

Histological Characteristics of Vanaraja, Aseel and Kadaknath Meat

V. P. Singh*, V. Pathak¹, Neeraj K. Gangwar² and S. K. Gupta³

Department of Livestock Products Technology, College of Veterinary and Animal Sciences, Sardar Vallabhbhaipatel University of Agriculture and Technology, Meerut, 250110, UP

¹Department of Livestock Products Technology, ²Department of Veterinary Pathology, ³Department of Veterinary Anatomy, College of Veterinary Science and Animal Husbandry, U.P. Pt. Deen Dayal Upadhyaya Veterinary University and Go Anusandhan Sansthan, Mathura, 281001, UP

ABSTRACT

Present study was conducted to assess the histological characteristics of Vanaraja, Aseel and Kadaknath meat. The muscle fibre thickness of breast and thigh muscles were separately assessed and found highest in Vanaraja and lowest in Kadaknath meat. Muscle bundle thickness was also found lowest in Kadaknath meat while no definite pattern was indicated in Vanaraja and Aseel. It was evident from the histological characteristics that the breast and thigh muscles showing highest muscle fibre thickness were lowest in perimysium diameter and vice versa. The muscle fibre and endomysium thickness both were lowest in Kadanath. The collagen, reticular and elastic fibres indicated the toughness in muscles and found highest in Vanaraja breed. On comparison of breast and thigh muscles, thigh muscles showed comparatively more number of collagen, reticular and elastic fibres.

Keywords: Muscles fibres, Muscle bundles, Collagen fibres, Reticular fibres, Elastic fibres

Received: 17/05/2020

Accepted : 14/07/2020

INTRODUCTION

In spite of broiler -in India, the meat of native chicken breeds is in high demand. It is due to inherent meat characteristics like taste and flavor. But, these native breeds are localized *i.e.* Kadaknath is localized in Jhabua and Dhar districts of western Madhya Pradesh, Aseel in state of Andhra Pradesh and in some areas of Rajasthan and Madhya Pradesh and Vanaraja in northeast rural/tribal areas. Aseel breed is characterized by its hardiness and ability to thrive under adverse climatic conditions, and its meat is considered to have a delicious flavor (Panda and Mahapatra, 1989). Mohan *et al.* (2008) reported that the meat of Kadaknath breed contains high protein (25.47%) and is believed to have aphrodisiac properties. Vanaraja is a dual purpose breed and has been developed by crossing random bred control population as the female line and Red Cornish population as the male line by Project Directorate on Poultry, Hyderabad (Chandra *et al.*, 2004).

Keeping in view the potential of these native breeds and to find out the possibilities to improve the quality of meat, a study was undertaken to assess the quality characteristics of these breeds at the age (42 days broilers commonly utilized for meat production. It is imperative to know that fresh meat quality is affected by various intrinsic and extrinsic factors.

Meat quality traits particularly texture are dependent on muscle structure and characteristics. Therefore, attempts were made to find out the histological differences in meat quality among these indigenous breeds and the quality parameters like muscle fibre and bundle diameters, contents of epimysium and perimysium as well as collagen, reticulin and elastin were measured.

MATERIALS AND METHODS

Source of materials: Ten birds each of Vanaraja, Aseel, and Kadaknath aged six weeks (42 days) were procured and slaughtered in Poultry Farm of the University. All the chemicals and glass wares used in the study were of standard make and quality.

Methodology: Samples were taken from the middle parts of left and right *Quadriceps femoris* muscle (leg muscles), and *Pectoralis superficialis* muscles (breast muscles). The samples were immediately fixed in 10% buffered neutral formalin solution for 24-48 hours, dehydrated and embedded in paraffin. Tissue samples (5 mm) were stained by hematoxylin eosin for general histological examination and muscle fibre thickness were measured as per the method outlined by Jeremiah and Martin (1982). Specific standard staining *i.e.* Van Gieson's staining for collagen fibers; Weigert's staining for

*Corresponding author E-mail address: drvplpt@gmail.com

elastic fibers and Gomori's for reticular fibres were done. Sections were observed under light microscope under 100x.

