Evaluation of the Volume Ratios of the Goose and Duck Liver Components by Stereological Methods

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Abstract: The morphologic and stereological studies are basic for comparative anatomical and experimental studies. The stereology is the best and accurate way to get information about the health status of an organ. On the other hand, the volume estimation is an important criterion for the health sciences. In this manner, this study aimed to get detailed morphologic knowledge about the liver tissue of winged through using stereologic methods. Liver tissue is examined two parts as parenchyma (hepatosit) and stroma (non-hepatosit). The stroma consisted of artery, vein, connective tissue and bile duct. Volume rates of these divisions are measured by point counting as stereological and indicated as percentage value. In this study, it was determined that the parenchyma tissue composed 89.3% of liver volume. Artery, vein, connective tissue and bile duct formed 1.5%, 60.4%, 35.1% and 3.0% of the stroma respectively. Consequently, the obtained values will be reference to anatomical and pathological researchers to be made about liver tissue in poultry.
Keywords: *Goose, duck, comparative anatomy, liver volume*

Stereolojik Yöntemlerle Kaz ve Ördek Karaciğer Bileşenlerinin Hacim Oranlarının Değerlendirilmesi

Özet: Morfolojik ve stereolojik çalışmalar karşılaştırmalı anatomik ve patolojik araştırmalar için temel çalışmalardır. Stereoloji, bir organın sağlık durumu ile ilgili bilgi almak için en iyi ve en doğru yoldur. Diğer yandan hacim hesaplaması, sağlık bilimlerinde önemli bir kriterdir. Çalışmada, ilk kez stereolojik metotlar kullanılarak kanatlı karaciğer dokusu ile ilgili ayrıntılı morfolojik bilgilerin elde edilmesi amaçlanmıştır. Karaciğer dokusu paranşim (hepatosit) ve stroma (non-hepatosit) olarak iki bölümde incelenir. Stroma arter, ven, konnektif doku ve safra kanalından oluşur. Bu bölümlerin hacim oranları nokta sayımı ile stereolojik olarak ölçülür ve yüzde değerler olarak ifade edilir. Çalışmada karaciğer hacminin %89.3'ünün paranşim dokusundan oluştuğu belirlendi. Arter, ven, konnektif doku ve safra kanalı sırasıyla stromanın %1.5, %60.4, %35.1 ve %3.0'ünü oluşturuyordu. Sonuç olarak elde edilen değerler kanatlılarda karaciğer dokusu ile ilgili yapılacak anatomik ve patolojik çalışmalara referans olacaktır.
Anahtar Kelimeler: *Kaz, ördek, karşılaştırmalı anatomi, karaciğer hacmi*

Introduction

The volume density of the structures in the liver like parenchyma, stroma and the subcomponents of the stroma could be measured by the stereological methods. These kinds of measurement methods are noted as simple and accurate, thus could contribute to the understanding of the organ structure. Practically, a regularly spaced point grid is placed randomly on the image on random sections. The required volumetric data is obtained by counting of the points lying on the interested structures (Aire, 1978).

The liver is a vital organ and in addition to its vital functions for the body, it is responsible for secreting the bile for absorption of dietary fats, processing the drugs and hormones, storing the glycogen, certain vitamins and minerals, phagocytizing the blood cells and some bacterias, activating the vitamin D and an active role on the carbohydrate, lipid and protein metabolisms (Altunkaynak and Ozbek, 2009). The impairment of the parenchyma density in the liver may be sign of some illness such as hepatic cirrhosis (Burity et al., 2004). Fatty diets may affect the parenchyma and cause the chronic liver disorders resulted with the carcinoma (Carrière et al., 1980). Some of the avian illnesses may influence the liver like marek, fowl typhoid, tuberculosis, aflatoxicosis and mycotoxicosis. The stereological and comparative research aimed to get more information about the organ hence facilitated the diagnosis and treatment of the illness. In the related literature stereological studies about the avian are few (Rohr et al., 1976; Rocha, 1996; Casotti and Braun, 2000;

Sahin et al., 2001; Karagoz and Haktanır, 2004). Moreover, detailed information about the parenchyma, stroma, bile duct, artery, vena and connective tissue would be crucial to understand the avian liver tissue in dept. Therefore, we aimed to measure several parameters forming the liver tissue native goose and duck, establish quantitative data and facilitate the future anatomical, pathological and clinical studies.

