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The Effect of Aging on Human Testis: Anatomical and Histological study

Saad AY Koprlo[1] *DVMS*, **Haider AR Ja'afar**[2] *MSc, PhD*, **Abdul-Karim S Mahood**[1] *MSc, PhD*

[1,3]Dept. of Anatomy, College of Medicine, Tikrit University, [2]Dept. of Anatomy, College of Medicine, Al-Nahrain University.

Abstract

Background Aging of testis is gradual dysfunction of body organs and tissues due to reduction of cell number and loss of cell capacity for reproduction and regeneration of its structural elements.

Objective To study the age related changes of the human testes anatomically and histologically.

Methods Forty testes of twenty adult Iraqi male cadavers, with age ranged from 20-69 years were taken from the Forensic Medicine Department of Tikrit and Azadi Teaching Hospital in Salahddin and Kirkuk province, respectively during the period from November 2006 to September 2007. The testicular specimens were divided into five groups according to cadaver age. Tunica albuginea was removed to investigate the lobular and tubular structures of the testis, by anatomical and histological procedures.

Results The gross metrical measurement and anatomical inspection revealed that a negative correlation was existed between the process of aging and the weight of the postmortem, testis. A strong directly proportional positive correlation was found between age and the thickness of basement membrane, interstitial spaces between seminiferous tubules. Tubular sclerosis was observed in some testis specimens of senescent cadavers associated with a decrease in the number of the lining epithelial cells of the seminiferous tubules and sclerotic changes in the walls of testicular blood vessels.

Conclusion The process of aging leads to testicular changes such as tubular diverticula and decrease in number of leydig cells.

Key words Aging, Leydig cells, senescent testis, seminiferous tubules.

Introduction

The aging is gradual dysfunction of body organs and tissues due to reduction of cell number and loss of cell capacity for reproduction and regeneration of its biochemical structural elements [1-4].

Testes as being small until puberty when they grow very quickly and reach maximum development in the period of the sexual activity and decreasing the volume and weight with age over 50 years old [3,5-7].

Age related change is a complex multifocal process as a result of gradual loss of the cell capacity for reproduction and regeneration of its biochemical structural elements [8-11]. Volume of the testis is a rough indicator of spermatogenesis remains constant over long period of life and reduction in paired testicular weights, total volume and seminiferous tubule volume, seminiferous epithelium and length of tubules [12-16]. Reduction of type-A dark spermatogonia, increased occurrence of multinucleated spermatogonia, megalospermtocytes as well as giant spermatids and leydig cells are the characteristic feature of senescence [17-19]. The average decreases in the production of testosterone in men usually over 40 years of

age, but in men the testes remains functional throughout life by spermatogenesis, as well as the synthesis of testosterone [20,21]. The diminished androgen production or spermiogensis might be reflected by a rise in gonadotropine serum levels and a testosterone decline with age [2224].

Aim of the study

To study the age related changes of the human testes anatomically and histologically.

Methods

Twenty normal adult Iraqi male cadavers, with age ranging from 20-69year's were taken during the period from November 2006 until June 2007 at the Forensic Medicine Department of Tikrit and Azadi Teaching Hospital in Salahddin and Kirkuk province, respectively. The agreement consent (permission paper) from the relatives of cadavers was performed for medico legal purposes. The cadavers were divided into five groups according to age and four in number for each group as follows:

- Group (A): ranged from 20–29 Years (control group).
- Group (B): ranged from 30–39 years.
- Group (C): ranged from 40–49 years.
- Group (D): ranged from 50–59 years.
- Group (E): ranged from 60–69 years.

Anatomical study: A longitudinal incision downwards through the skin of the anterolateral aspect of the scrotum. The testicular fascia and tunica albuginea were shelled out from the testis. This provides excellent exposure of testes, in order to examine extensions, Lobular structures of the testis (Figure 1).

The weight of the right and left testes was measured by an electronically weighing scale (Mettler AE 200, Japan). The total testis weight was then calculated by adding together the weight of both testes.

