_{2,20} = 5.06$, $P = 0.02$, Table 4) differed among treatments. Animals fed LSHF had higher serum Ca than those fed HSLF ($t = 4.86$, $P < 0.0001$) and MSLF ($t = 4.33$, $P = 0.0003$). As a result of higher

serum Ca, LSHF animals had higher serum Ca:P than animals fed HSLF ($t = 3.08$, $P = 0.006$) and MSLF ($t = 2.07$, $P = 0.05$, Table 4). However, serum phosphorus was similar in animals across diets ($F_{2,20} = 1.09$, $P = 0.36$, Table 4). Additionally, we found no differences among periods or d x p interactions for serum Ca (period: $F_{3,20} = 2.07$, $P = 0.14$; d x p: $F_{6,20} = 2.10$, $P = 0.10$, Table 4), serum P (period: $F_{3,48} = 1.10$, $P = 0.36$; d x p: $F_{6,48} = 0.76$, $P = 0.61$, Table 4) or serum Ca:P (period: $F_{3,48} = 0.73$, $P = 0.54$; d x p: $F_{6,48} = 1.30$, $P = 0.28$, Table 4).

Fecal Scores

Fecal consistency varied among treatment groups overall ($F_{2,21} = 7.74$, $P = 0.003$, Table 4) and across weeks ($F_{46, 681} = 2.28$, $P < 0.0001$, Figure 1), with a d x p interaction ($F_{92,681} = 1.45$, $P = 0.006$, Figure 1). Overall, animals fed MSLF had a lower fecal score than those fed LSHF ($t = 3.78$, $P = 0.001$) and HSLF ($t = 2.79$, $P = 0.01$), but animals fed LSHF and HSLF diets were not different ($t = 0.73$, $P = 0.48$). When evaluating across weeks, LSHF animals had higher fecal scores than those fed HSLF and MSLF at 22 wk ($F_{2,20} = 6.06$, $P = 0.0009$), and at 24 and 26 wk, LSHF animals had higher fecal scores than MSLF animals ($F \geq 3.5$, $P \leq 0.05$), but did not differ from HSLF animals ($P > 0.05$). However, at 31, 38, 51, and 65 wk, both LSHF and HSLF animals had higher fecal scores than MSLF animals (all $F \geq 4.82$, all $P \leq 0.03$) but did not differ from each other ($P \geq 0.05$). At 53, 60, and 63 wk, animals fed HSLF had higher fecal scores than those fed MSLF ($P \leq 0.05$) but did not differ from LSHF animals ($P > 0.05$).

Digestive Morphology

Digestive tissues collected from necropsied animals were similar histologically. Tissues from all animals showed a mild degree of inflammation, but no abnormal

characteristic in any tissue (Appendix D - F). However, the density ($F_{2,6} = 52.77$, $P = 0.0002$, Table 3) and length ($F_{2,6} = 10.11$, $P = 0.01$, Table 3) of rumen papillae varied among animals fed different diets. Animals consuming HSLF had denser papillae than animals consuming MSLF ($t = 9.97$, $P < 0.0001$) and LSHF ($t = 7.14$, $P = 0.0004$), and LSHF had denser papillae than MSLF ($t = 2.82$, $P = 0.03$). In addition, animals consuming MSLF had shorter papillae than both LSHF ($t = 4.47$, $P = 0.004$) and HSLF ($t = 2.67$, $P = 0.04$). HSLF and LSHF did not differ length of rumen papillae ($t = 1.80$, $P = 0.12$).

Growth and Body Condition and Bone Minerals

Animals fed the different diets had similar morphometrics and body condition (body mass: $F_{2,20} = 0.55$, $P = 0.58$; leg length: $F_{2,20} = 0.10$, $P = 0.90$; loin depth: $F_{2,20} = 1.65$, $P = 0.22$; MAXFAT: $F_{2,20} = 0.18$, $P = 0.84$; Table 6; Appendix G - K). However, as animals grew from an average $3.7 \text{ kg} \pm 0.4$ at birth to over $67.1 \text{ kg} \pm 8.5$ by 68 wk of age, there was significant period effects for body mass ($F_{4,20} = 130.42$, $P < 0.0001$), leg length ($F_{4,68} = 55.17$, $P < 0.0001$), loin depth ($F_{4,20} = 32.22$, $P < 0.0001$) and MAXFAT ($F_{4,20} = 35.28$, $P < 0.0001$). Likewise, lean body mass ($F_{2,6} = 0.95$, $P = 0.44$) and body fat ($F_{2,6} = 0.73$, $P = 0.52$) of 9 animals measured using DEXA scans at the end of the project were similar among diets. Also, total area ($F_{2,5} = 1.51$, $P = 0.49$) and total mass ($F_{2,5} = 1.04$, $P = 0.42$) of antlers were similar among diets.

DEXA scans of 9 animals revealed no differences in total body bone mineral composition ($F_{2,6} = 1.11$, $P = 0.39$, Table 6) or bone mineral density ($F_{2,6} = 1.51$, $P = 0.29$, Table 6) among diets. Likewise, DEXA scans of antlers revealed that bone mineral

composition ($F_{2,5} = 1.34$, $P = 0.34$, Table 6), and bone mineral density ($F_{2,5} = 2.76$, $P = 0.16$, Table 6) did not differ among diets.

The metacarpal bones of 9 animals did not differ in Ca ($36.73 \% \pm 0.50$, $F_{2,6} = 0.03$, $P = 0.97$), P ($18.52 \% \pm 0.25$, $F_{2,6} = 0.10$, $P = 0.90$), or Ca:P (1.98 ± 0.004 , $F_{2,6} = 2.00$, $P = 0.22$). Likewise, antlers of 8 males did not differ in Ca ($36.76 \% \pm 0.38$, $F_{2,5} = 0.23$, $P = 0.80$), P ($18.25 \% \pm 0.22$, $F_{2,5} = 0.43$, $P = 0.68$), or Ca:P (2.01 ± 0.008 , $F_{2,5} = 1.15$, $P = 0.39$).

DISCUSSION

As expected, the differences in levels of fiber, starch, and protein among three pelleted diets, HSLF (high starch, low fiber), LSHF (low starch, high fiber) and MSLF (moderate starch, low fiber) affected behavior and physiology of juvenile mule deer consuming them. However, growth and body condition of mule deer did not vary among dietary treatments because they provided similar digestible energy content.

Contrary to our general expectations, the higher NDF and lower starch content of the LSHF diet did not lower DMD or AED. Although the higher starch and lower fiber of HSLF resulted in a higher digestibility of cell solubles, (NDS), the digestibility of the fiber fraction (NDFD) was higher in the higher fiber LSHF. Additionally, LSHF had a higher NDF:STARCH of (17.33) compared to HSLF (1.65) and MSLF (2.12) because beet pulp was used as the fiber source in LSHF instead of a grain-based fiber source (e.g., wheat middlings). Beet pulp contains soluble pectin, which is readily but evenly fermented within the rumen to yield high levels of acetate and, therefore, energy (Van Soest et al., 1991). The ground aspen included in the LSHF contained a small amount of indigestible lignin, however, it did not increase the overall lignin content of the diet and therefore did not influence its digestibility.

The diets also differed in crude protein content and digestibility, leading to differences in DPI and BUN. First, unlike the other diets, MSLF contained ruminally-protected protein but at very low levels (0.5 %). This difference may explain higher CPD and CPI by animals consuming this diet relative to HSLF and LSHF. Ruminally-protected protein escapes microbial digestion in the rumen and is denatured in the

abomasum, ensuring direct metabolism and absorption by the animal rather than rumen microbial populations (Owens and Zinn, 1988).

Next, the higher level of CP (~ 19 %) in MSLF and HSLF than in LSHF also led to higher BUN for animals consuming these diets because dietary protein requirements are thought to be much lower (9 to 11% DM for young to mature cervids; NRC, 2007). Free-ranging white-tailed deer (*Odocoileus virginianus*) in Texas consuming natural forage (CP of rumen contents averaged 10.2 % in January to 17.0 % in June) had BUN averaging 8.8 mg/dL in October to 21.5 mg/dL in August (Waid and Warren, 1984). Animals consuming our lowest CP diet (LSHF, > 14 % CP) had BUN ranging from 15 to 35 mg/dL. In sheep and cattle, BUN concentrations above 18 and 19 mg/dL, respectively, were associated with reduced pregnancy rates and considered deleterious to embryonic development (Bishonga et al., 1994; Butler et al., 1996). Because lower dietary protein content did not adversely affect the growth and body condition of animals fed LSHF and may, in fact, have harmful consequences to reproduction, captive ruminants may benefit from pelleted rations containing CP lower than 15 %.

Despite differences in starch, fiber and crude protein, growth and body condition as measured by body mass, body fat, depth of the loin muscle, frame and antler size, did not differ among treatments. In fact, by the end of the trial in September 2008, nearly all animals had deposited at least 1 cm of rump fat, representing approximately 10 % body fat (Cook et al., 2007). Based on rump fat alone, our animals ranged from 7 to 20 % estimated body fat during period 5 (68 wk old), with the thinnest individual on HSLF and the fattest on LSHF. Mule deer with 7 to 12 % body fat have a 72 to 95 % chance of becoming pregnant, with a 50 to 80% chance of having twins (Tollefson, 2007). Based

on this information, our animals consuming HSLF, MSLF and LSHF had high chances of successful reproduction at the end of the trial.

