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Investigation of the morphologic and scanned electron microscopic properties of wild boar hairs in the Balıkesir region

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ABSTRACT

Objective: Determination of species from animal hair is an effective method in veterinary forensic investigations, research, endangered species and prevention of poaching. Since the hairs are resistant to deterioration, they can be stored as evidence for many years. In addition, pig hairs are often used in making brushes. When these brushes are used in the food industry, it raises questions about halal food. This study aimed to identify these hairs by examining the hair structure of wild pigs living in the Balıkesir region and revealing their characteristics.

Materials and Methods: The hairs of 3 wild boars obtained from the İvrindi (Balıkesir) region were used. After the hairs taken from different parts of the pigs were cleaned, stereomicroscopy and macroscopic examination were performed and routine procedures were applied for scanning electron microscopic imaging.

Result: In stereomicroscopy and macroscopic examination, it was determined that the length and thickness of the hairs in different regions varied significantly. In the study, the hairs were generally bifurcated from the upper 1/3 part. In the scanning electron microscopic images, the hardened cuticle patterns on the hair shaft, which have a scaly appearance, were detected, and their measurements were made. Scanning electron microscopic images determined that there were very small bifurcations from the hair shaft. However, it was thought that these hairs could not be used for species separation, since these parts would break off in the hairs used as brushes. Significant images could not be obtained in cross-sections.

Conclusion: It is thought that it will be used as a source for the identification of the hairs of wild boars in the Balıkesir region.

Keywords: *Hair, Pig, SEM, Stereo microscope*

INTRODUCTION

Hair serves many functions, including thermoregulation and protection. It maintains temperature in animals by retaining heat or preventing cold. The hair can also provide camouflage and act as sexual attraction (Breehl and Caban, 2022; Grubbs et al., 2022). The hair follicle also has a wide variety of functions, including thermoregulation, physical and immunological protection against external aggressions, sensory perception, social interactions (Welle and Wiener, 2016). There are many factors that affect the hair structure. Hormones, vitamins, glandular secretions, environment, genetics, nutrition and trauma can alter the normal state of hair growth. Thyroid hormones physiologically stimulate hair growth. Adrenocorticotropic hormone deficiency,

disease, injury, environmental factors and stress also have a negative effect on hair growth (Choudhary et al., 2012).

Analysis of animal hair can be used in veterinary forensic research and biology as an effective tool to deter the illegal trade, slaughter and poaching of animals, including endangered species. It can be taken as physical evidence, as the hairs show strong resistance to degradation and rot. Mammalian hairs can be easily collected, preserved and transported to the laboratory for species identification by microscopy (Nowak, 1998). The hair sample can be examined by direct observation of all hairs using a light microscope (Valente, 1983; Oli, 1993; Wallis, 1993; Taru and Backwell, 2013) where the scanning electron microscope provides the advantage of higher magnification (Andy and Tillman, 2006; Aris and George, 2008; Dehury et al, 2019).

Scanning electron microscopy can be used in wildlife forensics for species identification (Short, 1978). The surface pattern, cross-section, and medullary index provide information of the specimen that can be used as geographic region and species identification tool. The cuticle is a useful tool for distinguishing species between wild ungulates and for deer to separate juveniles from adults and winter from summer fur (Meyer et al., 2001).

Electron microscopic studies of mammalian hair have also been done with transmission electron microscopy (Muto et al., 1981; Slepecky et al., 1981; Weedon and Strutton, 1981; Maxwell et al., 1982) and scanning electron microscopy (Short, 1978; Riggott and Wyatt, 1980). Combinations of both methods have also been used (Hino et al., 1982; Raphael et al., 1982). Cross and longitudinal sections of hairs or sanded hairs were examined by scanning electron microscopic imaging (Hess et al., 1985).

Pigmentation and hair size are anatomical structures affected by different variables, such as seasonality and association with growth, so their diagnostic utility is limited. Pigmentation and hair size vary with age, season and body area. Also, under the influence of digestive enzymes, pigmentation can deteriorate, while hair size can be changed, for example, due to fragmentation. These are factors that make diagnosis difficult. Therefore, scanning electron microscopic examination is very important (Kennedy and Carbyn, 1981; Amerasinghe, 1983).

