IDENTIFICATION OF DOG BREEDS ON THE BASIS OF MEDULLARY INDEX, SCALE PATTERN AND SCALE COUNT

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Abstract: Presence of hair is a distinguishing character of Class-Mammalia. The microscopic characters, viz. scale pattern, scale count, colour of transverse section, shape of cross section, medullary index, pattern of medulla etc are specific for particular species. Thus hair taxonomic studies, called trichology, can be used in taxonomy, wildlife forensic, archaeology, food habitat studies etc. (*Kennedy, 1982; Dagnall et al, 1995*). Hair analysis not only identifies species, it can also discriminate the animal breeds. The use of trichology in above sited fields provides excellent reliability, low cost and local breed studies. These will enable the demonstration of adaptability, resistance, hardiness and conservation of breeds (*Felix et al., 2014*). The present study was carried out to study the potential use, application and develop an identification keys for identifying various breeds of dogs from microscopic analysis of hairs. Since animals are involved in any types of crimes (*Pilli et al., 2014*), and DNA typing from animals enabled homicide investigations and convictions in U.S. and Canada (Halverson, Forensic identification of canine hairs: is heteroplasmy significant?), hair examinations of dog breeds will enable convictions in wildlife crimes where dogs are most commonly used.

Keywords: Trichology, scale pattern, scale count, medullary index, identification keys, wildlife forensic.

Introduction:

Mammals are characterised by presence of hairs, an epidermal appendages of ectodermal origin, which are involved in thermoregulation, tactile sensation, insulation, physical attractiveness *(Terry, 1977*), skin healing *(Taylor et al., 2000),* sexual and social communication (Randall, 2001), protection from cold, sunlight and physical damage, diagnosis of psychoactive drugs metals *(Kelly et al., 2000: Khalique et al. 2005),* health disorders and identity of hair owner *(Gurden et al. 2004)* and as a container for sequestering and excreting unwanted compounds *(Stenn and Paus, 2001; Paus and Foitzik, 2004)*.

The hairs exhibit three distinct anatomical regions; cuticle, cortex and medulla from outer to inner. The cuticle consists of flattened scale like cells that protects hair from environmental insult. The free ends of scales point towards tip of hairs while basal end interlocks with root sheath cells. Human hairs have non-repeating, imbricate patterns while animal hairs have regular repeating pattern in their cuticular scales.

The cortex forms main body of the hair and consists of elongated, spindle shaped cells. The cortical granules, viz. Eumelanin and phaeomelanin, give characteristic color to hair. Eumelanin imparts brown to black while phaeomelanin gives yellow and red colour to hair. Organization, density, size and distribution of pigments in cortex are used to differentiate hairs between and within species. Cortical fusi, air spaces formed during keratinisation, are found near the root end of hairs. Cortex is also characterised by the presence of ovoid bodies, well defined, highly dense clumps of undispersed pigment *(Robertson, 1999)*.

The last innermost layer called medulla is continuous, discontinuous, fragmentary or absent. Medulla can give an idea about the family or species of animals. The diameter of medulla is also used for identification and comparison purpose.

The root hair may contribute to determination of species of origin and the growth phase of hair cycle. Root morphology also used to distinguish animals between humans as well as within animals.

Materials and Methods:

Collection of hair samples:

Hair samples were collected from mid dorsal region of fifteen breeds of dogs using a combing brush. Hair samples were stored in transparent zip-lock air tight plastic bags until used for analysis.

Histology of hair:

Hair samples were washed three times in acetone followed by deionised water *(Chakraborty, et al. 1996)*. The hairs were imbedded in paraffin wax; paraffin blocks were made and cut into 5 µm sections using microtome. The slides were prepared from the sections. Scale casting was done by clear nail polish method.Medullary diameter and shaft diameters were measured using micrometer scale while the hair colour was described after *(Ridgway, 1886)*. The medullary index was calculated by using formula as,

Medullary index = Diameter of medulla/Diameter of shaft.

Result:

Scale casting of hairs revealed that the scales had distinct pattern. The scales of cross Pomeranian, Rottweiler and Doberman exhibited irregular wavy shape. Transverse wavy pattern was demonstrated by scales of German shepherd, Great Dane, Cross Labrador and Dalmatian. Saint Bernard and Golden Retriever though had transverse wavy pattern, the scale edges were incomplete. The scales of Boxer and Labrador retriever were irregular wavy but with incomplete edges. Simple oblique wavy shaped scale pattern was shown by Pomeranian and Lhasa breeds. Pugs had spinout type of scale pattern.