Although not significantly different, retention time of food particles in the GIT tended to be longer for LSHF. In fact, the MRT of LSHF (21.3 h) was more similar to the MRT of mule deer fed natural grass and browse diets (21.4 to 23.6 h, Mautz and Petrides, 1971) than those fed pelleted diets (13.9 to 17.1 h; Mautz and Petrides, 1971). MRT of pelleted diets are normally shorter because foods are processed into smaller particles, which require less rumination and microbial fermentation to pass through the reticulo-omasal orifice (Van Soest, 1994).

Food intake during our experiment seemed to be more influenced by starch than fiber in the diet. Although animals on the LSHF diet spent more time feeding, presumably chewing, they ate the same mass of pellets per day as those on the MSLF during meal feeding, and the same as both MSLF and HSLF when given *ad libitum* access to pellets during the digestion trials. Animals fed high fiber diets that are less digestible often consume more food to maintain digestible energy intake until restricted by rumen fill and passage (Van Soest, 1994). However, DMD did not vary with fiber content in our pelleted diets, thus animals fed LSHF did not need to consume more to meet energy needs. On the other hand, high starch coupled with low fiber may have been responsible for differences in DMI of animals fed HSLF during meal-feeding. When fed 3 to 4 meals per day, animals fed HSLF consumed less food than did those fed LSHF and MSLF, yet animals fed HSLF *ad libitum* during digestion trials ate nearly twice as much per kg body weight and a similar amount as animals fed LSHF and MSLF. However, results of our digestion and preference trials indicate that diets in this trial were

highly digestible and equally palatable. Therefore, rumen fill and palatability probably did not play a role in regulating short-term DMI during meal feeding. Therefore, we suggest that biochemical cues caused by VFA concentration were the main regulators of short-term intake in our trials (Sjaastad et al., 2003; DeJong, 1985). Specifically, the production of propionic acid from starch-containing feeds has been shown to produce hypophagic response in many ruminants to a greater degree than acetic or butyric acids (Faverdin, 1999; Chaput, 2006; Bradford and Allen, 2007). In addition, the production of lactic acid from starch peaks during the first 1 to 2 h after ingestion, and because lactic acid has a lower pKa (3.9) than both acetic (4.8) and propionic (4.9) acids, its sudden appearance in the rumen could temporarily offset the osmotic balance and acid-base equilibrium, triggering the cessation of eating soon after ingestion of food (Van Soest, 1994; Faverdin, 1999). Unfortunately, our rumen fluid preservation methods did not allow for lactic acid analysis.

Not only did animals fed LSHF spend more time feeding, they also spent more time ruminating than animals fed higher starch, lower fiber diets. This result was expected because animals ruminate in direct proportion to the cell wall content of the diet to reduce particle size for microbial fermentation and passage through the rumen (Van Soest, 1994). In captive environments, the ability to induce rumination in exotic ruminants may prevent behavioral stereotypies by providing the opportunity for ruminants to perform innate behaviors (see Bashaw et al., 2001; Hummel et al., 2006a).

Rumination also induces chewing which stimulates saliva production, supplying the rumen with an aqueous buffering medium to stabilize pH against rapid fermentation characteristic of pelleted diets (Van Soest, 1994; Mertens, 1997). Because animals fed

LCHF ruminated longer, greater saliva production would be expected to have buffered the rumen against drops in pH resulting from VFA production. However, rumen pH of our mule deer did not differ among the 3 diets. Because pH of the rumen fluid samples obtained via gastric tubing (6.1 - 7.9) were much higher than those obtained directly from the rumen during necropsy (4.1 – 5.5), we suspect that rumen fluid samples obtained via tubing were contaminated with saliva, which has a pH around 8 to 8.5 (Van Soest, 1994). On the other hand, samples obtained during necropsy could have been influenced by the sedation or euthanasia drugs or the time from sedation to sample collection.

Besides increasing rumination and saliva production, LCHF also contained sodium sesquicarbonate (NaSC), an alkaline chemical that serves as a rumen buffer. Presently, this chemical's buffering capacity has not been extensively tested in exotic ruminants (Elizabeth Koutsos, PMI Nutrition, personal communication), but the inclusion in LCHF could have resulted in a protective buffering of the rumen against acidic end products, aiding in the development of a normally functioning GIT and thereby resulting in higher fecal scores at an early age. In our experiment, the presence of this chemical may have confounded the effects of increased fiber and chewing on rumen pH.

Many of our young mule deer consuming the higher starch diets had relatively poor fecal consistency when we began scoring feces at 20 wk and continued to 34 wk of age. However, over time, fecal consistency for all treatment groups improved which suggests that the animals' digestive tracts were adapting to the pelleted diets. Despite improvements in fecal consistency across the trial, 3 animals on HSLF and 1 animal on MSLF had to be removed from the trial because of severe, prolonged diarrhea that did not

respond to antibiotic treatment. In addition, animals on HSLF and MSLF frequently had significantly lower fecal scores than those on LSHF (Figure 1).

Low fecal consistency in deer that typically have firm, low-moisture feces is caused by increased passage rate, decreased absorptive surface area, or an osmotic imbalance (Smith and Magdesian, 2002). Perhaps young mule deer in this trial were particularly sensitive to consuming higher starch diets because the digestive system had not fully adapted to the diet and the fermentative end products could not be absorbed. The ruminant in early life functions primarily as a monogastric, shunting suckled milk directly to the abomasum for digestion and VFA production is minimal until consumption of solid food commences (Grover, 1988; Van Soest, 1994). Diets exhibiting faster fermentation and higher concentration of VFA's have the potential to irritate the walls of the developing digestive tract, thereby inhibiting proper absorption of digestive end products until the tissues adapt and repair themselves.

The NDF:starch ratio of our diets influenced the composition of VFA's in the rumen. As expected, in several of our sampling periods, LSHF had a higher acetate and higher A:P than the higher starch diets. Lower A:P ratios indicate ingestion of a higher proportion of non-structural carbohydrates which result in increased propionate production, and higher A:P values indicate consumption of more structural carbohydrates that result in higher acetate production (Van Soest, 1994). Additionally, the presence of 1 % NaSC has been shown to increase the concentration of acetate, A:P, rumen pH, and decrease the concentration of propionate and butyrate for dairy cattle consuming concentrate and corn silage diets (Ghorbani et al., 1989). The A:P of our animals on all the diets, however, were consistent with those of free-ranging deer. For example, white-

tailed deer in Texas consuming a mixed diet of browses and grasses had ruminal A:P of around 4.5 (Short et al., 1969), which is similar to the mean ruminal A:P of our mule deer fed LSHF (4.24). In contrast, free-ranging mule deer in Colorado consuming mostly woody browse had ruminal A:P ranging from about 2.9 during the summer to 3.4 in the winter (Short et al, 1966), similar to the mean ruminal A:P of our deer fed HSLF (3.89) and MSLF (3.60). However, A:P ratios of our mule deer exceeded those of cows fed a high grain diet (1 to 1.3; Van Soest, 1994).

Although we had expected animals fed higher starch diets to have longer and denser papillae to absorb rapidly-produced VFA's, we instead found that MSLF had the shortest and least dense papillae, whereas LSHF had the longest, and HSLF the densest, papillae. Although our sample sizes were small for this analysis (3 animals/diet), the length (7 to 8.5 mm) and density (107 to 154 papillae/cm²) of papillae in our animals were similar to wild deer. Average papillae length of mule and white-tailed deer consuming 100% forage was approximately 5 to 10 mm (Short, 1964; Zimmerman et al., 2006), and density for non-pregnant, non-lactating female mule deer consuming natural forage ranged from 103 – 119 papillae/cm² (Zimmerman et al., 2006).

Similar to rumen chemistry, the higher fiber in LSHF caused animals consuming that diet to have higher acetate in the blood. Blood acetate can provide as much as 50% of the energy supply of ruminant animals (Sabine and Johnson, 1964). Because dietary starch results in greater production of propionate, and propionate is a major precursor of gluconeogenesis, we expected that the higher starch diets would yield 1) greater plasma propionate concentrations and 2) greater concentrations of serum glucose. However, neither plasma propionate or serum glucose levels of animals in our experiment differed

among treatments. Similarly, in a study conducted by Lane and Jesse (1996), ruminal propionate infusion in lambs did not increase blood glucose concentrations. These results are probably caused by an increase in insulin secretion by the pancreas, resulting in increased rates of glucose uptake from the blood by the liver, muscle and adipose tissue (Sjaastad et al., 2003). An increase in circulating insulin is also triggered by ingestion of higher levels of protein (Cheema et al., 1991). Both HSLF and MSLF diets contained higher levels of protein which could serve as an additional stimulant for insulin production and release.

