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## The Statistical Analysis of Some Volumetric Measurements in the Japanese Quails' Head with Different Feather Color: A Computed Tomography Study

Mahmut DUYMUŞ<sup>1</sup> Günes ORMAN<sup>1</sup> Yasin DEMIRASLAN <sup>2</sup> Kadir ASLAN <sup>2</sup>

Yalçın AKBULUT <sup>3</sup> Sami OZCAN <sup>2</sup>

<sup>1</sup> Department of Radiology, Faculty of Medicine, University of Kafkas, TR-36100 Kars - TURKEY

<sup>2</sup> Department of Anatomy, Faculty of Veterinary Medicine, University of Kafkas, TR-36100 Kars - TURKEY

<sup>3</sup> Collage of Health, University of Kafkas, TR-36100 Kars - TURKEY

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#### Summary

The aim of this study was to contribute to quail head morphology and the formation of basic data sources for comparative measurement in the quail head. In this study, 60 Japanese quail (Coturnix japonica) heads, which were white, brown and wild type were used. Four different anatomical structures of the quail heads, varying in feather color, were measured manually by two independent radiologists. These anatomical structures were head volume (HV), brain volume (BV), parietooccipital air space volume (POAV) and calvarial bone volume (CBV). According to computed tomography (CT) measurements, the quail head with white feather color had the smallest volumetrical values. This condition is thought to be caused by genetic differences.

Keywords: Computed Tomography, Japanese quail, Morphometry

# Farklı Tüy Renklerine Sahip Japon Bıldırcınlarının Başlarında Bazı Hacimsel Ölçülerin İstatistiksel Analizi: Bilgisayarlı Tomografi Çalışması

### Özet

Bu çalışmanın amacı bıldırcın başlarında karşılaştırmalı ölçümler için temel veri kaynağı oluşturmak ve bıldırcın başı morfolojisine katkı sağlamaktı. Çalışmada beyaz, kahverengi ve yabani tip olmak üzere toplam 60 Japon bıldırcını başı kullanıldı. Farklı renklere göre bıldırcın başlarının dört farklı anatomik yapısı iki radyolog tarafından ölçüldü. Bu yapılar kafa hacmi, cavum cranii hacmi - beyin hacmi, parietooccipital hava alanı hacmi ve kafa kemikleri hacmi şeklindeydi. Bilgisayarlı Tomografi ölçülerine göre, beyaz tüy rengine sahip bıldırcın başınının en küçük hacimsel değerlere sahip olduğu belirlendi. Bu duruma genetik farklılıkların neden olduğu düşünülmektedir.

Anahtar sözcükler: Bilgisayarlı Tomografi, Japon Bıldırcını, Morfometri

## INTRODUCTION

The quail (Coturnix japonica) is a species categorized under Phasianidae family <sup>[1,2]</sup>. The quail is widely used as an *in vivo* <sup>[3-6]</sup> in physiological, pathological, toxicological and anatomical studies owing to its lower feed consumption, early sexual maturity and fast growth. In recent years, the increase in consumption of meat and eggs of the quail has also strengthened its economic aspect <sup>[1,2,7]</sup>.

Quail feather color is considered as a race or genetical

<sup>xxx</sup> İletişim (Correspondence)

- +90 474 2426807/5308
- yasindemiraslan@hotmail.com

feature. Differences in the Quail feather color are attributed to mutations in the color <sup>[8]</sup>. The feather colors are white <sup>[9]</sup>, brown, yellow <sup>[9,10]</sup>, wild type (gray) <sup>[11]</sup> and roux <sup>[10]</sup>.

