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# Histological Comparison Study of the Adult Human Heart between Male and Female in Kirkuk City in Iraq

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**ABSTRACT**

**Background**: The heart in human is normally a hollow muscular organ located within the chest in the middle mediastinum and is surrounded by a sac known as a pericardium.

**Methods**: The heart samples collected from 32 cadavers (male and female) taken from four age group ((≤ 29, 30-39, 40-49, ≥50) from the forensic center of Azadi_ teaching hospital of Kirkuk city from the March 2023 to August 2023.

**Results**: the general histological results shows the heart wall consist of the three layer the outermost layer called epicardium, middle layer called myocardium and the inner most layer called endocardium .

**The morphometric finding:** the minimum mean thickness of the epicardium of the LA was 103 .5 um in female and the maximum mean was 928.5 um. And the mean of the LV the minimum was 217.5 um and the maximum was 1377 um. The minimum mean thickness of the myocardium layer of the LA was1649um and the maximum was 3185 um while the minimum mean of the myocardium in the LV was 1666 um and the maximum was 4132 um . The minimum mean thickness of the endocardium of the LA was 152 um and the maximum mean was 312.5 um, while the minimum thickness in the LV was 60.5 um and the maximum was 170 um .The minimum mean diameter of the cardiocyto in the LA was 11 um and the maximum was 23.5 um, while the minimum mean in the LV was 13.8 um and the maximum was 27.5 um. The minimum mean diameter of the pruknjie fiber in the LA was 30.6 um and the maximum was 78 um, while the minimum mean in the LV was 38 um and the maximum was 91.5 um

**Conclusions**: The thickness of the epicardium and myocardium was thicker in the left ventricle than left atrium in all ages. The thickness of the endocardium in the left atrium was thicker than in the left ventricle The diameter of the pruknjie fiber was larger than the diameter of the cardiocyte in both atrium and ventricle.

**Introduction:** The heart is a muscular tube responsible for collecting blood from the tissues of the and the lungs, as well as pumping blood from the heart to the lungs and around the body. [1]

The heart has four chambers; Two atria are separated by an interatrial septum (IAS), and two ventricles are separated by an interventricular septum (IVS).Additionally, there is a short "septal segment" placed between IAS and IVS, which is known as the atrioventricular septum (AVS) due to the offset of the atrioventricular valve, since it is generally situated in between the sub aortic outlet section of the left ventricle and the right atrium. [2]

The wall of the heart is composed of three layer: endocardium (innermost layer), myocardium (middle layer) and epicardium (outer most layer).[3]

The heart's wall is divided into three layers: epicardium, myocardium, and endocardium. The endocardium is made up of the endothelium and a layer of sub endothelial connective tissue. The impulse-conducting system is located between the endocardium and the myocardium in the sub endocardium. A large part of the cardiac wall is made up of the myocardium. Cardio myocytes join together to make up this layer. These cardio myocytes are striated like myocytes found in skeletal muscle. However, unlike skeletal muscle cells, they are branched, contain intercalated disks, and are usually mononucleotide.[4] The epicardium is a single layer of mesothelial cells that lines the heart and makes key cellular contributions to embryonic heart formation.[5]

The epicardium layer raised from the proepicardium. [6]

The epicardium contain adipose tissue which is a type of the visceral adipose tissue it has both paracrine and endocrine effects.[7]

The myocardium layer is a type of striated muscle in the heart that is made up of a bundle of integrated cardiac muscle fibers that are organized into fascicles by a connective tissue sheath. [8]

The cardio myocyte of the myocardium contain a bundle of myofibrils and divided into sarcomeres, which contain several contractile proteins including actin and myosin.[9]

The myocardium supported by the intercalated discs, which is a structure that enable cell-to-cell mechanical connection and mediate electrochemical communication. These discs s generated by a specific sarcolemma area located between the ends of each abutted cell. [10]. The endocardium, the heart's endothelial lining, plays complex and critical roles in heart development, including the formation of cardiac valves and septa, the division of the truncus arteriosus into the aortic and pulmonary trunks, the development of Purkinje fibers that form the cardiac conduction system, and the formation of trabecular myocardium.[11]