Statistical analysis: The data obtained (n=6) were subjected to statistical analysis as per method prescribed by Snedecor and Cochran (1994) for Analysis of variance and Duncan test using SPSS statistics 16 software.

RESULTS AND DISCUSSION

Muscle fiber diameter

Breast muscle fiber: At the age of 42 days breast muscles of all three chicken breeds i.e. Vanraja, Aseel and Kadaknath showed the muscle fiber thickness in the range of 9.02 ± 0.06 to $9.49 \pm 0.15 \mu\text{m}$. The highest breast muscle fibre thickness was observed in Vanraja while lowest in Aseel breed. The muscle fibre thickness of breast muscles of Vanraja and Kadaknath was significantly ($P < 0.05$) higher than Aseel.

Thigh muscle fibers: Thigh muscles fibers of same age group of Vanraja, Aseel and Kadaknath showed the fiber thickness in the range of 10.11 ± 0.21 to $13.46 \pm 0.26 \mu\text{m}$. The highest thigh muscle fiber thickness was observed in Vanraja while lowest in Kadaknath. The muscle fibre thickness of thigh muscles of Vanraja, Aseel and Kadaknath were significantly ($P < 0.05$) different among each other. So on that basis it can be interpreted that Vanraja thigh meat has comparatively less tender meat than other chicken breeds as also depicted in the texture analysis of the product (Singh *et al.*, 2017).

Breast and thigh muscle fiber: On comparison of breast and thigh muscles of respective chicken breeds, Vanraja and Aseel thigh muscle fibers indicated significantly ($P < 0.05$) higher values than breast muscle fibers. However, no such differences were observed in Kadaknath breast and thigh muscle fibers. The difference could be due to breed specificity and growth characteristics.

Muscle bundle diameter

Breast muscle bundle: Breast muscles of all three native chicken breeds i.e. Vanraja, Aseel and Kadaknath showed the muscle bundle thickness in the range of 61.43 ± 0.25 to $77.60 \pm 0.27 \mu\text{m}$. The highest breast muscle bundle thickness was observed in Aseel while lowest in Kadaknath. The breast muscle bundle thickness of all three breeds differed significantly ($P < 0.05$) among each other. It could be indicative of differences in breed characteristics and growth pattern of muscles bundles.

Thigh muscle bundles: Thigh muscles bundles of same age group of Vanraja, Aseel and Kadaknath showed the fibre thickness in the range of 81.13 ± 0.17 to $87.31 \pm 0.30 \mu\text{m}$. The highest thigh muscle bundle thickness was observed in Vanraja while lowest in Kadaknath. The muscle fibre thickness of indicated that thigh muscles of Vanraja, Aseel and Kadaknath were none significantly ($P > 0.05$) different among each other. On that basis it can be interpreted that Vanraja thigh meat has comparatively less tender meat than other poultry breeds (Singh *et al.*, 2017).

Breast and thigh muscle bundles: On comparison of breast and thigh muscles of respective chicken breeds, Vanraja and Aseel thigh muscle bundles indicated significantly ($P < 0.05$) higher values than breast muscle fibres. However, no such differences were observed in Kadaknath breast and thigh muscle bundles.

The variation in muscle fibre thickness could be the indication of the breed variation and growth characteristics of a particular breed as suggested by Candek-Potokar *et al.* (1999) during histological study of pork meat quality. Botka-Petrak *et al.* (2011) reported variation in muscle structure in mechanically deboned meat from various species while Teusan *et al.* (2009) studied variation in muscle fibres of Cobb-500 on advancement of age.