Ratios of Ca and P in our diets were reflected in the animals' serum concentration. Although LSHF had lower Ca than the other diets, it also had lower dietary P, thus animals fed LSHF had a higher serum Ca:P. Excess P in the diet can reduce Ca absorption causing the formation of insoluble Ca complexes leading to bone disorders and urinary calculi (Emerick and Embry, 1963; Ammerman and Goodrich, 1983; Robbins, 1983). The serum Ca:P of animals fed all of our diets exceeded the minimum range suggested for young to mature cervids (1.11 to 1.67; NRC, 2007), and the bone mineral composition and density were equivalent among animals fed our diets.

In conclusion, pelleted diets with 14 % CP, 3% starch, and 50% NDF consisting of aspen fiber and beet pulp met the energy and protein requirements of growing mule deer, a medium-sized browsing ruminant, as well as grain-based diets with higher protein (19 %), higher starch (16 – 20 %), and lower fiber (33 – 35 % NDF). Animals on all 3 diets grew equally fast, with equivalent body mass, body fat, muscle size, and antler size. In addition, animals consuming a lower starch, higher fiber pelleted diet generally had better fecal consistency as they grew from 20 wk to 34 wk than those consuming higher

starch, lower fiber diets. Finally, lower starch, higher fiber pelleted diets increased the time animals spent ruminating and feeding without increasing total intake, thus providing behavioral enrichment for captive ruminants. Therefore, feeding exotic or wild ruminants pelleted diets containing lower levels of starch and higher levels of energy-yielding soluble fiber like beet pulp in place of grains provides a healthy, simple option for complete nutrition and a diet that more closely resemble forage consumed by these animals in the wild.

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Table 1. Diet composition of low-starch, high-fiber (LSHF); high-starch, low-fiber (HSLF); and moderate-starch, low-fiber (MSLF) pelleted diets fed to mule deer (*Odocoileus hemionus*) from birth in May 2007 to September 2008.

Ingredient	Diet composition (%)		
	LSHF	HSLF	MSLF
Alfalfa	0.00	30.42	14.08
Apple flavoring	0.30	0.30	0.00
Brewers dried yeast	1.00	1.00	0.00
Calcium carbonate	0.02	0.72	1.36
Calcium propionate	0.50	0.10	0.00
Calcium stearate	0.00	0.00	0.25
Canola meal	0.00	0.00	10.00
Dicalcium phosphate	0.93	0.25	0.01
DL-methionine	0.24	0.00	0.01
Dried whey	0.00	0.00	0.01
Flaxseed	1.00	0.00	0.00
Ground beet pulp	10.00	0.00	0.00
Ground corn	0.00	18.57	5.00
Ground oat hulls	5.75	0.00	15.00
Ground soy hulls	50.00	0.00	0.00
Ground whole aspen	6.00	0.00	0.00
Lactobacillus cultures	0.00	0.00	0.13
Lignin sulfonate	1.00	0.00	2.35
L-lysine	0.00	0.00	0.01
Magnesium oxide	0.26	0.09	0.00
Mixed tocopherols	0.06	0.06	0.01
Molasses	6.00	5.00	4.00
Salt	1.00	1.00	0.51
Sodium sesquicarbonate	0.40	0.00	0.00
Soybean hulls	0.00	0.00	9.32
Soybean meal (48%)	12.70	10.72	0.96
Soybean oil	2.11	1.00	1.56
Sucrose	0.10	0.10	0.00
Vitamin/mineral mix	0.63	0.47	0.43
Wheat middlings	0.00	30.20	35.00

Table 2. Nutritional composition of 3 pelleted diets fed to captive mule deer (*Odocoileus hemionus*) from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.

ITEM	Treatment		
	LSHF	HSLF	MSLF
% DM	90.9	90.7	91.2
% CP	14.81	19.6	19.3
%ADF	34.4	18.8	19.6
%NDF	51.6	33.3	35.8
% LIGNIN	2.4	3.9	4.0
% CF	28.9	14.1	14.6
% STARCH	3.0	20.0	16.6
NDF:STARCH	17.3	1.7	2.1
% TDN	71.0	73.7	72.9
% CRUDE			
FAT	3.9	3.9	3.8
% Ca	1.0	1.2	1.3
% P	0.4	0.6	0.6
Ca:P	2.6	1.9	2.0
% Mg	0.4	0.4	0.4
% K	1.3	1.4	1.4
% Na	0.6	0.4	0.4
Fe (ppm)	630.6	495.7	678.7
Zn (ppm)	253.6	403.7	205.0
Cu (ppm)	33.3	35.3	46.7
Mn (ppm)	273.1	263.7	264.4
Mo (ppm)	0.6	1.6	0.9
DIG.E (kJ/g)	15.3	15.5	15.6

DM = dry matter, CP = crude protein, ADF = acid detergent fiber, NDF = neutral detergent fiber, CF = crude fiber, TDN = total digestible nutrients, DIG.E = diet digestible energy.

Table 3. Nutritional composition of forage fed to captive mule deer (*Odocoileus hemionus*) from birth in May 2007 to September 2008.

	Forage		
	Grass Pasture	Alfalfa hay	Alfalfa hay cubes
% DM	92.0	91.6	91.5
% CP	17.2	19.6	19.9
%ADF	32.2	38.7	34.8
%NDF	51.9	48.2	44.1
% LIGNIN	4.0	8.7	8.4
% NFC	24.2	24.7	26.6
% CF	22.6	34.1	.
% STARCH	1.2	1.7	1.2
% TDN	65.9	57.2	60.7
%Fat	4.0	1.7	2.3
% Ca	0.3	1.2	1.4
% P	0.4	0.3	0.3
% Mg	0.1	0.2	0.3
% K	2.9	2.4	2.2
% Na	0.01	0.1	0.1
Fe (ppm)	215.3	247.2	707.3
Zn (ppm)	37.6	22.7	20.7
Cu (ppm)	7.8	8.1	7.7
Mn (ppm)	64.0	29.2	39.3
Mo (ppm)	0.5	0.8	1.2

DM = dry matter, CP = crude protein, ADF = acid detergent fiber, NDF = neutral detergent fiber, CF = crude fiber, TDN = total digestible nutrients, DIG.E = diet digestible energy.

Table 4. Means \pm standard errors of rumen and blood chemistry, fecal scores and papillae morphology of captive mule deer (*Odocoileus hemionus*) fed 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate -starch, low-fiber diet.

	Treatment			D x P ¹
	LSHF	HSLF	MSLF	
Rumen VFA, M% (gastric tubing)				
pH	6.9 \pm 0.1	6.8 \pm 0.2	6.8 \pm 0.1	-
Acetate	74.6 ^a \pm 0.8	71.5 ^b \pm 0.7	69.3 ^b \pm 0.8	+
Propionate	18.3 \pm 0.6	19.2 \pm 0.7	19.8 \pm 0.6	+
Butyrate	5.7 ^b \pm 0.2	7.9 ^a \pm 0.3	8.8 ^a \pm 0.5	-
A:P	4.3 \pm 0.2	3.9 \pm 0.2	3.6 \pm 0.1	+
Rumen VFA (necropsy)				
pH	5.4 \pm 0.1	5.2 \pm 0.2	5.4 \pm 0.0	
Acetate (mM)	123.7 ^a \pm 0.9	107.0 ^b \pm 3.5	92.8 ^c \pm 5.9	
M%	73.3 \pm 0.0	66.7 \pm 0.9	70.3 \pm 2.4	
Propionate (mM)	31.7 \pm 2.0	31.7 \pm 0.2	24.1 \pm 4.1	
M%	18.7 \pm 0.9	20.0 \pm 0.6	17.7 \pm 2.2	
Butyrate (mM)	13.9 \pm 1.6	21.7 \pm 1.2	15.1 \pm 3.1	
M%	8.3 ^b \pm 0.9	13.3 ^a \pm 0.9	12.0 ^a \pm 1.5	
A:P	3.9 \pm 0.2	3.4 \pm 0.1	4.0 \pm 0.5	
Blood VFA				
Acetate (mM)	0.5 ^a \pm 0.0	0.4 ^b \pm 0.0	0.4 ^b \pm 0.0	-
Propionate (mM)	0.05 \pm 0.0	0.04 \pm 0.0	0.04 \pm 0.0	-
Glucose (mg/dL)	148.1 \pm 4.9	131.9 \pm 7.3	140.2 \pm 5.1	-
BUN (mg/dL)	23.0 ^b \pm 0.7	30.0 ^a \pm 1.3	31.6 ^a \pm 1.1	+
Ca (mg/dL)	9.1 ^a \pm 0.1	8.3 ^b \pm 0.2	8.4 ^b \pm 0.1	-
P (mg/dL)	7.2 \pm 0.3	8.3 \pm 0.4	7.5 \pm 0.3	-
Ca:P	1.3 ^a \pm 0.0	1.1 ^b \pm 0.1	1.2 ^b \pm 0.0	-
Saliva pH	8.4 \pm 0.1	8.3 \pm 0.1	8.4 \pm 0.1	+
Fecal Score	91.1 ^a \pm 0.7	87.4 ^a \pm 1.3	80.5 ^b \pm 1.2	+
Papillae length (mm)	8.5 ^a \pm 0.1	7.9 ^a \pm 0.1	6.9 ^b \pm 0.1	
Papillae density (/cm2)	122.4 ^b \pm 8.5	154.5 ^a \pm 8.6	107.5 ^c \pm 8.4	

¹ D x P = diet x period interaction: - = not different, + = different ($\alpha = 0.05$).