Histological Study: Fixation of the specimens was made using 10% formalin saline (100 ml of 40% formaldehyde, 9gm Sodium chloride and 900 ml tap water) for 24-48 hour [25]: Routine staining of sections was performed using haematoxylin and eosin stains (H&E) and Periodic Acid Schiff's Technique "PAS Technique":

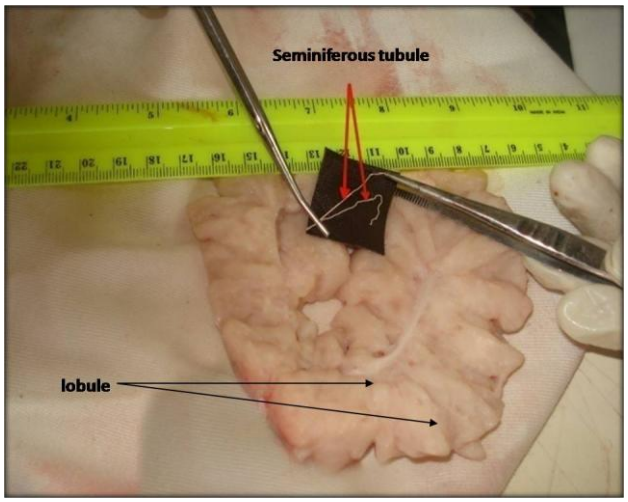

Figure 1. The highly convoluted seminiferous tubules (red arrows) and lobules (black arrows)

Measuring techniques by measuring the diameter of seminiferous tubules, and the interstitial space between the seminiferous tubules distance was measured by calibrated ocular lens and the calibrated stage was consisting of 100 minute lines is equal to (10 µm) and was performed, at 40 X objective lens, each line of the calibrated ocular lens is equal to 2.4 µm.

Results

Each testis consist of about (250- 300) lobules and each lobules has 1-4 seminiferous tubules which are highly convoluted tubules (40-70) cm length (Figure 1).

In groups B, C, D, and E the means weight of the right testes were 20.41±2.66, 20.64±2.24, 19.45±2.24 and 17.20±1.26 gm, respectively, whereas those of the left testes of the same groups were, 20.44±2.68, 20.60±2.25, 19.46±2.24 and 17.10±1.26 gm, respectively groups D and E had significant values compared with those of groups A (17.30 ± 1.53 gm) on right side, and 17.28±1.56 on left side

as control group (P<0.01) of both side (Table 1).

Table 1. Weight of testes according to age groups.

Group	No.	Rt. testis weight (gm)	Lt. testis weight (gm)
A	4	17.3 ± 1.53	17.28 ± 1.56
B	4	20.41 ± 2.66*	20.44 ± 2.68*
C	4	20.64 ± 2.24*	20.6 ± 2.25*
D	4	19.45 ± 2.24*	19.46 ± 2.24*
E	4	17.20 ± 1.26**	17.10 ± 1.26**

Group A is the control group
*= P<0.05, **= P<0.01 as compared to group A

It was appearing that the number of the lining epithelial cells (sertoli and spermatogonial cells) of the seminiferous tubule were decreased in testes specimens of groups D and E compared with those of groups A as control group (Figures 2&3).

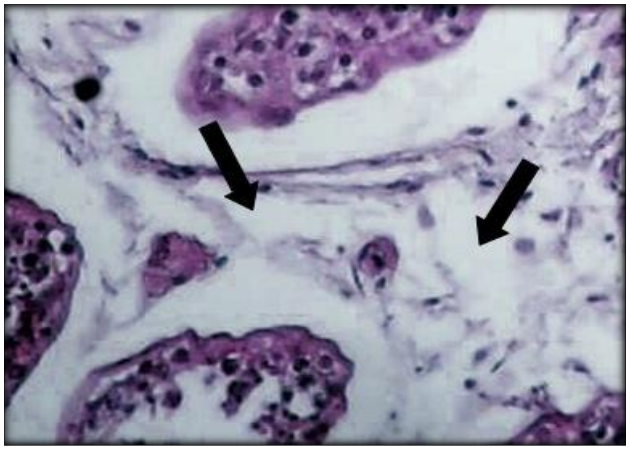

Figure 2. Testicular tissue showing the parenchyma with diffused small clusters of adipose cells (arrow), forming great part of testicular parenchyma in group D (H&E, 40X).

The adipose cells were few, scattered as a single cell in the parenchyma of groups A and B, while in group C, D and E were arranged as small clusters of adipose cells composed from many cells. The presence of adipose cells within the interstitial connective tissue gives a deep yellow appearance of testicular specimen section in these groups.