Table 1: Muscle fiber and bundle thickness in breast and thigh meat of different chicken breeds (mean \pm SE)

Breed	Muscle fiber thickness (μm)		Muscle bundle thickness (μm)	
	Breast	Thigh	Breast	Thigh
Vanraja	$9.49^{\text{aB}} \pm 0.15$	$13.46^{\text{aA}} \pm 0.26$	$63.58^{\text{bB}} \pm 0.23$	$87.31^{\text{A}} \pm 0.30$
Aseel	$9.02^{\text{cB}} \pm 0.06$	$11.30^{\text{bA}} \pm 0.19$	$77.60^{\text{aB}} \pm 0.27$	$85.07^{\text{A}} \pm 0.29$
Kadaknath	$9.25^{\text{ab}} \pm 0.12$	$10.11^{\text{c}} \pm 0.21$	$61.43^{\text{c}} \pm 0.25$	81.13 ± 0.17

Means bearing different superscripts (a, b, c, d) within column and (A, B, C, D) within row differ significantly ($P < 0.05$)

Muscle perimysium thickness

Breast muscle perimysium: Breast muscles of all three breeds i.e. Vanraja, Aseel and Kadaknath showed the muscle perimysium thickness in the range of 26.41 ± 0.34 to 35.22 ± 0.32 μm . The thickest breast muscle perimysium was observed in Aseel while thinnest in Vanraja. The muscle perimysium thickness of the broiler aged native poultry breeds indicated that breast muscles of Aseel and Kadaknath were significantly ($P < 0.05$) thicker than Vanraja (Singh and Pathak, 2017).

Thigh muscle perimysium: Thigh muscles perimysium thickness of same age group of Vanraja, Aseel and Kadaknath were in the range of 13.37 ± 0.16 to 17.28 ± 0.18 μm . The highest thigh muscle perimysium thickness was observed in kadaknath while lowest in Vanraja. The muscle perimysium thickness of the broiler aged native chicken breeds indicated that thigh muscles of Vanraja and Aseel were significantly ($P < 0.05$) lower than Kadanath. On that basis it could be interpreted that Kadaknath thigh meat was comparatively less tender than other chicken breeds.

Breast and thigh muscle perimysium: On comparison of breast and thigh muscles of respective chicken breed, breast muscle perimysium thickness in all breeds indicated significantly ($P < 0.05$) higher values than thigh muscles. The difference could be due to breed specificity and growth characteristics of breast and thigh muscle perimysium at this particular age. A negative correlation was observed in muscle fibre diameter and permysium thickness in same breed of the chicken.

Muscle endomysium thickness

Breast muscle endomysium: Endomysium thickness in breast muscles of all three breeds i.e. Vanraja, Aseel and Kadaknath of chicken were in the range of 3.70 ± 0.16 to 4.42 ± 0.14 μm . The highest breast muscle endomysium thickness was observed in Aseel while lowest in Kadaknath. The muscle endomysium thickness of the broiler aged native chicken breeds indicated that breast muscles of Aseel and Kadaknath were significantly ($P < 0.05$) different among each other. However, Vanraja breast muscle endomysium indicated values intermediary to Aseel and Kadaknath.

Thigh muscle endomysium: The endomysium thickness of thigh muscle of same age group of Vanraja, Aseel and Kadaknath were in the range of 3.92 ± 0.10 to 4.38 ± 0.19 μm . The level of significance and pattern of endomysium was almost similar as indicated in the breast muscle endomysium.

Breast and thigh muscle endomysium: On comparison of breast and thigh muscles of respective chicken breeds, Vanraja and Aseel breeds thigh muscle endomysium thickness indicated no significant ($P > 0.05$) differences among each other. Based on results and literature available it can be assumed that higher muscle perimysium thickness could be related to the higher shear force value or lower tenderness as suggested by Liu et al. (1996) in raw chicken muscles and Fang et al. (1999) in *Semitendinosus* (ST) muscles on Picro-Sirius staining. These findings were also supported by Lachowicz et al. (2004) and reported positive correlation between the width of perimysial and endomysial space and instrumental hardness of muscles from wild boars and domestic pigs.