Different superscripted letters within rows denotes significant differences among diet means ($\alpha = 0.05$). No superscript = no significant difference among diets.

Table 5. Means \pm standard errors of intake, feeding time, rumination time, digestibility, and mean retention time of 3 pelleted diets consumed by captive mule deer (*Odocoileus hemionus*) from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.

Trial type	Treatment			D x P ¹
	LSHF	HSLF	MSLF	
DMI of pellets (kg/day)				
Periods 1-5	1.1 ^a \pm 0.1	0.8 ^b \pm 0.1	1.1 ^a \pm 0.1	-
Digestion trial	2.0 \pm 1.1	1.7 \pm 0.2	1.9 \pm 0.1	
DMI of pellets (g/kg/day)				
Periods 1-5	23.4 ^a \pm 0.7	18.4 ^b \pm 1.0	24.4 ^a \pm 1.0	-
Digestion trial	38.8 \pm 1.6	35.4 \pm 1.3	38.1 \pm 1.3	
DMI of alfalfa cubes				
Periods 1-5	122.3 \pm 16.8	97.8 \pm 15.7	104.1 \pm 11.2	+
Pellet:alfalfa cube				
Periods 1-5	0.2 \pm 0.0	0.2 \pm 0.1	0.3 \pm 0.1	-
Feeding time (h/day)				
Scan sample	2.3 ^a \pm 0.2	1.7 ^b \pm 0.1	1.6 ^b \pm 0.2	
Rumination time (h/day)				
Scan sample	2.8 ^a \pm 0.3	1.4 ^b \pm 0.4	1.0 ^b \pm 0.1	
DMD (%)				
Digestion trial	65.0 \pm 0.7	67.6 \pm 0.8	64.4 \pm 1.1	
AED (%)				
Digestion trial	65.1 \pm 0.9	67.0 \pm 1.0	62.3 \pm 1.1	
CPD (%)				
Digestion trial	63.3 ^b \pm 1.7	66.2 ^b \pm 1.0	71.0 ^b \pm 1.2	
NDSD (%)				
Digestion trial	77.7 ^b \pm 0.3	82.4 ^a \pm 0.7	79.4 ^b \pm 0.9	
NDFD (%)				
Digestion trial	76.9 ^a \pm 0.3	60.8 ^c \pm 2.0	69.7 ^b \pm 1.3	
DEI of pellets (MJ/day)				
Period 1-5	10.6 ^a \pm 0.6	8.0 ^b \pm 0.6	10.9 ^a \pm 0.6	-
Digestion trial	19.6 \pm 1.0	17.3 \pm 1.7	19.4 \pm 0.9	
DEI of pellets (MJ/kg/day)				
Period 1-5	0.2 ^a \pm 0.0	0.2 ^b \pm 0.0	0.2 ^a \pm 0.0	-
Digestion trial	0.4 \pm 0.0	0.4 \pm 0.0	0.4 \pm 0.0	
DPI of pellets (g/day)				
Period 1-5	9.0 ^b \pm 0.5	8.3 ^b \pm 0.7	13.0 ^a \pm 0.7	-
Digestion trial	16.7 ^b \pm 0.9	18.0 ^b \pm 1.9	23.0 ^a \pm 1.3	
DPI of pellets (mg/kg/day)				
Period 1-5	198.8 ^b \pm 5.8	198.3 ^b \pm 10.5	293.1 ^a \pm 11.7	-
Digestion trial	329.6 ^b \pm 15.1	381.7 ^b \pm 15.1	460.2 ^a \pm 19.2	
MRT (h)				
Passage trial	21.3 \pm 1.8	19.6 \pm 0.9	18.8 \pm 1.7	

¹D x P = diet period interaction, - = not different, + = different ($\alpha = 0.05$)

DMD = dry matter digestibility, AED = apparent energy digestibility, CPD = crude protein digestibility, NDSD = neutral detergent solubles digestibility, NDFD = neutral detergent fiber digestibility, DEI = digestible energy intake, DPI = digestible protein intake

Different superscripted letters within rows denotes significant differences among diet means ($\alpha = 0.05$). No superscript = no significant difference among diets.

Table 6. Means \pm standard errors of body condition, DEXA, and antler measurements from captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.

	Treatment						D x P ¹
	LSHF		HSLF		MSLF		
Body condition							
Body mass (kg)	45.0	\pm 2.4	40.9	\pm 2.8	43.6	\pm 2.7	-
Hind leg length (cm)	45.7	\pm 0.5	45.1	\pm 0.6	45.5	\pm 0.6	-
Rump fat thickness (cm)	0.5	\pm 0.1	0.4	\pm 0.1	0.4	\pm 0.1	-
Thickness of lattissimus dorsi muscle (cm)	3.3	\pm 0.1	3.0	\pm 0.1	3.2	\pm 0.1	-
DEXA							
BMC (kg)	2.0	\pm 0.0	1.7	\pm 0.2	2.0	\pm 0.1	
BMD (g/cm ²)	1.3	\pm 0.0	1.2	\pm 0.1	1.2	\pm 0.0	
Fat (kg)	10.9	\pm 1.0	8.5	\pm 2.0	10.6	\pm 0.7	
Lean muscle (kg)	62.2	\pm 1.6	56.1	\pm 5.5	61.3	\pm 1.1	
Body mass (kg)	75.0	\pm 1.3	66.4	\pm 7.7	73.4	\pm 1.6	
% fat	14.5	\pm 1.3	12.5	\pm 1.8	14.3	\pm 0.8	
Antlers							
MBL (cm)	26.0	\pm 4.4	25.8	\pm 3.4	19.2	\pm 2.2	
Circ (cm)	7.3	\pm 0.9	7.0	\pm 1.3	6.1	\pm 0.6	
Tines	4.0	\pm 1.2	4.0	\pm 1.4	6.0	\pm 1.7	
BMC (g)	104.6	\pm 52.1	103.1	\pm 38.4	56.9	\pm 20.8	
BMD (g/cm ²)	1.3	\pm 0.2	1.2	\pm 0.1	1.0	\pm 0.1	

¹ D x P = diet x period interaction: - = not different, + = different ($\alpha = 0.05$).

BMC = bone mineral composition, BMD = bone mineral density, MBL = main beam length, CIRC = circumference

Different superscripted letters within rows denotes significant differences among diet means ($\alpha = 0.05$). No superscript = no significant difference among diets.

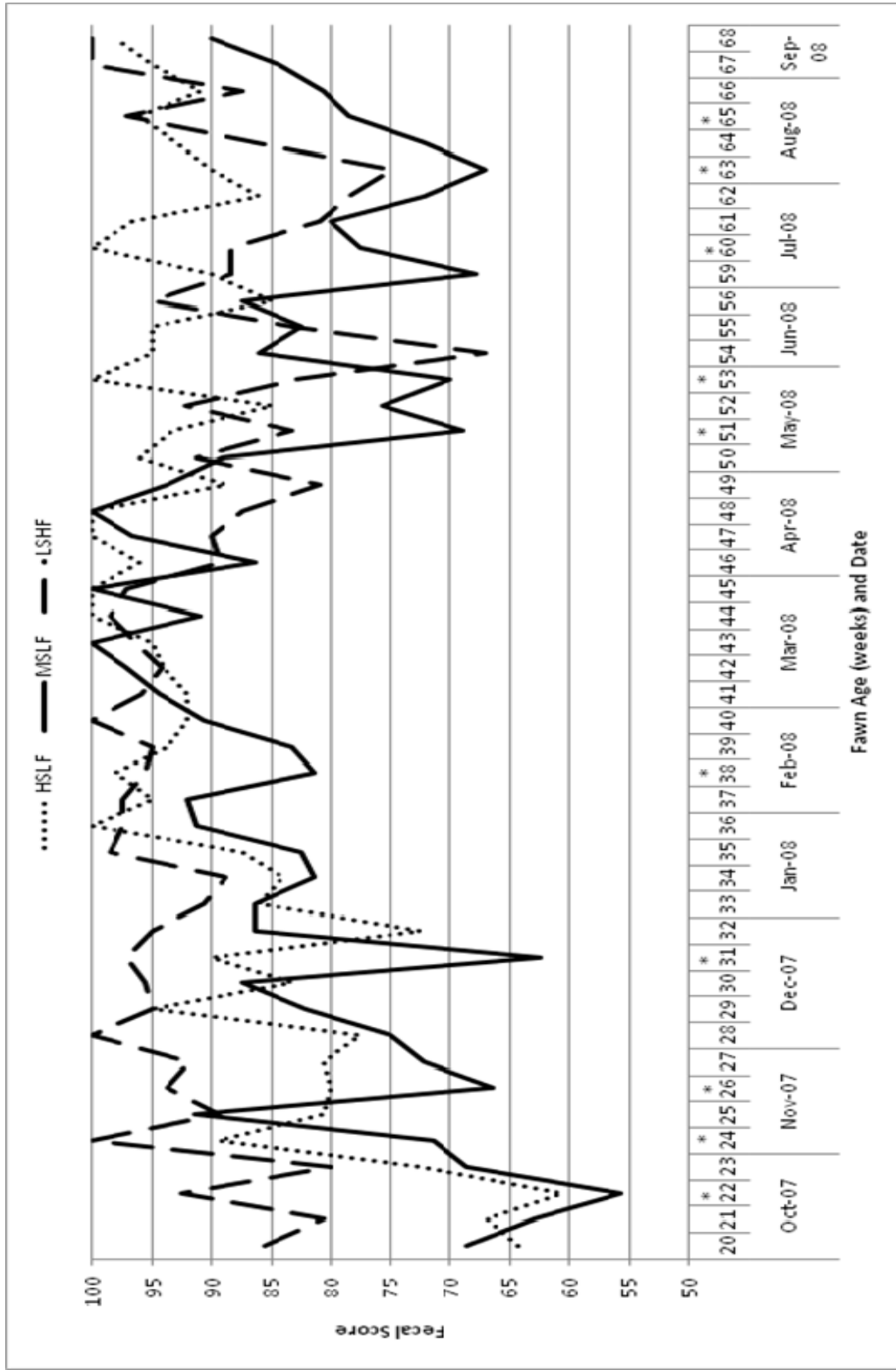


Figure 1. Mean weekly fecal scores for captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet. * indicates significant differences in fecal scores for that period.