Thickening of the intertesticular – arterial walls was found in tissue sections of group E which appeared as a homogenous pink hyaline thickening in about 50% of small testicular vessels associated with narrowing of lumen and there was obvious thickening of basement membranes of seminiferous tubules at same groups (Figure 3).

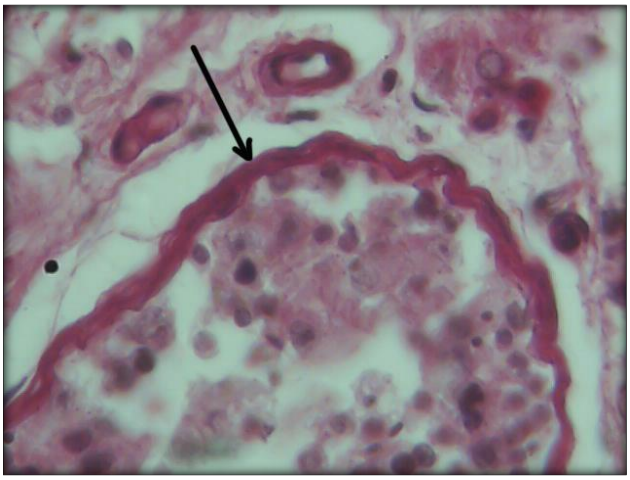

Figure 3. Thickening of the basement membrane of the seminiferous tubules in group E. (PAS, 100X).

The mean values of the seminiferous epithelial cells thickness in testis specimens were (52.27±2.74, 54.15±2.78 and 54.05±3.46 µm) in group A, B, and C, respectively, but means values reached values 44.10±4.35, 38.45±3.57 µm in groups D and E, respectively which is significant as compared with those of group A (control group) (P<0.01) as seen table 3.

Table 2. Mean and SD of the Thickness of the Sem. Epith. Cells and wall thickness of blood vessels according to age groups.

Group	No.	Thickness of the Seminephrous Epith. Cell	Wall thickness of Blood vessels
A	4	52.27 ± 2.74	11.58 ± 0.60
B	4	54.15 ± 2.78^{n}	12.85 ± 0.69*
C	4	54.05 ± 3.46^{n}	14.05 ± 0.75*
D	4	44.10 ± 4.35*	15.05 ± 0.49**
E	4	38.45 ± 3.57**	17.20 ± 0.42**

Group A is the control group
*= P<0.05, **= P<0.01 as compared to group A

The means of the interstitial spaces among the seminiferous tubules were, 27.78±1.63,

29.60±1.48, 39.22±3.47 and 44.72±1.75µm in group B, and C and D, respectively which revealed significant values as compared to group A (25.55±0.96) (P<0.01) as noticed in table 3.

Table 3. Mean and SD of the seminiferous Interstitial Space and Basement membrane` thickness of seminiferous Tubules according to age groups.

Group	No.	Seminiferous ISS between Tubules	BM thickness of Seminiferous Tubules
A	4	25.55 ± 0.96	2.97 ± 0.45
B	4	27.87 ± 1.63*	3.30 ± 0.20*
C	4	29.60 ± 1.58*	3.87 ± 0.29*
D	4	39.22 ± 3.47**	4.07 ± 0.40**
E	4	44.72 ± 1.75**	4.62 ± 0.38**

ISS = interstitial space, BM = basement membrane.
Group A is the control group
*= P<0.05, **= P<0.01 as compared to group A

The means of Basement membrane thickness in group B,C,D and E were, 3.30±0.20, 3.87±0.29, 4.07±0.40 and 4.62±0.38 µm, respectively which revealed significant values as compared to control group A (2.97±0.45), (P<0.01) as sown in table 3.

The measuring of blood vessels wall thickness revealed that there was an increment in the thickness of the blood vessels as, 12.85±0.69, 14.05±0.75, 5.05±0.49 and 17.20±0.42µm in groups B,C,D and E, respectively and statically significant as compared to control group A, (11.58±0.6) (P<0.01) as demonstrated in table 2.

The mean values of the diameter of seminiferous tubules in testes specimens of present study at magnification power 400 X, were 225.70±5 and 232.75±7.37 µm in groups B and C, respectively, but these means showed a marked decrease and reached significant values to 228.50±13.29, 187.75±11.47µm, in groups D and E, respectively, as compared to control group A, (190.50±1.2) (P<0.01) as shown in table 4.

Table 4. Diameters of seminiferous tubules according to age groups.