In terms of anatomical model preparation, computed tomography (CT) is a quite useful method for reliable topographical patterns <sup>[12]</sup>. CT is used for antropomorphometrical studies for creating macroscopic and microscopic models and revealing the phenotipic differences quickly in small animals [13-17]. The detailed imaging of structures that make up the skull and the minimum degree of overlapping images provides a great advantage to the CT compared to other radiological methods <sup>[18,19]</sup>. One of the advantages of the CT is the short-term applicability <sup>[20]</sup>. Using CT, high solution images are obtained from cross-sections of head, and subsequently anatomical or patological data could be evaluated [21-24]. Three-dimensional images of anatomical structures must be accurately obtained for the zoomorphological requirements, and functional, developmental, comparative studies <sup>[25]</sup>. Additionally, the development of high resolution imaging techniques such as magnetic resonance imaging (MRI) and CT facilitated to create a small animal model and anatomical evaluations <sup>[26-30]</sup>. Volumetric measurements of small organisms or non-living structures can be evaluated via the three-dimensional images obtained from CT<sup>[25]</sup>. Three-dimensional imaging method of animal materials can be gathered into two broad categories: the first one is based on reconstruction from serial section images and the second one is based on whole - volume imaging <sup>[25]</sup>. A volume image obtained from CT consists of a stack of reconstructed cross sections around the axis of rotation [31]. Therefore, the organism could easily be evaluated in terms of the development, growth and structural differences owing to obtained images [25]. In the next step, some morphometric and volumetric measurements are practised by using these images <sup>[20]</sup>.

Roentgen and CT are diagnosis methods which are frequently referred to in veterinary practice <sup>[32,33]</sup>. CT has just begun to be used in the field of avian medicine <sup>[20]</sup>. Therefore, the evaluation and interpretation of images are possible only by becoming familiar with a good level of macro - anatomy of the region and the cross - sectional anatomy <sup>[33]</sup>.

Turbinates, sinuses, nasal cavity, and other anatomical structures can be monitored by using CT in detail in the bird head <sup>[20]</sup>. In previous studies, anatomical CT images were excessively employed in imaging the head, nasal - paranasal sinuses, cranial cavity, nasolacrimal duct system, and turbinates have been analyzed in terms of and its three dimensional reconstructed models <sup>[24,34-39]</sup> in the differences animal specieses (reptile, sea turtle, donkey, rodentia, dog and cat <sup>[32-36,39]</sup>. However, there was no a study which reveals the anatomical data of the quail head by using CT in the literature.

The aim of this study was to contribute to quail head morphology and the formation of basic data sources for comparative measurement in the quail head using CT. Additionally, in this study, using the above-mentioned characteristics of CT was purposed to determined both status of imaging quality and parameters of method in quail head and to identify whether or not the difference term some volumetrical measurements (see materials and methods section) among quail heads, varying in feather color.

### **MATERIAL and METHODS**

In this study 60 quail heads were used. A total of 20 of them were white, 20 of them were brown and 20 of them were wild type. Quails were supplied from Quail Unit in the Education, Research and Application Farm of Kafkas University.Quails were six weeks, female and average 120-160 g (white: 130±5.3 g, brown: 148±7.6 g, wild type: 140±8.2 g) live weight. Animals were grown under the same live conditions (light intensity, feeding, the cage state, water etc.). Quails were killed by slaughtering for protect to wastable quilty of carcase and their heads were referred to the Research and Application Hospital Radiology Unit of Kafkas University to obtain CT images. To scan the heads 64 sliced CT (0.05 mm section) device (Aquilion 64<sup>®</sup>, Toshiba Medical Systems, 2011, Zoetermeer. The Netherlands) was used.

Four different anatomical structures of each group were measured manually by two independent radiologists with more than five-years experience. Radiologists were defined as observer 1 and observer 2. These anatomical structures were head volume (HV), brain volume (BV) (*Fig. 1-D*), parietooccipital air space volume (diverticula from recessus tympanicus dorsalis invade the occipital, parietal, prootic and squamosal bones <sup>[40]</sup>) (POAV) (*Fig. 1-A, Fig. 1-B, Fig. 2-F*) and calvarial bone volume (CBV). Aquarius iNtuition Edition ver. 4. 4. 6. software was used for measurements.

Different tissues have different densities in CT, so four different density ranges were used to calculate four different tissue volumes. To calculate the POAV from -1.000 to -850 Hounsfield unit (HU), HV 0-1000 HU, BV 0-150 HU and CBV 200-1000 HU ranges were <sup>[41]</sup> setted. Both 2D axial and coronal plane images were used to calculate all the volumes. In addition to 2D images, measurements were verified also on 3D volume rendered images. For the volume unit, cm<sup>3</sup> were used and all the data were recorded carefully.

