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RESEARCH ARTICLE

Assessing Thoracic Symmetry in Dogs

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A B S T R A C T

This study aimed to conduct a detailed statistical shape analysis of the thorax across various dog breeds, focusing on the identification and characterization of shape variations and asymmetrical features. For this purpose, 3D images of 39 thoracic samples were collected from various regions across Türkiye, ensuring a diverse representation of breeds and populations. In the analysis, 136 specific anatomical landmarks were meticulously identified and marked from a lateral view of the thorax for each sample. The study revealed that the first principal component explained 28.89% of the total variation in directional asymmetry, suggesting consistent size or shape differences on one side of the thorax. Additionally, it was found that the first principal component explained 27.82% of the total variation in fluctuating facet asymmetry occurs if one region is consistently greater or has a different shape compared to the opposite and indicates underlying genetic or functional differences. This study highlights the usefulness of geometric morphometric analysis in distinguishing thoracic shape differences among various dog breeds, providing insights into their morphological diversity. Moreover, the findings underscore the potential of geometric morphometric analysis for taxonomic purposes, enabling more precise classification and understanding of breed-specific characteristics. This approach can aid in identifying subtle morphological variations that may be linked to breed history, function, or environmental adaptation.

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INTRODUCTION

Dogs play an important role in society, both as companions and as workers. Domestic dogs are a good example to study the functional issue of body and shape variation with great morphological diversity, and according to Kennel clubs, domestic dogs include over 400 breeds. The differences in the development of the contemporary human chest structure compared to that of non-human primates have long been a subject of interest (García-Martínez *et al.,* 2013). Schultz (1961), in his groundbreaking review, offered a classificatory framework for describing and dichotomously categorizing the chest structure of various primate species, a method that remains in use today (Franciscus and Churchill, 2002; Sawyer and Maley, 2005; Bastir *et al.,* 2015). The thoracic skeleton is a bony-cartilaginous structure that encases and safeguards the thoracic organs and supports the mechanical breathing function. To fulfill its role in respiration, the chest framework provides a broad surface for muscle attachments (intercostal muscles, diaphragm, and supplementary respiratory muscles) (De Troyer *et al.,* 2004; Ozaydın and Olgun Erdikmen, 2023). The muscles function to elevate the ribs, which enlarges thoracic dimensions due to their angulation, shape, and articulations. This leads to decreased intra-thoracic pressure and facilitates inhalation (De Troyer *et al.,* 2004). Schultz (1961), employed the term "barrel-shaped" to characterize the ribcage structure of Homo sapiens and hylobatids (gibbons and siamangs), marked by a cylindrical chest with a broader upper thoracic region (i.e., thoracic cupola) and approximately parallel lateral thoracic walls. He also described the cranially converging (conical) ribcage configuration of great apes (Pan, Gorilla, and Pongo) as "funnel-shaped," featuring a narrower cupola and caudally diverging lateral thoracic walls.

Geometric morphometry (GM) is a shape analysis method based on anatomical points, curves, and contours examination, which uses data from two or three-dimensional Cartesian coordinates (Bookstein, 1997; Aytek, 2017; Gündemir *et al.,* 2021; İlayda *et al.,* 2023; Manuta *et al.,* 2024). In recent years, with the geometric morphometry method, shape analysis of biological samples has been carried out and shape variations between groups have been revealed. There are many studies on animals related to geometric morphometry (Abuelela *et al.,* 2021; Demircioğlu *et al.,* 2021; Jashari *et al.,* 2022; Mutlu *et al.,* 2022; Ozkadif and Haligür, 2022; Gündemir *et al.,* 2023a; Hadžiomerović *et al.,* 2023). Shape asymmetry also can be examined by using the geometric morphometry method. This review summarizes concepts and morphometric methods for studying shape and size asymmetry (Klingenberg, 2015: Gündemir *et al.,* 2023b). Numerous review articles have offered summaries of the biological principles associated with allometry (Gundemir *et al.,* 2024) and the statistical techniques for allometric assessments, primarily within the framework of conventional morphometric methods (Akçasız *et al.,* 2024; Gündemir *et al.,* 2024; Ozkan *et al.,* 2024).