Appendix A. Fecal consistency score sheet for captive mule deer (*Ocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008.

FECAL CONSISTENCY SCORE FOR MULE DEER

SCORE 0: DIARRHEA- WATERY, NO CONSISTENCY



SCORE 25: VERY SOFT, SOME CONSISTENCY BUT STILL PUDDLES ON GROUND



SCORE 50: SOFT, FORMS LOOSE PILES UPON HITTING THE GROUND, HAS DEFINITE TEXTURE AND SOME CONSISTENCY



SCORE 75: SOFT CLUMPED PELLETS IDENTIFIABLE BUT STILL LOSES SOME SHAPE UPON HITTING GROUND, MOIST



SCORE 100: FIRM INDIVIDUAL OR CLUMPED PELLETS, DRIER



Appendix B. Hematology and serum chemistry (means \pm SE) of mule deer (*Odocoileus hemionus*) fed 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.

Parameter	Treatment			Goats and Sheep (range only) ¹
	LSHF	HSLF	MSLF	
Leukocytes				
White blood cells ($\mu\text{L} \times 10^3$)				
$\bar{x} \pm \text{SE}$	4.4 \pm 0.7	6.0 \pm 1.1	4.2 \pm 0.2	
Range	1.9 - 27	1.5 - 33	2.1 - 9.1	4.0 - 13.0
<i>n</i>	36	29	36	
Basophils (μL)				
$\bar{x} \pm \text{SE}$	10.5 \pm 4.2	48.4 \pm 17.1	52.0 \pm 13.3	
Range	0 - 102	0 - 395	0 - 408	< 0.3
<i>n</i>	36	29	36	
Eosinophils (μL)				
$\bar{x} \pm \text{SE}$	229.6 \pm 33.0	124.0 \pm 20.0	124.4 \pm 19.6	
Range	0 - 943	0 - 404	0 - 462	< 1.0
<i>n</i>	36	29	36	
Lymphocytes ($\mu\text{L} \times 10^3$)				
$\bar{x} \pm \text{SE}$	1.3 \pm 0.1	1.6 \pm 0.2	1.5 \pm 0.1	
Range	0.4 - 2.5	0.8 - 4.8	0.5 - 3	2.0 - 9.0
<i>n</i>	36	29	36	
Monocytes (μL)				
$\bar{x} \pm \text{SE}$	81.4 \pm 11.1	105.7 \pm 19.3	155.1 \pm 51.4	
Range	0 - 240	0 - 378	0 - 1817	< 0.8
<i>n</i>	36	29	36	
Erythrocytes				
Red blood cells ($\times 10^6/\mu\text{L}$)				
$\bar{x} \pm \text{SE}$	8.6 \pm 0.2	8.7 \pm 0.3	8.5 \pm 0.2	
Range	7.1 - 13.5	7.2 - 13.0	6.7 - 12.7	8.0 - 16.0
<i>n</i>	36	29	36	
Hemoglobin (g/dL)				
$\bar{x} \pm \text{SE}$	12.4 \pm 0.2	12.7 \pm 0.3	12.2 \pm 0.2	
Range	10.9 - 17.2	10.2 - 17.3	10 - 14.7	8.0 - 16.0
<i>n</i>	36	29	36	
Mean corpuscular hemoglobin (pg)				
$\bar{x} \pm \text{SE}$	14.6 \pm 0.2	14.7 \pm 0.3	15.2 \pm 0.5	
Range	12 - 17	11 - 17	11 - 32	n/a
<i>n</i>	36	29	36	

¹Range for sheep and goats determined by WSU Clinical Pathology Laboratory

²Range for sheep and goats reported by NRC (2007)

Appendix B., cont.

Parameter	Treatment			Goats and Sheep (range only) ¹
	LSHF	HSLF	MSLF	
Erythrocytes, cont.				
Mean corpuscular hemoglobin concentration (% rbc)				
$\bar{x} \pm SE$	40.4 ± 0.2	41.1 ± 0.2	40.4 ± 0.8	
Range	38 - 44	39 - 43	13 - 43	30 - 38
<i>n</i>	36	29	36	
Mean corpuscular volume (μ ³)				
$\bar{x} \pm SE$	36.2 ± 0.6	35.7 ± 0.7	35.3 ± 0.7	
Range	29 - 43	27 - 42	26 - 43	16 - 48
<i>n</i>	36	29	36	
Mean platelet volume (μ ⁴)				
$\bar{x} \pm SE$	6.0 ± 0.2	6.0 ± 0.2	6.2 ± 0.2	
Range	4.9 - 8.1	4.8 - 8.4	4.1 - 11.6	n/a
<i>n</i>	35	29	36	
Packed cell volume (%)				
$\bar{x} \pm SE$	30.8 ± 0.6	31.0 ± 0.6	29.8 ± 0.5	
Range	27 - 43	25 - 42	24 - 37	22 - 50
<i>n</i>	36	29	36	
Platelets (/μL X 10 ⁵)				
$\bar{x} \pm SE$	4.2 ± 0.2	4.5 ± 0.2	4.9 ± 0.2	
Range	2.2 - 7.6	2.2 - 6.4	0.9 - 7.8	3.0 - 7.50
<i>n</i>	36	29	36	
Red blood cell distribution width (%)				
$\bar{x} \pm SE$	19.3 ± 0.4	19.9 ± 0.5	19.8 ± 0.4	
Range	16 - 24	16 - 25	16 - 25	n/a
<i>n</i>	36	29	36	
Protein				
Blood urea nitrogen (mg/dL)				
$\bar{x} \pm SE$	23.0 ± 0.7	30.0 ± 1.3	31.6 ± 1.1	
Range	15 - 35	18 - 48	23 - 47	13 - 36
<i>n</i>	29	22	29	
Creatinine (mg/dL)				
$\bar{x} \pm SE$	1.0 ± 0.0	1.1 ± 0.0	1.0 ± 0.0	
Range	0.8 - 1.2	0.9 - 1.4	0.8 - 1.3	0.3 - 1.3
<i>n</i>	29	22	29	

¹Range for sheep and goats determined by WSU Clinical Pathology Laboratory

²Range for sheep and goats reported by NRC (2007)

Appendix B., cont.

Parameter	Treatment			Goats and Sheep (range only) ¹
	LSHF	HSLF	MSLF	
Protein, cont.				
Globulin (g/dL)				
$\bar{x} \pm SE$	2.4 ± 0.1	2.7 ± 0.1	2.5 ± 0.1	
Range	1.6 - 3.7	2 - 3.8	1.6 - 4	2.2 - 4.8
<i>n</i>	29	22	29	
Albumin (g/dL)				
$\bar{x} \pm SE$	2.9 ± 0.1	2.8 ± 0.1	2.8 ± 0.1	
Range	2.3 - 3.4	1.8 - 3.3	2.1 - 3.2	2.9 - 4.0
<i>n</i>	29	22	29	
Total protein (g/dL)				
$\bar{x} \pm SE$	5.4 ± 0.1	5.5 ± 0.1	5.3 ± 0.1	
Range	4.7 - 6.4	5 - 6.7	4.6 - 6.8	5.5 - 9.0
<i>n</i>	29	22	29	
Enzymes				
Alkaline phosphatase (U/L)				
$\bar{x} \pm SE$	128.4 ± 16.0	95.0 ± 10.8	98.9 ± 7.6	
Range	44 - 407	40 - 238	24 - 198	28 - 274
<i>n</i>	29	22	29	
Aspartate aminotransferase (U/L)				
$\bar{x} \pm SE$	79.4 ± 3.3	106.5 ± 14.3	72.9 ± 2.8	
Range	51 - 134	59 - 387	47 - 101	54 - 143
<i>n</i>	29	22	29	
Creatine kinase (U/L)				
$\bar{x} \pm SE$	1.0 ± 0.0	117.6 ± 16.1	90.6 ± 8.2	
Range	38 - 233	41 - 334	0.8 - 1.3	n/a
<i>n</i>	29	22	29	
Glutaryl transferase (U/L)				
$\bar{x} \pm SE$	61.5 ± 8.4	77.3 ± 12.5	52.5 ± 4.3	
Range	25 - 278	36 - 310	30 - 128	19 - 87
<i>n</i>	29	22	29	
Sorbitol dehydrogenase (U/L)				
$\bar{x} \pm SE$	14.2 ± 1.3	13.8 ± 1.5	12.3 ± 0.8	
Range	7 - 46	8 - 39	7 - 24	15 - 79
<i>n</i>	29	22	29	

¹Range for sheep and goats determined by WSU Clinical Pathology Laboratory

²Range for sheep and goats reported by NRC (2007)

Appendix B., cont.

