The Role of Nutrition on the Canine Hair Follicle: A Preliminary Report

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Introduction

Ask any dog owner why they are willing to devote so much time and effort toward their pet's care and maintenance and there will be a variety of answers: companionship, protection, a living thing to care for, are all likely responses. However, we contend that two of the main reasons for dog ownership will not be stated. Simply put, dogs are enjoyable to touch and look at. These tactile and visual interactions between a human and a dog are among the greatest pleasures of "pet" ownership. As a consequence, when the hair coat is thinned or looks dry and unkempt, there is almost invariably a weakening of the human-animal bond. Dogs with coat problems are simply not handled as much.[1] This is why knowledge of the canine hair follicle and the attached adnexal glands is important. In this report, we present a review of the hair follicle and preliminary data collected in a study to define the effects of nutrition upon the canine hair follicle.

Structure and Function

The hair follicle is a deceptively simple structure. Although on first consideration growing a hair shaft should not be very difficult, in reality it is an extremely dynamic process requiring complex, yet to be understood interactions between epithelial and connective tissue elements, and phases of quiescence alternating with periods of dramatic growth. The easiest way to begin to understand the hair follicle is to view it when the hair follicle is actively growing (a period called anagen). The anagen follicle consists of a tube within a tube and growing through the center of these tubes is the hair shaft. The outer tube is known as the outer sheath, the inner tube as the inner sheath, and the swelling that supports the growing hair follicle at its base is called the hair bulb. Growing within the hair bulb is a nubbin of connective tissue known as the follicular papilla, which has a regulatory role in all aspects of hair growth, involution, and senescence (***Figure 1***).

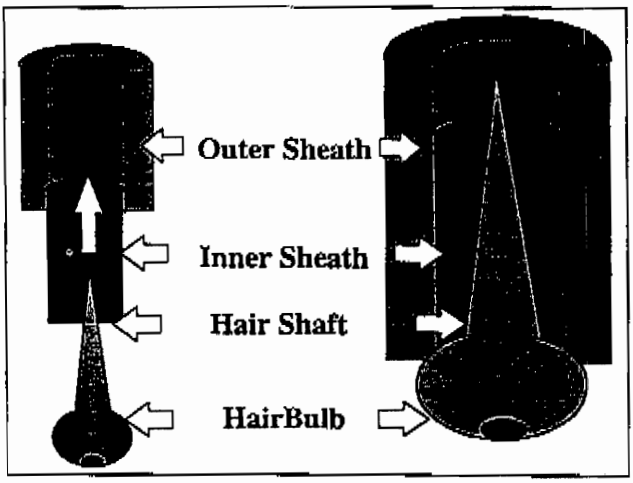


Figure 1. An anagen hair follicle.

Starting from the opening of the hair follicle, a region known as the follicular os, and working down to the hair bulb, the hair follicle can be divided into 3 anatomic segments: the infundibulum, the isthmus, and the inferior region. Traditionally, these areas have been divided based on the following sites of insertion: the infundibulum extends from the follicular os to the insertion of the sebaceous gland duct, the isthmus extends from the insertion of the sebaceous duct to the insertion site of the arrector pili muscle, and the inferior segment extends from the insertion of the arrector pili muscle to the base of the follicle (***Figure 2***).

There are several important differences in the structure of hair follicles across species. Most omnivores and herbivores have "simple" follicles. This means that each infundibulum contains a single hair shaft that exits through the os. Exceptions include fiber-bearing herbivores like goats, sheep, or llamas, which may have 2 to 3 hairs per infundibulum. Carnivores, in contrast, have "compound" follicles. That is, there are multiple hair follicles growing closely together that unite at the upper isthmic region, share a common infundibulum, and have multiple hairs exiting through a common os (***Figure 2***). By convention, the largest hairs in a compound follicle are called primary or guard hairs and the smaller hairs that make up the majority of the hair shafts in a compound follicle are called secondary or undercoat hairs. It should also be noted that some species (sheep, rats, mice) often retain an old hair shaft concurrent with a newly emerging hair. This does not constitute a compound follicle even though 2 shafts may be found in the infundibulum.

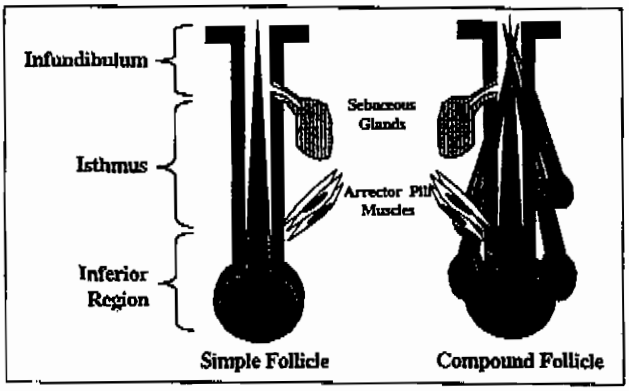


Figure 2. Histologic differences between a simple and compound hair follicle.