To calculate the BV and POAV region growing tool (*Fig.* 1-A, *Fig.* 1-D), CBV and HV single click tool (*Fig.* 1-C) was used. Because BV and POAV are spread to area, but CBV and HV are not.

Four different anatomical structure of three kind of quail heads were measured by two radiologists and many different parameters obtained. To proccess the findings easily, we named the variables as: head volume of observer 1 (HV1), head volume of observer 2 (HV2), brain volume of observer 1 (BV1), brain volume of observer 2 (BV2), parietooccipital air space volume of observer 1 (POAV1), parietooccipital air space volume of observer 2 (POAV2), calvarial bone volume of observer 1 (CBV1), calvarial bone volume of observer 2 (CBV2).

#### **Statistical Analysis**

Statistical analysis of the study was performed using

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aspect image of parietooccipital air space volume of quail head measured by region groving tool, B- 2D axial slice of parietooccipital air space volume of quail head measured by region groving tool, C- 3D volume rendered dorso-ventral aspect image of head volume of quail measured by single click tool, D- 3D volume rendered dorsoventral aspect image of brain volume of quail head measured by region groving tool

Şekil 1. A- Bıldırcın başındaki parietooccipital hava alanı hacminin bölge büyütme aracı kullanılarakölcümünündorso-ventralyönden 3B görüntüsü, B- Bıldırcın başındaki parietooccipital hava alanı hacminin bölge büyütme aracı kullanılarak ölçümünün 2B eksen kesit görüntüsü, C- Bıldırcın kafa hacminin tek tık aracı kullanılarak ölçümünün dorsoventral yönden 3B görüntüsü, D- Bıldırcın başındaki beyin hacminin bölge büyütme aracı kullanılarak ölçümünün dorso-ventral

Fig 2. Dorso - ventral view of transversal section through of the quail head. A- brain, B- cerebellum, C- occipital bone, D- oculus, E- rostrum, F- parietooccipital air space

Şekil 2. Bıldırcın kafasının transversal kesitinin dorso - ventral görünüşü. A- beyin, B- beyincik, C- occipital kemik, D- göz, E- gaga, F- parietooccipital hava alanı



Statistical Package for Social Sciences (SPSS) version 17.0 software package. Continuous variables were expressed as arithmetical mean±standard deviation.

Paired sample t test was used to determine the differences of the measurements of BV, HV, POAV and CBV between observer 1 and observer 2 regardless of the quail types and the groups were dependent according to observers.

Kruskal Wallis test used to calculate the differences of BV, POAV, CBV and HV of white, brown and wild type subgroups for observer 1 and 2 independently. Firstly three groups were analyzed, secondly white-brown, white-wild

and brown-wild groups were analyzed in pairs to find out the differences between the groups.

## RESULTS

According to the findings of the study, average values of the BV, POAV, CBV and HV of the quails (n:60) were 0.43, 0.23, 1.93 and 6.64 cm<sup>3</sup>, respectively.

According to the observers, the average results obtained from the study showed in *Table 1*. In this table, it seems that values of observer 1 are a little more than the values of observer 2.

Table 2 indicated that each volume of three groups of quail heads showed significant difference between each other, except BV2. Moreover, the same table showed significant difference between white and brown quail head measurements except BV2 and CBV2. Additionally, *Table 2* showed significant difference between wild type and brown quail head measurements except POAV1, BV2 and POAV2, and significant difference between white and wild type quail head measurements except BV2 and POAV2 volumes (P<0.05). This results indicated that approximately all of the four measurements of the three kind of quail heads were different from each other.

Paired samples t test demonstrates that there were no significant difference in CBV and HV measurements and there were significant difference in BV and POAV between the observers (P<0.05).

### DISCUSSION

One should consider the limitations of this study before using these data. We have firstly reported the volumetric measurements of head in the quail by using CT. Thus, we were not able to discuss the variation of morphometrical properties of the bird head. While this study provides new morphometrical data for four volumetric measurement in the quail head, volumetric morphometrical values for other species, such as the chicken, goose, and pigeon, are also needed. When we performed this study, there was not a method established in the birds. Therefore, we could not compare in terms of method.