Materials and Methods

This study was performed on six male native geese and ducks' liver tissue. All animals were adult and about two years old, averagely 2.3 kg living weight (geese: 2.5±0.21 kg, ducks: 2.2±0.35 kg). Geese and ducks livers were supplied from Avian Unit in the Education, Research and Application Farm of Kafkas University and immediately fixed by perfusion with the 10% neutral buffered formalin. The health status of livers was determined normal by physical and histopathological examination. The tissue fragments were taken from the liver systematically. All fragments were processed histologically and embedded into the paraffin. The paraffin blocks were sectioned into 6 µm thickness and taken onto glass slides and stained with haematoxylin. All glass slides were replaced under the light microscopes' 40x objective magnification and the image was reflected to the monitor of the computer.

Firstly, the liver tissue was divided into parenchyma (hepatocytes) and stroma (non-hepatocytes). The stroma involved of four different structures; artery, vein, connective tissue, and bile duct. The volume densities of all structures were determined by point counting. The point grid, M42 testing system was attached to the monitor and forty-five systematic random fields were evaluated for each specimen. The following formula of (Burity et al., 2004) was used for the calculation:

$$V_v = \frac{Pstructure}{Ptest} \%$$

The Pstructure symbolizes the number of points fall on the relevant structure and Ptest denotes the total number of the test points. Mann-Whitney *U*-test embedded in the Statistical Package for Social Sciences (SPSS) version 17.0 was used for the statistical analysis (mean, standard error and to compare two species).

Results

The parenchyma and bile ducts, venous and arterial vessels and the connective tissue in the stroma were clearly distinguished (Fig. 1).

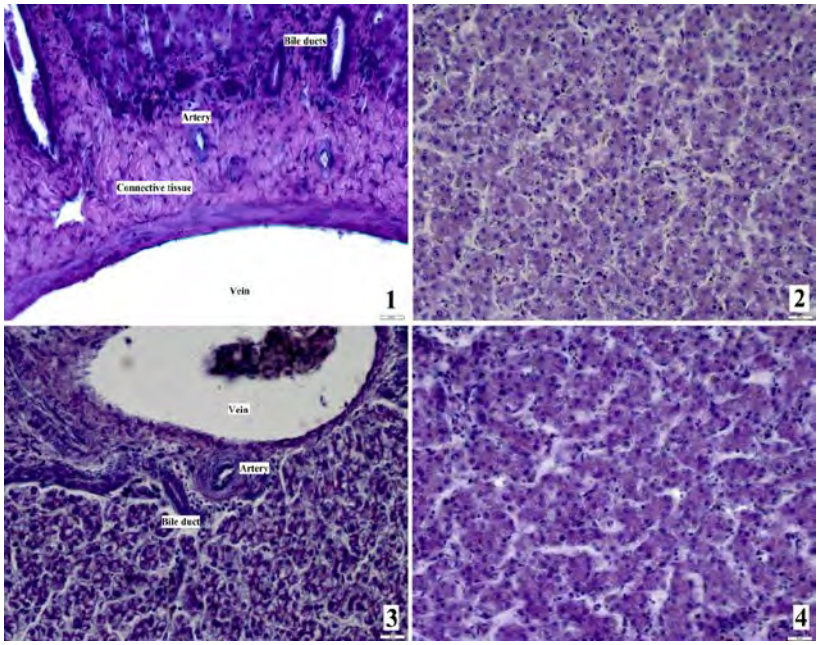

Figure 1. Overview of the livers under 40x objective: 1 and 2 the liver tissue of the goose, 3 and 4 the liver tissue of the duck.