In a study, surface scale patterns, transverse and longitudinal sections and sanded feathers of animals belonging to the families Tayassuidae and Suidae were examined by scanning electron microscopy. When the cross-sectional faces of Tayassu and Catagonus hairs were examined, cortical layers of varying thickness, protrusions arranged according to certain intervals and heights, and spongy areas were encountered, showing slight differences between genera and species (Hess et al., 1985).

Hairs are used to make some brushes. When these brushes are used in the food industry, it raises questions about halal food.

This study was carried out to examine the hair structure of wild pigs living in the Balıkesir region, to reveal their characteristics and to facilitate their diagnosis.

MATERIALS and METHODS

The study was carried out using the hairs of 3 wild boars (Sus scrofa) in İvrindi region (Balıkesir) in the spring season. No other wild pig breeds were found in Balıkesir region. The hairs taken from different parts of the pigs (inguinal, caudal and lumbar regions) were cleaned with detergent and rinsed with distilled water, and stereomicroscopy and macroscopic examination were performed, routine procedures were applied for scanning electron microscopic imaging.

For scanning electron microscopy studies, the hair samples were cut into 5 mm pieces, 3 mm from the root, and placed on the sample holder (on the adhesive tape) and prepared by gold plating for 20 seconds at 5 millibar vacuum and 5 mA current. The samples were examined under a scanning electron microscope. The hair pattern inter-scale distance for each animal species was measured using the SEM software digital scale.

For this study, research permission was obtained from the Ministry of Agriculture and Forestry with the number E-21264211-288.04-9829863 (10.05.2023).

RESULTS

It is important to identify the animal species from the hairs. Macroscopic analysis of hairs represents only the first step in the process of identifying an unknown hair sample. Among wild mammals, macroscopic hair species identification can only be definitively defined in wild boar hairs without the

aid of microscopic analysis. The boar's hairs are easily recognizable to the naked eye by the general appearance of its hairs, which have been bifurcated at least once. In piglets, bifurcation does not appear.

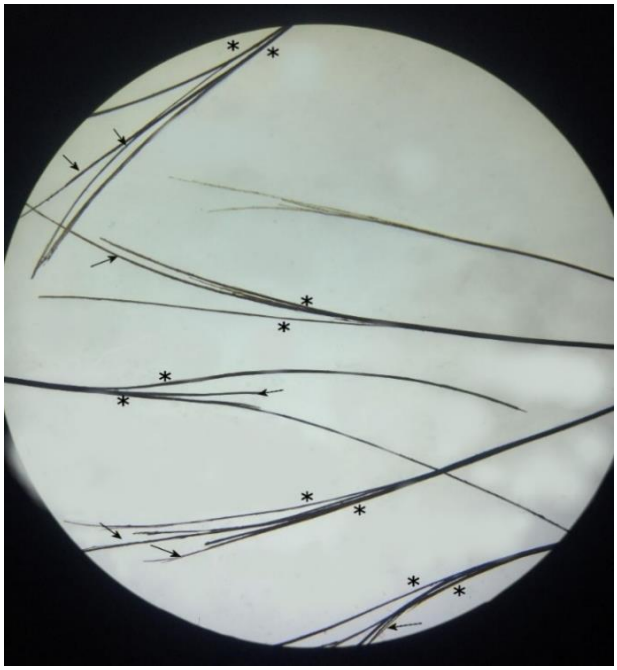

Figure 1. Stereo Microscope image, Asterisk: primary branches; arrow: secondary branches.

On a white background, bifurcations were clearly observed in the macroscopic examination of the hairs. These bifurcations could be from the same level as well as from different levels. In the study, it was observed that the hairs were generally bifurcated from the upper 1/3 part. This first bifurcation was called the primary bifurcatio, and the hair originating from this region was called the primary hair (Figure 1).

