Comparative Imaging Anatomical Study of the Heart and Selected Mediastinal Vessels in the Rabbit (Oryctolagus cuniculus)

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ABSTRACT

The aim of the study was to prove analogy of the results from ultrasonographic, computed tomographic and post mortem transverse study of the rabbit heart and select mediastinal vessels. Ten sexually mature, healthy New Zealand White rabbits, aged 12 months, with a body weight of 2.8 kg to 3.2 kg were investigated. Two - dimensional transthoracic echocardiography was performed in right and left lateral recumbency. The transducer was placed on the thorax for imaging the heart in standard planes (short and long axis). Transverse computed tomography of the thorax was carried out before and after intravenous contrast administration. The animals were positioned in ventrodorsal recumbency. The post mortem transverse frozen cuts of the thorax were 10 mm thick. By the ultrasonographic study the centrally situated hypoechoic lumen of the ascending aorta was found. The hypoechoic left and right atria (proventricles), parts of the right ventricle and pulmonary ostium with the pulmonary valve were visualized peripherally. The entire heart silhouette was observed via computed tomography. The atrioventricular septum was seen as a hypo attenuating structure. The heart ventricles, atria, ascending and descending aorta, esophagus and trachea were visualized. The four heart cavities and major vessels were marked by the post mortem transverse frozen study. The comparative analysis of the data from the ultrasonographic, computed tomographic and post mortem transverse frozen study of the rabbit heart and its mediastinal vessels showed that the results could be applied in the interpretation and diagnosis of the heart and vascular lesions in this species.

Key Words: Ultrasonography, computed tomography, cadaver anatomy, heart, rabbit

ÖZET

TAVŞANLARDA KALP VE SEÇİLMİŞ MEDIASTINAL DAMARLARIN KARŞILAŞTIRMALI GÖRÜNTÜLEMELİ ANATOMİK ARAŞTIRMASI

Çalışmanın amacı, tavşan kalbinin ve seçilmiş mediastinal venaların ultrasonografik, bilgisayarlı tomografik ve post mortem enine araştırma sonuçlarının benzerliğinin ortaya konulmasıdır. 10 adet, 12 aylık yetişkin, vücut ağırlığı 2,8-3,8 kg olan sağlıklı beyaz Yeni Zelanda tavşanı incelenmiştir. İki boyutlu transtorasik ekokardiyografi, sağ ve sola lateral yatırmak suretiyle gerçekleştirilmiştir. Transducer toraksa, standart düzlemde kalbin görüntülenmesi amacıyla yerleştirilmiştir. Toraksın enine bilgisayarlı tomografisi intravenöz kontrast uygulaması öncesi ve sonrasında gerçekleştirilmiştir. Hayvanlar ventrodorsal konumda yatırılmıştır. Toraks, post mortem enine dondurulmuş olarak 10 mm kalınlığında kesilmiştir. Ultrasonografik çalışmayla aort çıkışında, merkezi bir konumda yer alan hipoekoik lümen bulunmuştur. Hypoekoik sol ve sağ atriyumlar (proventriküller), sağ ventrikül ve pulmoner ostium ile pulmoner kapak kısımları periferik olarak görüntülenmiştir. Tüm kalp silüeti bilgisayarlı tomografi ile gözlenmiştir. Atriyoventriküler septum bir hipo azaltıcı yapı olarak görülmüştür. Kalp ventrikülleri, atriumlar, aort çıkışı ve girişi, esophagus ve trachea görüntülenmiştir. Post mortem enine dondurma çalışması ile dört kalp boşluğu ve büyük damarlar işaretlenmiştir. Çalışmada elde edilen veriler, tavşan kalbi ve mediastinal damarlarının ultrasonografik, bilgisayarlı tomografik ve post mortem enine dondurma çalışmasına ait verilerinin karşılaştırmalı analizi sonuçlarının, bu türlerde kalp ve damar lezyonlarının teşhisi ve yorumlanmasında uygulanabilir olduğunu göstermiştir.

