Ministry of Higher Education and Scientific Research University of Qadisiyah College of Veterinary Medicine



## Morphological and Histological study Aortic arch in rabbits

Set By

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## **Dedication**

Personally, I'd like to thank all those who have

helped with their advice and efforts...

I'd like also to thank all the All veterinary

professors, especially my supervisor

Dr. Maha for his valuable advices

For my parents friends and everyone, I offer my research.

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## **Abstract:**

The study aimed is to investigate the morphological and histological variations of the aortic arch of local breed rabbits (*Oryctylagus conniculus*).knowledge of these variations is important for other basic sciences.

Nine adult local rabbits were obtained to study the aortic arch(four animal for corrosion cast technique ,two injected with colored latex and three for histological study) all the rabbits were euthanized and exsanguinatd, for the cast of the aortic arch they injected by self polymerizing resin via the left ventricle of the heart and the specimens were macerated in (KOH %25) for two weeks and the cast of the aortic arch were prepared, two main branches identified from a ortic arch(except in one cadaver has three branches), the brachiocephalic trunk firstly and the left subclavian artery, the two common carotid artery (right and left) and the right subclavian artery arising from brachiocephalic trunk, the right and left subclavian arteries gave the same branches :-(double vertebral arteries, costocervical trunk, cervical profund artery internal. thoracic artery, external thoracic artery, superficial cervical artery, and continous as axillary artery).

Histologically the wall of the aortic arch is consist of three layers from internal to external :-tunica intima, it's the thinnest layer (1±0.166)  $\mu m$ ,tunica media, it's the thickest layer (12±0.0149) $\mu m$  and tunica adventitia, the thickness of this layer is (5±0.166) $\mu m$ . tunica intima is consist of single layer of endothelial cells,tunica media consist of numerous elastic lamina in acircular arrangment and the tunica adventitia is consist of connective tissue,smooth muscle fibers,collagene fibers,few elastic fibers ,fibroblast cells, mast cells vasavasorum , the aortic arch has alarge diameter (98±0.2019) $\mu m$  and thickness of the wall is ( 18±0.0282) $\mu m$  to convey the high blood pressure in aortic arch the conclusion of this study, there is avariation in the branches of the aortic

arch in different species of animals when the results compared with another studies and also, the diameter, thickness of the wall of aortic arch is variable from one animal to another.

**Keywords :-**Aortic arch ,Histology,Rabbits,Brachiocephalic trunk,left and right subclavian arteries.

## الخلاصة: ـ

ان الهدف من الدراسة الحالية هو بيان الاختلاف النسييجي والشكلياني للقوس الابهري في العلوم الارانب المحلية (Oryctylagus conniculus) وان معرفة هذا الاختلاف لة اهمية في العلوم الاساسية الاخرى. اجريت دراسة القوس الابهري في تسع ارانب محلية ،اربعة ارانب استخدمت التحضير قالب القوس الابهري،اثنان حقنت بمادة اللاتكس الملون . واستخدمت ثلاثة للدراسة النسيجية ، مجميع الحيوانات تم تخديرها واستنزافها وتم تحضير قالب القوس الابهري وذلك بحقن مادة الراتنج عن طريق البطين الابسر للقلب، ثم تمت عملية التعطن لباقي انسجة الحيوان وذلك بوضعة في محلول هيدروكسيد البوتاسيوم بتركيز 25% ولمدة اسبوعان. ينشأ من القوس الابهري فرعان رئيسان هما الجذع العضدي الدماغي اولا والشريان تحت الترقوي الايسر ويعطي الفرع الاول الشريانان السباتيان السباتيان الايمن والذي يعطي نفس الفروع التي يعطيها الشريان تحت الترقوي الايمن والذي يعطي نفس الفروع التي يعطيها الشريان تحت الترقوي الايمن الجذع العنقي الضلعي، شريان ضلعي غائر، ،شريان صدري داخلي،،شريان صدري خارجي،،شريان عنقي سطحي و يستمر باسم الشريان الابطي) .نسيجيا يتكون حدار القوس الابهري من ثلاث طبقات وهي من الداخل الى الخارج (الغلالة الداخلية وهي اقل الطبقات سمكا (166.0±1) مايكروميتر، الغلالة الوسطى وهي اكثر الطبقات سمكا (166.0±2) مايكروميتر.