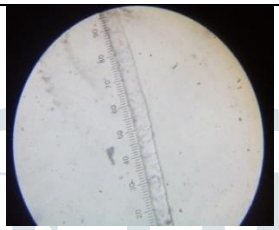
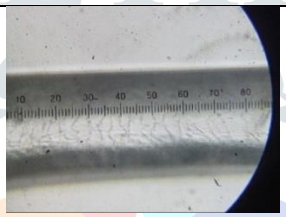
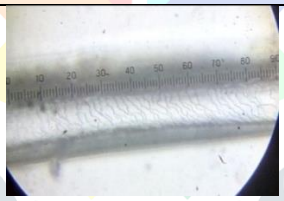
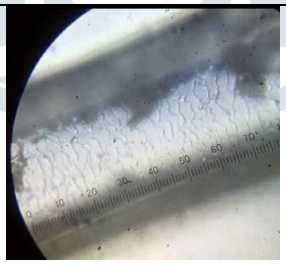
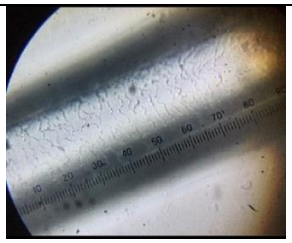
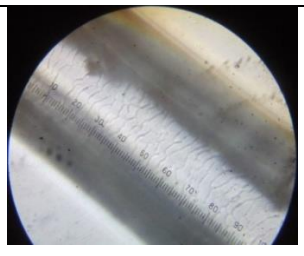
Scale count (number of scales/mm) were highest in Saint Bernard (50) while the Pug exhibited lowest (14) scales/mm. Scale count for other breeds was 27 (Cross Pomeranian), 24 (German Shepherd), Pomeranian (37), Great Dane (31), Golden retriever (28), cross Labrador (30), Dalmatian (40), Boxer (38), French Mastiff (46), Rottweiler (25), Labrador retriever (32), Doberman (29) and Lhasa (32).

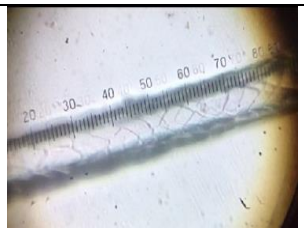
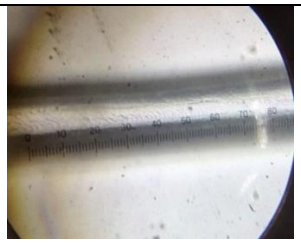
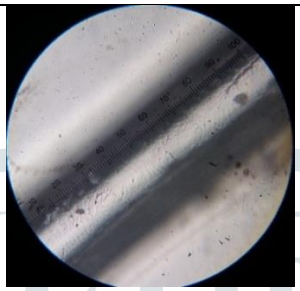
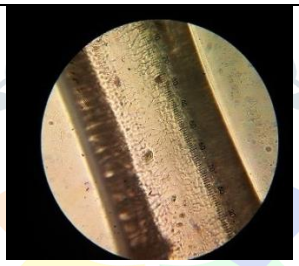
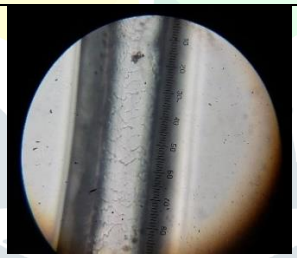
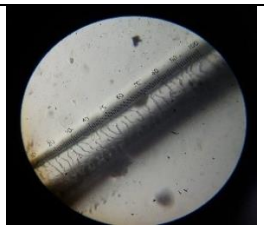
The hairs from cross Pomeranian, Pomeranian, Golden retriever and cross Labrador had transparent transverse sections. German shepherd, Saint Bernard and Great Dane exhibited brown coloured transverse section. Black colored sections were present in Dalmatian, Rottweiler and Labrador retriever. Boxer and Doberman had yellow coloured transverse sections. Lhasa had gray coloured transverse section.

Shape of transverse section also varied from one breed to another. Cross Pomeranian, German shepherd, Great Dane, Boxer, Labrador retriever, Saint Bernard and Lhasa had rounded transverse sections. Oval shaped sections were present in Pomeranian, Golden Retriever, cross Labrador, Dalmatian and Rottweiler. Doberman exhibited elliptical transverse sections.

Medullary index was highest in Doberman (0.81) while Lhasa had lowest medullary index (0.33). medullary index of remaining breeds was as Saint Bernard (0.56), Labrador retriever (0.57), Rottweiler (0.46), Boxer (0.62), Dalmatian (0.45), cross Labrador (0.63), Golden retriever (0.54), Great Dane (0.57), Pomeranian (0.40), German Shepherd (0.55) and cross Pomeranian (0.34).

