ORIGINAL ARTICLE

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CT anatomy of cervical vertebrae of Asian elephant (*Elephas maximus*)

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Abstract

Background: Elephants are currently the largest mammals on earth. A comprehensive examination of the anatomy of this animal to diagnose various disorders is required. In addition, due to the heavy head of these animals, adaptations have been made in the anatomical structure of the neck that is worth studying.

Objective: This study aimed to investigate a standard morphologic and morphometric description of the elephant cervical spine. Another aim of this study was to compare the changes in the cervical skeleton of elephants with horses and cattle.

Methods: For this study, the cervical vertebrae of the Asian elephant, cattle and horse were examined. CT Images were obtained using Somatom Spirit II CT Machine. Statistical analysis was done by SPSS 24 software.

Results: Two dorsal tubercles and a groove between them were observed on the dorsal arch of the atlas vertebra of the Asian elephant. In elephant samples, the variation of vertebral body height, spinous process height, transverse process width, vertebral body length and vertebral foramen volume indices were statistically significant. The volume of the vertebral foramen in the elephant decreases in the second vertebra compared to the first vertebra, decreases in the third vertebra, decreases in the fourth, increases in the fifth, decreases in the sixth and increases in the seventh.

Conclusions: In this study, the structure of the cervical vertebrae of the Asian elephant was examined, and certain features were observed. One of the main features was the reduction of the length of the vertebrae, which leads to the decrease of the ratio of neck length to the size of the body. This condition can be due to the high weight of the head in the elephant. To maintain this weight, it is necessary to reduce the length of the neck and confer less mobility.

KEYWORDS

Asian elephant, cattle, cervical vertebrae, CT scan, horse

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1 | INTRODUCTION

Currently, elephants are the largest mammals on the earth. Moreover, these animals are the most important animals in zoos, circuses and wildlife parks. In Asia, they are also used in agriculture. Elephants are not native to Iran, but with the existing connections between Iran and countries such as India, elephants have entered the country for various purposes. The Sassanian Persians (The Sasanian or Sassanid Empire, officially known as the Empire of Iranians. it endured for over four centuries, from 224 to 651 AD, making it the longest-lived Persian imperial dynasty) used elephants on various occasions against the armies of the Later Roman Empire. The elephants were Asian elephants from southern provinces of Iran and India (Charles, 2007). Therefore, in archaeological studies, it is possible to find ancient specimens of elephant bones. Moreover, conservation of these animals in zoos provides the necessity of knowing the basic information such as the anatomical structure of these species to diagnose and treat different disorders. One of the most important features of elephants is their short neck structure, which must tolerate the heavyweight of the head. The main weight-bearing part of the neck is the vertebral column. Diagnostic imaging techniques are one of the most important methods for diagnosing clinical disorders; in addition, these studies can be used for anatomical studies.

The elephant is the name of a large animal of the genus *Proboscidea* in the family *Elephantidae*, which is native to India and Africa. Apart from the size of elephants, two other things that distinguish them from other mammals are their trunks and tusk. Elephants are the heaviest land animals (Shoshani & Tassy, 2005).

Currently, there are species of African and Asian elephants. Elephants weigh up to 7 tons and live for about 70 years. The African elephant is the name of a genus of elephants (Loxodonta) that lives mostly in the Republic of the Congo, the Democratic Republic of the Congo and Kenya. The Asian elephant (Elephas maximus) is a species of elephant that lives in India, Myanmar, the Malay Peninsula, Sri Lanka and Sumatra. The Asian elephant has a swollen forehead and lighter tusk and smaller ears. Also, the height and ivory length of the Indian elephant are smaller than the African elephant (Shoshani & Tassy, 2005). The elephant's neck is too short to allow them to breathe through their mouths while swimming, so the trunk is used like a snorkel to allow proper breathing. In addition, this attribute allows elephants to open their mouths and feed under water. Asian and African elephants can be distinguished by comparing their ears, head shape and tusks. In general, Asian elephants are smaller than African elephants, shorter in height and lighter in weight (Asier, 2016).

In 1997, Wilke et al. (1997) examined the anatomy of a sheep's spine and compared it to a human spine. In this study, five complete vertebrae were used to study pedicles, spinal canal, transverse processes, spinal processes, articular processes, end plates and disc. The results showed that the vertebrae of sheep and humans are very similar in the thoracic and lumbar regions. As a result, they stated that the sheep vertebral column is a suitable model for experimental studies. In 2010, Sheng et al. scrutinised the spinal anatomy of large animals and compared it to the human spine (Sheng et al., 2010). The animals studied in this study included sheep, calves, deer, monkeys and pigs. Significant differences in the anatomy of the spine have been observed between humans and the vertebrae of large animals. In the cervical region, the vertebrae of apes and humans were more similar than those of other organisms. In the thoracic and lumbar regions, the average pedicle height of all animals was higher than that of humans. The mean pedicle widths between animals and humans were similar except in the thoracic vertebrae of sheep. The human spine was wider than other animals. The average height of vertebral bodies was shorter than that of all animals.

In 1979, Jeffcott et al. evaluated the natural radiographic anatomy of a horse's thoracic vertebrae from birth to maturity. Radiographic anatomy of 149 horses was performed to examine the sacral and caudal regions. The spine of the newborn sphere is more curved in the middle region than that of the adult animal and is convex from the dorsal surface. The transverse dorsal appendages are shorter than the length of the vertebral bodies and there is no visible ossification centre from the T2–T7 vertebra. In the middle part of the thoracic cavity, the spinous process had rough ends. In 1993, Mattoon et al. performed a quantitative radiographic evaluation of the lumbosacral vertebrae of healthy dogs and dogs with degenerative lumbar-sacrum stenosis (Jeffcott, 1979; Mattoon & Koblik, 1993).

In 1995, Jones et al. (1995) investigated the anatomy of the healthy dog's lumbar-sacral vertebrae with CT scan images. All CT scan sections were compared with the same sections in anatomy and radiographic evaluations. Anatomical features observed in CT scans of dogs consist of vertebral bodies, pedicels, laminae, articular processes, spine processes, transverse processes, mammary processes, intervertebral foramina, sacrum wings, sacroiliac joints, nerve roots and spinal nerves.