In numerous investigations, fluctuating asymmetry is broadly recognized as an effective indicator (biomarker) of the phenotypic reaction to environmental pressures (Benitez *et al.,* 2020). In this research, we will examine the techniques for analyzing allometry in geometric morphometrics. Consequently, the concept of fluctuating asymmetry measurement is described as minor non-directional deviations from ideal bilateral symmetry (Van Valen, 1962). In bilateral models, both sides are anticipated to share a similar genetic and environmental background, and deviations from perfect bilateral symmetry are expected to reflect the impacts of developmental noise and stability (Palmer *et al.,* 1993). Directional asymmetry, when one side is always larger than the other, or also antisymmetry, when the two sides are always different but without a predicted direction to the differences (Palmer, 2005), is genetically influenced and is thought to be unrelated to developmental stability. (Palmer, 1994). Recent research has indicated that phylogenetic and ecological factors may play a significant role in determining many allometric relationships (Nee *et al.,* 1991; Toryan *et al.,* 2024). Less is known about the differences in thorax shape among various dog breeds.

The thorax is critically important both clinically and morphologically. Considering the asymmetrical position of the heart and the symmetrical distribution of the lungs, asymmetrical development in the structure of the thorax is a plausible phenomenon. In cases of thoracic asymmetry, more detailed research could provide answers to various clinical and morphological questions, potentially leading to improved understanding and management of thoracic conditions in dogs.

The purpose of this research is to investigate thoracic asymmetry across various dog breeds by employing 3D geometric morphometrics analysis. This study aims to perform a comprehensive statistical shape examination of the thorax, focusing on the hypothesis that shape variation between the right and left sides of the thorax is influenced by the vertebral heart score. By evaluating these asymmetries, we seek to enhance our understanding of thoracic morphology and its potential implications for canine health and breeding practices. Our findings may contribute to the broader field of veterinary medicine and provide valuable insights into the structural variations among different dog breeds.

MATERIALS AND METHODS

Animals: In this study, were used 39 samples of the thorax of different dog breeds from different parts of Türkiye. In order to examine the effects of asymmetry on breed, the animals participating in the study were required to be pedigree. Mixed breeds were not used. Dogs that had completed their development and had no pathological findings were used in the study. 3D geometric morphometrics on virtual 3D thorax models obtained from computed tomography (CT) scans of living mix dog breeds. Stratovan Checkpoint was employed to encompass the thorax shape with 136 anatomical landmarks placed on five ribs on each side of the thorax. As a reference, we performed a clinical CT scan at the Istanbul University-Cerrahpasa, Faculty of Veterinary Medicine. Approval was obtained from the Local Ethics Committee of the Faculty of Veterinary Medicine, Istanbul University-Cerrahpasa (2022/38) for the study to be carried out. All images were obtained from archive images of Istanbul University-Cerrahpasa, Faculty of Veterinary Medicine, Animal Hospital. Each dog was scanned at 110 kV, 28 mA, and 0.6 mm section thickness using a Siemens (Somatom Scope vc30b) Multi-Detector Computed Tomography (Akcasiz *et al.,* 2024). The specimens were collected from adult animals; bones showing any signs of pathological abnormalities, deformities, or other damage were omitted from this research. We performed examinations on the upper ribcages of thoracic datasets, which consist of thirteen ribs. Consequently, we opted to exclude the 13th rib pair, the final set of rib pairs.

Landmarks: The landmarks we performed were done from a lateral view of the thorax (Fig. 1). Landmarks were placed from the upper border of the ribs to the lower border. The right and left sides of the thorax were landmarked in the same order. Then the landmark coordinates for each specimen were recorded by using the extension "morphologica". A total of 136, 3D landmarks were used. While placing the landmarks, the lengths of the ribs were taken into consideration. 4 to 7 landmarks were placed according to their lengths. we placed 4 LM on ribs 1-2, 5 LM on ribs 3-5, 6 LM on ribs 6-9, and 7 LM on ribs 10-12. The landmarks were placed on the outer surface of the ribs. Therefore, we positioned markers at the highest point of the rib or its head, one at the lowest point of it, one at the central point on the anterior interarticular crest, one at the central point of the shaft, one marker at the lowest point of the costal angle, and one at the lowest points of the sternal end.

