

GROSS ANATOMICAL, HISTOLOGICAL AND ECHOCARDIOGRAPHIC STUDIES ON AORTA OF BUFFALO

HIMANSHU BHASIN, ANURADHA GUPTA*, JITENDER MOHINDROO¹, NEELAM BANSAL
and VARINDER UPPAL

Department of Veterinary Anatomy, ¹Department of Veterinary Surgery and Radiology
Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana-141 004, India

Received: 19.04.2019; Accepted: 15.06.2019

ABSTRACT

The present study was conducted on twenty four adult buffalo heart (12 male and 12 female) collected from slaughter house to study the anatomical structure of blood vessels by comparing biometric, ultrasonographic and micrometric parameters. The biometrical studies revealed that the average diameter of ascending aorta was 1.96 ± 0.10 cm in males and 2.03 ± 0.10 cm in females. The average diameter of descending aorta was 2.62 ± 0.06 cm and 2.44 ± 0.09 cm in males and females, respectively. Histologically, aorta was made up of three layers i.e. tunica intima, tunica media and tunica adventitia which were analogous to endocardium, myocardium and epicardium of heart. The micrometrical data revealed that thickness of wall of ascending aorta was 0.56 ± 0.03 cm and 0.62 ± 0.02 cm in males and females, respectively whereas the thickness of descending aorta was 0.65 ± 0.04 cm and 0.76 ± 0.02 cm in males and females, respectively. However, the echocardiographic evaluation showed that the mean thickness of wall of ascending aorta was 0.65 ± 0.03 cm in males and 0.78 ± 0.07 cm in females. The mean thickness of wall of descending aorta was 0.84 ± 0.04 cm in males and 0.89 ± 0.06 cm in females. It was found that the diameter of descending aorta was more than ascending aorta. It was concluded that the diameter of ascending aorta was more in female buffalo while the diameter of descending aorta was more in male buffalo.

Keywords: Aorta, Echocardiography, Gross biometry, Micrometry

Ultrasonography is a good choice for imaging and describing the majority of heart diseases. It is non-invasive, straight forward method for assessment of bovine heart. Furthermore, cardiac ultrasound examination can be performed easily in field conditions with high sensitivity and specificity (Braun, 2009).

Two-Dimensional (2D) and M-mode echocardiography has been used to evaluate the normal as well as pathological heart by various authors (Muzzi *et al.*, 2006). However, in any species, for echocardiography to distinguish between normal subjects and those suffering from cardiac disease, it is essential to establish reliable reference values for the species. The methods and normal values for echocardiography in normal dairy cattle (Hallowell *et al.*, 2007) and dromedary camel (Tharwat *et al.*, 2012) were reported but the normal echocardiographic values of heart in male and female buffaloes have not been reported till now.

Gross anatomical studies on aorta have been reported in buffalo (Panhwar *et al.*, 2007) and ox (Mohan and Prakash, 1997) but detailed comparative gross anatomical, micrometrical and echocardiographic studies on aorta of buffalo have not been reported till now in healthy buffalo which can form the baseline for diagnosing various heart diseases.

MATERIALS AND METHODS

The gross morphological, biometrical, micrometrical and echocardiographic studies were conducted on adult buffalo aorta (n=24) collected from slaughter house. The

echocardiographic studies were also done on adult male and female buffaloes presented to the teaching Veterinary hospital for elective surgeries like castration, supra-numerary teats, medial patellar desmotomy and post fracture repair follow up etc. at GADVASU, Ludhiana.

The gross anatomical studies were conducted on aorta in adult male (12) and female (12) buffaloes. Immediately after slaughtering, the heart was collected by opening the thoracic cavity and external and internal structures of all the four chambers of heart of adult male and female buffaloes were studied. After gross anatomical studies aorta were subjected to biometrical analysis.

Echocardiographic examination of aorta was done on standing animals from left side (third to fifth intercostal space) using 3.5-MHz convex transducers. In preparation for ultrasonography, hair were clipped and shaved on both sides from the 3rd to 8th intercostal space. The area was scrubbed with alcohol to remove excess oil and coupling gel was applied. In the cardiac area, the heart was thoroughly imaged. Echocardiography of heart sample was performed *in vitro* collected from slaughter house and Veterinary Clinics, GADVASU, Ludhiana. The hearts were collected immediately after slaughtering and their ultrasound were taken after formalin fixation. For ultrasonography, after removing blood clots from atria and ventricles, the heart were imaged under water immersion at room temperature using B mode Real Time ultrasound scanner fitted with a 7.0 MHz sector transducer from a distance of 1.5-2 cm.