Anahtar Kelimeler: Ultrasonografi, bilgisayarlı tomografi, kadavra anatomisi, kalp, tavşan

Introduction

The rabbit heart's shape is oval, and its apex is more rounded than that of the carnivores. The rabbit heart is positioned cranially (from second to fifth rib), compared to other domestic mammals. There are two cranial venae cavae and one caudal. There are two pulmonary veins - left and right. The ascending aorta begins from the level of the fifth rib. The right azygos vein enters the right cranial vena cava at the level of the third rib (Barone, 1996; McCracken et al., 2008).

Many authors (Thomas et al., 1993) recommend planes for two dimensional transthoracic echocardiography of the carnivores' heart. They have recommended three positions of the transducer: right parasternal, left cranial parasternal and left caudal (apical) parasternal.

The rabbit pericardial adipose tissue has been identified and measured by Yonkova et al. (2010). The authors have suggested using rabbit as a model for study the human adipose lesions.

A lot of authors (Falkner et al., 2005) perform transthoracic echocardiographic research of sheep aortic ring regarding its preopearive screening before experimental implanting of aortic valve prosthesis.

A comparative anatomical and echocardiographic analysis of the human aortic and tricuspidal valve is carried out by Wang et al. (1992), Espaniola-Zavaleta et al. (2006) and Muñoz-Castellanos et al. (2009). The authors have proved the correspondence between the objects, studied by anatomical and ultrasonographic methods. Many researchers (Vladova et al., 2005) have made a comparative ultrasonographic, computed tomographic and transversal anatomical post mortem frozen study of the feline heart. The group have compared the results, obtained by all the used methods and have proved the correlation between them.

Comparative computed tomographic and anatomical investigations of the feline and canine thoracic organs, performed by Samii et al. (1998) and De Rycke et al. (2005) has demonstrated complete correspondence between the observed anatomical findings.

Similar studies have been done with goats by Alsafy (2008). The author has obtained transverse computed tomographic cuts of the thorax and frozen transverse post mortem sections, 10 mm thick, from the sternal manubrium to the xyphoid process. The author (Alsafy, 2008) has compared the soft tissue and bone findings and proved analogy between the results.

The rabbit heart has been studied via computed tomography to be used as an animal model for the human atherosclerotic research (Zhao et al., 2011).

The rabbit thoracic cavity has been investigated in sternal recumbency by Zotti et al. (2009) using computed tomography and frozen cadavers. Tomographic transversal slices' thickness was 5 mm and this of the frozen cuts - 10 mm. The authors have made a comparative analysis of the corresponding structures.

Our aim is to prove analogy of the results from ultrasonographic, computed tomographic and post mortem transverse study of the rabbit heart as well as some selected mediastinal vessels. The data could be used as a suitable morphological standard for interpretation of the cardiovascular lesions in the animals.

Materials and Methods

Object

Ten sexually mature and healthy New Zealand White rabbits, aged 12 months with a body weight of 2.8 kg to 3.2 kg were used. The animals were anesthetized with 15 mg/kg IM Zoletil[®] 50 (tiletamine hydrochloride 125 mg and zolazepam hydrochloride 125 mg in 5 mL sterile isotonic solution) Virbac, France.

The study was approved by the institutional committee of animal care (Approval № 25, published in Government Gazette, № 59, 2003). The experiments were made in strict compliance with European convention for protection. vertebrate animals' used for experimental and other scientific purposes (Starsbourg /16th May 1986), European convention for companion animals' protection (Starsbourg /13th November 1987) and animal protection's law in Republic of Bulgaria (section IV-Experiments with animals, art. 26, 27 and 28, received on 24th January 2008 and published in Government Gazette, № 13, 2008).