وتتكون الغلالة الداخلية من طبقة واحدة من الخلايا البطانية. اما الغلالة الوسطى فتتكون من طبقات من الألياف المرنة التي تترتب بشكل دائري، اما الغلالة البرانية فتتكون من النسيج الضام،الألياف العضلية الملساء،الألياف الغراوية،القليل من الألياف المرنة،الأرومات الليفية والأوعية الدموية المغذية لجدار الوعاء الدموي، ويبلغ قطر القوس الأبهري(0.2019±98) مايكروميتر ويبلغ سمك جدار القوس الأبهري (18±0.0282) مايكروميتر لكي يستطيع تحمل ضغط الدم العالى في القوس الأبهري .

ونستنتج من الدراسة الحالية ان فروع وقطر وكذلك سمك جدار القوس الابهري تختلف باختلاف نوع الحيوان وذلك عند مقارنة نتائج هذة الدراسة مع باقي الدراسات الاخرى .

الكلمات المفتاحية :- القوس الابهري، نسيجيا، ارانب، الجذع العضدي الدماغي، الشريانان تحت الترقوي الايمن والايسر.

## Chapter one Introduction

Chapter one Introduction

## Introduction:-

Aorta is the major elastic artery of circulatory system and the arteries branched from it which supply all tissues of organs in the body, aorta is divided into four segments according to the region, ascending aorta, aortic arch, descending thoracic and abdominal aorta (1,2).

Aortic arch or arcus aortae is the convexity of aorta toward caudo-dorsal, exiting the heart . this segment of aorta was very important it gave several vessels which supplied head, neck, fore limbe and the proximal half of thoracic region like right and left common carotid arteries ,vertebral artery, axillary artery, internal thoracic, costocervical trunk and dorsal scapular artery respectively (3).

Three vessels origionated from aortic arch is studied in several researches like in domestic mammals(4), hamster(5),chinchilla(6),guinea pig (7),fox (8), human (9) and korean native goat (10).the major branches of aortic arch are brachiocephalic trunk and left subclavian artery which described in equine, ruminants, pig dog and cats (3).

Histologically the wall of the aortic arch consist of three tunica from internal to external (intimea, media, adventitia). tunica intima is relatively thick and is lined by single layer of endothelial cells beneath it subendothelial layer which consist of loose connective tissue that contain elastic fibers and few smooth muscle cells.tunica media is the thickest layer which consist of numerous elastic laminae As asheet in a circular and oplique arrangment and between them ,there is a thin layer of connective tissue contain collagen fibers and smooth muscle cells,tunica intima and tunica media separated from each other by internal elastic lamina.

Tunica adventitia is the relatively A thin layer contain A bundles of collagen fibers and few elastic fibers, fibroblasts, mast cells and vasa vasorum.

the aortic wall diameter and thickness of each layer of the wall is change in four segments of the aorta with the variation in variable species (11, 12) guinea pig and rat.

The research work aimed for study aortic arch in rabbits in order to providing information for other basic science.

# Chapter two Materials and Methods

Chapter two Materials and Methods

## Materials and Methods:-

Nine adult local breed rabbits (*Oryctylagus Conniculus*) aged 6-12 months and weighted (1.5-2 Kg) were used for morphological and histological study of the aortic arch, (four for preparing aortic cast ,two injected with colored latex and three for histological study).

All the animals were euthanized with (Ketamine 50 mg/k.g ml and Xylazine 10 mg/k.g), the thoracic cavity of all the animals were opened. A cannula was inserted into the left ventricle to exsanguinated the animals after that injected with normal salin 0.9% into the left ventricle to prevent formation of blood clotting.

For the cast technique we used the self polymerizing resin to study the aortic arch and the arteries arises from it ,repared resin rapiddly selfcuring powder(5 mg) mixed with (15 ml) of self polymerizing liquid and injected into the left ventricle of the heart of cadaver by using the disposable syringe (50 ml) after that the rabbit left (24)hours and transferred into 25% KOH bath for two weeks for maceration. (13,14,15).