The number of primary bifurcations ranged from 2 to 9. This number was found to be less in fine hairs. Hairs that did not show bifurcation were also observed. It was observed that there were re-bifurcations from these bifurcated fragments in some large hairs. This is called secondary bifurcation, and the hair originating from this region is called secondary hair (Figure 1).

It was determined that the hairs separated from the primary bifurcatio were divided into two or more parts. It was observed that these bifurcations were directed towards the tip of the hair, but bifurcations directed in the opposite direction were also detected (Figure 2). It was observed that the hairs especially in the caudal and dorsal region were quite thick.

Table 1. Macroscopic features of the hair of wild boars.

Hair pattern position	Surface condition	Distance between consecutive patterns	Pattern	Appearance
Transverse	Smooth (Except secondary bifurcatios)	Narrow	regular wave	Bright

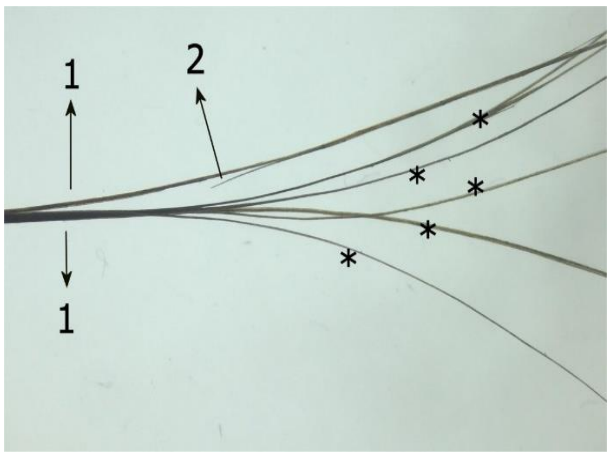

Figure 2. Stereomicroscopic hair image, 1- Primary branch, 2- A posteriorly directed secondary branch, Asterisk: secondary branch.

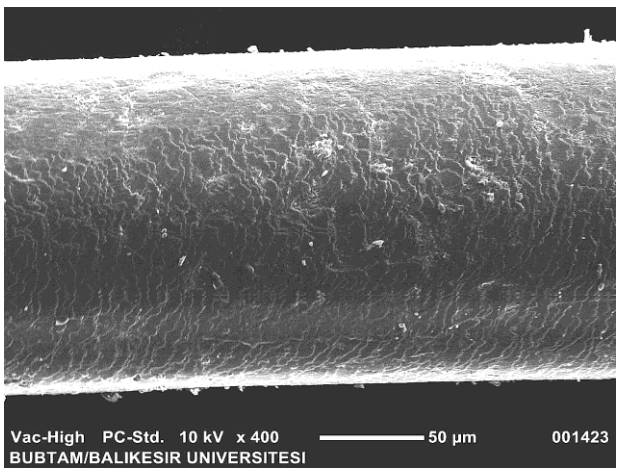

Figure 3. Hair surface, scanning electron microscopic image.