Table 2: Perimysium and Endomysium thickness of breast and thigh meat of different chicken breeds (Mean \pm SE)

Breed/strain	Perimysium thickness (μm)		Endomysium thickness (μm)	
	Breast	Thigh	Breast	Thigh
Vanraja	$26.41^{bA} \pm 0.34$	$13.37^{bB} \pm 0.16$	$4.07^{ab} \pm 0.09$	$4.22^{ab} \pm 0.07$
Aseel	$35.22^{aA} \pm 0.32$	$13.66^{bB} \pm 0.31$	$4.42^a \pm 0.14$	$4.38^a \pm 0.19$
Kadaknath	$34.73^{aA} \pm 0.26$	$17.28^{aB} \pm 0.18$	$3.70^b \pm 0.16$	$3.92^b \pm 0.10$

Means bearing different superscripts (a, b, c, d) within column and (A, B, C, D) within row differ significantly ($P < 0.05$)

Collagen fibers

In Vanraja breast muscle bundles, collagen was present in bunches of wavy threads in perimysium and only wavy threads in thigh muscle bundles. But it was located as threaded structures in endomysium of both breast and thigh muscle. The collagen content in Aseel breast and thigh muscle bundles were in form of thread like fibrils in perimysium and very few collagen threads in endomysium. The quantity of collagen in total was perceived lesser than Vanraja both in breast and thigh muscle bundles. The breast muscle bundles of Kadaknath were showing the collagen in the form of very few threads like fibrils in perimysium and in traces in endomysium but quantity seemed lesser than all other breeds. However, thigh muscle bundles of Kadaknath were showing thread like collagen structures in perimysium but quantity was greater than breast muscle but lesser than Vanraja thigh muscles bundles. In Vanraja and Kadaknath breeds thigh muscles were having more collagen threads than breast muscles. So it could be perceived that due to the presence of higher collagen in thigh they were less tender than breast muscles. The reason might be the tendency of more collagen deposition to provide the strength to thigh than breast. The variation in age and its collagen towards higher side with advancement of age was reported by Coro *et al.* (2000) in *Pectoralis superficialis* muscles of poultry and Nakamura *et al.* (2004) in broiler chickens.

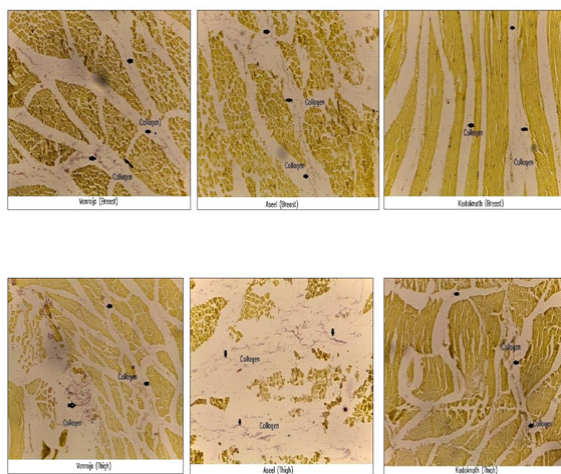


Fig. 1 Collagen content in breast and thigh meat of different chicken breeds

Elastin fibres

The quantity of elastin fibres in thigh muscle bundles were more than in the breast muscle bundles. They were surrounding the blood vessels and in form of small fibrils in perimysium, smaller thread like structures in endomysium. In Aseel, elastin fibres were in form of thin thread like structures

in the perimysium and in form of fine thread like structure in endomysium. Elastic fibres in Kadaknath were in the form of small fibrils and around the blood vessels in perimysium both in breast and thigh muscles. Quantity wise, the elastin fibres in Kadaknath were perceived lesser than Vanraja but greater than Aseel muscles.