Echocardiography

The ultrasonographic transthoracic two dimensional study was performed with CHISON 600 VET (China) ultrasonic equipment, a 7 MHz multi frequent microconvex transducer C20605 with front length 20 mm. The findings were documented on Mitsubishi P91E printing device. The two dimensional echocardiography of the rabbit heart was done in left recumbency and right lateral position. The transducer was put on the following places for visualizing of some standard planes from the heart image:

1. Left parasternal plane for imaging the short axis of the heart, showing the aortic valve, left and right ventricle on different levels etc. (the transducer was inclined consecutively at the third and fourth intercostal spaces, close to the

sternum) (Tomov and Naumov, 1992; Thomas et al., 1993).

2. Right parasternal plane for imaging the longitudinal axis of the heart, showing the four heart cavities, parts of them etc. (the transducer was inclined consecutively at the third and fourth intercostal spaces, laterally to the sternum) (Tomov and Naumov, 1992; Thomas et al., 1993).

3. Apical plane (left caudal parasternal) showing the heart cavities (the transducer was inclined in the fifth left intercostal space) (Tomov and Naumov, 1992; Thomas et al., 1993).

Computed tomography

The bodies of the thoracic vertebrae were used as bone markers for the computed tomographic study. The experimented animals were positioned in ventrodorsal recumbency. As a contrast medium of the vascular structures, **OPTIRAY**[®] 350 (ioversol 741 mg/mL, Healthcare Ltd., UK was used. It was applied parenterally (1 mL/kg, I V) in the cephalic vein. The study was performed via axial computed tomography SIEMENS, SOMATOM, ARTX with 125 cm table height, FOV=250, filter 1, anode intensity 70 mA, anode tension 110 kV and scanning time 3 sec. A high resolution -512 and gantry (GT)-0°, window width (W) 280H and center (C) + 33H were employed. Scans of the thoracic cavity were obtained with 3 mm slice thickness.

Post mortem transverse frozen study

Five animals were euthanized with 150 mg Thiopental[®] (50 mg/kg, IV) (thiopental sodium 1000 mg) Biochemie, Austria (Posner and Burns, 2009). The manipulation was complied with the requirements of the American Veterinary Association on euthanasia. The cadavers were frozen at - 20°C (Alsafy, 2008; Zotti et al., 2009). The thoraces were then cut transversely into 10 mm thick slices. They were used for comparing the ultrasonographic and features tomographic computed of the investigated structures (Zotti et al., 2009).

Results

At the ultrasonographic study of the heart in the left parasternal plane in its short axis the transverse image of the centrally situated ascending aorta's hypoechoic lumen was found. The hypoechoic left and right atria, parts of the right ventricle and pulmonary ostium and valve were visualized peripherally (Figure 1). The walls of the heart cavities were hyperechoic, compared to their lumen. The pulmonary trunk's origin was observed to the left of the aorta and to the right – the hypoechoic parts of the right ventricle and atria. The left atrium was observed behind the ascending aorta. The tricuspid and pulmonary valves were identified (Figure 1).



- Figure 1. Left parasternal plane of the heart in short axis orientation: ascending aorta (ao), pulmonary ostium (op), pulmonary valve (vp), right ventricle (vd), tricuspidal valve (vtc), right atrium (ad), left atrium (as).
- Şekil 1. Kısa eksen yönünde kalbin sol parasternal düzlemi: aorta çıkışı (ao), pulmoner ostium (op), pulmoner kapak (vp), sağ ventrikül (vd), triküspital kapak (vtc), sağ atrium (ad), sol atrium (as).

When imaging in the right parasternal plane in the longitudinal axis, the aortic valve and ascending aorta's origin were visualized as a centrally situated hyperechoic structure, surrounded by the hypoechoic left and right atria, parts of the right ventricle, origin of the pulmonary trunk and pulmonary valve. The pulmonary valve was observed as a hyperechoic structure in front of the aortic beginning (Figure 2).



- Figure 2. Right parasternal plane of the heart in long axis orientation: ascending aorta (ao), pulmonary ostium (op), pulmonary valve (vp), pulmonary artery (ap), right ventricle (vd), right atrium (ad), left atrium (as).
- Şekil 2. Uzun eksen yönünde kalbin sağ parasternal düzlemi: aorta çıkışı (ao), pulmoner ostium (op), pulmoner kapak (vp), plumoner arter (ap), sağ ventrikül (vd),), sağ atrium (ad), sol atrium (as).