The muscular arrangement of The left atrial wall, particularly the myocardial sleeves that extend onto the pulmonary veins,[12]

The left ventricle myocardium extension into pulmonary artery and aorta beyond the ventriculo-arterial junction.[13]

**Methods:** The current study has been done on autopsy of 32 cadavers including male and female and divided in four age groups (≤ 29, 30-39,40-49, ≥50) has been referred to forensic centers according to the official papers from police stations according the court pre orders., at forensic medicine departments at Kirkuk and Salah-Aldin governorates, cause of death was due to trauma like car accidents, shooting, stabbing, fall from height and so on. On history from their family they approved their dead has no any known pathology or medicine taking.

**Microscopically inspection and the descriptive histology:**

The microscopic investigation of the heart section include the Morphometric and the descriptive histology. Using of the light microscope to perform the microscopical investigation.

**Morphometric technique:**

Study the three layer of the heart epicardium, myocardium and endocardium, measure the thickness of the each layer in the left atrium and ventricle, measure the diameter of the cardiocyte and pruknjie fiber of the left atrium and ventricle and compare it between male and female by the microscope.

**Preparation of the heart sample:**

The heart dissected into small pieces which representing all the layers (epicardium, myocardium and endocardium).

The first step of preparation of heart sample is fixation by using 10% buffer formalin saline for

48hour. [14]

The specimens were washed in running tap water, labeled by the number given to the specimens to identify the tissue through all stages of processing and then dehydrated through gradual concentrations of ethanol alcohol. The Clearing was performed by two changes of xylene (one hour for each change). The specimens were then embedded in melted paraffin inside an embedding block for 2 hours and blocked in paraffin wax. [15]

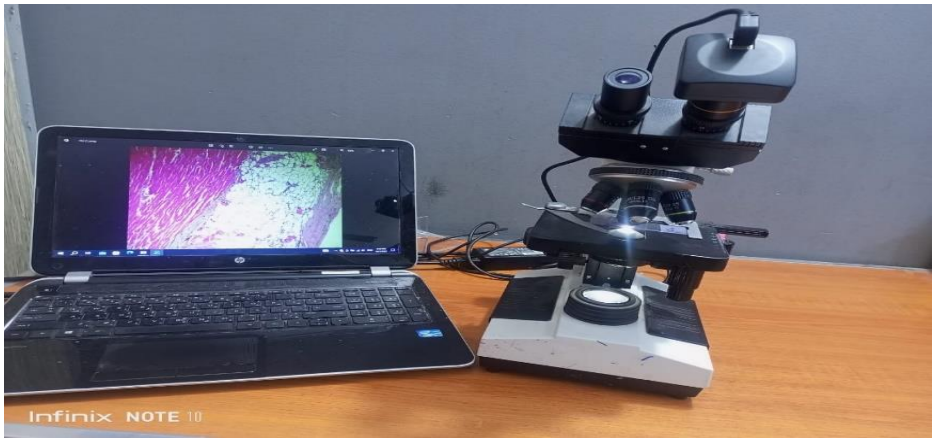
Tissue blocks were serially sectioned at 5 µm thickness using a manual microtome, mounting sections from warm water on the slide to make histological slides ready for staining. All sections were then immersed in xylene for 30 minutes, rehydrated through graded ethanol alcohol for 15 minutes for each change, and finally dipped in tap water for 5 minutes.[16]

Routine staining with Hematoxylin and Eosin

**Microscopic inspections:**

| Device used in this study | Figure |
|---|---|
| Light microscope type(Leica) | 1 |
| Light microscope type(XSZ-107BN) | 2 |

**Figure(1):): shows the light microscope type (Leica) measuring the thickness of the heart layer and the diameter of the cardio cyte and purknjie fiber.**




**Figure(2): shows light microscope type (XSZ-107bn) and camera AM scope mu 300.**

**Morphometric Histology:**

**1. Measuring technique of the thickness of the heart layers.**

The thickness of the layers of the heart (epicardium, myocardium, and endocardium) counted by the standard counting unit (µm) by using Leica microscope and XSZ-107bn microscope. For each specimen the 50-x objective lens was used in the Leica microscope and 40-x objective lens was used in XSZ-107bn microscope. The thickness of the each layer of the heart was measured

at different three points and the mean value was taken.