Geometric analysis: For Geometric analysis, Morphoj v1.06d software program was used as described by Klingenberg, (2011). MorphoJ is a software suite for geometric morphometric assessment of two- and three-dimensional landmark data (Klingenberg *et al.,* 2002). Files in "morphological" format are opened in MorphoJ with the "symmetry" option (Boz *et al.,* 2023). Then the symmetrical landmarks were matched with each other. Grouping operations were performed on the thorax shape of the different dog breeds. Geometric morphometric analysis of morphological features by collecting a series of coordinate data.

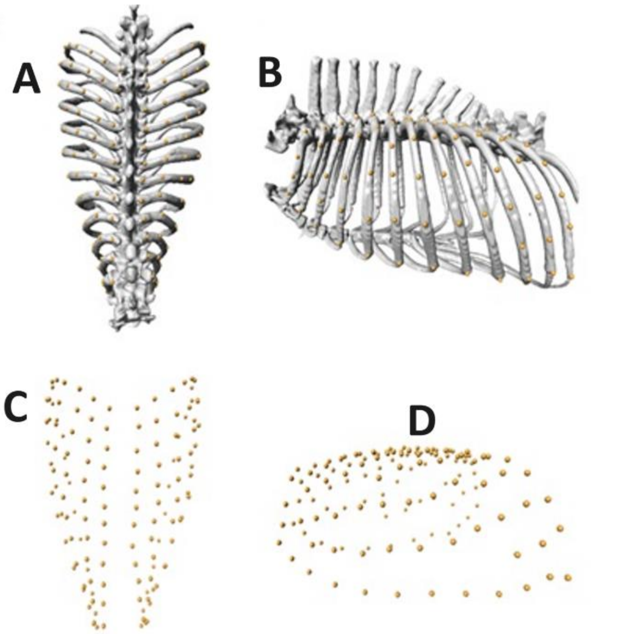

Fig. 1 3D landmarks of the grand mean shape of 12 thoracic levels and the relationship of the landmarks to rib and thorax morphology. a) Dorsal and b) lateral view with surface model. c) 3D landmarks in dorsal view and d) in left lateral view without surface.

After getting the Cartesian x,y coordinates for all landmarks were extracted the shape data by using full Procrustes fit (Rohlf and Slice, 1990), and then taking into account the symmetry of the thorax was evaluated, and Generalized Procrustes Analysis (GPA) was applied to the imported landmark data before Principal Component Analysis (PCA). This Procrustes superimposition method involves three steps: first, translating all landmark configurations to a common centroid; second, scaling all configurations to a uniform centroid size; and third, iteratively rotating all configurations to minimize the summed squared distances between the landmarks and their corresponding sample averages. In the study, a fairly large data set was obtained, and this large data set was analyzed using PCA. PCA is a dimensionality reduction method that is often used to reduce the dimensionality of large data sets, by transforming a large set of variables into a smaller one that still contains most of the information in the large set. Subsequently, Principal Component Analysis was conducted to uncover the shape variation of the thorax across all samples, where the first principal component (PC) represented the shape pattern with maximum variance in the sample and was revealed as a shape deformation, the second principal component (PC) is geometrically orthogonal or perpendicular to the first one, also accounted for the second most variance. The change of directional asymmetry according to PCA analysis is given in Fig. 2, and the shape change according to PCA analysis is given in Fig. 3. Fluctuating asymmetry Fig. 4 shows the change in PCA analysis, and Fig. 5 shows the shape change according to PCA analysis. Centroid size and shape among samples were compared with Procrustes ANOVA.

RESULTS

Principal component analysis: The shape variation between samples was analyzed by principal component analysis using 136 landmarks in 3 dimensions in different dog breeds (Table 1). Accordingly, the first principal component (PC1) explained 28.89 % of the total shape variance, PC2 22.65%, and PC3 11.87% of the total shape variance, therefore, the first three principal components (PC1 + PC2 + PC3) explained the rest of 63.41%. In this study, the following analysis was focused on PC1 and PC2. The PCA distributions revealed that the breeds did not exhibit distinct groupings. Instead, asymmetry appeared to be more of an individual characteristic than a breed-specific one. This underscores the importance of considering individual variations when studying asymmetric formation.

Table 1: Principal components for directional asymmetry and fluctuating asymmetry of the thorax.