In 1996, Feeney et al. (1996) investigated the lumbosacral vertebrae of healthy dogs by CT scan. In 1979, Wright et al. (1979) investigated the radiographic anatomy of the dog's cervical vertebrae. In this study, longitudinal diameters of neural canals were studied by radiography.

In 1985, Penning et al. analysed the movement of dog cervical vertebrae with human cervical vertebrae. In this study, it was stated that the rate of movement of dog cervical vertebrae is greater than that of humans and mainly depends on the degree of flexion and elongation of the atlanto-occipital joint. The patterns of curvature and tension of the dog's cranial and caudal cervical vertebrae are similar to the patterns of these cases in humans.

In 1979, Jeffcott et al. investigated the horse's spine radiography. This study was designed to meet the demand of producing a method for minimising the secondary x-rays that increase the quality of radiography by limiting the initial X-ray radiation through the collimator.

In Gilad and Nissan (1985) examined the geometric sections of the human spine in the longitudinal section. Healthy individuals aged 20–38 years with an average height of 174 cm participated in this study. Radiographs of 141 cervical and 157 lumbar vertebrae were studied under the same conditions and standard procedure. The radiographic findings were as follows: The C1 vertebra was covered by the skull, making it impossible to identify and measure geometrically. Also, the thoracic vertebrae were not evaluated because the longitudinal section of the ribs covered the thoracic vertebrae. In some cases, approximately 10% of the C2 vertebrae were not sufficiently recognisable, and approximately 10% of the C7 vertebrae were covered by the clavicle, arm and ribs. Furthermore, the average width of the vertebral bodies increases from C2 to C7. The average height of the vertebral body in C2 is higher than other cervical vertebrae, while the rest of the vertebrae are almost the same height.

In 2017, Nganvongpanit et al. evaluated the morphology and elemental composition of Asian elephant (*Elephas maximus*) bones (humerus, radius, ulna, femur, tibia, fibula and rib). Computerised tomography was used to image the intraosseous structure, compact bones were processed using histological techniques and elemental profiling of compact bone was conducted using X-ray fluorescence (Nganvongpanit et al., 2017).

In 2019, Cossu et al. observed five atlases of an extinct species, the late pleistocene mammoths (M. primigenius), and compared them with five atlases of a closely related existent species, the African elephant (L. africana). Their study was on the presence or absence of arcuate foramen (Cossu et al., 2019). In the human, the groove of the vertebral artery on the posterior arch of the atlas (sulcus arteriae vertebralis) may become a complete or partial osseous foramen: the arcuate foramen. The presence of a complete or partial arcuate foramen is a rare anatomical variant described in a minority of patients and it seems to be associated with vertigo, vertebrobasilar insufficiency. posterior circulation strokes and musculoskeletal pain. They observed this foramen in all the mammoths they studied, but it was not found in any of the African elephant specimens (Cossu et al., 2019). In their study, Hutchinson et al. (2008) also reconstructed and examined the skeletal structure of elephant limbs, but did not do anything about the spine.

Gunji et al. (2014) report an abnormality of the neck in a bedridden Asian elephant. To diagnose this calf, a CT scan of the neck and thorax was performed and these cases were observed: articular facets of the cervical and first three thoracic vertebrae possessed intricately rough surfaces and the anterior articular processes of C4, C5 and C6 intruded to the adjacent processes. This study concludes that a long-term lying posture under the bedridden condition might cause the over-dorsalisation of the neck and the deformations of the articular processes in large mammals (Gunji et al., 2014). Marchant and Shoshani (2007) studied the head muscles of Loxodonta africana and Elephas Maximus and also studied their anatomical differences. They have mentioned that differences in musculature between elephant species exist and are reflected in their physical appearances. In this study, differences and anatomical features related to skeletal features were not mentioned (Marchant & Shoshani, 2007). Bezuidenhout and Seegers (1996) studied the osteology of the African elephant (Loxodonta africana), in this study, they examined the vertebral column, ribs and sternum. In this study, the bones of a 27-year-old female sample were examined. This study was morphological and no measurements were made.

Due to the value and importance of elephant and various risks such as fractures, joint problems, etc. that can threaten their health, overall examination of the anatomy of these animals to diagnose various disorders seems necessary. According to the available information, an accurate and standard description of the structure of elephant cervical vertebrae has not been performed so far.

The aim of this study was to provide an accurate description of the anatomy of elephant cervical vertebrae based on CT scan findings and anatomical studies of existing specimens and also to compare these features with the structure of cervical vertebrae in horses and cattle.

It seems that through the CT scan method and common anatomical and morphometric studies, anatomical features and modifications of elephant cervical vertebrae in comparison with horses and cattle can be investigated.

2 | MATERIALS AND METHODS

2.1 Bone samples

Cervical vertebrae of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) were studied (five sets of cervical vertebrae from each animal). Each of the seven cervical vertebrae in each series of specimens belonged to a specific animal. All five specimens were adult male. Due to the closed ossification centres of the specimens, they were all mature in terms of bone growth. The average weight of the elephants was 4540 \pm 0.52 kg (mean \pm standard deviation), the average weight of the horses was 440 \pm 0.44 kg and the average weight of the cattle was 690 \pm 0.44 kg.

To separate the bones pieces, the conventional process of bone separation including soft tissue separation, boiling, degreasing and bleaching has been used. Before boiling the samples, the bones removed from the soft tissue were placed in cold water for 24 h and a small amount (much less than 10%) of household ammonia was added to the water. Ten per cent hydrogen peroxide was used for bleaching. Due to the large volume of samples and the high cost of common solutions for the degreasing stage, super gasoline was used for this stage.

All of the samples were prepared with the same process. It is noteworthy that elephant specimens have been selected from various available collections, including the Anatomy Department of the Faculty of Veterinary Medicine, University of Tehran; Pardisan Park Museum, Department of Environment etc., and several private collections. The anatomical structure of the samples has been evaluated. The elephant samples with horses and cows were compared and specific characteristics of the elephant were recorded.



FIGURE 1 The positioning of elephant vertebrae for CT scan imaging

2.2 | CT Scan study

In this step, the samples were transferred to the Radiology Department of the Faculty of Veterinary Medicine, University of Tehran Images were taken using a Somatom Spirit II CT Scan Machine. CT images were transversely taken to the axis of the vertebral column (Figure 1).