**2. Measuring technique of the diameter of the cardio myocyte and purknjii fiber.**

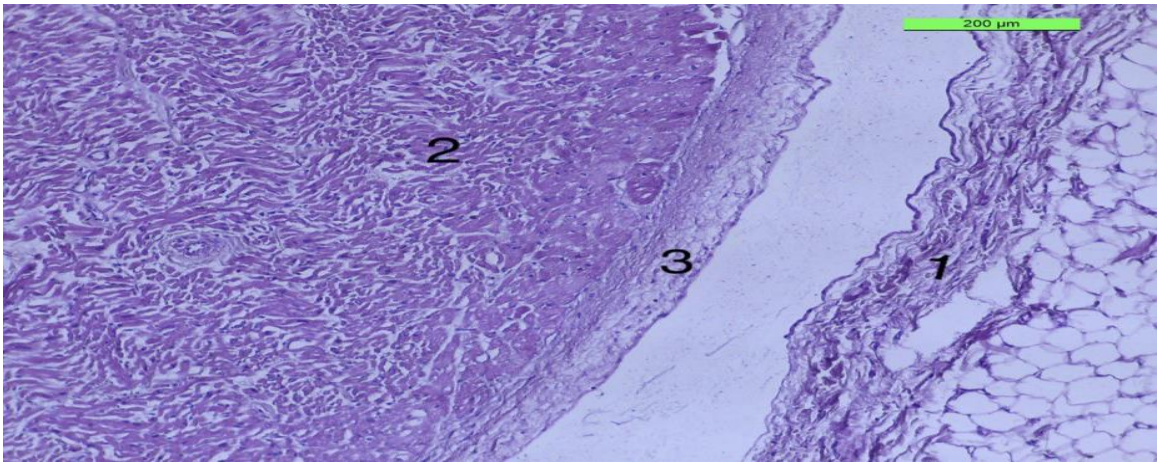
Measuring the diameter of the cardio myocyte by the Leica microscope, measured of the 10 different cardio myocyte and plural them the division in 10. And measured the diameter of the five different purknjii fiber and plural them and division in 5. And measuring the diameter of the cardio myocyte and purknjii fiber by the XSZ-107bn microscope, measured of 10 different cardio myocyte and plural them and division in 10 then multiplied by correction factor 2.5. measured of the 5 different purknjii fiber and plural them and division in 5 then multiplied by correction factor 2.5.

**Results:** Light microscope study shows normal histological finding. The heart section in different groups in both females and males, it showed that the wall surrounding its four chambers does not differ in its tissue structure, as its consists of the following layers figure (3)arranged from outside to inside as follows

**Epicardium layer:** Consist of the single layer of the mesothelial cells and beneath it there is a layer of the connective tissue rich in fat cells, blood vessels and nerve supply figures.

**Myocardium layer:** The current study has shown that the thickness of the heart wall is mainly due to the thickness of its middle layer in both males and females, the myocardium layer which consist of the cardiac muscles arranged in a regular directions. The muscles appeared branched striated muscles, connected to each either via intercalated disc some of these cells bi nucleated cell figure (4), between the cardiac muscles there was connective tissue rich in blood, nerve and fat cells.

**Endocardium:** The current study also showed that the layer lining the heart cavities is the thinnest layer of the heart wall. This layer consists of the single layer of the endothelial cells, which are based on a basement membrane. Underneath this is a layer of loose connective tissue, and beneath it is a layer of connective tissue rich in nerve and present of an important purkinje cells figure (5), related to the conducting system and this layer called sub endocardial layer.