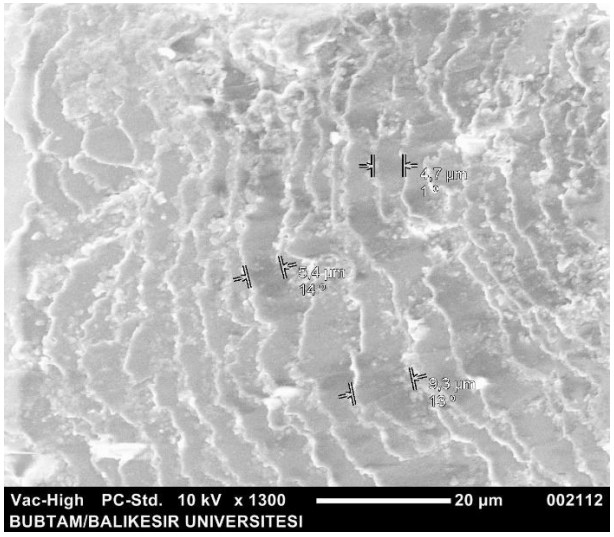

Figure 4. Hair surface, scanning electron microscopic image

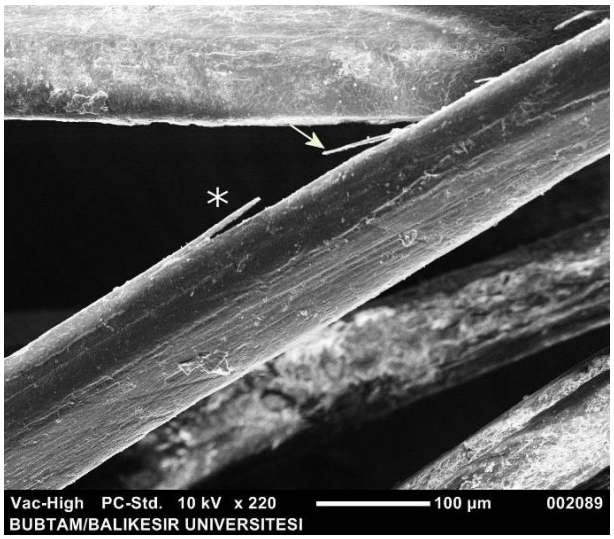

Figure 5. Asterisk: secondary hair extending towards the hair tip, Arrow: Secondary hair extending towards the hair root -in the opposite direction.

The pattern position, surface condition, and distance between successive patterns of the hairs taken in the study were examined. In the pattern position, the direction of the pattern was determined. In our study, it was determined that the pattern direction was transverse, and when the surface condition was examined, it was generally a smooth surface. However, considering the primary bifurcations separated from the hair body and the secondary bifurcations separated from them, these parts were not considered as smooth surfaces.

The distance between successive patterns was measured in electron microscopic images. This range is set very narrow. The pattern is determined as regular wavy. Overlapping, regular wave patterns were observed on the edges of the flakes on all surfaces examined. The general appearance was determined as bright (Table 1).

In stereomicroscopy and macroscopic examination, it was determined that the length and thickness of the hairs in different regions varied significantly. It was observed that the hairs especially in the caudal and lumbar regions were quite thick. No difference in pattern was observed.

In the scanning electron microscopic images, the hardened cuticle patterns in the hair shaft, which have a scaly appearance, were detected and their measurements were made (Figure 3-4.). It was also determined that there are very small bifurcations from the hair body (Figure 5). In the cross-sections, a gap was observed only in the body section.

DISCUSSION

Scaly patterns of plumage of domestic pig (Sus scrofa domesticus), European wild boar (Sus scrofa scrofa), and Warty pig (Phacochoerus aethiopicus) have been reported. It has been noted that it seems impractical to try to distinguish species and subspecies based solely on scale patterns (Wilford et al., 1985). With the pattern determined in our study, it was determined that the patterns of Domestic pig (Sus scrofa domesticus), European wild boar (Sus scrofa scrofa) and Warty pig (Phacochoerus aethiopicus) were basically the same.

The plumage of the three subspecies of Tayassu pecari has been reported to be relatively straight, short and flattened, with dark hair colors and frayed tips of most (Hess et al., 1985). In addition, the fact that the hair is the same color from one end to the other is consistent with the findings of our study. In this respect, the hairs in the study are similar to tayassu pecari. However, in our study, it was observed that the ends of most hairs were not frayed.

The feathers of the Tayassu tajacu are relatively long, not as straight as those of the Tayassu pecari, and color variations are noted from one end to the other. It has also been reported that the feathers exhibit characteristic fluctuations, some of which are not frayed at the ends, and are variable in length and diameter (Hess et al., 1985). In the presented study, it was determined that all hairs were relatively short, straight, unwavering and without color differences from one end to the other.

In a study comparing the summer plumage of C. Wagneri with the winter plumage, it was reported

that the winter plumage is larger and longer, but the morphological features are otherwise the same (Hess et al., 1985). Since the study examined the hairs in the first spring season, such a comparison could not be made.