Parameter	Treatment			Goats and Sheep (range only) ¹
	LSHF	HSLF	MSLF	
Enzymes				
Glucose (mg/dL)				
$\bar{x} \pm SE$	148.1 ± 4.9	131.9 ± 7.3	140.2 ± 5.1	
Range	104 - 224	70 - 183	83 - 187	46 - 96
<i>n</i>	29	22	29	
Minerals				
Calcium (mg/dL)				
$\bar{x} \pm SE$	9.1 ± 0.1	8.3 ± 0.2	8.4 ± 0.1	
Range	8.3 - 10	5.3 - 9.3	7.7 - 9.4	8.5 - 10.6
<i>n</i>	30	22	28	
Phosphorus (mg/dL)				
$\bar{x} \pm SE$	7.2 ± 0.3	8.3 ± 0.4	7.5 ± 0.3	
Range	4.7 - 10.2	4.9 - 10.9	4.2 - 10.1	2.9 - 14.5
<i>n</i>	30	22	28	
Magnesium (mg/dL)				
$\bar{x} \pm SE$	1.9 ± 0.0	1.8 ± 0.1	1.8 ± 0.0	
Range	1.6 - 2.2	1.5 - 2.5	1.6 - 2.4	2.2 - 4.2
<i>n</i>	30	22	28	
Chloride (mEq/L)				
$\bar{x} \pm SE$	100.4 ± 0.4	101.3 ± 0.7	100.1 ± 0.4	
Range	96 - 106	92 - 106	96 - 104	95 - 111
<i>n</i>	30	22	28	
Potassium (mEq/L)				
$\bar{x} \pm SE$	5.3 ± 0.4	5.4 ± 0.5	4.8 ± 0.4	
Range	3.5 - 11.6	3.8 - 11	3.3 - 9.9	3.7 - 5.6
<i>n</i>	30	22	28	
Sodium (mEq/L)				
$\bar{x} \pm SE$	142.2 ± 0.6	143.1 ± 0.7	139.3 ± 0.7	
Range	136 - 151	136 - 149	132 - 148	140 - 152
<i>n</i>	30	22	28	
Enzymatic carbonate (mmol/L)				
$\bar{x} \pm SE$	24.6 ± 0.3	24.6 ± 0.5	22.9 ± 0.5	
Range	21 - 28	20 - 29	15 - 26	21 - 30
<i>n</i>	30	22	28	

¹Range for sheep and goats determined by WSU Clinical Pathology Laboratory

²Range for sheep and goats reported by NRC (2007)

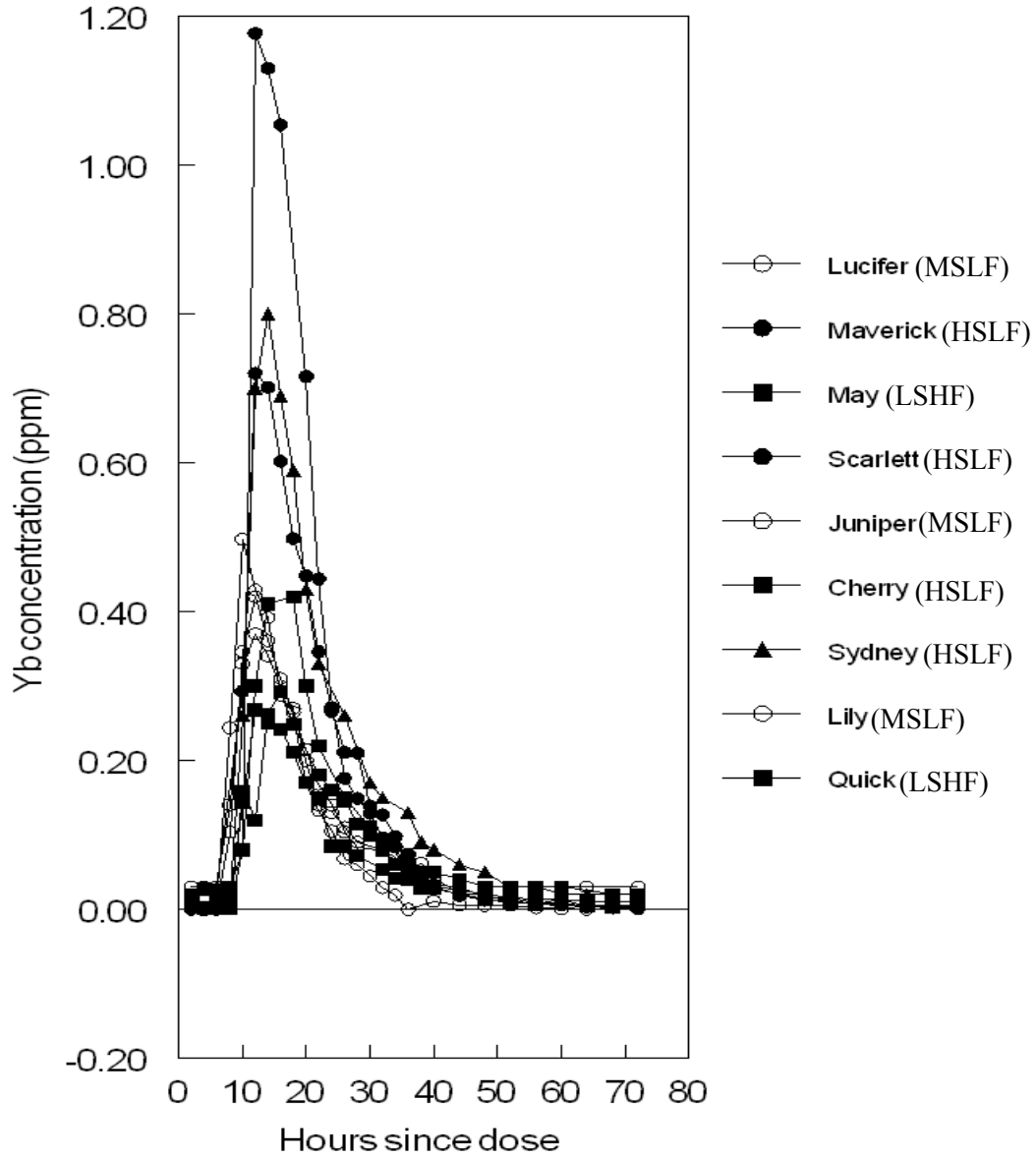
Appendix B., cont.

Parameter	Treatment			Goats and Sheep (range only) ¹
	LSHF	HSLF	MSLF	
Minerals, cont.				
Iron (µg/mL)				
$\bar{x} \pm SE$	176.3 ± 6.7	143.2 ± 8.6	133.8 ± 7.1	
Range	103 - 254	81 - 249	50 - 209	
<i>n</i>	30	22	27	
Zinc (µg/mL)				
$\bar{x} \pm SE$	0.8 ± 0.0	0.7 ± 0.1	0.7 ± 0.0	
Range	0.4 - 1.4	0.4 - 1.3	0.5 - 1.1	0.8 - 1.1 ²
<i>n</i>	30	22	29	
Copper (µg/mL)				
$\bar{x} \pm SE$	1.0 ± 0.0	0.9 ± 0.0	0.8 ± 0.0	
Range	0.7 - 1.3	0.7 - 1.2	0.6 - 1.2	0.9 - 1.4 ²
<i>n</i>	30	22	29	
Selenium (ng/mL)				
$\bar{x} \pm SE$	80.5 ± 1.6	90.4 ± 5.7	104.2 ± 2.1	
Range	65 - 105	61 - 194	79 - 129	
<i>n</i>	30	22	29	
Manganese (ng/mL)				
$\bar{x} \pm SE$	2.3 ± 0.6	3.8 ± 1.7	2.0 ± 0.4	
Range	0.5 - 18.9	0.3 - 35	0.4 - 10.4	
<i>n</i>	30	22	29	
Molybdenum (ng/mL)				
$\bar{x} \pm SE$	9.4 ± 0.9	21.5 ± 2.0	5.2 ± 0.4	
Range	4.1 - 22.8	2.1 - 42.9	1.7 - 9.7	
<i>n</i>	30	22	29	
Cobalt (ng/mL)				
$\bar{x} \pm SE$	0.9 ± 0.0	0.8 ± 0.1	10.9 ± 1.2	
Range	0.4 - 1.2	0.4 - 1.7	2.9 - 28.4	
<i>n</i>	30	22	29	

¹Range for sheep and goats determined by WSU Clinical Pathology Laboratory

²Range for sheep and goats reported by NRC (2007)

Appendix C. Excretion curves for mule deer (*Odocoileus hemionus*) given a pulse dose of neutral detergent fiber particles of either a low-starch, high-fiber (LSHF), a high-starch, low-fiber (HSLF), or a moderate-starch, low-fiber (MSLF) diet. Individual animal identifications precede diet assignments in key.