**Figure(3): shows the layers of the heart wall(epicardium, myocardium, endocardium), stained with Hematoxylin and Eosin Magnifications power =200um**

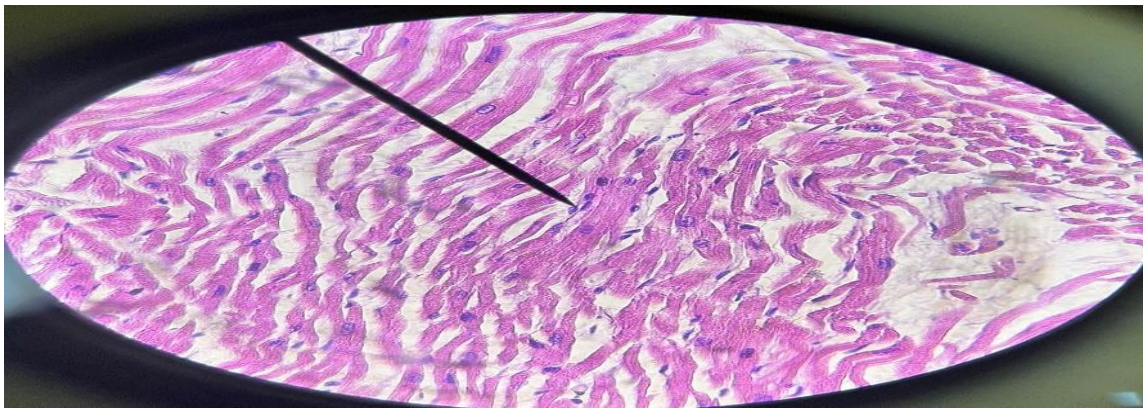


**Figure (4): shows the binucleated cardiocyte. Stained with hematoxylin and Eosin, magnifications power=40**

**Morphometric histological results:**

**Table (1) : shows the thickness of the epicardium layer in the left atrium and ventricle**

| Age Class (years) | Sex | Minimum Thickness of epicardium of left atrium(um) | Maximum thickness of the epicardium of the left atrium (um) | Mean | Minimum Thickness of the epicardium of the left ventricle | Maximum thickness of the epicardium of the left ventricle | Mean |
|---|---|---|---|---|---|---|---|
| ≤ 29 | Male | 150 | 162.5 | 156.3 | 450 | 470.7 | 460.4 |
| | Female | 99 | 108 | 103.5 | 209 | 226 | 217.5 |
| 30-39 | Male | 187.5 | 190.5 | 189 | 687 | 703 | 695 |
| | Female | 162 | 170 | 166 | 354 | 362 | 358 |
| 40-49 | Male | 343 | 368 | 355.5 | 858.7 | 872 | 865.4 |
| | Female | 237 | 242 | 239.5 | 429 | 440 | 434.5 |
| ≥50 | Male | 925 | 932 | 928.5 | 1371 | 1384 | 1377 |
| | Female | 825 | 835 | 830 | 1225 | 1244 | 1234 |

**Table (2) : shows the the thickness of the myocardium layer of the left atrium and ventricle in both male and female.**

| Age Class | Sex | minimum Thickness of myocardium of the left atrium (um) | Maximum thickness of the myocardium of the left atrium | Mean | Minimum Thickness of the myocardium in the left ventricle | Maximum thickness of the myocardium of the | Mean |
|---|---|---|---|---|---|---|---|
| ≤ 29 | Male | 2700 | 2730 | 2715 | 2805 | 2818 | 2811 |
| | Female | 1641 | 1658 | 1649 | 1660 | 1672 | 1666 |
| 30-39 | Male | 2755 | 2774 | 2764 | 3100 | 3118 | 3109 |
| | Female | 1777 | 1792 | 1784 | 1880 | 1892 | 1671 |
| 40-49 | Male | 2923 | 2937 | 2930 | 3846 | 3864 | 3855 |
| | Female | 2015 | 2026 | 2020 | 3650 | 3666 | 3658 |
| ≥ 50 | Male | 3167 | 3185 | 3176 | 4123 | 4142 | 4132 |
| | Female | 2190 | 2204 | 2197 | 3725 | 2736 | 3730 |