The parenchyma was restraining the general tissue of the liver and the stroma forms of residual part. In the stroma, the highest percentage corresponded to the venous vessels and followed by the connective tissue, bile ducts, and the arteries. The mean values of both species for the

parenchyma, stroma, artery, vein, bile ducts, and connective tissue are 89.3, 10.7, 1.5, 60.4, 3.0, 35.1% respectively. The parameters and comparisons between species are showed in the Table 1. The volume densities of the liver components of both species were not statistically significant (*P*>0.05).

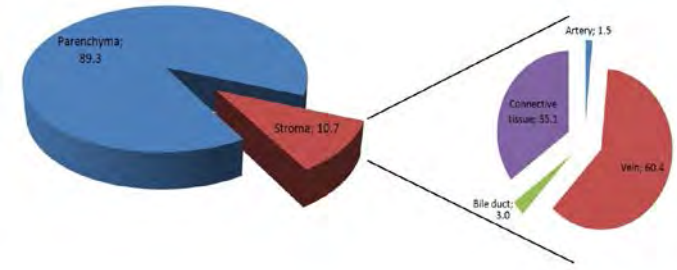

Figure 2. The graph represents the mean value of liver components in poultry.

Table 1. The stereological values of the liver components.

Stereological values	Goose (n = 6)	Duck (n = 6)
Liver		
$V_{v[parenchyma]}$ (%)	89.1±1.3	89.5±3.4
$V_{v[stroma]}$ (%)	10.9±1.3	10.5±3.4
Stroma		
$V_{v[arterial\ vessels]}$ (%)	1.10±2.1	1.80±4.0
$V_{v[venous\ vessels]}$ (%)	57.6±7.2	63.3±6.3
$V_{v[ducts]}$ (%)	2.90±1.8	3.10±2.3
$V_{v[connective\ tissue]}$ (%)	38.4±6.6	31.8±5.6

The compared parameters concerning to the species were not statistically significant (P>0.05). Data are expressed as mean ± standard error of the mean.

Discussion

Gaining more information about the tissue has beneficial effects for determining the healthy or illness conditions. The volume density of the liver tissue components was previously studied extensively. In humans, parenchyma and the stroma corresponded to the 79.3 and 20.7% of the liver tissue. This density was measured from the liver biopsies (Tortora and Derrickson, 2012). It is clearly seen that the stroma of the human is nearly twice more than the avian stroma. It is known that the parenchyma consists the largest part (about 95%) of the trout liver. The remaining part is made of the stroma.

The veins are the largest part in the stroma with nearly 76% of the trout while the bile ducts has more volumetric density (17%) and the connective tissue level albeit lower than the other animals (arteries and the connective tissue composed of the just 6% part of the stroma) (Warui and King, 1985). Parenchyma consists the 84.9% part of the liver in baboons (Burity et al., 2004). This value found to be smaller in the present study. In another comparative study on three species of primates, the volume density of the parenchyma and stroma, beside stroma components (bile duct, connective tissue, arterial and venous vessels) were evaluated (Weibel et al., 1966). It was reported that the parenchyma corresponded to the 92.6% part of the liver. In the stroma, the venous vessels have the greatest percentage and the connective tissue 37%, bile ducts 3.8%, and arterial vessels 1.6%.

The findings of above mentioned study is in agreement with the findings presented in our study and both results deviate from the data reported in other animal groups. To sum up, quantitative values of the liver structures on the avian were investigated to the extent of our knowledge for the first time.

The volume densities of the structures, composing the livers, were measured and evaluated. The volume densities of the liver components of the avian were found to be relatively closer to those reported in primates. The parenchyma and stroma volume density were found to be different from fishes and humans.

Authors believed that these results would ultimately contribute to future research on the anatomical and pathological studies.

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