The tissue samples were fixed in 10% neutral

*Corresponding author: anugadvasu@gmail.com

Table1
Biometrical data on aorta of male and female buffalo

S. No.	AA (cm) male		AA mean (cm) male	DA (cm) male		DA mean (cm) male	AA (cm) Female	AA mean (cm) Female	DA (cm) Female	DA mean (cm) Female
1	1.2	1.2	1.2	2.3	2.1	2.2	2.6	2.4	2.5	2.4
2	2.5	1.7	2.1	3.0	2.8	2.9	2.3	2.1	2.2	3.5
3	1.8	1.6	1.7	2.4	2.1	2.2	2.2	1.6	1.9	3.0
4	2.2	2.2	2.2	2.5	3.0	2.7	3.0	2.8	2.9	2.2
5	1.6	2.0	1.8	3.1	2.7	2.9	2.0	1.6	1.8	2.3
6	2.2	2.0	2.1	2.5	2.9	2.7	1.8	2.2	2.0	2.7
7	1.8	2.1	1.9	2.7	2.6	2.6	2.1	2.0	2.0	2.1
8	2.2	2.5	2.4	2.6	2.4	2.5	2.1	1.8	1.9	2.2
9	1.4	1.6	1.5	2.3	2.5	2.4	2.1	2.1	2.1	2.8
10	2.1	2.2	2.1	2.8	2.7	2.7	1.7	1.7	1.7	2.1
11	2.2	2.4	2.3	2.6	2.8	2.7	1.8	1.8	1.8	2.2
12	2.2	2.3	2.2	2.8	2.9	2.8	1.5	1.6	1.5	1.8
Mean			1.9			2.6				2.0
Standard deviation			0.3			0.2				0.36
Standard error			0.1			0.06				0.10

buffered formalin (NBF) and Bouin's fixatives immediately after collection. Once the fixation was achieved, the tissues were processed for paraffin block preparation by acetone-benzene schedule (Luna, 1968). The blocks were prepared and sections of 5-7 μ m thickness were cut and obtained on clean glass slides with rotary microtome. The paraffin sections were stained with haematoxylin and eosin stains to study the histomorphology and micrometry.

Micrometrical observations were recorded on haematoxylin and eosin stained sections with help of image analysis system loaded in digital microscope micrometer. The thickness of left and right atrial wall, thickness of wall of aorta and pulmonary artery were measured.

All the parameters were statistically evaluated for correlation to heart weight using Microsoft Excel version 2007 and SPSS software having inbuilt function for statistical analysis. Arithmetic mean, range, standard error and coefficient of variation for morphometric measurements were computed and statistically analysed for their significance (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

Aorta arose from the base of left ventricle and its opening was guarded by three semilunar cusps (anterior, right posterior and left posterior). From the anterior and left posterior, sinuses of valvular coronary arteries arose (Fig. 1). These findings were in close proximity with the observations of Anuradha *et al.* (2013) in human and mammalian species. Anuradha *et al.* (2013) found that at the union of ascending aorta with the aortic arch, the caliber of vessel was increased and termed as the bulb of aorta.