Appendix D. Qualitative histology report on digestive tissues for 3 male (JU, AR, and QK) mule deer (*Odocoileus hemionus*) consuming a low-starch, high-fiber pelleted diet from birth in May 2007 to September 2008.

	JU	AR	QK
Rumen	1 + lp	1 + lp, 1 + pustules	1 + pustules
Reticulum	1 + lp	Norm	Norm
Omasum	Norm	1 + pustules	1 + pustules
Abomasum	Norm	Norm	Norm
Duodenum	1 + lp	Norm	1 + lp
Jejunem	1 + lp	1 + eos	1 + lp
Ileum	1 + lp	1 + lp	1 + lp and eos
Cecum	1 + lp	1 + lp	1 + lp
Colon	Norm	Norm	Norm
Kidney	Norm	Norm	Norm
Liver	Focal portal lp	Norm	Norm

The grading scale for lesions includes 1+ = minimal, 2+ = mild, 3+ = moderate, 4+ = severe

Key:

Norm = Normal

1+ = minimal

2+ = mild

Pustules = aggregates of neutrophils in epithelium (only forestomachs)

lp = lymphoplasmacytic

eos = eosinophils

interstitial lp = interstitial lymphoplasmacytic inflammation (kidney only)

portal lp = lymphoplasmacytic inflammation in portal areas (liver only)

HISTOLOGIC DIAGNOSES:

1. Essentially normal tissues

Appendix E. Qualitative histology report on digestive tissues for 2 male (MV and BV) and 1 female (BC) captive mule deer (*Odocoileus hemionus*) consuming a high-starch, low-fiber pelleted diet from birth in May 2007 to September 2008.

	MV	BV	BC
Rumen	1 + lp	1 + lp and neuts	1 + pustules
Reticulum	Norm	Norm	Norm
Omasum	Norm	1 + lp and neuts	Norm
Abomasum	Norm	Norm	Norm
Duodenum	2 + lp, 1 + eos	1 + lp and eos	2 + lp
Jejunem	1 + lp	1 + lp and eos	Norm
Ileum	Norm	1 + eos	1 + lp and eos
Cecum	1 + lp	1 + lp	Norm
Colon	Norm	Norm	1 + lp and eos
Kidney	1 + interstitial lp	Norm	Norm
Liver	Multifocal portal lp	Norm	Focal portal lp

The grading scale for lesions includes 1+ = minimal, 2+ = mild, 3+ = moderate, 4+ = severe

Key:

Norm = Normal

1+ = minimal

2+ = mild

Pustules = aggregates of neutrophils in epithelium (only forestomachs)

lp = lymphoplasmacytic

eos = eosinophils

interstitial lp = interstitial lymphoplasmacytic inflammation (kidney only)

portal lp = lymphoplasmacytic inflammation in portal areas (liver only)

HISTOLOGIC DIAGNOSES:

1. Essentially normal tissues

Appendix F. Qualitative histology report on digestive tissues for 3 male (RI, LU, IR) mule deer (*Odocoileus hemionus*) consuming a moderate-starch, low-fiber pelleted diet from birth in May 2007 to September 2008.

	RI	LU	IR
Rumen	1 + lp	1 + lp and 1 + pustules	Norm
Reticulum	Norm	Norm	Norm
Omasum	1 + lp	1 + lp	Norm
Abomasum	Norm	Norm	Norm
Duodenum	1 + lp	Norm	1 + lp
Jejunem	1 + lp	1 + lp	1 + lp and eos
Ileum	1 + lp	1 + lp	1 + lp
Cecum	1 + lp	1 + lp	1 + lp
Colon	Norm	Norm	Norm
Kidney	Norm	Norm	Norm
Liver	Norm	Norm	Norm

The grading scale for lesions includes 1+ = minimal, 2+ = mild, 3+ = moderate, 4+ = severe

Key:

Norm = Normal

1+ = minimal

2+ = mild

Pustules = aggregates of neutrophils in epithelium (only forestomachs)

lp = lymphoplasmacytic

eos = eosinophils

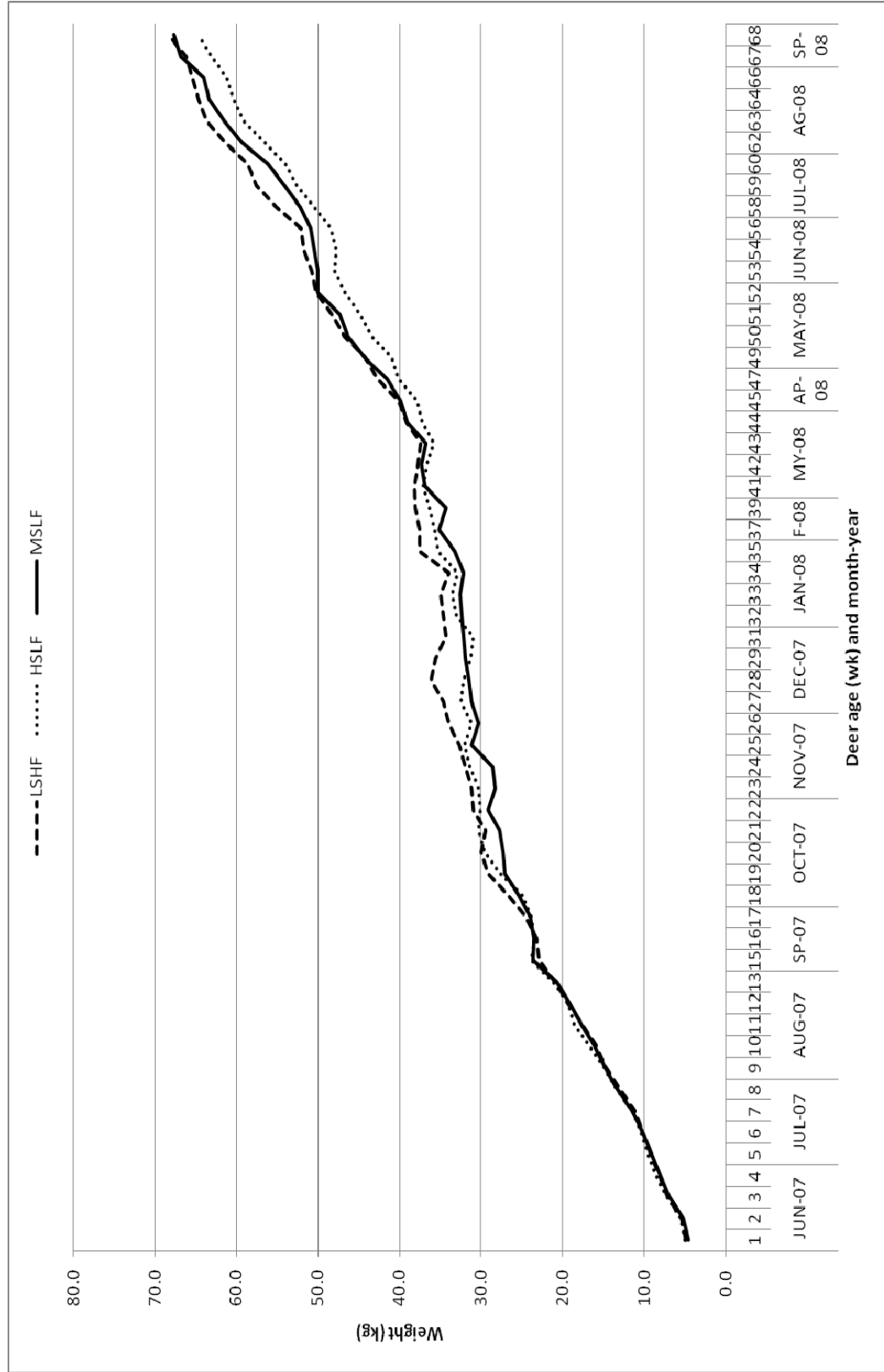
interstitial lp = interstitial lymphoplasmacytic inflammation (kidney only)

portal lp = lymphoplasmacytic inflammation in portal areas (liver only)