There is a striking symmetry to the orientation of the hair follicles of carnivores. Rather than being randomly dispersed over the skin, a primary hair with its secondary hairs generally occurs in groups of 3. We call this grouping of follicles a *follicular unit*. Thus, if one looks with a magnifying lens at the surface of the skin of a dog whose hair has been closely clipped, the follicular unit will typically appear as 3 openings close together, then a space, and then another 3 openings close together. In addition, there is also a distinctive orientation of the primary and secondary hairs. Primary hairs are always the most cranial (toward the head) with the largest primary hair present in the center and somewhat in front of the 2 flanking primary hairs. Secondary hairs are caudal (toward the rear) of the primary hairs. The secondary hairs that are closest to the primary hairs are the largest and become progressively smaller the more caudally they are positioned. The arrector pili muscles segregate the 3 compound follicles within the group. In this way, the hair follicles are designed so that the hairs will lie down smoothly, with the guard hairs lying on top of the fine undercoat. The common concept is that dog hair consists of 2 distinct hair types, primary and secondary. This is not true. In reality, there is a continuum of diameters and the only way to separate a primary from a secondary hair is to either establish an arbitrary cutoff point diameter or to define primary hairs anatomically; the most anterior hair in each group of follicles can be considered a primary hair regardless of its diameter (***Figure 3***). The ratio of secondary to primary hairs can be >10:1.

In most mammals, the follicles begin to develop prenatally in one location, such as the crown of the head and spread in a wave over the body. In many species with a dense haircoat, such as dogs, the primary follicles emerge first and the smaller, secondary follicles grow from the primary follicles.[2]

Several large groups of proteins are involved in the formation of hair and an estimated 50–100 individual proteins comprise the hair fiber. The 2 major groups of proteins that make up the bulk of the hair are (1) hair-specific keratin intermediate filaments that are also referred to as "low sulfur" or "hard" keratins to distinguish them from the epidermal keratins and (2) the proteins that organize the keratins into a matrix, called keratin-associated proteins or intermediate filament associated proteins. As in the epidermis, the follicular assembly of keratin filaments requires that 2 different but structurally related protein molecules, one being acidic (type 1 keratin) and one being neutral to slightly alkaline (type 2 keratin), polymerize to

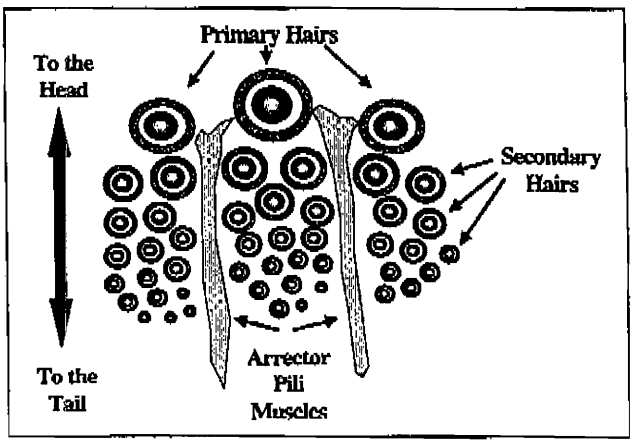


Figure 3. The canine follicular unit examined by transverse sectioning.

ultimately form 8–10 nm diameter filaments. The stability of the keratin filaments depends on a number of interactions, including covalent cystine cross links, interactions between side group chains, hydrogen bonds, and hydrophobic interactions.[3] The keratin-associated proteins consist of 3 large families, the "high sulfur" group, the "ultra high sulfur" group and the "high glycine/tyrosine" group, each containing many sub-families.

The Hair Cycle

The growth of hair is cyclical and most of the morphologic changes associated with the cycle occur in the lower half of the hair follicle. The active growth phase of the hair cycle is called "anagen" and is characterized histologically by a hair bulb surrounding an inverted follicular papilla and a fully formed inner sheath. The involutionary phase between active and no growth is known as "catagen". In catagen, there is loss of the hair bulb and separation from the follicular papilla. Although a thickened follicular sheath and increased numbers of apoptotic cells in the outer sheath are present, the most defining feature of catagen is the partial replacement of the inner sheath by trichilemmal cornification as the hair bulb is lost and the follicular papilla evaginates. Telogen is the stage of senescence and is characterized by complete replacement of the inner sheath with trichilemmal cornification. Telogen follicles are approximately one-third the length of anagen follicles. The portion of the outer sheath at the base of the telogen hair is composed of a small, compact nest of basaloid cells known as the hair germ. This cluster of cells sits on the follicular papilla which is poorly defined when not invaginated within the hair bulb. Telogen is a physiologic, not pathologic process and is as much a part of the hair cycle as anagen. Telogen only becomes pathologic when a new anagen hair does not soon replace a telogen hair and the follicle continues to involute.