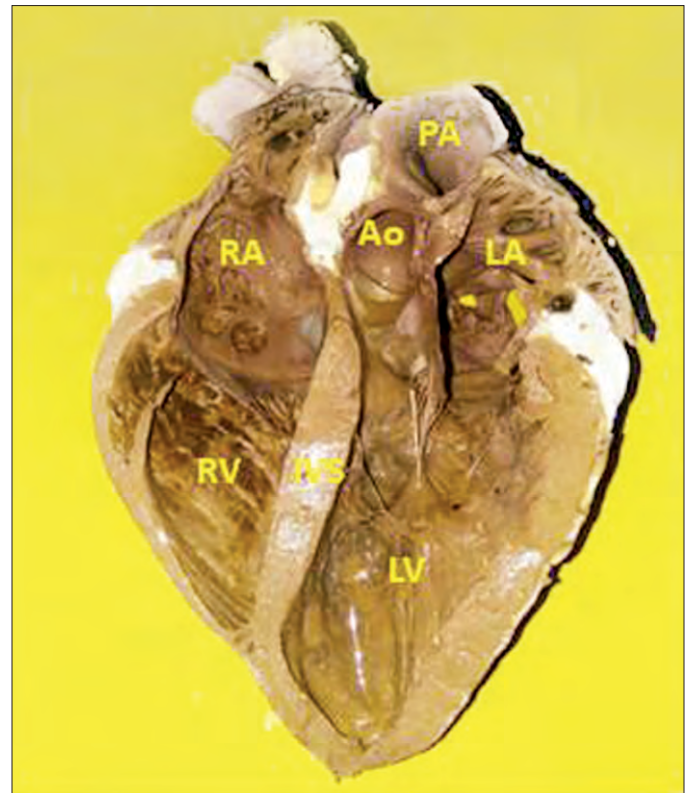


Fig. 1. Longitudinal section of heart showing pulmonary artery (PA), aorta (Ao), left atrium (LA), right atrium (RA), left ventricle (LV), right ventricle (RV) and interventricular septum (IVS)

The diameter of ascending aorta varied from 1.2 cm to 2.35 cm with the mean of 1.96 ± 0.10 cm in males (Table 1) whereas same parameters varied from 1.55 cm to 2.9 cm with the mean of 2.03 ± 0.10 cm in females (Table 1). The diameter of descending aorta varied from 2.2 cm to 2.9 cm with the mean of 2.62 ± 0.06 cm in males (Table 1) whereas same parameters varied from 1.8 cm to 3.05 cm with the mean of 2.44 ± 0.09 cm in females. It was concluded that

descending aorta had bigger diameter. It was further observed that the diameter of ascending aorta was more in female buffalo while the diameter of descending aorta was more in male buffalo than ascending aorta.

The thickness of wall of ascending aorta varied from 0.56 cm to 0.94 cm with the mean of 0.65 ± 0.03 cm in males whereas same parameters varied from 0.39 cm to 1.33 cm with the mean of 0.78 ± 0.07 cm in females (Table 2 & Fig. 2). The thickness of wall of descending aorta varied from 0.64 cm to 1.19 cm with the mean of 0.84 ± 0.04 cm in males whereas same parameters varied from 0.67 cm to 1.49 cm with the mean of 0.89 ± 0.06 cm in females (Fig. 3). Yamaga and Too (1986) measured aortic root dimensions as 70 mm in normal cows. Hallowell *et al.* (2007) measured the aortic diameter in diastole as 5.0 ± 0.26 cm in Jersey cows and 6.4 ± 0.62 cm in Holstein-Friesian cows. Tharwat *et al.* (2012) found the aortic diameter in diastole as 7.0 cm in adult camels. Torad *et al.* (2016) found the aortic diameter at the level of valve leaflets as 3.11 ± 0.10 cm in small buffaloes and 3.96 ± 0.31 cm in large buffaloes while the aortic diameter at the level of sinus of valsalva was 3.37 ± 0.19 cm in small buffaloes and 4.15 ± 0.29 cm in large buffaloes. He also observed the aortic diameter at the level of sino-tubular junction as 2.94 ± 0.07 cm in small buffaloes and 3.72 ± 0.31 cm in large buffaloes. It was concluded that thickness of wall of ascending and descending aorta was more in female buffalo than male buffalo.

The thickness of ascending aorta wall was 0.56 ± 0.03 cm and 0.62 ± 0.02 cm in males (Table 3) and females, respectively whereas the thickness of descending aorta was 0.65 ± 0.04 cm and 0.76 ± 0.02 cm in males (Table 3)



Fig.2. B-mode echocardiographic images showing the thickness of ascending aorta in (a) males and (b) females buffalo

and females, respectively (Table 3). The tunica intima was lined by a simple squamous epithelium whose nuclei bulge into the lumen of the vessel. The internal elastic lamina