	Directional Asymmetry		Fluctuating Asymmetry	
Components	Eigenvalues	Variance	Eigenvalues	Variance
PC1	0.00431733	28.897	0.00056850	27.829
PC2	0.00338452	22.654	0.00026625	13.034
PC3	0.00177399	11.874	0.00019209	9.404
PC4	0.00162992	10.909	0.00013821	6.766

Fig. 2 shows the directional asymmetrical changes of the samples because of PCA, while Fig. 3 illustrates how these changes affect shape according to PC1 and PC2. The same schematic for fluctuating asymmetry is presented in Fig. 4 and 5.

Wire-frame warp plots of changes in the skull shape of PC1 and PC2 in Fluctuating Asymmetry are represented in Fig. 4. For PC1, most changes to the thorax occur at the most inferior points of the sternal end. Additionally, in the dorsal view, changes are observed at the most superior point of the rib and the medial point at the anterior interarticular crest. These shape changes result in a wider thorax, which is more common in larger dog breeds. The results of linear regression are given in Table 2. The regression between both directional asymmetry and fluctuating asymmetry and shape on centroid size was not proved as significant (P value: 0.5229).

Procrustes ANOVA findings for breeds are displayed in Table 2. Despite the extent of overlap among dog breeds observed in PCA, the distinctions in centroid dimensions and morphology differences between the various dog breeds overall were statistically significant ($p<0.0001$). The ANOVA tests whether the individual variation is significantly broader than the error. It was found that significant individual variation (Mean Squares = 0.0000766307 and $p<0.0001$) which is larger than measurement error (Mean Squares = 0.0000423354). Procrustes ANOVA for dog thorax was statistically significant, showing individual variability ($p<0.05$).

DISCUSSION

In this study, the directional and fluctuating asymmetry of the thorax of different dog breeds was examined. Anthropogenic impacts over the past century have resulted in biodiversity and extinction of species (Chapin *et al.*, 2000). Through this research, we aim to investigate these impacts and uncover potential insights into their effects on canine morphology. Fluctuating asymmetry, which signifies minor developmental variations between the right and left sides, can be caused

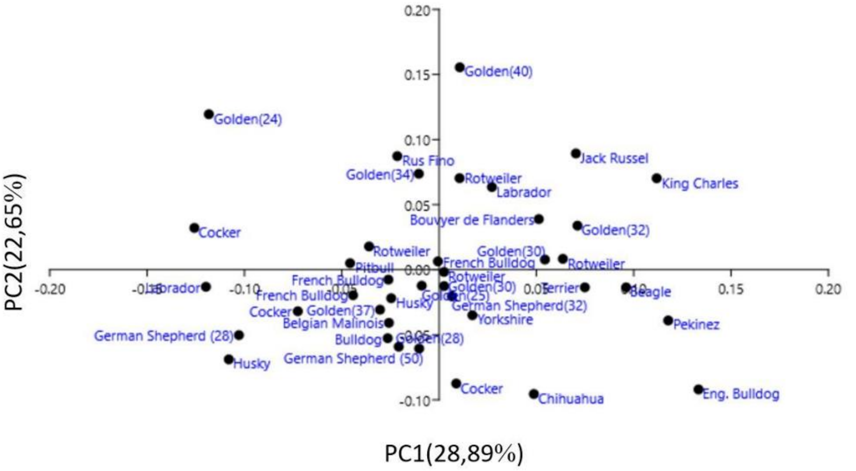

Fig. 2. Scatter plot of PC1 and PC2 for thorax data. Results of the principal component analyses performed on the directional asymmetrical component of the dog thorax. Accordingly, to PC1 and PC2, the dog thorax with the highest value is represented by King Charles, and the lower value is the Cocker dog breed.