**Table (3): shows the thickness of the endocardium of the left atrium and left ventricle**

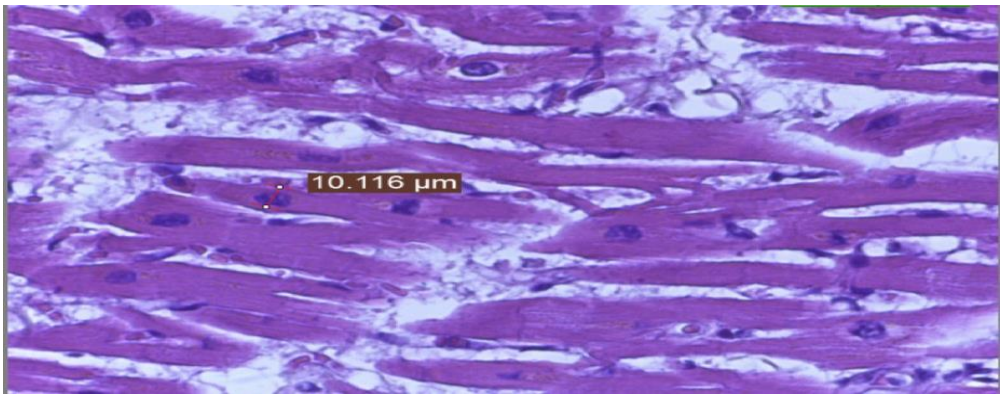
| Age Class | Sex | minimum thickness of endocardium of the left atrium | Maximum thickness of the endocardium in the left atrium | Mean | Minimum Thickness of the endocardium of the left ventricle | Maximum thickness of the endocardium of the left ventricle | mean |
|---|---|---|---|---|---|---|---|
| ≤29 | Male | 162 | 168 | 165 | 62 | 70 | 66 |
| | Female | 150 | 154 | 152 | 58 | 63 | 60.5 |
| 30-39 | Male | 162.5 | 169.2 | 165.6 | 78 | 89 | 83.5 |
| | Female | 156 | 160.5 | 158.2 | 62 | 90.3 | 76.15 |
| 40-49 | Male | 275 | 282 | 278.5 | 162 | 165.3 | 163.6 |
| | Female | 160 | 165 | 162.5 | 75 | 92 | 82 |
| ≥50 | Male | 310 | 315 | 312.5 | 165 | 175 | 170 |
| | Female | 250 | 256 | 253 | 196 | 100 | 148 |

**Table (4): shows the diameter of the cardio cyte of the left atrium and ventricle**

| Age Class (year) | sex | The minimum Diameter of cardio cyte of the left atrium | The maximum diameter of the cardiocyte of the left atrium | Mean | The minimum Diameter of the cardio ocyte of the left ventricle | The maximum diameter of the cardiocyte of left ventricle | Mean |
|---|---|---|---|---|---|---|---|
| ≤ 29 | Male | 15 | 17 | 16 | 17 | 22 | 19.5 |
| | Female | 10 | 12 | 11 | 12.5 | 15 | 13.8 |
| 30-39 | Male | 22.5 | 27 | 24,8 | 22.5 | 28 | 25.3 |
| | Female | 12.5 | 15.5 | 14 | 15 | 17 | 16 |
| 40-49 | Male | 25 | 30 | 27.5 | 25 | 30 | 27.5 |
| | Female | 15.4 | 19 | 17.2 | 20 | 23 | 21.5 |
| ≥50 | Male | 23 | 24 | 23.5 | 27.2 | 27.7 | 27.5 |
| | Female | 22 | 23.2 | 22.5 | 27 | 27.4 | 27.2 |

**Table (5): shows the diameter of purknjie fiber of the left atrium and ventricle in both sexes.**

| Age Class (year) | sex | The minimum Diameter of pruknjie of the left atrium | The maximum diameter of purknjie of the left atrium | Mean | The minimum Diameter of the pruknjie of the left ventricle | The maximum diameter of the pruknjie fiber of left ventricle | Mean |
|---|---|---|---|---|---|---|---|
| ≤ 29 | Male | 30 | 31.2 | 30.6 | 70 | 76 | 73 |
| | Female | 30 | 34.2 | 32.1 | 37 | 39 | 38 |
| 30-39 | Male | 27 | 42 | 34.5 | 75 | 77 | 76 |
| | Female | 42 | 44 | 34 | 70 | 72 | 71 |
| 40-49 | Male | 50 | 50.3 | 50.2 | 82 | 87 | 84.5 |
| | Female | 50 | 50.2 | 50.1 | 75 | 79 | 77 |
| ≥50 | Male | 70 | 72 | 71 | 90 | 93 | 91.5 |
| | Female | 75 | 78 | 76.5 | 82.5 | 85 | 83.8 |