Technical factors for this study were as follows: rotation time, 1 s; slice thickness, 1 mm; reconstruction interval, 0.5–1 mm; pitch, 1; X-ray tube potential, 120 kV and X-ray tube current, 130 mA. In the images obtained from the CT scan, different structures of the vertebral column were analysed. Appropriate window width (WW) and window level (WL) were selected for each image. A bone window was used to examine the images.

2.3 | Morphometric study

After analysing the CT scan images and identifying the different sections, the parameters were measured in the CT scan images of the spine. The measured parameters are described in Table 1. Measured parameters include vertebral body height (VBH), spinous process height (SPH), transverse process width (TPW), vertebral body length (VBL), cervical length (C-Length), length of the dens in the C2 (Dens) and volume of the vertebral foramen (Vol). The results of the measurements are shown in Tables 2–4 (in these tables, the volume is the same as the volume of the vertebral foramen). Morphometric measurement from digital CT images was performed with Syngo MMWP VE40A software. Hounsfield unit – 120 to +1900 was selected for volumetric measurements in this software (Hounsfield unit for soft tissue and bone). In this study, the vertebral foramen volume of each vertebra was also measured.

2.4 | Statistical analysis

Statistical analyses were done by SPSS software version 24.0. The descriptive statistics are described by mean \pm standard error of the mean (SEM). Repeated measures analysis with 95% confidence interval was carried out by general linear model (GLM) procedure for trends measures. The comparison of ratios among species was done by one-





way ANOVA and Tukey's post hoc tests. A *p*-value less than 0.05 was considered statistically significant.

3 | RESULTS

3.1 | Morphological characteristics of Asian elephant cervical vertebrae

Seven cervical vertebrae were observed in the elephant. In this paper, further explanation will be given, but in general, the length of the cervical vertebrae was short. Figures 2 and 3 show 3D reconstruction of the CT scan of the elephant cervical vertebrae. Figure 4 shows the transverse CT scan images of the cervical vertebrae of the Asian elephant. Figure 5 shows the 3D reconstruction of the CT scan of the horse and cattle cervical vertebrae.

3.1.1 | First cervical vertebrae (atlas)

The cranial articular cavities were not deep. The fovea dentis was hollow and considerably enlarged. Lateral vertebral foramen was observed, but alar foramen or alar notch was not observed. The wings

TABLE 1 Morphometric parameters of the vertebral column, the measurement of the parameters (red line) is shown in the CT images

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Abbreviation	Parameter	Description	Visual description in the elephant cervical vertebra
VBH	Vertebral body height	Distance from the base of the vertebrae to the vertebral canal in the transverse view; the maximum distance was measured	VBH
SPH	Spinous process height	Distance from the base to the apex of the spinal process in the midsagittal view; the maximum distance was measured	SPH
TPW WPW (C1)	Transverse process width	Distance between the end of the right and left transverse processes in the transverse view; maximum distance was measured	TPW
VBL	Vertebral body length	The length of the vertebral body in the midsagittal view; the maximum distance was measured	VBL Carly M
C-Length	Cervical length	The length of the cervical part of the vertebral column in the midsagittal view; the maximum distance was measured	C-Length
Dens	Length of the dens in the C2	The length of the dens in the C2 in the midsagittal view; the maximum distance was measured	Dens
Vol	Volume of the vertebral foramen	Volumetric measurement from digital CT images was performed with Syngo MMWP VE40A software. Hounsfield unit –120 to +1900 was selected for volumetric measurements in this software (Hounsfield unit for soft tissue and bone). In this method, range of vertebral foramen of each cervical vertebrae had to be specified in different images of transverse, sagittal and dorsal views.	Vol

Note: The CT images on which the morphometric measurements were performed are related to the Asian elephant,



FIGURE 3 Cranial (a), caudal (b) and left lateral (c) views of the first cervical vertebra of the Asian elephant. 1. Cranial articular cavity, 2. fovea dentis, 3. dorsal tubercle, 4. ventral tubercle, 5. wing, 6. lateral vertebral foramen, 7. transverse foramen. 8. caudal articular surface, 9. vertebral foramen



FIGURE 4 Cranial (a), caudal (b) and left lateral (c) views of the second cervical vertebra of the Asian elephant. 1. Spinous process, 2. caudal articular process, 3. vertebral foramen, 4. transverse foramen, 5. vertebral body, 6. dens, 7. cranial articular surface, 8. lateral vertebral notch, 9. transverse process



FIGURE 5 Cranial (a), caudal (b) and left lateral (c) views of the third cervical vertebra of the Asian elephant. 1. Vertebral body, 2. spinous process, 3. vertebral foramen, 4. cranial articular process, 5. caudal articular process, 6. transverse process, 7. transverse foramen

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TABLE 2 Measurements performed on elephants' cervical
 vertebrae (cm and cm³), Asian elephants (*Elephas maximus*)

TABLE 3	Measurements performed on cattle cervical vertebrae
(cm and cm ³)	, cattle (<i>Bos taurus</i> , Holstein Friesian)