The length of time required to complete the hair cycle varies between different species, and is different among dog breeds. In human scalp hair, anagen is the longest phase of the cycle; thus the hair grows almost constantly, resulting in the need for haircuts. In most mammals, the hair cycle is telogen based: the hairs grow to a preordained length and then enter a long period of inactivity in which the telogen hair follicle firmly retains the hair shaft. A good example of telogen-based hair cycles occurs in quilled mammals. In 1996, we described the morphologic features of quill follicles in porcupines, hedgehogs, and echidnas. We found that after the quills had grown to a set length, the follicles entered into a "haired telogen" state in which the quill was tightly annealed to the outer sheath. These quills were neither shed nor grew. In hedgehogs, the quills could remain for years and perhaps for the life of the animal. The value of growing hair and then retaining it is likely in the conservation of protein and the energy required for their synthesis.[4] We believe that the hair cycle in most canine breeds is more like that of quilled mammals than humans.[5] In short, dogs have telogen-predominant hair cycles in which the hair shafts are retained in telogen follicles for long periods of time. How long the follicle remains in telogen is a breed-specific phenomenon. In some canine breeds such as the Nordic breeds, the hair appears to be held in a telogen state, sometimes for years. In other breeds of dogs, such as Poodles, the hair cycle is anagen based and these dogs, like humans, need to have haircuts (***Figure 4***).

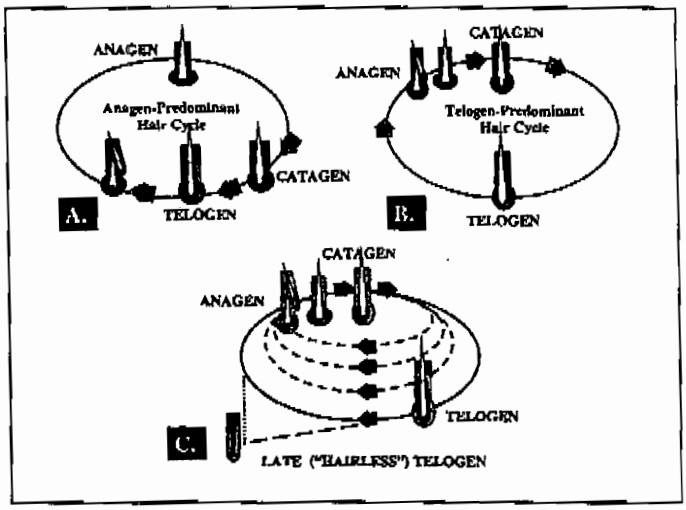


Figure 4. Species and canine breed differences in the mammalian hair cycle. Stereotypically, there are 2 major hair cycle patterns, one in which the predominant phase of the hair cycle is anagen (A) and one in which the predominant phase of the hair cycle is telogen (B). We believe most dogs have telogen-based cycles but the length of the anagen phase appears to have considerable breed variability (C).

Factors Controlling the Hair Cycle

All hair follicles have an intrinsic rhythm that can be altered by systemic factors. The interplay between local (intrinsic) and systemic (extrinsic) factors in hair cycle control has been illustrated by grafting studies in rats. Flaps elevated on the flanks of rats and rotated 90–180 degrees and then replaced, continue to shed in the rhythms of their original sites for long periods. Similarly, grafts between syngeneic mice of different ages also retain the shedding pattern of the donor. Eventually, however, the hair cycles in the grafts and flaps synchronize with the surrounding skin.[6]

In the hair follicle, intrinsic factors (known as growth factors or cytokines) are produced by and act on a variety of different cell types including the cells of the hair matrix, inner sheath and outer sheath, mesenchymal cells of the follicular papilla, fibroblasts surrounding the follicle, and endothelial cells of the blood vessels that supply the follicles (***Table 1***).[7-25]

Extrinsic factors are those that are produced in another organ and are transported to the follicle via the peripheral blood. Hormones are the best-defined extrinsic factors. Those that effect hair growth include melatonin that acts synergistically with prolactin, gonadal and adrenocortical sex hormones that can inhibit or stimulate hair follicles depending on hormone and body location, glucocorticoids that inhibit hair growth, and thyroid hormones that stimulate hair growth.[26-37]

Both intrinsic and extrinsic factors work together to control the length of the cycle, and therefore, the genetically determined length of the hair. The molecular

basis for the morphologic changes that occur in the hair follicle cycle is largely undefined and there are still no regulator molecules identified that are unique to the hair follicle.

Table 1. Selected growth factors and the action they have on hair growth (see references 7–25)

Intrinsic Factor	Site(s) of Production	Site(s) of Receptor(s)	Effect on Hair Follicle
Epidermal growth factor	• Outer sheath	• Outer sheath • Hair bulb	In vitro: Stimulates hair growth In vivo: Retards hair growth
Transforming growth factor-Alpha	• Outer sheath	• Outer sheath • Hair bulb	In vitro: Stimulates hair growth In vivo: Retards hair growth
Fibroblast growth factors (FGF) FGF-1,2	• Outer sheath • Inner root sheath • Hair bulb basement membrane	• Outer sheath • Inner root sheath • Hair bulb basement membrane	In vitro: Stimulates hair growth In vivo: Inhibits hair growth
Transforming growth factor-beta	• Entire follicle	• Entire follicle	• Inhibits hair growth
Insulin-like growth factor	• Follicular papilla	• ?	Stimulates hair growth
Interleukin-1	• Follicular papilla	• Entire follicle	• Inhibits hair growth
Vascular endothelial growth factor	• Follicular papilla • Hair bulb	• ?	Stimulates hair growth
Hepatocyte growth factor/scatter factor	• Follicular papilla	• Matrix cell	• Stimulates hair growth