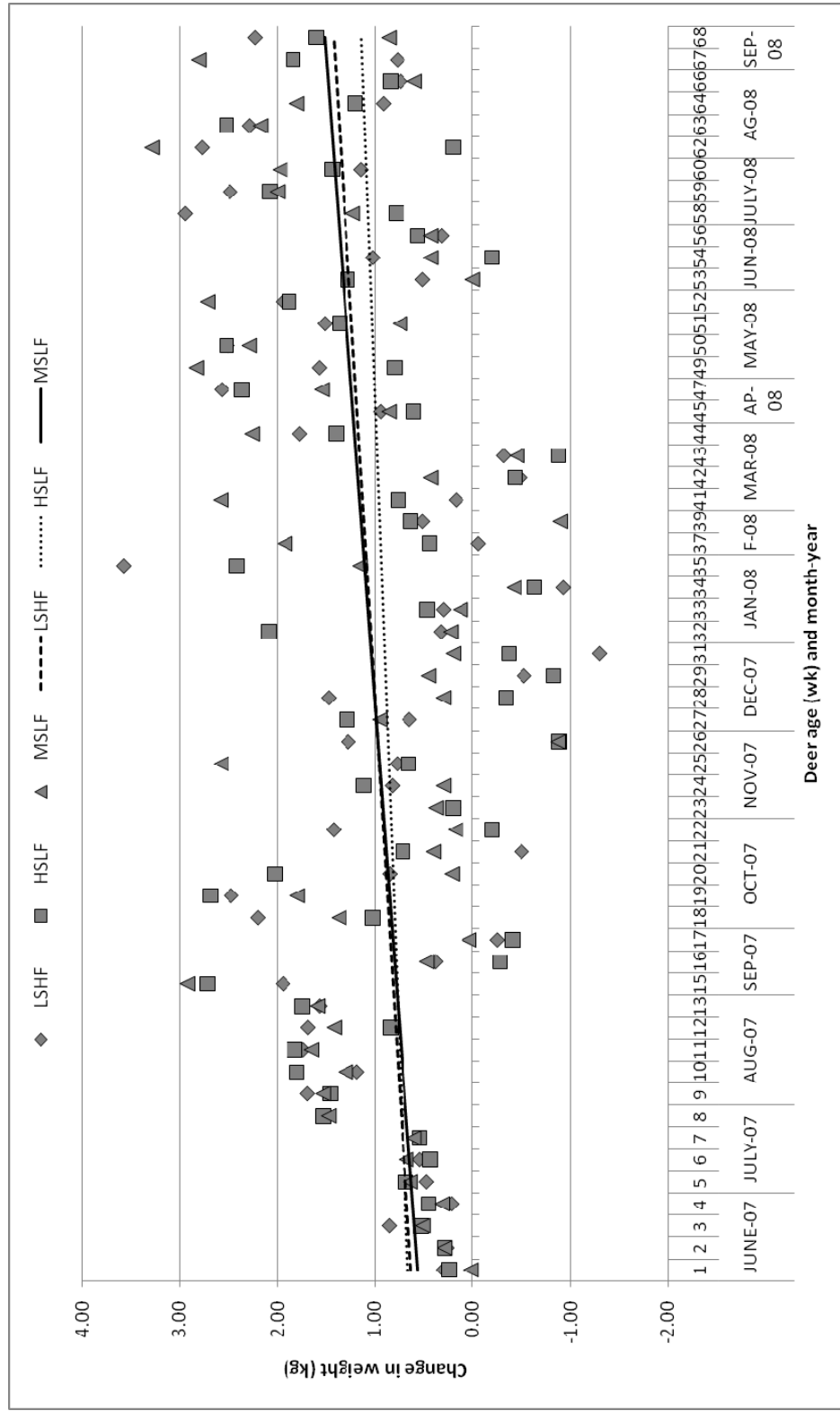
HISTOLOGIC DIAGNOSES:

1. Essentially normal tissues

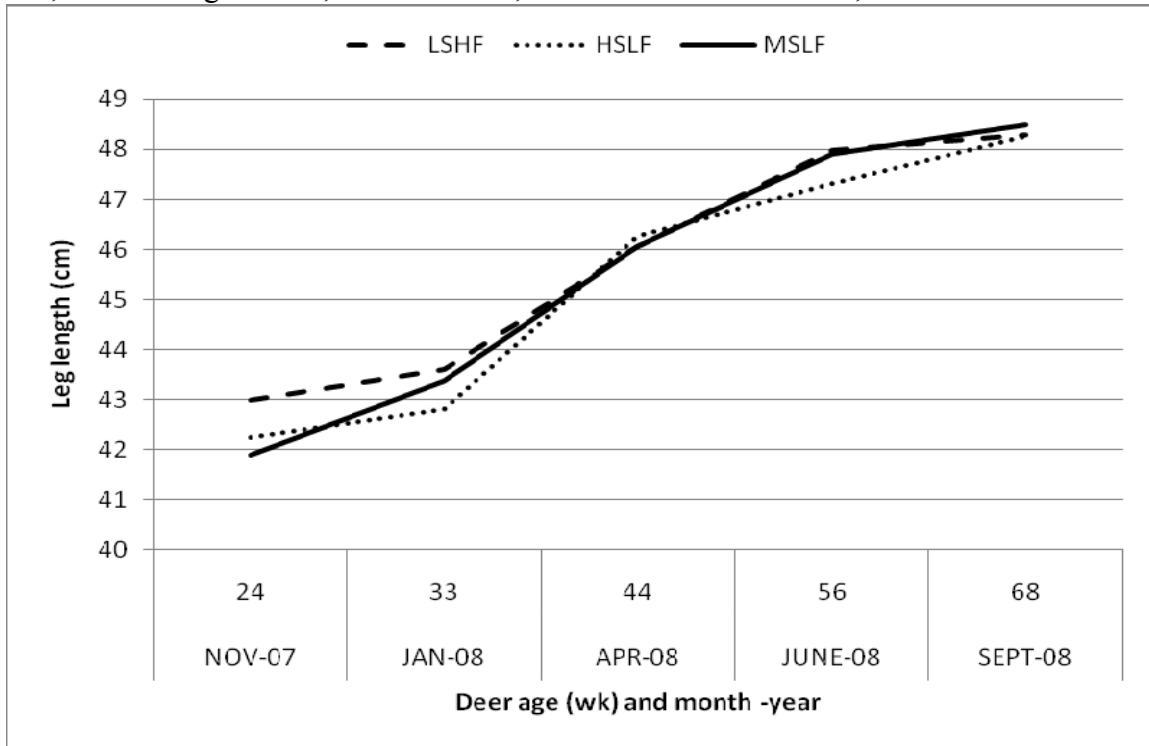
Appendix G. Body mass of captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.



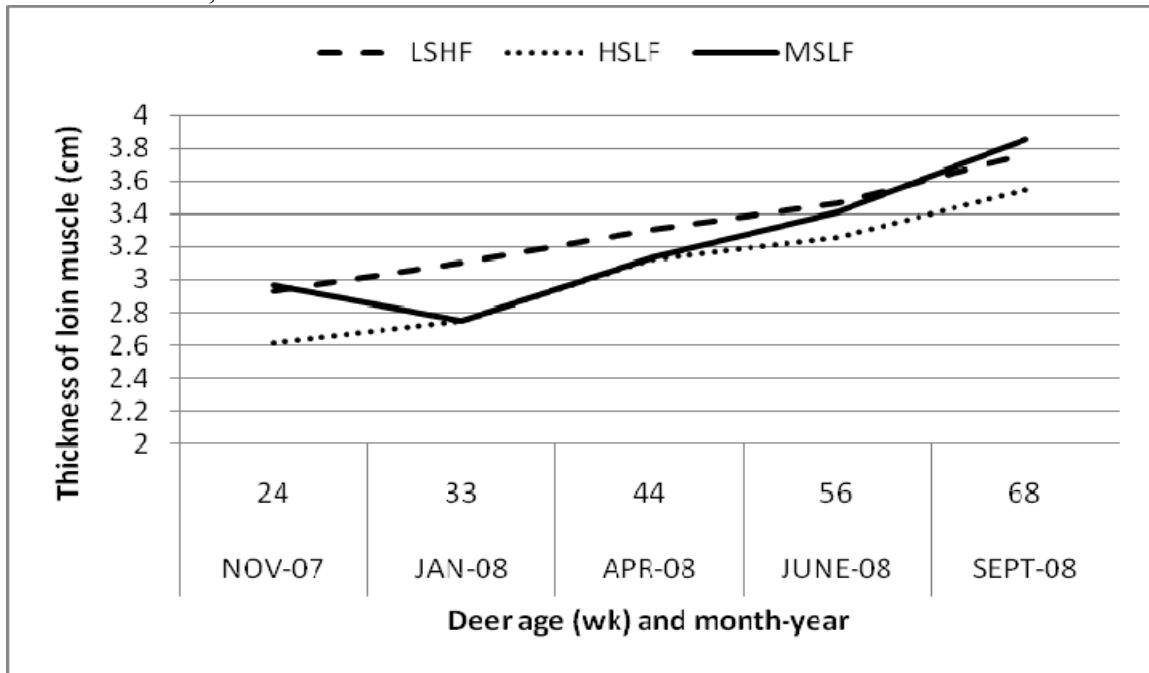
Appendix H. Change in body mass of captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.



Appendix I. Hind-leg length of captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.



Appendix J. Thickness of *longissimus dorsi* (loin) muscle of captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.



Appendix K. Rump fat thickness (MAXFAT) of captive mule deer (*Odocoileus hemionus*) consuming 3 pelleted diets from birth in May 2007 to September 2008. LSHF = low-starch, high-fiber diet; HSLF = high-starch, low-fiber diet; MSLF = moderate-starch, low-fiber diet.