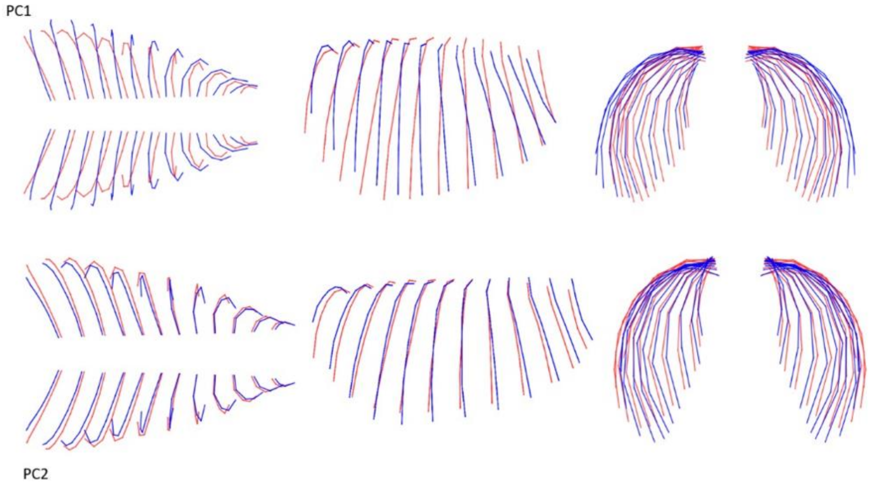

Fig. 3. Shape changing for PC1 and PC2 in dorsal, lateral, and frontal view of the dog thorax on the directional asymmetry. Wire-frame warp plots of changes in the dog thorax, as mapped by 136 landmarks in 3 dimensions. Blue outlines represent the mean shape configuration, while the red outlines show the shape changes associated with the positive extremes of the PC axes. Wire-frame warp plots of changes in the skull shape of PC1 and PC2 in Directional Asymmetry are shown in Fig. 3. For PC1, most changes to the thorax are at the most inferior point of the angulus costae and at the most inferior points of the sternal end, where is broadest, this can be related to the difference between small and bigger dog breeds.

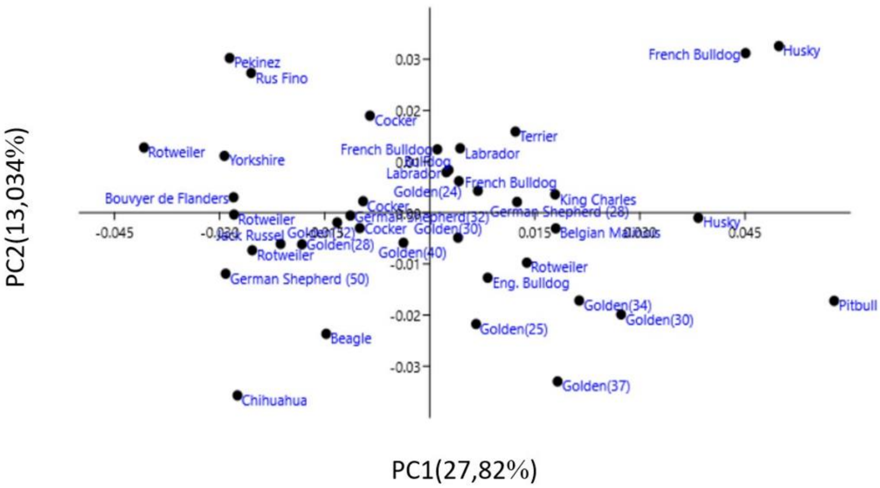

Fig. 4. Fluctuating asymmetry. Scatter plot of PC1 and PC2 for thorax data. According to PC1 PC2 values, on fluctuating asymmetry higher value has the Husky dog breed and the lowest value has Rotweiler dog breeds.