Effects of Nutrition on the Hair Follicle

Because of the dynamic nature of hair growth and because hair is composed primarily of proteins held together with lipids, a large quantity of the diet is required to maintain a normal pelage. Buffington has stated that 30% of the daily protein requirement of dogs and cats can be utilized for the synthesis of keratin in skin and hair.[36] In studies using pigs and rats, periods of chronic protein deprivation resulted in protein synthesis decreasing more in the skin than in any other major organ.[37,38] Unfortunately, the authors of these studies do not speculate as to where the decrease in cutaneous protein synthesis occurred. Considering that the epidermis is essential for life, it is unlikely that biochemical corners could be substantially cut there. In addition, the dermis is a relatively stable tissue with a low protein turnover rate. Thus, it is difficult to conceive that decreasing the production of the dermal protein and glycosaminoglycans would result in substantial protein conservation. The only site in the skin where protein can be conserved with little or no harm to the dog, is the hair

follicle. For this reason, dietary deficiencies in amino acids, carbohydrates, essential fatty acids, or any of the vitamins, metals, or minerals required for protein or lipid synthesis can all affect the hair follicle. These changes are best defined in profound deficiency states. Starvation is associated with hair loss and/or poor hair quality as is a deficiency in copper, zinc, vitamin A, vitamin C (in species that cannot synthesize it), vitamin D, vitamin E, riboflavin, nicotinic acid, biotin pyroixidine, pantothenic acid, cyanocobalamin, zinc, and copper.[39-42] However, there is evidence that even mild changes in diet can cause a change in the diameter of the hair follicle. For example, in angora goats, there is a positive correlation with body weight and hair diameter.[43]

Much of what we know concerning the effects of nutrition on the hair follicle is based on studies in humans, rodents, or animals used for fiber production (sheep and goats).[39-44] Whether knowledge based on these species can be applied to dogs is unknown; however, there is evidence to suggest that interspecies comparisons may not be valid. First, the basic anatomy of canine hair differs from humans, mice, sheep, and goats. Mice and humans have simple follicles and sheep and goats have some follicles that are simple and others that are compound. None of these species have the number of hair shafts exiting through a follicular os as the dog. Furthermore, only dogs have the symmetry that allows for distinguishing anatomic primary hairs (the 3 anterior follicles in each of the grouping of follicles that makes a follicular unit). In addition, with the exception of breeds such as the Poodle that require their hair be trimmed, for most canine breeds, the telogen stage is a long, if not the longest, stage of the hair cycle. This is not true for humans, mice, or animals used for fiber. We theorize that the reason why many canine breeds have such a long telogen phase is because it conserves both energy and proteins. By this same logic, Nordic breeds have a longer haired telogen stage than other breeds because they need a thick hair coat to survive the cold and need to conserve the protein within hair during the winter months when sources of protein may be difficult to obtain. Similarly, Poodles, a breed that constantly grows its hair, may have different nutritional requirements than a breed that can maintain its hair in a state of metabolic "suspended animation".

We are currently involved in a study to define the effects of nutrition on the skin. A major objective of this study is to determine if changing the diet can influence the growth and quality of the hair coat.

Techniques to Study the Hair Follicle

Over the past 15 years, there have been great technological advances in the morphologic analysis of the hair of humans and fiber-producing animals; however, none of these methods have been applied to companion animals.

DermScope analysis of hair. The DermScope is a portable, external microscope that produces a high-quality digital image. To date, the instrument has been used primarily by dermatologists to evaluate surface changes of human skin. We have adapted its use to serve as an external scanning microscope allowing us to examine the hair coat at magnifications of up to 600X.

Analysis of horizontal sections of skin biopsy samples. In 1984, Headington described a remarkably simple idea that revolutionized the way dermatopathologists looked at the hair follicle.[45] Rather than trimming a punch biopsy of skin vertically (from the epidermis to the subcutis), he suggested that the best way to examine the hair follicle and its diseases was to trim the biopsy transversely (through the dermis on

a plane parallel with the epidermis). The resultant histologic sections were more informative because they demonstrated all the hair follicles in a punch biopsy. For the first time serial sections were not needed to determine the severity of an alopecia or to determine the number of hair follicles that were in an actively growing or an atrophic state. In addition, transversely sectioned hair shafts were relatively easy to measure, thereby allowing for morphometric analysis. The Headington technique resulted in hundreds of publications associated with quantitative assessment of hair loss and remains a widely used technique in diagnostic dermatopathology in humans.[46,47] As useful as this technique is to examine human hair follicles, it is arguably of greater value in evaluating canine compound follicles. Dogs can have more than 30 hair shafts/follicular infundibulum. Trying to see all these hairs with vertical sectioning is almost impossible without making dozens of slides. In addition, only by transverse sectioning can the pattern of hair growth be defined and morphometically analyzed. This method also allows for the determination of accurate anagen to telogen ratios.