For colored latex (5 ml of latex mixed with red carmine stain ) injected into the left ventricle by using syringe (50 ml ) after (24 ) hours the arteries arises from the aortic arch were dissected carefully (13).

For histological study the specimen of aortic arch was removed and fixed in 10% formalin , then washing and dehydrated by ascending series of ethanol 70% ,80% ,90% 100% in which two times 1-2 hour. Clearing by xylol(15 min.) and embeded in paraffine wax (2-4 hour) after that blocks sectioned by rotary microtome with (five-six) micrometer in thickness and staining by Hematoxyline- Eosine stain, the diameter of aortic arch and the thickness of each layer of aortic arch wall was measured by using ocular micrometer and documented by flourecent- microscope.

## Chapter three Results and Discussion

Chapter three Result and discussion

### Result and discussion:-

## Morphological results:-

The aortic arch of local breed rabbits was the second segment of aorta fig.(1) which present between ascending and descending segments at the level of vertebral coloumn at fourth to fifth intervertebral disc over the bifurcation of pulmonary trunk near left pulmonary artery and it passing in caudo-dorsal course ,it end when the descending thoracic aorta begins,this result in agreement with(16) in human,(17) in rabbits in Slovak (18)in red squirrels( *Scicurus Vulgaris*).

The aortic arch give two main branches fig.(2,3) brachiocephalic trunk and left subclavian artery and this in agreement with researchers in another species like in ruminants, equine, pigs, carnivores and domestic mammals (3,4), red squirrels (18),(19) in ground squirrels (*Spermophilus citellus*), chinchilla (6) guinea pigs (7) and fox (8), while disagreement with other studies (the aortic arch give arise to three branches, brachiocephalic trunk, left common carotid artery and left subclavin artery) in human (20), rat (21), porcupine(*Hystrix Cristata*) (22), in mole-rats (*Spalax Leucodon*(23), and in Turkish spiny mice and in adult Cyprus spiny mice (24), but in some human cadavers showed variations in branching pattern some of them gave two branches, left subclavian artery and common trunk which gave brachiocephalic trunk, left common carotid artery and another give aris for four branched are brachiocephalic trunk,lef and rightt common carotid arteries and left vertebral artery(25).

Brachiocephalic trunk branched to left common carotid artery and common trunk from which the right common carotid and right subclavian artery arising fig. (4) and this result is corresponding with (7,18 and 19) in guinea pig, red and ground squirrels .right common carotid artery passing to right side of head and neck region, then the right subclavian artery give branches supply the

right limb and right side of thoracic region, while the left subclavian artery passing to the left limb and left part of thoracic region and it respected with (3, and 16) in domestic animals and human.

The branched ramified from the left and right subclavian arteries are :- two vertebral arteries, common trunk (costocervical trunk ,cervical profund artery ,internal thoracic artery) , external thoracic artery ,superficial thoracic artery and it continuous as axillary artery fig,(4,5) and this noticed is similar to (6 and23) in the study of chinchilla and mole-rats, and differs with (7) in the study of guinea pig.

## Histological results :-

The wall of the aortic arch is composed of three layers from internal to external is tunica intima, tunica media and tunica adventitia fig. (6a) and this in agreement with (11,26,27,28).

Tunica intima is consist of single layer of flattened endothelial cells, few elastic fibers and few smooth muscle cells ,between tunica intima and tunic media there is internal elastic lamina, the thickness of tunica intima in aortic arch is  $(1\pm0.166 \text{ table})\mu\text{m}$  (1) fig,(6b,6c)

Tunica media is composed of elastic concentric laminea with circular arrangement and in the space between elastic laminae are thin layer of connective tissue contain collagen fibers, smooth muscle cells ,tunica media is the thickest layer (12±0.0149)µmm table (1) fig.(6b,6c)

Tunica adventitia (fig.6b,6c) is relatively thin and contain bundles of collagen fibers and few elastic fibers ,also present of fibroblasts,mast cells ,smooth muscle cells and vasa vasorum. The thickness of Tunica Adventitia is(5 $\pm$ 0.0166) µm table (1).the diameter of aortic arch is( 98 $\pm$ 0.2019)µm and thickness of aortic arch wall is (18 $\pm$ 0.0282)µm so the aortic arch has alarge

diamater and thick wall to convey the high blood pressure of the blood that passing throught it, our above result is differ from the result of (11,23) in the (guinea pig and rat) and spiny mice due to the nature of histological structure of aortic arch wall also in the study of hamster (28) they can observed ER strees on the aortic arch wall under normal condition

Table 1.Mean diameter and thickness(  $\mu m$ ) of the intimal,medial, adventitial layers and thickness of the wall of aortic arch in local breed rabbits .