Table 2
Echocardiographic data on aorta of male and female buffalo

S. No.	AA thickness (cm) Male			DA thickness (cm) Male			AA thickness (cm) Female			DA thickness (cm) Female		
	AA mean (cm) Male	AA mean (cm) Male	AA mean (cm) Male	DA mean (cm) Male	DA mean (cm) Male	DA mean (cm) Male	AA mean (cm) Female	AA mean (cm) Female	AA mean (cm) Female	DA mean (cm) Female	DA mean (cm) Female	DA mean (cm) Female
1	0.51	0.51	0.51	0.64	0.64	0.64	0.63	1.08	0.74	0.91	0.83	0.85
2	0.54	0.54	0.54	0.71	0.71	0.71	0.50	0.80	0.66	0.73	0.66	0.69
3	0.54	0.63	0.58	0.74	0.85	0.79	0.63	1.04	0.74	0.91	0.83	0.85
4	0.59	0.62	0.60	0.81	0.63	0.72	0.56	0.90	1.03	0.94	0.88	1.14
5	0.51	0.42	0.46	0.75	0.60	0.67	0.50	0.62	0.59	0.60	0.83	0.78
6	0.80	0.66	0.73	0.85	1.03	0.94	0.85	0.53	0.61	0.57	0.96	0.74
7	0.65	0.65	0.65	0.73	0.77	0.75	0.50	0.93	0.93	0.93	0.95	1.21
8	0.91	0.98	0.94	1.21	1.18	1.19	0.72	0.90	0.93	0.91	0.94	0.96
9	0.69	0.75	0.72	0.92	0.99	0.95	0.86	1.28	1.39	1.33	1.54	1.45
10	0.65	0.72	0.68	0.89	0.90	0.89	0.44	0.49	0.58	0.53	0.68	0.75
11	0.61	0.65	0.63	0.78	0.81	0.79	0.61	0.63	0.69	0.66	0.78	0.82
12	0.72	0.79	0.75	0.95	1.15	1.05	0.4	0.41	0.38	0.39	0.67	0.72
Mean			0.65			0.84			0.78			0.89
Standard deviation			0.12			0.16			0.25			0.22
Standard error	0.03			0.04			0.07			0.06		

Table 3

Micrometrical data on aorta of male and female buffalo

S. No.	AA(cm) Male	DA(cm) Male	AA(cm) Female	DA(cm) Female
1	0.43	0.56	0.54	0.77
2	0.56	0.43	0.50	0.54
3	0.69	0.94	0.67	0.89
4	0.52	0.88	0.69	0.70
5	0.76	0.65	0.51	0.65
6	0.40	0.52	0.74	0.71
7	0.65	0.76	0.85	0.93
8	0.75	0.67	0.60	0.68
9	0.48	0.50	0.64	0.77
10	0.51	0.62	0.67	0.75
11	0.53	0.56	0.55	0.68
12	0.47	0.75	0.53	0.64
Mean	0.56	0.65	0.62	0.72
Standard Deviation	0.12	0.15	0.10	0.10
Standard Error	0.03	0.04	0.03	0.03