Optical-based Fiber Density Analyzer (OFDA). The OFDA is essentially a computerized microscope that is positioned over a moving sample of cleaned hair shafts that have been cut in 2 mm lengths. The microscope magnifies and captures video images of individual hair shafts. The width of each identified fiber image is then measured. In addition to fiber diameter, fiber curvature is also assessed. After measurement, the data are presented in a final histogram showing the fiber diameter and curvature distribution. Standard deviation and coefficient of variation for both diameter and curvature are calculated. The advantages of using the OFDA to evaluate hair shafts lie in its accuracy (the instrument is capable of a resolution of 1 micron and can accurately determine diameters in the range of 4 to 300 microns) as well as its speed (>10,000 fibers can be measured/minute).[44] In addition, we believe each canine breed will have an OFDA-generated histogram that is either characteristic or, at a minimum, will allow for the classification of a given breed into one of a small number of breeds with similar coat types.

Materials and Methods

All procedures dealing with animal care, handling and treatment in this study were reviewed, approved, and subjected to continuous monitoring by the Animal Use Committee, Texas A&M University.

Animals used in study and diets. This study was performed on the same population of animals used to study the effects of sebum on diet. Refer to chapter by Dunstan et al. in this proceedings entitled "The Role of Nutrition on Canine Sebum Secretion: A Preliminary Report" for details about the animals and diets used.

Sample collection. Samples were collected the day before the initiation of feeding the experimental diets and at 3-week intervals for 18 weeks. To avoid any problem with diurnal rhythm, all samples were obtained between 9:30AM and 11:30AM. Hair samples were obtained from the dorsal lateral trunk in an area extending from the lumbar area to caudal cervical region. Because of size limitations of the puppies, different areas were used for different assays. For DermScope evaluation, the dorsal lumbar area was used. For rate of hair growth studies and OFDA analysis, the dorsal anterior thoracic/caudal cervical area was used. Skin biopsies were obtained from the dorsolateral thoracic area.

DermScope evaluation. To prepare the clipped site for the DermScope evaluation, the area was wiped gently with an alcohol-soaked cotton pad. Multiple images were obtained using the 200X lens and compared to vertical and transverse histologic sections.

Rate of hair growth determination. Rate of hair growth was determined after the 18-week study was completed. An area over the right lateral caudal cervical/anterior thoracic region was used for sampling. The entire region was clipped closely using a #50 blade. Hair was saved to serve as a baseline to define "normal" length. The regrowing hairs were sampled at days 3, 7, 14, 21, 28, and 42 with each sample being obtained just anterior to the previous sample site. The clipped samples were affixed to a glass slide using 2-sided tape and coverslipped. Fifty of these hairs were digitized with a calibrated Sony® camera to insure that the same magnification was used for each measurement. The length of the hair on these images was determined by tracing and the data were converted to millimeters using NIH Image.

OFDA analysis. For OFDA analysis, an area approximately 4cm X 4cm was clipped. These clippings were briefly degreased and trimmed in 2mm lengths ("snippers"). Snippets were placed on an automatic spreader that distributed the hairs evenly and randomly on a hinged slide. The prepared slides were then placed under the microscope and measured for diameter and curvature.[18]

Histologic and morphometric evaluation of skin biopsy samples. Two 6mm skin biopsies were used for morphologic and morphometric assessment of the hair follicles. Biopsies were obtained under local anesthetic using standard surgical techniques and samples were fixed in 10% neutral buffered formalin. After fixation, one biopsy sample was trimmed vertically. The other biopsy sample was sectioned horizontally. Samples were routinely processed, embedded cut side down, sectioned at 5 µm, and stained with hematoxylin and eosin. Routine descriptive morphologic evaluation was performed on the vertically-sectioned samples. The horizontally-sectioned samples were step-sectioned until 5 complete follicular units were identified. The horizontally-sectioned samples were evaluated routinely and morphometrically. The morphometric analysis took 2 forms: (1) the stage of the hair cycle was defined for each follicle in the 5 follicular units evaluated and (2) the number of hairs/follicular unit was counted. A hair follicle was considered in anagen when an inner root sheath surrounded the hair shaft at the level of the follicular isthmus. A hair follicle was considered in physiologic telogen when trichilemmal cornification surrounded the hair shaft at the isthmic region. A follicle was considered in "end stage" or pathologic telogen when the hair follicle at the level of the isthmus was devoid of a hair shaft and either had luminal trichilemmal cornification but no hair shaft or was simply an epithelial tube with no luminal cornification. Because catagen hairs are impossible to consistently recognize on transverse sections and are so uncommon (< 2% of all follicles), no attempt was made to segregate them in this study. The anagen and telogen hairs were then categorized depending on whether they were anatomic primary or anatomic secondary hairs.