Group	No.	Age (year)	Diameter of seminiferous tubules (µm)
A	4	20-29	190.50±1.29
B	4	30-39	225.70±5.00**
C	4	40-49	232.75±7.37**
D	4	50-59	228.50±13.29**
E	4	60-69	178.75±11.47*

Group A is the control group
*= P<0.05, **= P<0.01 as compared to group A

Discussion

There was a tendency of human testes to contain more adipose with aging and they could be a factor that would accelerate aging process, this observation agrees with other authors [4,8], stated that there is an increase in connective tissue and adipose tissue by aging .

Regarding the weight of the testes specimens, it has been found that a good negative correlation was existed between age progress and the testes weights. other authors [3,5-7,16], mentioned that there was a slight decrease of absolute testicular weight starting at the age of 42.5 years, due to tubule involution which associated with an enlargement of the tunica propria leading to progressive sclerosis parallel to a reduction of the seminiferous epithelium with complete tubular sclerosis as an end point.

The histological finding showed that the diverticula appeared in some seminiferous tubules in aged groups as evagenations of the seminiferous epithelium towards testicular interstitium. The diverticula were connected to the seminiferous tubules by a narrow neck or by a wide base. Other study which mentioned that peritubular myoid cells may be affected by hormone alterations that take place with increasing age [18, 34], whereas Laporte and Gillet [35] stated that besides the peritubular cells, Sertoli cells might also be involved in the formation of diverticula. Since human sertoli cells undergo morphologic alterations with aging leading to a progressive decline, it is

probable that these alterations may compromise sertoli cell function [28].

In addition to the diverticula appearance in some seminiferous tubules in aged groups, is a reduction of the seminiferous epithelium (spermatogonium and sertoli cells). This finding was in agreement with other authors [29, 30] who mentioned that the thickening of tunica propria related to the tubular involution leading to the progressive sclerosis parallel to a reduction of seminiferous epithelium with complete tubular sclerosis as an end point.

The histomorphometric study of the testicular tissue specimens showed a distinct negative correlation between age and number of seminiferous tubules. This finding can be attributed to the tubular involution of the testes as age- related dependent change which was shown in present study and was generally similar to the finding of other authors [8,10,24,30], they reported that there was a reduction of the seminiferous tubules due to age-related reduction of blood supply to the testicular parenchyma, and sclerotic changes in the walls of the testicular blood vessels.

A marked decrease in thickness of the seminiferous epithelial cells was observed in testis specimens of groups D and E. According to Johnson [17] and Holstein [20] this finding was because of degenerative changes due to physiological germ cells loss observed in the germinal epithelium of elderly men. Sertoli cells also can undergo degenerative changes by accumulation cytoplasmic lipid droplet and multinucleated patterns. Harbitz [21] and paniagua *et al* [22] who mentioned that the process of aging leads to decrease in number of the seminiferous epithelial cells.

A highly significant positive correlation was found between the process of aging and the interstitial space between seminiferous tubules and due to tubular involution, and other reason that underlies this increase in tubule-interstitial spaces was the interstitial fibrosis which occurs mainly due to age-related increased content of collagen in parenchyma of testis [8, 15, 29, 31].

Regarding the thickening of basement membrane of seminiferous tubules of testes in specimens of the present study showed a distinct positive correlation with age, this can be attributed to the increase of collagen, and the increased content of various laminin isoforms with in the basement membrane of seminiferous tubules and gets thicker with age [33].

The sclerotic changes seen in the walls of testicular blood vessels in correlation to age can be attributed to the age-related sclerotic changes in the arteries wall of the testis parenchyma take place; This finding was in consistent with findings of authors which mentioned that testicular sclerosis was associated with defective vascularization of testicular parenchyma and with systemic arteriosclerosis of aged men with arteriographic studies [10,28].

It was evident in the present study a good negative correlation between age and the diameter of the seminiferous tubules. other authors [15, 34], with respect to old age revealed collapsed seminiferous tubules lined by Sertoli cells, incomplete spermatogenesis and the seminiferous tubules, and efferent ductules were devoid of spermatozoa. Whereas Wolf [33] cited that, mean diameter of seminiferous tubules in young men was (189±2.8 µm), and it reduced in the elder men to (150±3.7 µm).

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Correspondence to: Dr. Haider AR Ja'afar
E-mail: haiderabid@yahoo.com

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