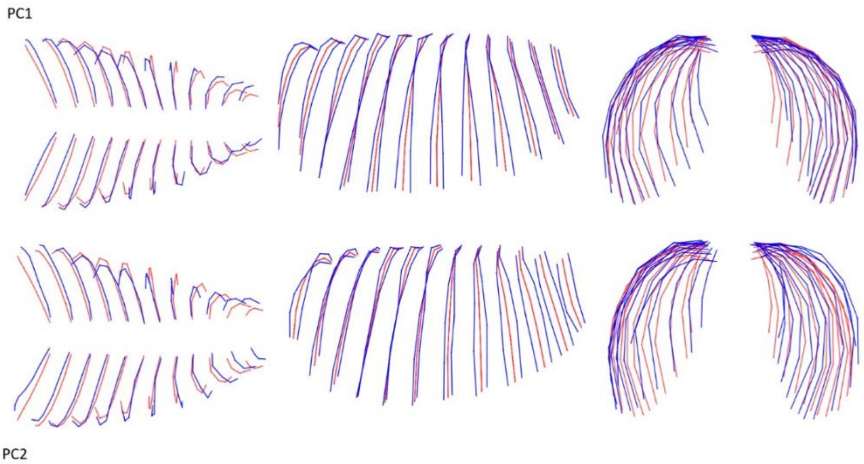

Fig. 5. Shape changing for PC1 and PC2 in dorsal, lateral, and frontal view of the dog thorax on the fluctuating asymmetry. Wire-frame warp plots of changes in the dog thorax, as mapped by 136 landmarks in 3D. Blue outlines represent the mean shape configuration, while the red outlines show the shape changes associated with the positive extremes of the PC axes.

Table 2: Results of linear regression analysis of relationships between size (centroid size) and both directional asymmetric components and fluctuating asymmetric components of the dog thorax.

	Regression	Predicted SS	Residual SS	% predicted	P-value
Dog thorax	Directional asymmetry	0.03143412	0.53630036	5.5368%	0.0390
	Fluctuating asymmetry	0.00115864	0.07646747	1.4926%	0.8999

Table 3: Shape and Centroid size, Procrustes ANOVA for the thorax of the dog breeds asymmetry: Sums of Squares (SS) and mean squares (MS) are in dimensionless units of Procrustes distance. Degrees of freedom (DF).

	Effect	SS	MS	DF	F	P
Centroid Size	Individual	14197,658576	709,882929	20	0.98	0.5229
Shape	Individual	0.30652295	0.0000766307	4000	9.26	<.0001

by chemical pollution, forest deprivation, and environmental factors (Anciaes and Marini, 2000). Gonzales-Rivas *et al.* (2023) reported in their study on reptiles that there are variations in shape among those that develop in a shaded area and are exposed to anthropogenic stresses. There is now a lot of information available indicating that external factors can produce changes in an organism's shape, including the effects of urbanization, parasitism, and contamination. In addition, some studies in reptiles reported that stress and environmental factors cause glucocorticoid increase and cause disease and pathological cases (Romero, 2004). However, it has been reported that this hormonal increase could result in asymmetry in species (Zhelev *et al.,* 2019).

In the study of directional asymmetry in Araucana horses, Parés-Casanova *et al*. (2020) demonstrated that mechanical pressures of varying intensities during mastication significantly influence the shape and internal structure of the bone. This structure is especially true for the parts where masticatory muscles are attached because bone formation and resorption processes are affected by mechanical stress factors. In our study, we observed differences in directional and fluctuating asymmetry of the thorax between species.

Based on findings from other studies, we hypothesize that the lifestyles and environmental factors associated with different regions may lead to symmetrical changes in the thorax of dogs. The PCA distributions revealed that the breeds did not exhibit distinct groupings, indicating that asymmetry is not a consistent characteristic within specific breeds. Instead, asymmetry appeared to be more of an individual characteristic than a breed-specific one. This finding underscores the importance of considering individual variations when studying asymmetric formation. It suggests that while breed characteristics might influence general morphology, the asymmetry of the thorax is largely influenced by individual factors. Therefore, future studies should focus on a more personalized approach, taking into account the unique anatomical features of each dog. One potential avenue for research is investigating the genetic basis of thoracic asymmetry. Understanding the hereditary factors that contribute to asymmetry could provide insights into its prevalence and variability among individuals. Additionally, exploring the functional consequences of thoracic asymmetry is crucial. Studies could examine how asymmetry impacts respiratory function, physical performance, and susceptibility to thoracic conditions. This perspective is essential for understanding the underlying causes of asymmetry and could have significant implications for veterinary practice, particularly in the diagnosis and treatment of thoracic conditions. Ultimately, by considering both genetic and functional aspects, we can develop more effective strategies for managing and mitigating the impacts of thoracic asymmetry in dogs. It has been proposed that deviation from Directional asymmetry in an animal group might be comparable to Fluctuating asymmetry and that there are methods to "tune" the assessed Directional asymmetry to transform it into Fluctuating asymmetry (Graham *et al.,* 1998). However, in an asymmetry that is subjected to (or recently subjected to) Directional selection, it is unclear whether a positive-signed deviation from the expected form (e.g., left larger than right) is equivalent to a negative-signed deviation (e.g., left larger than right). We can make this assumption with Fluctuating asymmetry, but with Directional asymmetry, it is much more challenging because Directional selection in asymmetry reveals a difference in reproductive success for left-sided and right-biased traits. Consequently, deviation from the average Directional asymmetry cannot generally be interpreted in the same manner as variation in fluctuating asymmetry (Leamy *et al.,* 2000). In this study, the difference between dog breeds was revealed by using directional asymmetry and fluctuating asymmetry. The similarity of directional asymmetry and fluctuating asymmetry mentioned by the researchers in their studies is not very suitable for our study. When looking at dog breeds, dogs of different breeds reveal different results according to two different types of asymmetries. In other words, the fact that the results of directional and fluctuating asymmetries can be zoomed out contradicts our study. With the knowledge that the two asymmetry types can be interpreted in different ways, our work is in the same direction.