According to Anna Maria De Marinis and Alessandro Asprea, fawns and wild cattle can be distinguished from adults by having an irregularly wavy cuticular pattern along the length of their feathers. It has been reported that all hair samples of young animals examined show this feature from birth to 3-4 months of age (Jedrzejewski et al., 1992). During the first molt, it has been observed that the offspring transform their natal fur to a fur similar to that of the adults (Ryder, 1960; Johnson and Hornby, 1980; Jedrzejewski et al., 1992). Since adult animals were used in our study, no comparison could be made.

Although it has been reported that holes made by ectoparasites are evident in the hair surface images (Hess et al., 1985), these holes caused by ectoparasites were not observed.

Adults and offspring of sheep have been reported to show wavy and dull hair (De Marinis and Alessandro, 2006). In our study, no wavy hair was found and the hairs were brightly colored. Also, no fragmented hairs like those found in carnivores were found.

Worn, bifurcated guard hairs characterize domestic and wild boars and crossbreeds (Marchinton et al., 1974; Hess et al., 1985; Mayer and Brisbin, 1991). Boar subspecies and other Suidae species typically have hairs with frayed ends (Koppiker and Sabnis, 1977; Amerasinghe, 1983; Hess et al., 1985). In addition, Tayassuidae species have bifurcated hairs at the tips (Hess et al., 1985). The degree of wear can be correlated with the chronology of hair regeneration. The significance of this character of hair morphology is unknown (Hess et al., 1985). In our study, no significant wear was observed on the hair tips, but bifurcations were detected.

It has been determined that the cross-sectional faces of the hairs of Tayassu and Catagonus species have cortical layers of different thicknesses, protrusions arranged according to certain intervals and heights, and spongy areas that show slight differences (Hess et al., 1985). In the study, however, such regular and intermittent images could not be obtained, it was determined that there was only a gap in the middle of the hair.

Although Hess et al. (1985) stated in their study that parasites or their effects were observed in the hair, ectoparasites and their effects were not observed in our study.

The location of the scales relative to the longitudinal axis (transverse- middle) of the hair, the structure of the free edges of the scales (straight - wavy), the distance between the scales edges (distant - near), and the scaled pattern (regular mosaic, regular and irregular wave, Ω-shaped) diagnosis of the hair (De Marinis and Alessandro, 2006). In our study, it was determined that the pattern direction of the hair is transverse, the distance between the hair flake edges is narrow, the structure of the flake free edges is regularly wavy as a scale model, and the flakes are overlapped.

CONCLUSION

In the study, the hairs of wild boars living in the Balıkesir region were examined. Their most important bifurcation, which is used to distinguish them from other hairs macroscopically, was determined in detail. Micro bifurcations were also detected in scanning electron microscopic images, which are not encountered in the literature. The posterior ones of the bifurcations that normally tend towards the tip of the hair were also observed.

REFERENCES

Amerasinghe FP. The structure and identification of the hairs of the mammals of Sri Lanka. Ceylon J Sci. 1983; 16:76-125.

Andy A, Tillman C. Surface scanning electron microscopy of suri alpaca fiber and other members of the camel family. Science. 2006; 311:85-171.

Aris FP, George C. Morphology of the hair in the Goat breed Capra prisca. J Anim Vet Adv. 2008; 7(9):1142-1145.

Breehl L, Caban O. Physiology, Puberty. Treasure Island: StatPearls StatPearls Publishing; (FL):2002.

Choudhary OP, Dhote BS, Bharti SK, Sathapathy S. The advantages of the scanning electron microscope in the forensic studies of hair in domesticated and wild animals. J Forensic Sci. 2012; 3(2): 191-192

De Marinis AM, Asprea A. Hair identification key of wild and domestic ungulates from southern Europe. Wildlife Biol. 2006; 12(3):305-320.

Dehury S, Mishra UK, Hembram A, Sahoo N. Light and scanning electron microscopy analysis of hair samples of some wild animals for individual identification. J Entomol Zool Stud. 2019; 7(6):900-904

Grubbs H, Nassereddin A, Morrison M. Embryology, Hair. Treasure Island: StatPearls StatPearls Publishing; 2022.