In the apical plane heart study, the hypoechoic cavity of the left ventricle was found, surrounded by its hyperechoic myocardium (Figure 3).





Şekil 3. Sol ventrikülün apeks apical düzlemi: miyokard (mc), sol ventriküler boşluk (vs).

With computed tomographic study of the thorax in the transverse plane through the fourth vertebral body, parts of the heart silhouette were visualized with portions from the left and right atria, right ventricle and hyper attenuating ascending aorta, situated between them. The lungs' apical lobes were observed laterally and between them were both cranial venae cavae. The trachea was visualized centrally (with hypo attenuating lumen and hyper attenuating wall). Dorsally to the last were the arch of azygos vein (to the right) and the esophagus (to the left). Left and ventrally to the fourth thoracic verterba, the hyper attenuating aortic arch was visible (Figure 4).



- Figure 4. Transverse computed tomographic image of the rabbit thorax through the fourth thoracic vertebra (th4) (right side - R, left side - L): sternum (s), rib (r), scapula (sc), right ventricle (vd), right atrium (ad), left atrium (as), azygos vein (vz), ascending aorta (aa), aortic arch (ao), right cranial vena cava (vcd), left cranial vena cava (vcs), right lung (pd), left lung (ps), trachea (t), esophagus (o). (window width (W) - 280H; center (C) + 33H)
- Şekil 4. Tavşanın toraksı ile dördüncü torasik vertebrasının (th4) enine bilgisayarlı tomografik görüntüsü (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sağ atrium (ad), sol atrium (as), azigos damar (vz), aorta çıkışı (aa), aortik kemer (ao), sağ cranial vena cava (vcd), sol cranial vena cava (vcs), sağ akciğer (pd), sol akciğer (ps), trachea (t), esophagus (o). (pencere genişliği (W) – 280H; merkez (C) +33H)

At the level of the fifth thoracic vertebra, the heart was incompletely visualized. Parts of the right and left atria were visible with the right ventricle and centrally situated hyper attenuating ascending aorta. Dorsally, the trachea, azygos vein and esophagus were observed. Ventral and to the left of the fifth thoracic vertebra the hyper attenuating descending aorta appeared, and to the right and ventrally – left pulmonary artery (Figure 5).



- Figure 5. Transvese computed tomographic image of rabbit thorax through the fifth thoracic vertebra (th5) (right side R, left side L): sternum (s), rib (r), scapula (sc), right ventricle (vd), right atrium (ad), left atrium (as), interventricular septum (si), azygos vein (z), ascending aorta (aa), descending aorta (ad), left pulmonary artery (aps), right lung (pd), trachea (t), esophagus (o). (window width (W) 280H; center (C) + 33H)
- Şekil 5. Tavşanın toraksı ile beşinci torasik vertebrasının (th5) enine bilgisayarlı tomografik görüntüsü (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sağ atrium (ad), sol atrium (as), intraventriküler septum (si), azigos damar (vz), aort çıkışı (aa), aort girişi (ad), sol pulmoner arter (aps), sağ akciğer (pd), sol akciğer (ps), trachea (t), esophagus (o). (pencere genişliği (W) – 280H; merkez (C) +33H)

In the transverse plane through the sixth thoracic vertebra, the whole heart was visualized. Atrioventricular septum was observed as a hypo attenuating structure. The cardiac atria and ventricles were best visible. The descending aorta and esophagus were seen at this level, and ventrally was found the hyper attenuating right pulmonary artery (Figure 6).

The transverse post mortem cut through the fourth vertebra showed the right ventricle and interventricular septum, localized in the ventral part of the middle mediastinum. The ascending aorta and both atria were situated dorsally. Above them the right and left cranial venae cavae were visualized. In the dorsal part of the cranial mediastinum the trachea, esophagus and the aortic arch were observed (Figure 7).