Variables	Mean ± SE(μm)
Diameter of aortic arch	98 ± 0.2019
Tunica intima	1 ± 0.166
Tunica media	12± 0.0149
Tunica adventitia	5 ± 0.166
Thickness of wall of aortic arch	18± 0.0282

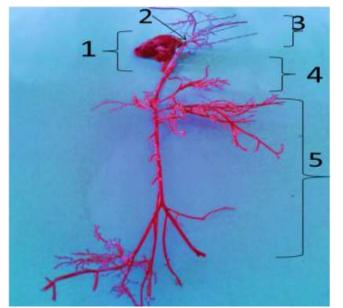


Fig 1:-The cast of the aortic arch in local breed rabbits showing (1)heart (2) ascending aorta (3)aortic arch(4) descending thoracic aorta (5) abdominal aorta.



Fig.2 Aortic arch insitue in rabbit showing:-(A) aortic arch (BT) Brachiocephalic trunk (LS) left subclavian artery (LC) Left common carotid artery (RC) right common carotid artery (RS)Right subclavian artery .



Fig.3:-The cast of the aortic arch and it's major branches in rabbits (dorsal view) showing(A) Aortic arch (1)Brachiocephalic trunk (2)right common carotid artery (3)Right subclavian artery(4)left common carotid artery(5) left subclavian artery.

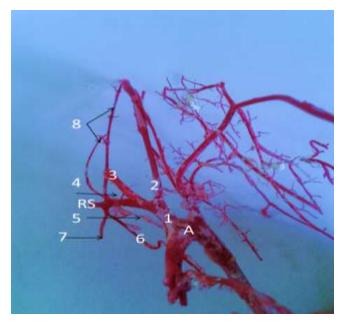


Fig .4: The cast of the aortic arch in rabbits (right lateral view)showing branches of Brachiocephalic trunk (A)aortic arch (RS) Right subclavian artery (1) brachiocephalic trunk (2)left common carotid artery (3) Right common carotid artery (4) Cervical profund artery (5) Costocervical artery (6)intrnal thoracic artery (7) external thoracic artery (8)Vertebral arteries.

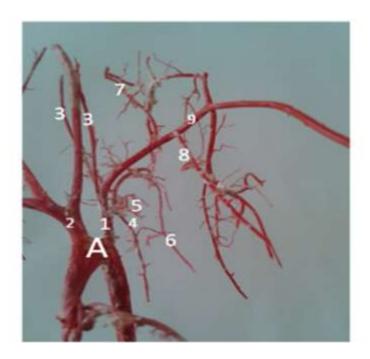


Fig.5:-The cast of the aortic arch in rabbits (left lateral view) showing, (A)aortic arch (1) Left subclavian artery (2)Brachiocephalic trunk (3)Vertebral arteries(4) Costocervical artery (5) Cervical profund artery (6) Internal thoracic artery (7) superficial thoracic artery (8) External thoracic artery (9) Axillary artery.

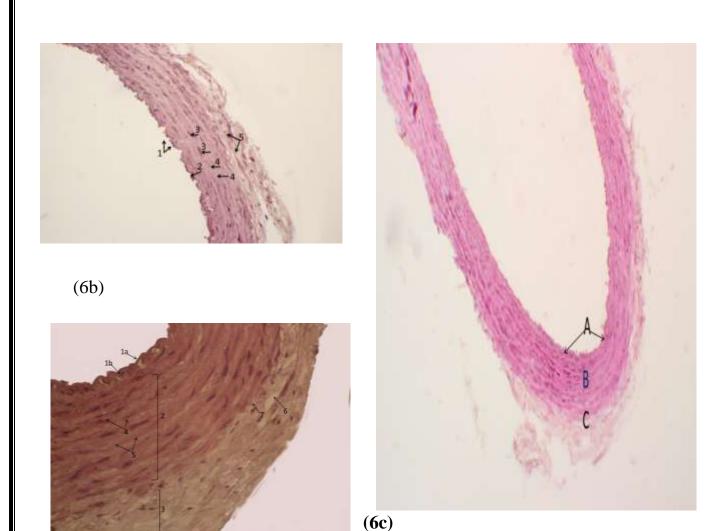


Fig.6:-The histological structure of Aortic arch in rabbits showing :-(6a) (A) Tunica intima (B)Tunica media (C)Tunica adventitia (100x) H&E stain.