Index	Number	Minimum	Maximum	Mean	SEM	Index	Number	Minimum	Maximum	Mean	SEM
EVBHC2	5	8.12	11.23	9.69	0.52	CVBHC2	5	2.13	5.21	3.76	0.52
EVBHC3	5	10.22	13.41	11.75	0.53	CVBHC3	5	2.21	5.22	3.75	0.51
EVBHC4	5	10.37	13.07	11.69	0.45	CVBHC4	5	2.33	5.32	3.92	0.50
EVBHC5	5	10.32	13.21	11.92	0.48	CVBHC5	5	2.12	5.11	3.65	0.50
EVBHC6	5	9.56	12.51	11.17	0.49	CVBHC6	5	2.11	5.12	3.70	0.53
EVBHC7	5	9.55	12.35	11.09	0.46	CVBHC7	5	2.21	5.15	3.70	0.50
ESPHC2	5	6.31	9.31	7.90	0.50	CSPHC2	5	2.79	5.73	4.34	0.49
ESPHC3	5	1.04	4.62	3.05	0.59	CSPHC3	5	1.61	4.62	3.12	0.50
ESPHC4	5	1.34	4.51	3.12	0.52	CSPHC4	5	1.49	4.49	3.05	0.50
ESPHC5	5	3.41	6.31	4.85	0.49	CSPHC5	5	1.81	4.78	3.37	0.50
ESPHC6	5	6.54	9.52	8.17	0.49	CSPHC6	5	2.11	5.06	3.63	0.50
ESPHC7	5	12.36	15.16	13.89	0.49	CSPHC7	5	3.32	6.33	4.86	0.51
ETPWC1	5	31.75	34.82	33.41	0.51	CTPWC1	5	10.21	15.16	12.12	0.83
ETPWC2	5	18.98	21.87	20.49	0.49	CTPWC2	5	6.15	11.21	8.15	0.85
ETPWC3	5	19.31	22.25	20.87	0.49	CTPWC3	5	6.25	11.27	8.24	0.84
ETPWC4	5	19.18	22.31	20.87	0.54	CTPWC4	5	6.17	11.15	8.16	0.83
ETPWC5	5	19.44	22.41	20.98	0.50	CTPWC5	5	6.28	11.32	8.26	0.84
ETPWC6	5	20.89	23.89	22.45	0.50	CTPWC6	5	5.41	10.34	7.34	0.83
ETPWC7	5	21.51	24.51	23.02	0.50	CTPWC7	5	5.88	10.91	7.86	0.84
EVBLC2	5	4.47	10.25	7.90	0.99	CVBLC2	5	4.77	7.71	6.34	0.49
EVBLC3	5	2.78	5.86	4.05	0.57	CVBLC3	5	4.21	7.15	5.74	0.50
EVBLC4	5	2.75	5.62	3.94	0.54	CVBLC4	5	4.13	7.11	5.63	0.50
EVBLC5	5	2.91	5.88	4.10	0.56	CVBLC5	5	4.22	7.21	5.70	0.51
EVBLC6	5	2.79	5.71	4.07	0.55	CVBLC6	5	1.87	4.91	3.45	0.51
EVBLC7	5	3.75	6.91	5.06	0.58	CVBLC7	5	1.91	5.11	3.57	0.53
EVolC1	5	243.12	277.21	257.62	6.34	CVolC1	5	56.21	86.28	71.89	5.05
EVolC2	5	221.31	251.14	234.14	5.49	CVoIC2	5	23.33	53.27	38.93	5.03
EVolC3	5	163.71	193.71	175.89	5.81	CVoIC3	5	12.15	42.04	27.71	5.03
EVolC4	5	162.37	194.34	174.95	6.12	CVoIC4	5	10.42	40.38	26.03	5.04
EVolC5	5	162.11	193.31	175.97	5.51	CVoIC5	5	10.36	40.44	26.02	5.04
EVolC6	5	164.61	192.67	175.87	5.57	CVoIC6	5	7.62	37.53	23.17	5.02
EVolC7	5	165.69	195.67	180.33	5.45	CVoIC7	5	5.21	35.16	20.82	5.04
EDensC2	5	2.07	4.47	3.24	0.48	CDensC2	5	0.51	2.21	1.58	0.33
EC-Length	5	32.75	35.77	34.08	0.56	CC-Length	5	28.41	49.36	39.28	3.51
ETPWC1/ EC-Length	5	0.97	1.02	0.98	0.01	CTPWC1/ CC-length	5	0.29	0.36	0.31	0.01
EDensC2/ EVBLC2	5	0.23	1.00	0.47	0.14	CDensC2/ CVBLC2	5	0.11	0.33	0.24	0.04

Note: At the beginning of the symbol of each parameter, the letter E indicates the elephant and at the end of it, the symbol of the vertebral number and the letter C have been added.

were also somewhat cranial. Caudal articular surfaces were not very elongated and was almost square. The ventral surfaces of the wings were prominent, and there was no depression. The transverse foramen was horizontal to the wing and extended along with it. Two dorNote: At the beginning of the symbol of each parameter, the letter C indicates the cattle and at the end of it, the symbol of the vertebral number and the letter C have been added.

sal tubercles and a groove between them were observed on the dorsal arch of the atlas. A ventral tubercle was located on the ventral arch of the atlas that tended posteriorly (Figures 6 and 7). Arcuate foramen was not observed in any of the atlases.



FIGURE 6 Cranial (a), caudal (b) and left lateral (c) views of the fourth cervical vertebra of the Asian elephant. 1. Vertebral body, 2. spinous process, 3. vertebral foramen, 4. cranial articular process, 5. caudal articular process, 6. transverse process, 7. transverse foramen



FIGURE 7 Cranial (a), caudal (b) and left lateral (c) views of the fifth cervical vertebra of the Asian elephant. 1. Vertebral body, 2. spinous process, 3. vertebral foramen, 4. cranial articular process, 5. caudal articular process, 6. transverse process, 7. transverse foramen

3.1.2 | The second cervical vertebrae (axis)

The proximal part of the spinous process was prominent and semicircular. The process was divided into two parts, which consist of a groove between the two parts. Each of these two sections reached the top of the caudal articular process. The articular surface of the caudal articular process was divided into two parts, a lateral and a medial part.

Transverse foramen was observed in the transverse process. Transverse process was inclined towards the caudal. The caudal portion of the vertebral body was large and provided a considerable surface for articulation with the next vertebra. The dens also had a prominent position at the dorsal and the cranial surfaces. Cranial articular surfaces were square; these two surfaces did not meet under dens. Lateral vertebral foramen was not observed and there was an incision in this section. Also, no crest was observed on the ventral surface (Figures 6 and 8).

3.1.3 | Third, fourth and fifth cervical vertebrae

The length of the vertebral body was short. The height of the spinous process increased from the third to fifth vertebrae. This process was almost vertical in the third and fourth vertebrae but it was leaned anteriorly in the fifth vertebrae. The transverse process was inclined to the caudal and a transverse foramen was observed. Articular surfaces of the anterior and posterior articular process had two sections, one located more internal and one more lateral (Figures 6, 9–11). At the ventral surface of all of these three vertebrae, a crest-like protrusion with slight eminence was observed.