- Figure 6. Transverse computed tomographic image of the rabbit thorax through the sixth thoracic vertebra (th6) (right side - R, left side - L): sternum (s), rib (r), scapula (sc), right ventricle (vd), left ventricle (vs), right atrium (ad), left atrium (as), atrioventricular septum (sav), descending aorta (ad), right pulmonary vein (vpd), right lung (pd), left lung (ps), esophagus (o). (window width (W) - 280H; center (C) + 33H)
- Şekil 6. Tavşanın toraksı ile altıncı torasik vertebrasının (th6) enine bilgisayarlı tomografik görüntüsü (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sol ventrikül (vs), sağ atrium (ad), sol atrium (as), atriyoventriküler septum (sav), aort girişi (ad), sağ pulmoner damar (vpd), sağ akciğer (pd), sol akciğer (ps), esophagus (o). (pencere genişliği (W) – 280H; merkez (C) +33H)

At the level of the fifth thoracic vertebra, the right and left ventricles and the interventricular septum were visualized. The difference between the left and right myocardial thickness was visible. Both atria were located dorsally. Below the body of the fifth thoracic vertebra (on the left) the beginning of the descending aorta, azygos vein (on the right), the esophagus with trachea (ventrally) were observed (Figure 8).

At the level of the sixth thoracic vertebra the four heart cavities and the ascending aorta (ventrally) were seen, the azygos vein, descending aorta, esophagus and trachea were situated dorsally (Figure 9).

Discussion

Recently, the rabbit has been used as an animal model in the study of the human ischemic heart disease, unlike in the past, when the swine and dog were utilized for research (Van der Laarse and Van der Wall, 2009).



- Figure 7. Transversal post-mortem frozen section of the rabbit thorax through the fourth thoracic vertebra (th4) (right side - R, left side - L): sternum (s), rib (r), scapula (sc), right ventricle (vd), right atrium (ad), left atrium (as), interventricular septum (si), azygos vein (vz), ascending aorta (aa), aortic arch (ao), right cranial vena cava (vcd), left cranial vena cava (vcs), right lung (pd), left lung (ps), trachea (t), esophagus (o).
- Şekil 7. Tavşanın toraksı ile dördüncü torasik vertebrasının (th4) transversal post mortem dondurulmuş kesiti (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sağ atrium (ad), sol atrium (as), intraventriküler septum (si), azigos damar (vz), aort çıkışı (aa), aortic kemer (ao), sağ kranial vena cava (vcd), sol kranial vena cava (vcs), sağ akciğer (pd), sol akciğer (ps), trachea (t), esophagus (o).

Our results showed that the two dimensional transthoracic echocardiography in the rabbit viable modality, allowing the was а visualization of the normal heart and vascular structure in certain planes, depending on the probe's location. With axial computed tomography and transverse post mortem frozen sections, the image was limited to one plane; however, these methods provide greater detail of normal rabbit cardiovascular anatomy.

The rabbit heart's topography, similar to the carnivorous and human one (Barone, 1996) allowed us to use a combination of ultrasonographic approaches (Tomov and Naumov, 1992; Thomas et al., 1993) typical of the two-dimensional transthoracic echo-cardiography.



- Figure 8. Transversal post-mortem frozen section of the rabbit thorax through the fifth thoracic vertebra (th5) (right side - R, left side - L): sternum (s), rib (r), scapula (sc), right ventricle (vd), left ventricle (vs), right atrium (ad), left atrium (as), interventricular septum (si), left myocardium (lm), right myocardium (rm), azygos vein (vz), descending aorta (ad), right lung (pd), left lung (ps), trachea (t), esophagus (o).
- Şekil 8. Tavşanın toraksı ile beşinci torasik vertebrasının (th5) transversal post mortem dondurulmuş kesiti (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sol ventrikül (vs), sağ atrium (ad), sol atrium (as), intraventriküler septum (si), sol miyokardiyum (lm), sağ miyokardiyum (rm),azigos damar (vz), aort girişi (ad), sağ akciğer (pd), sol akciğer (ps), trachea (t), esophagus (o).