(6b) ( (1)Endothelial layer cells (2)Internal elastic lamina (3)Smooth muscle cells (4)Elastic fibers lamina (5)connective tissue. (200 x) H&E Stain .

(6c)(1a)Endothelial cells layer (1b)Internal elastic lamina (2)Tunica media (3) Tunica adventitia (4)Smooth muscle cells (5)Elastic fibers lamina (6) vasa vasorum (7) external elastic lamina (400 x) H&E stain.

## References:-

- 1-Mclaughlin,CA. and Chiasson, RB.(1987). Labrotary Anatomy of Rabbit. W.C. Brown Company, Lowa.41.
- 2-Barone,R;Pavaux.C; Blin PC. and Guq, p.(1973). Atlas of Rabbit Anatomy, Masson and Cie, Paris 121-122.
  - 3-Getty,R. (1975): Sisson and Grossman's. The Anatomy of the domestic animal, 5th ed. W.B. Saundres Company Philadephia Equine vol.1 and vol. 2 (565-568),Ruminants (960-966) pig (1306-1310) Dog (1595-1598) Cat (1599 -1602).
- 4-Schummer, A; Wilkenes, H; Vollmerhaus, B. and Habermehl, K. H. (1981).organs of circulation system, the skin and the cutaneous organs of the domestic mammals. Verlag Paul Parey, Berlin Pp:72-77.
- 5-Popesko, P;Rajtova, V. And Horak, J. (2003). Colour Atlas of the anatomyof small laboratary animal rat, mouse, hamster , wolf. publishing, London pp:48,132,198.
- 6-Ozdemir,V;Cevik-Demarkan ,A. And Turkmenoglu (2008).Subgross and macroscopic investigation of blood vessels orginating from aortic arch in the chinchilla (Chinchilla Lanigera). Anat.Histol. Embryol. , 37 (2):131-133.
- 7-Kabak,M. And HazirogluRM. (2003).subgross investigation of blood vessels originating from arcus aorta in guinea-pig (Cavia Porcellus) Anat.Histol.Embryol., 32(6):362-366.
- 8-Emine, E. And Özean, Ö (3013). Amacroanatomical study of the arcus arch in the fox (Vulpes Vulpes). Turkish Journal of veterinary and Animal sciences 37:672-674.
- 9-M.Sri, HB.; Vasanthi, A. and Asha ,L.D.(2015). Aortic arch variations-A rare finding. IOSR Journal of Dental and Medical Sciences .Vol.14 Issue 3 :52-55.

- 10-Lee,HS.and Lee,IS. (1984). Anatomical stydies on patterns of the branches of the aortic arch in the Korean native goat ,Korean Journal of Vet,Res, 24:1-7.
- 11-Josian,MM;Antononio,MO.;Carlos ,R.P; Selma,M.M; Maria L.E. (2004). Structure of the aortic wall in the guinea pig and rat .Braz.J.Morphol. Sci. 21(1):35-38.
- 12-Nigar ,V ;Feral ,Ö,Ersin,F;Ali ,O. And Murat,Y.(2003). Histological change in the rat thoracic aorta after chronic nitric oxide synthese inhibition.Turk.J.Med.Sci.33:141-147.
- 13-Ahasan, ASML; Islam, MS; Kabria, ASMG; Rahma, MM and Uddin, M (2012). Major variation in branches of the abdominal aortain New Zealand White rabbit (Orycotolagus Cuniculus). International Journal of natural Sciences 2(4):91-98.
- 14-Vincent,PE; Pata, A.M; Hunt, AAE.; Weinberg, P.D. and Sherwin, S.J.(2011). Blood flow in the rabbit aortic arch and descending thoracic aorta. J. R. Soc. Interface 8:1708-1719.
- 15-Luna L.G. (1968). Manual of Histological staining methods of armed forces institute of pathology third ed.Mc Graw-Hillbook company New York.
- 16-Darke,RL;Vogl,Wayn,A;Michell,Adam,W.M.(2010). Gray's Anatomy for Student
- (2nd ed.) .Churchill Livingstone (Elsevier). ISBN 978-0-443-06952-9.
- 17-Mazenky,D;Vdoviakova,K; Petrovova, E. And Luptakova, L. (2009). Variability origin of some branches of aorta thoracic in rabbit. Slovak J.Anim. Sci. 42(1):60-62.
- 18-Aydin,A.(2011).the arteries orginating from the aortic and the branches of these arteries in red squirrels (Scicurus Vulgaris) Veterinarni Medicine 56(3):131-134.