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FIGURE 8 Cranial (a), caudal (b) and left lateral (c) views of the sixth cervical vertebra of the Asian elephant. 1. Vertebral body, 2. spinous process, 3. vertebral foramen, 4. cranial articular process, 5. caudal articular process, 6. transverse process, 7. transverse foramen



FIGURE 9 Cranial (a), caudal (b) and left lateral (c) views of the seventh cervical vertebra of the Asian elephant. 1. Vertebral body, 2. spinous process, 3. vertebral foramen, 4. cranial articular process, 5. caudal articular process, 6. transverse process, 7. facet for head of the first rib



FIGURE 10 Cranial views of the vertebral foramen range of the seventh cervical vertebra of the Asian elephant; the number of vertebrae indicated the blue part in the first image is related to fovea dentis

3.1.4 | Sixth cervical vertebra

In this vertebra, the spinous process was longer, unlike the former vertebra, and it inclined more to the cranial. In these vertebrae, as

the previous one, the transverse process was inclined to the caudal and transverse foramen was seen on this process. Furthermore, the articular surface of the cranial and caudal articular processes was divided into two parts (Figures 6 and 12). In the ventral surface of these vertebrae, a crest-like tuberosity with light eminence was observed.

3.1.5 | Seventh cervical vertebra

In this vertebra, the spinous process was longer than the previous one and more inclined to the cranial. In this vertebra, as in the previous ones, the transverse process was positioned caudally, and there was no transverse foramen on it. Also, the articular surface of the cranial and caudal processes was two part. In the caudal part of the body of this vertebra, on both right and left sides, there was a joint surface for the head of the first rib (costal facet) (Figures 6 and 13). In the

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FIGURE 11 3D reconstruction images (osseous-shaded-vp) of the cervical vertebra of the Asian elephant. Left lateral (a) and cranial (b) views



FIGURE 12 Longitudinal (a) and dorsal (b) sections of reconstructed 3D CT scan images of the cervical vertebra of the Asian elephant

ventral surface of these vertebrae, a crest-like tuberosity with light prominence was observed.

3.2 Morphometric characteristics of cervical vertebrae of Asian elephants, horses and cattle

The results of measurements performed on elephants, horses and cattle are shown in the tables in this section. In each table, the meaningfulness of the statistical differences is alphabetically marked. Changes in the measured parameters of the cervical vertebrae of the elephant, horse and cattle are shown in Charts 1-6.

3.2.1 Elephant

In elephant samples, the variation of VBH, SPH, TPW, VBL and vertebral foramen volume indices were statistically significant (p < 0.05). The trend of parameter changes and whether the changes are statistically significant are shown in Table 2.

3.2.2 | Cattle

In the cattle samples, the variation of VBH, SPH, TPW, VBL and vertebral foramen volume indices were statistically significant (p < 0.05).











The trend of parameter changes and whether the changes are statistically significant or not are shown in Table 3.

3.2.3 | Horse

In the horse samples, the variation of VBH, SPH, TPW, VBL and vertebral foramen volume indices were statistically significant (p < 0.05). The trend of parameter changes and whether the changes are statistically significant or not are shown in Table 4.

3.2.4 | Ratio of some parameters

Using Tukey's post hoc test, there was a statistically significant difference in the ratio of width of transverse process to neck length (TPW/C-Length) (Chart 7) between elephant and cow (p < 0.001). The ratio of width of transverse process to neck length between elephant and horse was statistically significant (p < 0.001) by Tukey's post hoc test. There was a statistically significant difference in the ratio of width of transverse process to neck length between cow and horse (p < 0.05) by Tukey's post hoc test. Using one-way ANOVA test, there were not any



CHART 2 SPH changes in the cervical vertebrae of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) (unit: cm)



CHART 3 TPW changes in the cervical vertebrae of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) (units: cm)

statistically significant differences in Dens/VBL C2 between species (p > 0.05). The change in the shape and volume of the vertebral foramen is seen in Figure 14.

Changes related to each of the parameters in the different animals relative to each other are shown in the Charts 1–5. As shown in the Chart 6, the largest cervical length among the animals we studied was the horse; it should be noted that as shown in the Chart 4, the largest amount of VBL was observed in horse (in each vertebra), which are definitely related to these two issues. The TPWC1/ C-Length ratio in elephants is close to one, and it can be said that the width of the atlas is approximately equal to the length of the neck.

4 DISCUSSION

In this study, the anatomy of the cervical vertebrae of Asian elephants is morphologically and morphometrically evaluated. In the following, we will analyse these characteristics and compare them with horses and cattle. In domestic animals, there are seven cervical vertebrae, and the difference in neck length in different animals does not affect the number of vertebrae (König et al., 2007), while the size of the vertebrae is different in various animals. Consequently, in Asian elephants, seven cervical vertebrae were relatively short.

Lateral vertebral foramen has been reported in the first cervical vertebrae of horses and cattle, while in elephants this structure was observed as a notch (König et al., 2007). It has also been noted that in the cranial part of the dorsal arch in horses and cattle, alar foramen was observed; in the elephant this structure was neither observed in the form of notch nor foramen (Sisson & Grossman, 1975).

In horses and cattle, it is mentioned that there is a lateral vertebral foramen that leads into the vertebral foramen, which, in this study, was also observed in elephants. Regarding the ventral surface of the wing



CHART 4 VBL changes in the cervical vertebrae of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) (units: cm)



CHART 5 Volume changes in the cervical vertebrae of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) (units: cubic centimetres)

of the atlas vertebrae of horses and cattle, it has been mentioned that there was a depression called atlantal fossa (Sisson & Grossman, 1975); in the elephant, a convex surface was detected in this part instead of the fossa.

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In horses, it is mentioned that there is a transverse foramen in the wing of the Atlas, but this foramen is not seen in cattle (König et al., 2007). In Asian elephant, it should be noted that this foramen is seen in the wing of the atlas. Transverse foramen is located horizontally in the wings of the elephant.

In the case of horses and cattle, it has been reported that there is a tuberosity called the dorsal tubercle on the dorsal arch of the atlas (Sisson & Grossman, 1975). Furthermore, in this study, two large tuberosities above the dorsal arch were observed.

In the Asian elephant, it was mentioned that the ventral tubercle in the atlas was not recognisable (Dehury et al., 2016). On the contrary, in our study, this structure was quite clear.

The upper incision between the two cranial articular cavities is wider than the lower incision, which was also observed in our study (Dehury



CHART 6 C-Length (cervical length) of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian) (units: cubic centimetres)



CHART 7 TPWC1/C-Length ratio of Asian elephants (*Elephas maximus*), horses (*Equus ferus caballus*, Thoroughbred) and cattle (*Bos taurus*, Holstein Friesian)

et al., 2016). In the study of Dehury et al. (2016), as in our study, two tubercles are observed above the dorsal arch.