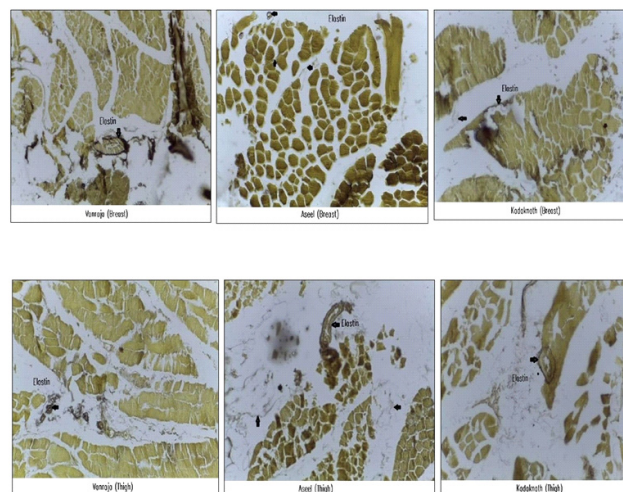


Fig. 2 Elastic fiber content in breast and thigh meat of different chicken breeds

Reticular fibres

In Vanraja breed of chicken breast muscle bundles showed long thread like chains of reticular fibres in perimysium and as very small threads in the endomysium. In thigh muscle bundles of Vanraja reticular fibres appeared as coiling threads and bunch in the perimysium and like small threads in the endomysium. The total quantity of reticular fibres in the thigh muscles bundles of Vanraja was more than the breast muscle bundles. The reticular fibres in Aseel breast muscle was seen as chain like threads in the perimysium and small thread like structure in the endomysium. The contents of reticular fibres in breast muscle of Aseel were lesser as compared to Vanraja. The thigh muscle bundles of Aseel appeared as thread like structures in the perimysium and as small fine threads in the endomysium. The total quantity of reticular fibres in thigh muscle bundles of Aseel was more than breast muscle. In Kadakanth breed of chicken breast and thigh muscle bundles showed small coiled thread and thread like reticular fibres respectively in the perimysium whiles they were small threads like in the endomysium. Quantity wise reticular fibres in Kadaknath breast muscle were greater than Vanraja and Aseel breast muscle bundles. The reticular fibres of Kadaknath breast were greater in quantity than in its thigh muscle bundles. The higher elastic and reticular fibers in thigh muscles might be the reason of toughness in muscles as suggested by Voutila (2009) and Mobini (2015) in various species.

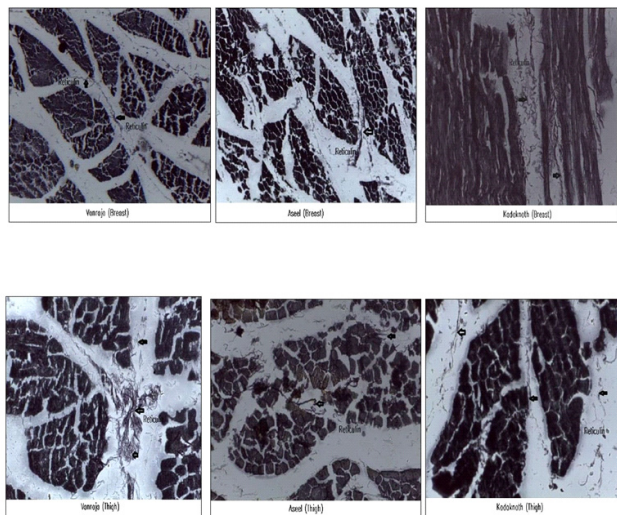


Fig. 3 Reticular fibres in breast and thigh meat of different chicken breeds

CONCLUSION

The muscle fibre diameters of breast and thigh muscles were highest in Vanraja and lowest in Kadaknath meat. Muscle bundle diameter was also found lowest in Kadaknath meat while highest in Aseel breast and thigh both. It was evident from the histological characteristics that the breast and thigh muscles showing highest muscle fibre diameters were lowest in permysiumthickness and vice versa. However, no such pattern was evident in endomysium. The muscle fibre and endomysium diameters both were lowest in Kadanath. The collagen, reticular and elastin fibres were highest in Vanraja breed than others. So it can be concluded that Vanraja meat could be tough than Assel and Kadaknath.