Hess WM, Flinders J, Pritchett CL, Allen JV. Characterization of hair morphology in families Tayassuidae and Suidae with scanning electron microscopy. J Mammal. 1985; 66(1):75-84.

Hino H, Ammitzboll T, Moller R, Asboe-Hansen G. Ultrastructure of skin and hair of an Egyptian mummy. J. Cutaneous Pathol. 1982; 9:25-82.

Jędrzejewski W, Jędrzejewska B, Okarma H, Ruprecht AL. Wolf predation and snow cover as mortality factors in the ungulate community of the Białowieża National Park, Poland. Oecologia. 1992; 90(1):27-36.

Johnson E, Hornby J. Age and seasonal coat changes in long haired and normal Fallow deer (Dama dama). J Zool.1980; 192(4):501-509.

Kennedy AJ, Carbyn LN. Identification of wolf prey using hair and feather remains: with special reference to Western Canadian National Parks. Canadian Wildlife Service, Western and Northern Region. 1981.

Koppiker BR, Sabnis JH. Further studies on the identification of hairs of some Indian mammals. JBNHS. 1977; 74:50-59.

Marchinton RL, Aiken RB, Henry VG. Split guard hairs in both domestic and European wild swine. J Wildl. 1974; 38(2): 361-362.

Maxwell DJ, Bannatyne BA, Fyffe REW, Brown AG. Ultrastructure of hair follicle afferent fibre terminations in the spinal cord of the cat. J Neurocytol. 1982; 11:571-582.

Mayer JJ, Brisbin IL. Wild Pigs of the United States. Their history, morphology and current status. Athens: University of Georgia Press; 1991. pp.313.

Meyer W, Pohlmeyer K, Schnapper A, Hülmann G. Subgroup differentiation in the Cervidae by hair cuticle analysis. Zeitschrift für Jagdwissenschaft. 2001; 47:253-258.

Muto H, Ozeki N, Yoshioka I. Fine structure of the fully keratinized hair cuticle in the head hair of the human. Acta Anat. 1981; 109:13-18.

Nowak B. Contents and relationship of elements in human hair for a non industrialized population in Poland**.** Sci Total Environ. 1998; 209(1):59-68.

Oli MK. A key for the identification of the hair of mammals of a snow leopard (Pantheraunica) habiat in Nepal. J Zool. 1993; 231(1):71-93.

Raphael KA, Chapman RE, Frith PA, Pennycuik PR. The structure of hair and follicles of mice carrying the naked (N) gene. Gertpt Res. Cambridge. 1982; 39:139-148.

Riggott JM, Wyatt EH. Scanning electron microscopy of hair from different regions of the body of the rat. J Anat. 1980; 139:121-12.

Ryder ML. A study of the coat of the Mouflon Ovis musimon with special reference to seasonal change. UK: Blackwell Publishing; 1960. p.387-408.

Short HL. Analysis of cuticular scales on hairs using the scanning electron microscope. J Mammal. 1978; 59(2):261-268.

Slepecky N, Hamernik R, Henderson D. The consistent occurrence of a striated organelle (Friedmann body) in the inner hair cells of the normal chinchilla. Acta Otolaryngol. 1981; 91:189-198.

Taru P, Backwell L. Identification of fossil hairs in Parahyaena brunnea coprolites from Middle Pleistocene deposits at Gladysvale cave, South Africa. J Archaeol Sci. 2013; 40(10):3674-3685.

Valente A. Hair structure of woolly mammoth mammuthus primigenius and the modern elephant, elephas maximus and loxodont. J Afr Zool. 1983; 199(2):271-274.

Wallis RL. A key for the identification of some Ontario mammals. Can J Zool. 1993; 71(3):587-591.

Weedon D, Strutton G. Apoptosis as the mechanism of the involution of hair follicles in catagen transformation. Acta Dermato (Stockholm). 1981; 61:335-369.

Welle MM, Wiener DJ. The hair follicle: a comparative review of canine hair follicle anatomy and physiology. Toxicol Pathol. 2016; *44*(4):564-574.

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