Fig. 1: Scale casting of hairs showing scale pattern and scale count

Sr. No.	Name of Breed	Scale casting	Scale pattern	Scale count/mm
1	Cross Pomeranian		Irregular wavy shape	27
2	German Shepherd		Transverse wavy shape	24
3	Pomeranian		Simple Oblique wavy shape	37
4	Great Dane		Transverse wavy shape	31
5	Golden Retriever		Transverse wavy shape with incomplete edges	28
6	Cross Labrador		Transverse wavy shape	30

7	Pug		Spinouts	14
8	Dalmatian		Transverse wavy shape	40
9	Boxer		Irregular wavy shape with incomplete edges	38
10	French Mastiff			46
11	Rottweiler		Irregular wavy shape	25
12	Labrador Retriever		Irregular wavy shape with incomplete edges	32
13	Doberman		Irregular wavy shape	29

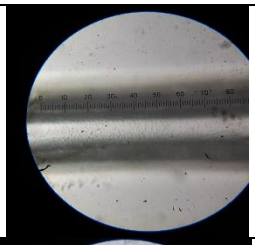
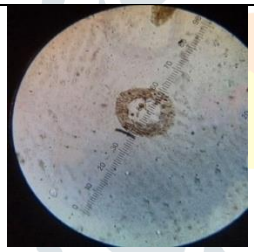
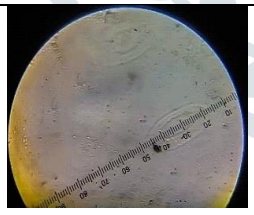
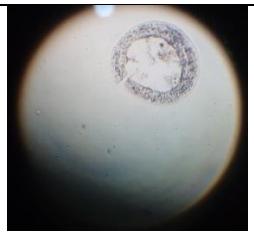
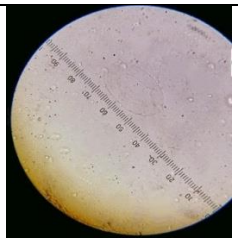
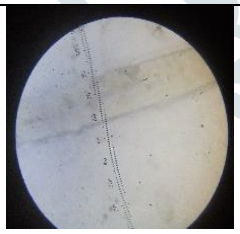
14	Saint Bernard		Transverse wavy shape with incomplete edges	50
15	Lhasa		Simple Oblique wavy shape	32

Fig. 2: Cross section of hairs showing colour, shape and size.

Sr. No.	Name of Breed	Cross section of hair	Color of cross section	Shape of cross section	Medulla size (µm)	Shaft size (µm)	Medullary index (µm)
1	Cross Pomeranian		Transparent	Rounded	28.8	82.8	0.34
2	German Shepherd		Brown	Rounded	270	490.68	0.55
3	Pomeranian		Transparent	Oval	32.4	80.68	0.40
4	Great Dane		Brown	Rounded	39.6	68.4	0.57
5	Golden Retriever		Transparent	Oval	10.8	19.8	0.54

6	Cross Labrador		Transparent	Oval	9	14.4	0.63
7	Dalmatian		Black	Oval	88.2	198	0.45
8	Boxer		Yellow	Rounded	54	86.4	0.62
9	Rottweiler		Black	Oval	88.2	192.6	0.46
10	Labrador Retriever		Black	Rounded	43.2	75.6	0.57
11	Doberman		Yellow	Elliptical	234	432	0.81
12	Saint Bernard		Transparent and Brown	Rounded	34.776	61.2	0.56
13	Lhasa		Gray	Rounded	9	27	0.33

Discussion:

Since hairs are readily lost during its cycle and can be readily transferred, it is the most common evidence transferred from culprit to victim at the time of crime (Locard, 1930). The telogen dog hairs are commonly found at crime scene. Though animal hairs are commonly encountered at crime scene, they are often overlooked as a source of forensic evidence. Western countries maintain pet animals like dogs and cats in close proximity as a result pet hairs are commonly found on clothing, cars, home of owners. As a result, the pet hairs can be used to link suspect to crime (Halverson and Basten, 2005).

The two hypervariable regions of mitochondria (HV1 and HV2) rather than nuclear DNA is used for individualization of dogs which are present between 15458 and 16727 (Okumura et al. 1996). Canine STR loci can be used in pedigree verification, criminal investigation and homicide trails. The medullary fraction can be effectively used to distinguish the dogs and cats (Peabody et al. 1983). Microscopic characteristics of hairs can also be used to discriminate animals most commonly encountered at crime scene (Hicks, 1977). The microscopic analysis and comparison of animal hairs are not as significant as human hairs for individualisation of dog. But it is potential useful to associate suspect's dog with crime scene (Oien, 2009). Domestic dogs exhibit extraordinary genetic variations across breeds and also have parallel with humans with respect to disease.

Microscopic hair examination, although, does not result in firm identification, it provides strong basis for association and certainly provides strong exculpatory evidence (Oien,). Peabody et al (1983) and Hicks (1977) used microscopic characters to distinguish dogs and cats and animal discrimination respectively. It must, however, be noted that the dog hairs do not possess enough individual microscopic characters to associated a questioned hair to a particular dog from other dogs of same breed (FBI Lab.).

Conclusion:

Dogs are commonly used in wildlife crimes as well as they are favourite pet animals. Association of dogs and man can be useful in linking suspect to crime scene. The hairs from various breeds of dog exhibited differences in scale pattern, scale count, colour and shape of transverse section and medullary index which can be used for identifying the dog breeds.

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