The dorsal border of the spinous process is somewhat flat in cattle and somewhat prominent in horses. In this study, we also observed that this border is somewhat prominent and, like a horse, bifurcated in the posterior part and continues to the top of the caudal articular processes (Sisson & Grossman, 1975).

Dens in horses and cattle are almost hollow, while in Asian elephants it was prominent and rod-shaped (Sisson & Grossman, 1975). Lateral vertebral foramen is present in horses and cattle, but in Asian elephants, this structure was observed as a notch (Sisson & Grossman, 1975) In Asian elephants, like horses, where the cranial articular surfaces do not meet below the dens, they do not meet here, unlike cattle (Sisson & Grossman, 1975). In the study of Dehury et al. (2016), it was mentioned about dens, which has a conical structure, and it should be noted that the same state was observed in our study.

In elephants, the length of the body was short, while in horses and cattle, the length of the vertebral bone body was significant (Sisson & Grossman, 1975). In these three vertebrae in the elephant, the cranial and caudal articular processes had two lateral and medial joint surfaces, which was not the case of horse and cattle. A gradual increase in the height of the spinous process from the third to the fifth

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FIGURE 14 3D reconstruction images (osseous-shaded-vp) of the cervical vertebra of the horses (*Equus ferus caballus*, Thoroughbred) (a) and cattle (*Bos taurus*, Holstein Friesian) (b)

vertebrae is also observed in elephants, horses and cattle (Sisson & Grossman, 1975).

In horses and cattle, the spinous process of the sixth vertebrae is higher than the previous vertebrae, which was also observed in the Asian elephant (Sisson & Grossman, 1975). The transverse process of the sixth vertebra in horse and cattle has a different shape in comparison with other vertebrae, which is wider at the ventral part. This feature was very prominent in cattle, but in elephants, the shape of this process was not different from other vertebrae and did not have a wider ventral part (Sisson & Grossman, 1975). In this elephant vertebrae, also, the two-part shape of the articular surface of the cranial and caudal articular process was observed, which is not the case in horse and cattle (Sisson & Grossman, 1975).

In horse and cattle, the seventh vertebral spinous process was longer than the previous vertebrae, which was also observed in the Asian elephant (Sisson & Grossman, 1975). Just in as there is no transverse foramen in the seventh cervical vertebrae of horses and cattle, this foramen was not observed in the elephant (Sisson & Grossman, 1975).

Based on the statistical results observed in this study, the following conclusions can be reached about the changes in the measured parameters in three species: The size of VBH (vertebral body height) in elephants from the second to the sixth vertebra has changed in such a way that it increases from the second to the third vertebra, decreases from the third to the fourth vertebra, increases again from the fourth to the fifth vertebra and from the fifth to the seventh vertebra remains constant. But in cattle it is constant between the second and third vertebrae, it increases in the fourth vertebra, it decreases again in the fifth vertebra and it is fixed from the fifth to the seventh vertebra. In horses, it is larger in the third vertebra than in the second vertebra, it decreases in the fourth vertebra to the third, it decreases in the fourth vertebra compared to the third, it decreases in the fourth vertebra compared to the third, it decreases in the fourth vertebra compared to the third.

the fifth vertebra and it decreases in the sixth vertebra and does not change in the seventh vertebra compared to the sixth. Shateri et al. (2020) studied the cervical spine of rabbits in 2020 by CT scan. According to a study of the cervical vertebrae of the rabbits, VBH did not change from C2 to C7.

The size of SPH (spinous process height) in elephants decreases in the third vertebra compared to the second, increases in the fourth to the third and does not change in the fifth to the fourth, in the sixth compared to the fifth, and in the seventh compared to the sixth increases. In cattle it increases in the third vertebra compared to the second, in the fourth vertebra, it decreases compared to the third and in the fifth to the fourth, it does not change, and it also increases in the sixth and seventh. In the horse, it decreased in the third vertebra compared to the second, increased in the fourth, decreased in the fifth compared to the fourth, increased in the sixth compared to the fifth and did not change in the seventh compared to the sixth vertebrae. In the case of the SPH of the rabbits, it did not change from C2 to C7 (Zehtabvar et al., 2020). It should be noted that the length of the spinous process in the C6 and C7 seems longer, but the angle of this process is such that SPH has not changed; in this study, the angle of the spinous process was not measured.

The size of TPW (Transverse process width) in the elephant decreases in the second vertebra compared to the first vertebra, increases in the third to the seventh vertebra. In cattle, it decreases in the second vertebra compared to the first vertebra, increases in the third, decreases in the fourth, increases in the fifth, decreases in the sixth and increases in the seventh compared to the sixth vertebrae. In the horse, it decreases in the second vertebra compared to the first vertebra, increases in the third and fourth, decreases in the fifth and increases in the sixth and seventh vertebrae. Tan et al. (2004) in the case of TPW in humans, indicated that this param-

TABLE 4 Measurements performed on horses' cervical vertebrae (cm and cm³), horses (*Equus ferus caballus*, Thoroughbred)

Index	Number	Minimum	Maximum	Mean	SEM
HVBHC2	5	3.41	6.22	4.88	0.48
HVBHC3	5	3.49	6.45	5.07	0.50
HVBHC4	5	3.36	6.33	4.95	0.50
HVBHC5	5	3.22	6.21	4.75	0.50
HVBHC6	5	3.15	6.11	4.63	0.50
HVBHC7	5	3.31	6.19	4.80	0.48
HSPHC2	5	3.48	6.39	5.01	0.49
HSPHC3	5	0.15	3.12	1.67	0.50
HSPHC4	5	0.42	3.28	1.95	0.49
HSPHC5	5	0.31	3.32	1.85	0.50
HSPHC6	5	0.54	3.37	2.02	0.48
HSPHC7	5	0.52	4.51	2.67	0.65
HTPWC1	5	14.41	17.28	15.92	0.49
HTPWC2	5	6.32	9.41	7.93	0.52
HTPWC3	5	9.65	12.72	11.24	0.51
HTPWC4	5	9.68	12.68	11.27	0.51
HTPWC5	5	9.35	12.43	10.95	0.52
HTPWC6	5	9.46	12.49	11.08	0.51
HTPWC7	5	9.49	12.48	11.13	0.51
HVBLC2	5	8.69	11.71	10.28	0.50
HVBLC3	5	7.55	10.65	9.17	0.52
HVBLC4	5	7.41	10.29	8.91	0.49
HVBLC5	5	7.31	10.31	8.90	0.50
HVBLC6	5	7.29	10.29	8.86	0.50
HVBLC7	5	6.31	10.36	7.69	0.69
HVolC1	5	132.04	161.21	146.92	4.92
HVoIC2	5	42.07	74.11	59.74	5.47
HVoIC3	5	25.52	56.71	42.31	5.17
HVoIC4	5	25.21	56.33	41.93	5.16
HVoIC5	5	25.38	56.41	42.07	5.14
HVoIC6	5	25.33	56.34	42.01	5.14
HVolC7	5	25.43	56.61	40.97	5.25
HDensC2	5	1.55	4.79	3.26	0.53
HC-Length	5	52.49	73.31	63.21	3.49
HTPWC1/ HC-length	5	0.24	0.27	0.25	0.01
HDensC2/ HVBLC2	5	0.18	0.41	0.31	0.04