**Figure(6): shows the diameter of the cardiocyte. stained with hematoxylin and Eosin, magnifications power= 500 um**

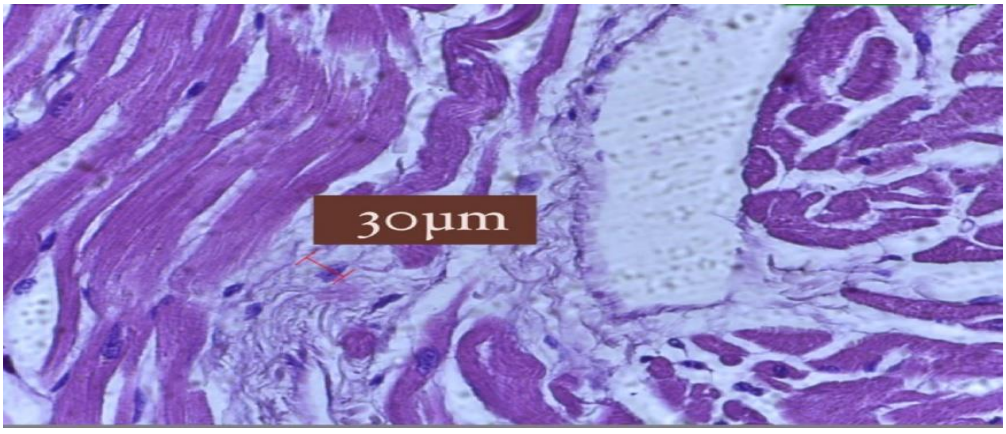



**Figure (6): shows the diameter of the purknjie fiber. stained with hematoxylin and Eosin, magnifications power=500 um**

**Discussion:**

- **Thickness of heart layers**

There was no contradict data on the measuring of the thickness of heart layers (epicardium, myocardium and endocardium) in adult human. The current study confirms theses result which finding the myocardium layer was the thickest layer and the endocardium was the thinnest layer. All layers correlated with the age and sex.

- **Diameter of the cardiocyte and pruknjie fiber of the left atrium and left ventricle:**

The finding of the diameter of the cardiocyte of this study was ranged from 10-30 um these had agreement with the pervious study M.P.Nash. [17]. The minimum diameter of the cardiocyte in the atrium was 10 um and in ventricle was 12.5 um these was agree with the S.D Bird...etc. [18] while the mean diameter of the pruknjie fiber in both atrium and ventricle ranged from 30-80 um this was agree with the Eliska O. [19]

The diameter of the cardiocyte in the ventricle in both male and female increased with age this results agree with the Iliev A A..etc. [20]

In the this study the diameter of the pruknjie fiber HASDEM˙IR in both left atrium and ventricle in both male and female in all age group was larger than the cardiocyte this finding result had strong correlation with the *Fosgren S.* [21]

**Conclusions:**

The study finding, the myocardium layer in all age groups and in both sexes was the thickest layer, the thickness of the epicardium layer was thicker in the left ventricle than the left atrium in

all ages. The endocardium layer was the thinnest layer of the heart wall and it was thicker in the left atrium than the left ventricle. The diameter of the cardio cyte in the left ventricle was larger than in the left atrium and the diameter of the purknjie fiber was larger than the cardiocyte in both atria and ventricle and in all age groups.

**Competing interest :**

The author declare that there is no conflict of interest.

**Author Contributions :**

Noor Abbas Ramadan: the research article proposal, experiment design, explaining the finding, and article writing and preparing materials, data curation, review and editing

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