Results

DermScope evaluation. DermScope examination defined differences in the number and pattern of hair shafts erupting through the follicular os. As a rule, the compound follicles were arranged in groups of 3, although some compound follicles were solitary and groups of up to 5 were occasionally identified. DermScope evaluation

was detailed enough to allow for assessment of the cuticular pattern of erupting hair. In all breeds, no differences in the structure of the hair shafts were noted and no differences could be attributed to diet.

The DermScope was also used to evaluate the epidermis. In all dogs, the epidermis was nonpigmented and opaque, allowing visualization of the dermis and to some extent, the base of pigmented hairs. Occasionally, tylotrich pads, a localized neuroreceptor organ associated with follicles were identified. These appeared as slightly raised, white, thickenings of the epidermis. No differences in the epidermis based on diet were identified using the DermScope.

Histologic and morphometric evaluation of skin biopsy samples. The vertically-sectioned samples defined a dermis that became somewhat thicker with time from the beginning of the study to the end. Sebaceous glands also became larger. The overall follicular morphology was difficult to discern by this method of trimming. As was noted with the DermScope examination, most follicular units were arranged in groups of 3 but occasionally, follicular units consisted of a single compound follicle and sometimes follicular units had 4 or more compound follicles. As a rule, the larger the number of compound follicles in a follicular unit, the larger the central anatomic primary follicle and the hair shaft it produced. No discernable differences could be attributed to diet.

Analysis of the horizontally sectioned skin biopsies is presented in ***Table 2***. Siberian Huskies fed the high-quality diet had an increased number of hair follicles per follicular unit and hair follicles with hair shafts when compared to those fed the low-quality diet at weeks 9 and 18. In contrast, Labrador Retrievers and Miniature Poodles fed the high-quality diet had numerically fewer follicles/follicular unit at week 18, but not week 9. As a percentage of the week 0 value, Labrador Retrievers fed the high-quality diet had more follicles/follicular unit at weeks 9 and 18, while Miniature Poodles did not appear to be affected by diet at either week 9 or 18 relative to week 0. In neither Labrador Retrievers nor Miniature Poodles was a difference noted in the number of hair follicles with hair shafts/follicular units. In all breeds, substantive differences related to diet were difficult to demonstrate. Overall, there was little to distinguish the dogs fed the high- and low-quality diets; however, this data is somewhat altered because there was a moderate increase in the number of follicles in telogen in the Siberian Huskies and Miniature Poodles fed the high-quality diet but a marked decrease in the number of telogen follicles in the Labrador Retrievers fed this diet.

Table 2. Morphometric analysis of horizontally-sectioned skin biopsy samples

	Low-quality Diet			High-quality Diet		
	Week 0	Week 9	Week 18	Week 0	Week 9	Week 18
Huskies						
Hair follicles/follicular unit	35	38.5	43.2	28.4	55.3	52
Hair follicles with hair shafts/follicular unit	24.9	29.4	35.2	22.2	49	45.5
Hair follicles in anagen/follicular unit	24.8	17.6	25.4	20.6	35	25.6
Hair follicles in telogen/follicular unit	13.6	22	17.7	7.9	20.4	26.4
Poodles						
Hair follicles/follicular unit	16	19.7	23.6	14.7	20.3	21.8
Hair follicles with hair shafts/follicular unit	11	15.4	21.3	10.3	15	18.8
Hair follicles in anagen/follicular unit	11	15.4	21	10.3	15	18.8
Hair follicles in telogen/follicular unit	5.1	4.3	2.5	5.5	5.3	3.2
Labrador Retrievers						
Hair follicles/follicular unit	25.8	21.7	31.5	17.5	32.2	24.4
Hair follicles with hair shafts/follicular unit	13.7	13.5	19.5	11.7	29.2	19.8
Hair follicles in anagen/follicular unit	10.4	5.1	9.7	7.2	8.8	11.2
Hair follicles in telogen/follicular unit	15.5	16.6	21.8	10.3	23.3	13.1

OFDA Analysis. The data obtained from OFDA analysis was interpreted by analyzing the diameter and curvature of all the hairs measured based on histograms and by analyzing the mean diameter and curvature. Overall, the 2 diets had no demonstrable effect on mean diameter or curvature over the time frame of the study for the Labrador Retrievers and Miniature Poodles; however, the Siberian Huskies fed the high-quality diet had slightly greater curvature than those fed the low-quality diet (***Table 3***).

Rate of hair growth. As defined in ***Table 4***, there was no substantive difference in the rate of hair growth based on diet.

Table 3. Optical-based Fiber Density Analyzer (OFDA) analysis of mean hair diameter and degree of curvature based on the effects of breed and diet over time. Diet 1 = low-quality diet; Diet 2 = high-quality diet. Each bar represents the mean of 6 dogs ± SEM.