The ultrasonographic study was performed at the left 3^{rd} , 4^{th} and 5^{th} left intercostal spaces and the 3^{rd} and 4^{th} spaces on the right side, because of the rabbit heart's cranial position compared to the rest of the animals (Barone, 1996; Falkner et al., 2005).

Our ultrasonographic results, as the data of Vladova (2006) regarding feline echocardiography, support the positioning of the transducer in a parasternal position in the rabbit echocardiography.

The ascending aorta's visualization in right parasternal position of the heart longitudinal axis and in left parasternal one of the short heart axis can be used as a model for the aorta's valvular apparatus screening, like the experiment, described by Falkner et al. (2005) in the sheep.



- Figure 9. Transversal post-mortem frozen section of the rabbit thorax through the sixth thoracic vertebra (th6) (right side - R, left side - L): sternum (s), rib (r), scapula (sc), right ventricle (vd), left ventricle (vs), right atrium (ad), left atrium (as), ascending aorta (aa), descending aorta (ads), azygos vein (vz), right lung (pd), left lung (ps), trachea (t), esophagus (o).
- Şekil 9. Tavşanın toraksı ile altıncı torasik vertebrasının (th6) transversal post mortem dondurulmuş kesiti (sağ taraf - R, sol taraf - L): sternum (s), kaburga (r), scapula (sc), sağ ventrikül (vd), sol ventrikül (vs), sağ atrium (ad), sol atrium (as), aort çıkışı (aa), aort girişi (ad), azigos damar (vz), sağ akciğer (pd), sol akciğer (ps), trachea (t), esophagus (o).

In accordance with the human heart studies of Espaniola-Zavaleta et al. (2006), Muñoz-Castellanos et al. (2009), Vladova et al. (2005) and Wang et al. (1992), we performed a comparative anatomical echocardiographic analysis of the rabbit heart and some mediastinal vessels. The results of the transverse post mortem study, computed tomography and ultrasonographic research corresponded.

In comparison with Yonkova et al. (2010) and Zhao et al. (2011), who studied the rabbit heart's adipose alterations, we investigated imaging anatomical features of this organ in healthy rabbits.

In our research the appearance of the thoracic portion of the descending aorta, the both cranial venae cavae, right azygos vein and

26

both pulmonary veins toward the thoracic vertebra corresponded with our anatomical transverse section and those results shown from the groups of Barone (1996) and McCracken et al. (2008).

Similarly to the computed tomographic investigation of feline and canine heart (De Rycke et al., 2005; Vladova et al., 2005; Vladova, 2006), we used the thoracic vertebrae as a bone marker to define the investigated heart and vascular structures' location in our computed tomographic and transverse post mortem study. Unlike Vladova (2006) we confirmed that at the level of 2^{nd} and 3^{rd} rabbit thoracic vertebrae only vascular structures were observed. The whole heart appeared at the level of the 5th thoracic vertebra, and the descending aorta was visualized at 6th thoracic vertebra. The fact confirms the more cranial location of the rabbit heart within the thorax when compared to the cat (Vladova, 2006).

The comparative analysis of the post mortem transverse sections and computed tomographic results corresponded with the studies of Alsafy (2008), De Rycke et al. (2005), Samii et al. (1998) and Vladova (2006).

Unlike the study of Zotti et al. (2009) where the rabbit is placed in sternal position, we received transversal cuts by placing the animals in ventrodorsal recumbency. Our computed tomographic slices were with lesser thickness (3 mm), but the frozen transverse sections were with the same thickness.

We suggest that the comparative data from the ultrasonographic, computed tomographic and transverse post mortem investigation of the rabbit heart and its mediastinal vessels are applied as a norm in the interpretation and diagnosis of cardiac and vascular lesions in this species. The results of our study allow conclusion that the rabbit is a suitable experimental model for morphological studies of the heart, in connection with the research of some craniological problems in animals and human.

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