CONFLICT OF INTEREST STATEMENT: Authors declare no conflict of interest in preparing this manuscript.

ETHICS STATEMENT: The approval for slaughtering of experimental birds was obtained from research committee of the University.

REFERENCES

- Botka-Petrak K, Luciae H, Gottstein •, Martina D, Gomerèiae J, Savica J and Anetrak K (2011). Histological and chemical characteristics of mechanically deboned meat of broiler chickens. *Vet. Archiv.* 81(2): 273-283.
- Candek-Potokar M, Lefaucheur L, Zlender B and Bonneau M (1999). Effect of slaughter weight and/or age on histological characteristics of pig *Longissimus dorsi* muscle as related to meat quality. *Meat Sci.* 52: 195-203.
- Chandra CVS, Rao S, Rao GN, Sharma RP, Reddy BLN, Gupta BR and Satyanarayana S (2004). Genetic study on juvenile traits of Vanaraja chickens. *Ind. J. Anim. Sci.* 74:1229-1231.
- Coro FA, Youssef EY and Shimokomaki M (2000). Age related changes in breast poultry meat collagen crosslink, hydroxyllysylpyridinium. *Proceedings of 46th International Congress of Meat Science and Technology*, Buenos Aires, Argentina, pp. 432-433.
- Fang SH, Nishimura T and Takahashi K (1999). Relationship between development of intramuscular connective tissue and toughness of pork during growth of pigs. *J. Anim. Sci.* 77(1): 120-130.
- Jeremiah LE, Martin AH (1982). Effect of prerigor chilling and freezing and subcutaneous fat cover upon the histological and shear properties of bovine *Longissimus dorsi* muscle. *J. Anim. Sci.* 62: 353-361.
- Lachowicz K, Zochowska J and Sobczak M (2004). Comparison of the texture and structure of selected muscles of piglets and wild boar juveniles. *Polish J. Food Nutr. Sci.* 13/54(1):75-79.
- Liu A, Nishimura T and Takahashi K (1996). Relationship between structural properties of intramuscular connective tissue and toughness of various chicken skeletal muscles. *Meat Sci.* 43(1): 43-49.
- Mobini B (2013). Histological differences in intramuscular connective tissues composition between dark and light colored muscles in broiler chickens. *Global Veterin.* 10(3): 360-364.
- Mohan J, Sastry KVH, Moudgal RP and Tyagi JS (2008). Performance profile of Kadaknath desi hens under normal rearing system. *Int. J. Poultr. Sci.* 43: 379-381.
- Nakamura YN, Iwamoto H, Shiba N, Miyachi H, Tabata S and Nishimura S (2004). Growth changes of the collagen content and architecture in the *pectoralis* and *iliotibialis lateralis* muscles of cockerels. *Brit. Poultr. Sci.* 45(6): 753-761.
- Panda B, Mahapatra SC (1989). Common breeds of poultry. In: ICAR (Eds), *Poultry Production*, pp. 6-18 (New Delhi, India, ICAR, Publication).
- Singh VP, Pathak V (2017). Physico-Chemical, colour and textural characteristics of Cobb-400, Vanajara, Aseel and Kadaknath meat. *Int. J. Livest. Res.* 7(11): 98-106.
- Snedecor GW, Cochran WG (1994). *Statistical Methods*. 8th ed. Affiliated East-West Press, New Delhi, India, and Iowa State University Press, Ames.
- Teusan V, Radu-Rusu RM and AncaTeusan (2009). Investigations on the histological structure of the superficial pectoral muscle in cobb-500 commercial meat-type hybrid hen. *Cercetări Agronomice În Moldova*, XLII (4) (140): 76-83.
- Voutila L (2009). Properties of intramuscular connective tissue in pork and poultry with reference to weakening of structure. *Ph.D. Thesis*, Faculty of Agriculture and Forestry of the University of Helsinki.