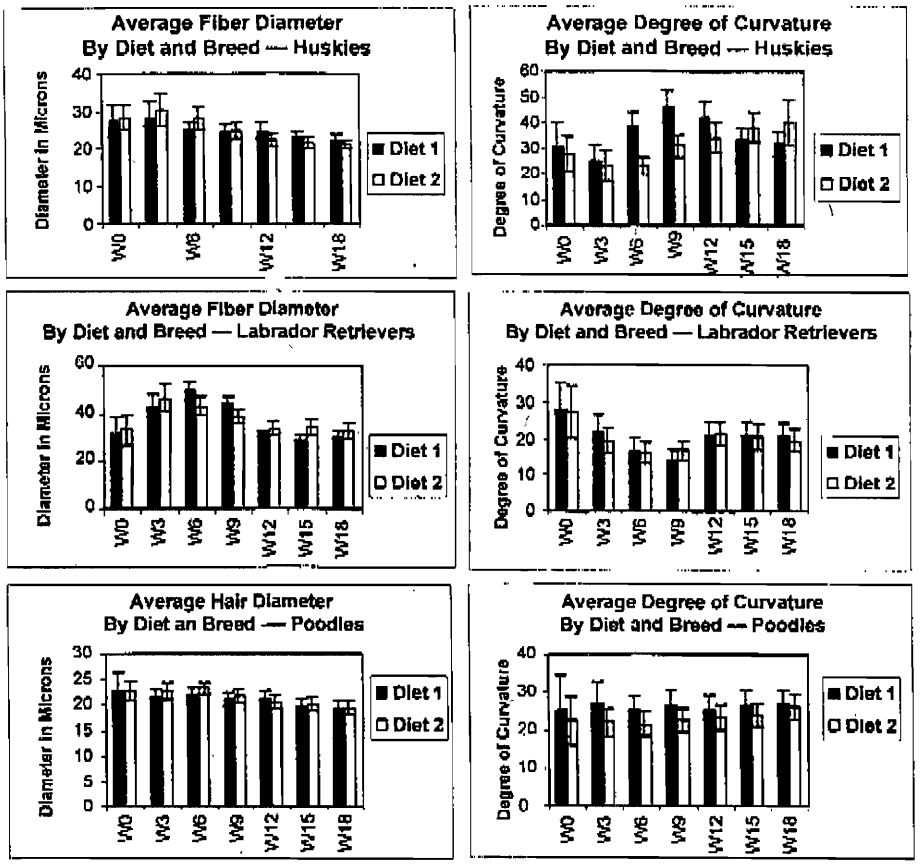
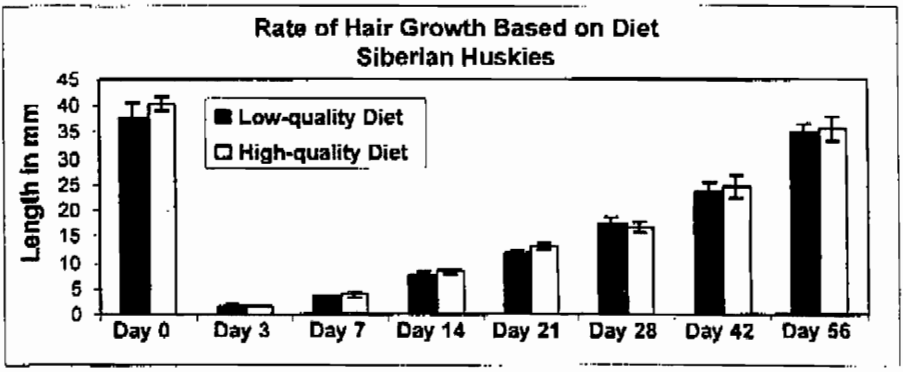
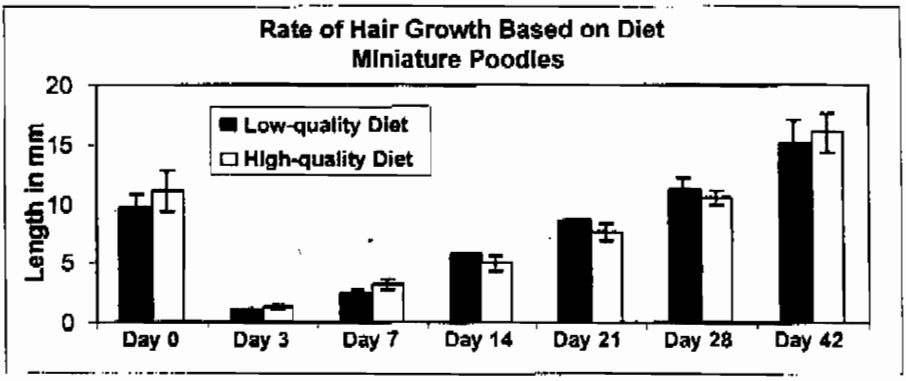
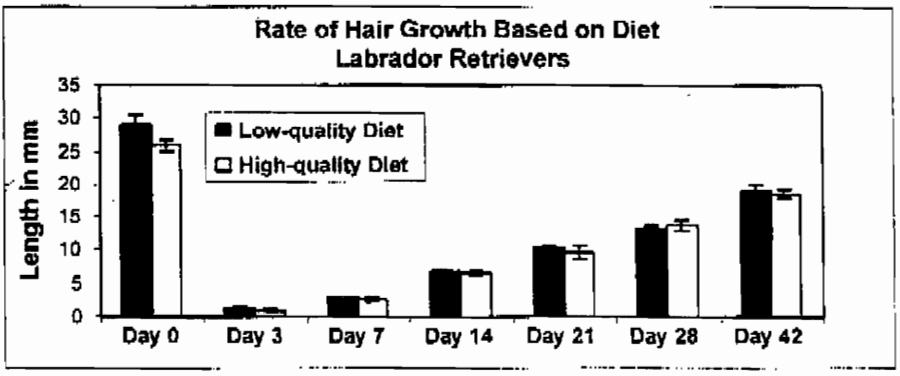