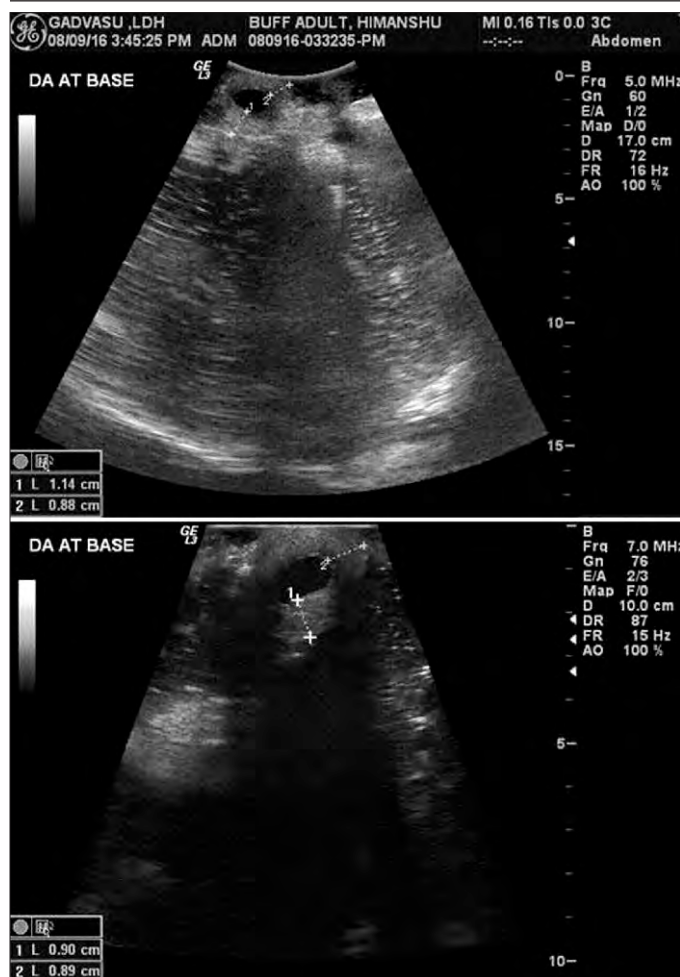


Fig. 3. Two-dimensional images showing the thickness of descending aorta in (a) males and (b) females buffalo

was not readily identifiable, because the intima was rich in elastic fibres. The tunica media was composed of smooth muscle cells whose nuclei were clearly evident. These

smooth muscle cells laid in the spaces between the concentrically layered fenestrated membranes, composed of elastic tissue. The outermost coat of the aorta, the tunica adventitia, was composed of collagenous and elastic fibres interspersed with connective tissue cells and blood vessels, the vasa vasorum. Similarly, Banks (1993) described that aorta and its branches were distinguished by their great elasticity and made up of three layers from i.e. tunica intima, tunica media and tunica adventitia which were analogous to endocardium, myocardium and epicardium, respectively. Tunica intima had a large sub-endothelial layer which grew with age and its border was delineated by internal elastic membrane. Tunica media was thickest of three layers and smooth muscle cells were arranged in spiral around the long axis of the vessel. Tunica adventitia was relatively thin connective tissue layer with fibroblast, macrophages, collagen fibres and blood vessels were present in this layer.

REFERENCES

- Anuradha, Bansal, N., Mohindroo, J. and Uppal, V. (2013). Gross biometrical and ultrasonographic studies on the heart of sheep, goat, pig and buffalo: A Clinical Approach. *Vet. Pract.* **14**(1): 146-148.
- Banks, W.J. (1993). *Applied Veterinary Histology* (4th Edn.), Mosby Year Book, St. Louis, USA. pp. 429-432.
- Braun, U. (2009). Ultrasonography of the gastrointestinal tract in cattle. *Vet. Clin. North Am. Large Anim. Pract.* **25**: 567-590.
- Hallowell, G.D., Potter, T.J. and Bowen, I.M. (2007). Methods and normal values for echocardiography in adult dairy cattle. *J. Vet. Cardiol.* **9**: 91-98.
- Luna, L.G. (1968). *Manual of Histological Staining Methods of the Armed Forces Institute of Pathology* (3rd Edn.), McGraw Hill Book Company, New York. pp. 258.
- Mohan, M. and Prakash, B.S. (1997). Observations on the anatomy of the heart of water buffalo (*Bubalus bubalis*). *J. Anim. Vet. Adv.* **2**: 259-270.
- Muzzi, R.A.L., Muzzi, L.A.L., Araujo, R.B.D. and Cherem, M. (2006). Echocardiographic indices in normal German Shepherd dogs. *J. Vet. Sci.* **7**: 193-198.
- Panhwar, S.P., Rind, M.M., Khan, H., Rind, M. and Rind, R. (2007). Gross anatomical studies on normal heart of buffalo (*Bubalus bubalis*). *Int. J. Agri. Biol.* **9**(1): 162-166.
- Snedecor, G.W. and Cochran, W.G. (1994). *Statistical Methods* (8th Edn.), Oxford & IBH Publishing Co., Calcutta, India.
- Tharwat, M., Sobayil, F., Ali, A. and Buczinski, S. (2012). Echocardiography of the normal camel (*Camelus dromedarius*) heart: Technique and cardiac dimensions. *BMC Vet. Res.* **8**: 130.
- Torad, F.A., Amer, M.S., Shamaa, A.A. and Elsherpieny, E.A. (2016). Echocardiographic measurements and indices in normal adult buffalo (*Bubalus bubalis*). *J. Appl. Anim. Res.* **45**(1): 336-341.
- Yamaga, Y. and Too, K. (1986). Echocardiographic detection of bovine cardiac diseases. *Jap. J. Vet. Res.* **34**(3-4): 251-267.