Note: At the beginning of the symbol of each parameter, the letter H indicates the cattle and at the end of it, the symbol of the vertebral number and the letter H have been added.

eter increases from C3 to C7. The same trend was observed in Asian elephants. TPW in the rabbit did not change from C2 to C4, it increased at C4 and did not change up to C7, though it was the largest in C7 and C5 of the sheep. TPA was not measured in the

sheep. SCW in the rabbit was invariable from C2 up to C7. In the sheep, SCW was the greatest in the cervical region. PDW in the rabbit showed an invariable measure from C2 to C7, while it had the smallest size in the sheep between C2 and C6 and increased at C7 (Shateri et al., 2020).

The size of the VBL (Vertebral body length) in an elephant in the third vertebra decreases compared to the second. Decreased in the fourth, increased in the fifth, decreased in the sixth and increased in the seventh. In cattle, it decreases in the third vertebra compared to the second. Decreased in the fourth, increased in the fifth, decreased in the fourth and increased in the seventh compared to the sixth. In the horse, it decreases in the third vertebra compared to the second and decreases in the fourth to seventh.

Abiodun et al. performed an anatomic evaluation of the sub-axial cervical spine in humans. Among the parameters they have studied, VBH is similar to the VBL we have studied in elephant specimens. It should be noted that this is due to the difference in naming anatomical aspects in humans and animals, and both refer to the same parameter, but the naming is different. They have mentioned that this parameter increased insignificantly from C3 to C7, although the case was different at C6 vertebral level where a decrease was noticed (Abiodun et al., 2020).

Tan et al. did a study on quantitative three-dimensional anatomy of cervical, thoracic and lumbar vertebrae of Chinese Singaporeans. The cervical vertebrae studied in that study were C3–C7. Some of the parameters studied in humans were similar to the parameters we studied in elephants in terms of measurement method. Tan et al. (2004) have indicated that VBH is fairly constant from C3 to C5 before the increase to T1. It should be noted that based on the method of measuring and placing the elephant on the ground, this parameter is equivalent to VBL in our study. According to a study of the cervical vertebrae of the rabbits, VBL did not change from C2 to C7 (Shateri et al., 2020).

The volume of the vertebral foramen in the elephant decreases in the second vertebra compared to the first vertebra, decreases in the third vertebra, decreases in the fourth, increases in the fifth, decreases in the sixth and increases in the seventh. In cattle, it decreases in the second vertebra compared to the first and it decreases in the third to seventh vertebrae. In the horse, it decreases in the second vertebra compared to the first, it decreases in the third and fourth and it increases in the fifth and decreases in the sixth and seventh.

The ratio of transverse process width to neck length (TPW/ C-Length) in the elephant is greater than in the cattle and horses and higher in the cattle compared to the horse. In 2017, Dehury et al.'s investigation on the Asian elephant, the width and length of the atlas were less than the second cervical vertebra, which in our study was much wider than the second vertebra, and it seems to be due to measurement problems in that study (Dehury et al., 2016).

Cossu et al. have indicated that they have not seen the arcuate foramen in any of the African elephant atlas vertebrae that they have studied, 'we did not see such a foramen in the case of the Asian elephant'. Therefore, the Asian elephant does not have the problems caused by this foramen (Cossu et al., 2019). ¹⁷⁶⁶ │ WILE

Linden et al. state that in smaller mammals, compression of the cervical vertebrae usually occurs. In Asian elephants, compression of the cervical vertebrae also occurred despite the large body size, which is somewhat consistent with our findings on relative neck length (Linden et al., 2019).

Considering the observed features of the morphology and morphometry of the cervical vertebrae of the Asian elephant, some possible features of the muscles associated with them can be considered. There are old studies on the muscular structure of the elephant, for example, the musculature of Indian elephant. Musculature of the trunk, neck and head was noted by Mori and Shindo (1956). One of the problems with Mori's study of Asian elephant's muscles in 1956 is that muscle naming is based on human anatomy naming, not veterinary anatomy terminology. This problem makes it difficult to compare the structure of elephant muscles with other animals whose information is available in veterinary anatomy references.

It is noted that some muscles are attached to the atlas vertebra in the Asian elephant; some of these muscles include a small part of the M. cutaneous trunci crosses over the scapula, goes in neck region and passes into a thin, flat tendon to be inserted into the transverse process of the atlas. M. levator scapulae is small, and lies beneath the neck fascia, arising from the transverse process of the atlas by means of a thin, flat, tendinous slip (Mori & Shindo, 1956). Veterinary anatomy references indicate that M. serratus ventralis cervicis, which is divided into several converging bundles, reaches the surface of the neck between the trapezious and the brachiocephalicus. It is homologous to the levator scapulae in humans (Sisson & Grossman, 1975). M. serratus ventralis cervicis originates from the transverse processes of the cervical vertebrae. The muscle bellies are very strong and have numerous tendinous intersections. The muscle fibres converge to insert on the medial aspect of the scapula (facies serrata) and the scapular cartilage. This muscle supports the trunk and carries scapula and trunk forward (König et al., 2007).