According to the findings of the study, average values of the BV, POAV, CBV and HV of the quail (n:60) were 0.43, 0.23, 1.93 and 6.64 cm<sup>3</sup>, respectively. According to this situation, BV was forming %6.47 of HV, POAV was forming %3.46 of HV, and CBV was forming %29.07 of HV.

BV and POAV differences were due to the subjective results of the region growing tool and method. CBV and HV anology were due to the single click method based on our results. Single click is a useful tool that allows measuring the volume by clicking only once to calculate the volume. However, region growing is time consuming and relatively difficult. In this regard, the results of single click are more objective than region growing method.

In this study, feather color having the race property for quails was primary exit point. Therefore, the heads of quails

Table 1. Shows all the data of each groups based on observers											
Table 1. Araştırıcılara göre grupların verilerinin tamamı görünmektedir											
Measurement		Observer 1		Observer 2							
	White	Brown	Wild	White	Brown	Wild					
BV (region growing)	0.45±0.07	0.56±0.07	0.50±0.05	0.36±0.67	0.37±0.05	0.34±0.10					
POAV (region growing)	0.21±0.07	0.33±0.07	0.29±0.04	0.15±0.05	0.19±0.06	0.20±0,19					
CBV (single click)	1.56±0.27	1.79±0.32	2.49±0.29	1.63±0.26	1.79±0.32	2.35±0.36					
HV (single click)	5.92±0.41	6.60±0.65	7.50±0.65	5.88±0.34	6.50±0.81	7.45±0.68					
The second											

The unit of the mean values and the standard deviations are cm<sup>3</sup>

<b>Table 2.</b> Kruskal Wallis Test of the white, brown and wild type subgroups (W-White, B-Brown, WT-Wild Type) <b>Table 2.</b> Beyaz, kahverengi ve yabani tipin Kruskal Wallis Testi (W-Beyaz, B-Kahverengi, WT-Yabani Tip)											
Test	HV1	BV1	POAV1	CBV1	HV2	BV2	POAV2	CBV2			
Chi-Square (W, B&WT)	*33.75	*22.1	*19.71	*36.83	*32.76	2.84	*7.07	*26.63			
Chi-Square (W&B)	*11.71	*18.62	*15.81	*6.4	*10.11	0.27	*6.94	3.05			
Chi-Square (WT&B)	*13.14	*9.51	3.59	*22.55	*11.35	2.64	3.64	*16.15			
Chi-Square (W&WT)	*26.56	*5.10	*10.36	*27.27	*27.83	1.32	0.05	*20.91			
* Means within a row are significantly different (P<0.05)											

in three different feather colors were used. There are many quails with different feather colors in the world. Some of them are white, brown, roux, yellow and gray or wild type <sup>[8-11]</sup>. In this study, different races of quails were used to compare morphological and morphometric differences. Thus, both a method for subsequent studies and a data base were formed.

CT measurements shows that the wild type quail has the biggest head and then brown and white quails were ranking lower, respectively. The brown quail has the biggest cranial cavity volume and then it is followed by wild type and white had smallest. Moreover, the wild type quail has the biggest head bone volume and then brown and white quails have respectively. According to these results, quail race with white feather color had the smallest volumetrical values. Although more compherensive studies are needed to determine the cause of the above mentioned situation, we could speculate that the applied selection pressure in the quail unit and genetic differences can be effective.

When data of head morphometry in the quails are evaluated study material is provided from single sex and equal number of members. Female quails constituted the animal material of this study. Hence, the data obtained from female quails was analyzed.

The lack of previous CT based studies in the avian literature hampered the comparisions. For this reason, the results that we have reported here have potential to contribute not only to the assessments of species but also the comparative and topographic anatomy of birds.

In conclusion, this study showed us that CT can be used for morphological and morphometric studies in quail head, additionally birds and differences term some volumetrical measurements among quail heads, varying in feather color. Furthermore, we concluded that more compherensive studies are necessary to determine the exact relationship between feather color (or race) and some morphometric measurements in the Japanese quails.

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