Table 4. Graphs represent the rate of hair growth in 3 canine breeds over a 42-day period, based on diet. Each bar represents the mean of 6 dogs ± SEM.







Discussion

The effects of diet on the canine hair follicle were subtle when compared to the effects of breed and/or time. DermScope and OFDA analysis failed to identify differences with the exception of the greater curvature for Siberian Huskies fed the high-quality diet. On evaluation of horizontal sections, there was a greater increase in the number of follicles/follicular unit in Labrador Retrievers and Siberian Huskies fed the high-quality diet. Diet quality had no effect on Miniature Poodles. We have no good explanation for the disparity of results in the different breeds studied.

The OFDA analysis also failed to demonstrate differences in mean diameter of hair shafts based on diet. Siberian Huskies fed a high-quality diet had a slightly greater curvature than dogs of the same breed fed a low-quality diet.

The percentage of telogen follicles/follicular unit decreased in Labrador Retrievers fed the higher quality diet but increased slightly in Siberian Huskies and Miniature Poodles fed the higher quality diet.

There was no evidence that the 2 diets fed these dogs resulted in a different rate of hair growth.

How do we explain the lack of changes due to diet? One conclusion could be that the low-quality diet was not so nutritionally wanting and/or the high-quality diet may not have been so beneficial to the hair follicle to induce definable changes. However, we contend that an equally likely and currently being tested reason for the lack of discernible differences in the parameters evaluated is that the 18-week feeding period used in this investigation was not enough time to induce dietary-related changes.

A feature of hair follicles of most mammalian species is their ability to retain hair shafts for long periods of time in a prolonged telogen state. Analogous to follicular hibernation, such maintenance of the hair coat requires little energy or protein expenditure. We have proposed that many dogs have a telogen-based hair cycle, meaning the majority of the hair follicles are retained in this haired telogen state. Based on data from this study, Siberian Huskies and Labrador Retrievers fall into this category. At the end of the 18-week dietary trial, Labrador Retrievers and Siberian Huskies fed the high-quality diet had 56% and 50% of their hair follicles in telogen, respectively. How the hair follicles in these breeds respond to differences due to dietary quality is unknown. We have shown that Beagle dogs with ^{131}I-induced hypothyroidism do not become alopecic but do not regrow clipped hair because the majority of their follicles are in a prolonged haired telogen state. Based on this study, we hypothesize that only healthy young dogs shed their hair because they are in a state of nutritional and metabolic health that would allow for lost hairs to be replaced. If our theory is correct, then over a prolonged period the dogs fed a poor diet will have a gradual increase in the number of hairs in telogen and these hairs will not be shed until these dogs are put on a higher nutritional plane. A study that will allow us to prove, disprove, or modify our hypothesis is now in progress.

Why the hair cycle of Miniature Poodles seems unaffected by the differences in diet remains unanswered. Because the hair of this breed, like human scalp hair, has an anagen based cycle (ie, the majority of the hair follicles are in anagen) and Miniature Poodle hair follicles do not "hibernate" for a prolonged period of time, we expected this breed to be the most sensitive to dietary changes. Clearly, this was not the case in this study. Recently, genes associated with continuous hair growth have been identified in the mouse (*angora*).[17] The possibility exists that in Poodles, a canine angora

gene or, an analogue, may inhibit the normal regulatory mechanism for conserving hair growth by putting hairs in telogen. Thus, the moderate differences in the high- and low-quality diets used may not be enough to shut off the messages that tell the hair follicle to continue growing. Regardless of the reason, hair growth in Miniature Poodles is distinctly different than in Siberian Huskies and Labrador Retrievers.

Finally, tempting as it would be to conclude that external evaluation by the DermScope, OFDA analysis, and use of hair clipping to determine the rate of hair growth *in toto* could be used to replace the skin biopsy to assess the health of the hair coat, at present, this is premature. The value of the skin biopsy sample, especially when horizontally sectioned, in the evaluation of the canine hair follicle cannot be understated. The skin biopsy remains the best way to determine the number of hair shafts/follicular unit and remains the only way to determine the anagen:telogen ratio.

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