In the Asian elephant, *M. semispinalis* cervicis originates from transverse processes of the thoracic and cervical vertebrae except the atlas. The muscle fibres move anteriorly and into the dorsal tubercle of the atlas (Mori & Shindo, 1956). Veterinary anatomy references indicate that semispinal muscle of the head occupies the space between the occiput, the cervical vertebrae and the nuchal ligament, covered on its lateral aspect by the longissimus and the splenius muscles. The semispinal muscle of the head raises the head, when acting bilaterally and flexes the head and neck laterally, when acting unilaterally. The semispinal muscle raises the head when acting bilaterally and bends the head and neck laterally when acting unilaterally. In these anatomical references, there is no mention of attaching this muscle to the atlas vertebra (König et al., 2007).

Origins of the semispinalis muscles are the spinocostal transverse fascia, transverse processes of the 5th to 8th thoracic vertebrae and articular processes of the 2nd to 7th cervical vertebrae. Insertion of these muscles is the squamous part of occipital bone (König et al., 2007).

It should be noticed that a few differences can be found in the connection site of semispinalis muscle to the atlas and other cervical vertebrae. In addition, the atlas vertebra in the elephant is wide. Totally, these findings reveal some differences in the function of mentioned muscle.

Rectus capitis posterior minor muscle in the Asian elephant, which originates from the dorsal tubercle (upper of its side), is small and covered by rectus capitis posterior major muscle. This muscle fibre-bundles, which diverge upward and end under the insertion of rectus capitis posterior major muscle (Mori & Shindo, 1956). It should be noticed that another name by which rectus capitis posterior minor and major muscles are known in the veterinary anatomy references is rectus capitis dorsalis. In these references, rectus capitis dorsalis minor muscle in the domestic animals has been introduced as a short flat muscle, the origin of which is in dorsal arch of the atlas and the insertion is on the skull above the foramen magnum. Dorsal tubercle of atlas in the elephant is large and doubled.

Obliquus capitis superior muscle, which is fan-shaped in the Asian elephant, originates from the transverse process of the atlas vertebra (back of its upper part). About obliquus capitis inferior muscle, the origin is spine of axis vertebra (upper part of its side). The fibrebundles, which insert to the transverse process of the atlas (lower part of its tip), diverge and form a fusiform belly (Mori & Shindo, 1956). A noticeable point about the animals whose body faces the ground is that cranial and caudal terms are used instead of superior and posterior terms. Therefore, in the veterinary references, these tow muscles are known as obliquus capitis cranialis and obliquus capitis caudalis.

The atlas and axis vertebrae have been covered by obliguus capitis caudalis muscle, which arises from the axis (spinous process) and obliquely (craniolaterally) ends on the wing of atlas. The contraction of this muscle in one side (unilaterally) results in rotation of the atlas and the head around the dens of axis vertebra. However, bilaterally contraction of this muscle can fix the head at its position (König et al., 2007). Obliquus capitis cranialis muscle mainly originates from the wing of atlas (lateral and ventral portion) and ends on the mastoid process in the temporal bone and to the nuchal crest. This short muscle covered atlanto-occipital joint obliquely (craniolaterally), while itself is covered by the splenius muscle and some parts of the brachiocephalic muscle. Contraction of this muscle in one side extends the insertion joint and bends the head towards that side (König et al., 2007). As mentioned, the width of atlas vertebra in the elephant is relatively large, which can affect the function of these two muscles.

Rectus capitis lateralis muscle in the Asian elephant originates from the atlas (dorsal surface). About the longus cervicis muscle or longus colli as a strong muscle, the insertion is the ventral tubercle of atlas vertebra (Mori & Shindo, 1956). These two muscles can be found in the other animals to. The first one as a small muscle covers the alar fossa of the atlas in the ventral arch and ends to the jugular processes of the occipital bone. The action of rectus capitis lateralis muscle in the other animals is like the Asian elephant (König et al., 2007). About the second muscle (longus colli) in the other animals, it passes during the ventral surface of cervix from the first thoracic vertebrae to the atlas. Long muscles of the head continue this muscle cranially. The origin of this muscle in the cervical part is the transverse processes of the 3rd to 7th cervical vertebrae by the separate muscle bundles. These bundles run craniomedially to insert on the bodies of the more cranial cervical vertebrae near the midline. Thoracic part of this muscle ends to the last two cervical vertebrae up to the 6th thoracic ones on their bodies (König et al., 2007). It should be noted that for bone studies similar to ours, the CT scan is a better choice than radiography. Some of the drawbacks of radiography, such as image magnification and the fact that the image can only be taken from certain angles, since CT scans do not have these problems, make CT scans a better choice for morphometric studies (Thrall & Widmer, 2018).

In this study, the structure of the cervical vertebrae of the Asian elephant was examined, and certain features were observed. One of the main features was the reduction of the length of the vertebrae, which leads to the decrease of the ratio of neck length to the size of the body. This condition can be due to the high weight of the head in the elephant. To maintain this weight, it is necessary to reduce the length of the neck and confer less mobility. Another noteworthy point about the elephant is the width of the first cervical vertebra. The width of the atlas is approximately close to the length of the neck. Moreover, the ratio of the width of the atlas to the length of the neck is greater in elephants compared to horses and cattle.

ACKNOWLEDGEMENTS

The authors wish to express their appreciation to everyone that assists us in this study. We would like to thank Mr. Saboori and Mr. Chavoshi Poor Naeini for their efforts in maintaining the elephant bone specimens of the Faculty of Veterinary Medicine, University of Tehran for several years. Authors would like to acknowledge to fully financial support of Faculty of Veterinary Medicine, University of Tehran for this research.

CONFLICT OF INTEREST

The authors declare that they have no conflict of interest.

ETHICAL STATEMENT

This study was a DVM thesis and all experimental procedures were approved by the Faculty of Veterinary medicine, University of Tehran Local Ethics Committee (30704/6/6).

DATA AVAILABILITY STATEMENT

Data available on request from the authors.

PEER REVIEW

The peer review history for this article is available at https://publons. com/publon/10.1002/vms3.837.

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How to cite this article: Zehtabvar, O., Vajhi, A. R., Akbarein, H.-a., Ahmadian, F. S., khanamooeiashi, M., Soflaei, R., & Borgheie, F. (2022). CT Anatomy of cervical vertebrae of Asian elephant (*Elephas maximus*). *Veterinary Medicine and Science*, *8*, 1750–1768. https://doi.org/10.1002/vms3.837