Manning and Chamberlain (1993) studied the asymmetry of canine teeth in primates. They found that this asymmetry is associated with measures of sexual selection in dogs. Specifically, it is linked to sexual dimorphism, canine size, mass dimorphism, and intra-male competition (Selba, 2020). Visual inspection of the endocasts of brachycephalic and normocephalic dogs reveals a distinct variation in the overall shape of the endocast symmetrically. As anticipated, it has been reported that the Cephalic index was significantly associated with canine endocast morphology. In our study, asymmetry differences in the costa differ in dog breeds, as in other studies. The differences in the asymmetries of the anatomical structures can be explained by many reasons, as in the studies, the different asymmetric results of the costa in dogs are related to the weight, gender, etc. of the dogs in our study.

According to Selba (2021), brachycephalic dogs, which have a high cephalic index (CI), score low on the first principal component (PC1). In contrast, normocephalic dogs, with a low CI, score high on PC1. Gündemir (2023c) reported that in the shape analysis of the mandible of cats and dogs, the PC1 value was positive in large dogs and PC2 in most of the brachiocephalic dogs. According to Manuta *et al*. (2023), the rise in PC1 and PC2 values reflects different anatomical features. An increase in PC1 signifies a narrower acetabulum, while an increase in PC2 indicates changes in the margin of the acetabulum. In the research by Hadžiomerović (2023) involving ear ossicles, elevated PC1 was observed in the caput mallei of the malleus, while PC2 also increased in the caput mallei of the malleus. Szara *et al*. (2024) found that in their research on Japanese quails, variations in PC1, PC2, and PC3 values—both increases and decreases—across the same and different anatomical structures reveal distinct differences. Additionally, this study found that small-sized dogs were associated with positive PC1 values.

Anatomical changes were observed in the formations of the costa (ribs) in these dogs. These findings align with changes reported in anatomical structures in other studies, showing consistency with previous research. Manuta *et al*. (2023), in their research on crossbred cats, noted that the disparity between the pelvises of females and males was not statistically significant. In their study, Gündemir *et al*. (2023d) found no statistical difference in centroid size across cat species when viewed from the dorsal and lateral aspects. However, there was a pronounced statistical difference in shape between cat species in both the dorsal and lateral views. Similar and different results were obtained in this study, as in other studies. In dog breeds, while the centroid size is not statistically significant, it is quite significant in shape.

The sample size of 39 thoracic samples may pose limitations. A smaller sample size can affect the reliability and generalizability of the findings, particularly when analyzing breed-specific differences. With a limited number of samples, there is a greater potential for variability and less power to detect subtle differences between breeds. This could lead to less precise estimates of thoracic asymmetry and potentially overlook breed-specific patterns that may be present in a larger sample.

Conclusions: In this study, it was desired to reveal the differences between two different types of asymmetries of the thorax of different dog breeds and to set the differences between them as an example and useful for other studies. Working with different dog breeds will not only shed light on different studies but will also benefit the studies of other departments.

Conflict of interest: The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the study.

Data availability statement: The data that support the findings of this study can be requested from the corresponding author.

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Authors contributions: ZM, YA, and BCG conceived and supervised this study. ZM and YA collected of the data. BCA carried out statistical analysis. All authors contributed to the critical revision of the manuscript and have read and approved the final version.

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