- 19-Aydin,A; Ozkan,Z.E; Yilmaz,S. And Ilgun,R.(2011). The arteries originating from the aortic arch and the patterns of their branches in ground squirrels (Spermophilus Citellus) Veterinarni Medicina 56 (9): 469-472.
  - 20-Shahcheraghi,N; Dwyer,H.A; Cheer,A.Y; Barakat,A;and Rutaganira, T.(2002).Unsteady and three-Diamentional stimulation of blood flow in the human aortic arch .Journal of biomechanical Engineering Vol. 124:378-387.
  - 21-Monnereau,L; Carrretero,A.; Bergges S; Navarro M; Leonard M; Layzhi,F; Sautet J.and Rubert, J. (2005). Morphometric study of the aortic arch and it's major branches in rat fetuses on the 21st day of gestation. Anat. Embryol. (Berl) 209(5):357-369.
  - 22-Atalar,O.(2011). Specific anatomical structure of some wild animals in East Anatolin of Turkey. Journal of Animal and Veterinary Advances 10(23):3080-3084.
  - 23-Aydin,A; Ozkan, ZE.and Ilgun,R. (2013). The morphology of the arteries originating from the arcus aorta and the branches of these arteries in mole-rats (Spalax Leucodon). Veterinarni Medicina, 58(7):373-376.
- 24-Cagdas,O; Seda , K;Hatice , M.E, Erkut,K.and Merih,H.(2010).Subgross investigation of the blood vessels orginating from aortic arch (Arcus aortae) in spiny mouse.journal of animal and veterinary Advances 9 (20):2665-2667.
- 25-Virendra,B; Rakhi,R ;Vaishall,J;Vishal,B andShiv, R.(2013).Anatomical variations in the Branching Pattern of Human Aortic Arch:Acadaveric study from Central India. Hindawi publishing Corporation.ISSN Anatomy Volum 2013,Articl ID 828960 5 Pages .
- 26-Fernando, CADS; Lara,SC; Rodrigo, PL. Rejane M.G. and Sebastiao ,R.T.(2004). Amodified method for the selective stianing of elastic system fibers in methacrylate tissue section. Braz. J. Morphol. Sci. 21(2):73-79.

- 27-Nereide,FC; Winston, BY; Sergio, S.M; Julio, L.S; Antonio, D.S.R. and Carlos, R.P (2005). Morphological and biomechanical study of abdominal aorta of rats submitted experimental chronic alcoholism. Acta.Cir. Bras. J. Vol.20 No.3.
- 28-Masaki, K; Makoto, H; Yoshiyuki ISHII; Shigeru, K. And Yasuhiro ,Y.(2009).Aortic ER strees in streptozotocin-induced diabetes mellitus in APA hamsters.Exp.Anim.58(2):113-121.



{وَقُلْ رَبِّ أَدْخِلْنِي مُدْخَلَ صِدْقِ وَأَخْرِجْنِي مُخْرَجَ صِدْقِ وَأَخْرِجْنِي مُخْرَجَ صِدْقٍ وَأَخْرِجْنِي مُخْرَجَ صِدْقٍ وَاجْعَلْ لِي مِنْ لَدُنْكَ سُلْطًانًا نَصِيرًا}

صدق الله العظيم سورة الإسراء: رقم الآية (٨٠)