

Effect of Age on Histological Changes of Urogenital Tract in Albino Rats**Fatima Qasim Mohammed Al-hayyali**

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<http://creativecommons.org/licenses/by/4.0/>.**Abstract**

This study was conducted to investigate the histologic changes of kidney, urinary bladder, testis, seminal vesicle and prostate gland of rats of various ages (6,12 and 18 months old). Results illustrated variable changes of kidney represented by congestion of renal capillaries, acute tubular necrosis, shrinkage of glomerular tuft, degeneration in tubular epithelium of the 6,12 and 18 months old rats, the morphometric changes of the glomerular diameter decreases with advanced age. The lesions of urinary bladder characterized by hyperplasia of the epithelial lining, papillae into the lumen, hypertrophy of muscle fibers, desquamation of epithelium, cystitis in addition to thickening of the mucosa and hyalinization of muscle fibers of the 6,12 and 18 months old rats. The thickness of the bladder wall showed a significant changes increase at ($P \leq 0.05$) with age, while morphometric analysis did not show any age related changes in the bladder muscle thickness. In testis there were congestion of blood vessels, a spermia, degeneration of the spermatocytes and spermatozoa, thickening of interstitial and hyperplasia of leydig cells of the 6,12 and 18 months old rats. The wall thickness of Seminiferous tubule increases with age and basement membrane thickness and tubular diameter decreased with age. The seminal vesicle of the 6,12 and 18 months old rats revealed hyperplasia of glandular epithelium. Lesions of the prostate gland of 6,12 and 18 months old rats showed epithelial hyperplasia which extended as finger-like projections and presence of variable amounts of colloid substance.

Keywords: Histological changes, Morphometric changes, Aging, Rats, Seminal vesicle, Prostate

تأثير العمر على التغيرات النسجية لأعضاء الجهاز البولي والتناسلي في الجرذان البيضاء

الخلاصة

تضمن البحث معرفة التغيرات النسجية الشكلية لنسيج كل من الكلية، المثانة البولية، الخصية، الحوصلة المنوية والبروستات للجرذان وبأعمار مختلفة (6 و12 و18 شهر). وأوضحت النتائج تغيرات متباينة في الكلية تمثلت بأحتقان الأوعية الشعرية الكلوية، تنخر نبيبي حاد، أنكماش اللمة الكبيبية، تنكس في الظهارة النبيبية في المجاميع الثلاث (6 و12 و18 شهر)، وبيئت التغيرات الشكلية صغر قطر الكبيبة مع تقدم العمر. وتميزت آفات المثانة البولية بفرط تنسج البطانة الظهارية، وجود حليمات داخل تجويف المثانة، ضخامة الألياف العضلية، توسف في ظهارة المثانة، تجمع مواد نضحية في تجويف المثانة بالإضافة الى تثخن في الطبقة المخاطية والألياف العضلية مصابة بتنكس زجاجي في مجاميع الجرذان الثلاث 6 و12 و18 شهر. وسبب تثخن جدار المثانة تغيرات معنوية عند مستوى ($P \leq 0,05$) مع تقدم العمر في حين لم يظهر التحليل الشكلي لسمك الطبقة العضلية للمثانة أي تغيرات مرتبطة بالعمر. ووجد في نسيج الخصية أحتقان في الأوعية الدموية، فقدان الحيامن، تنكس الخلايا المولدة المنوية والخلايا المنوية الأولية، تثخن النسيج الخلالي للخصية وفرط تنسج خلايا ليديك في المجاميع الثلاث 6 و12 و18 شهر. وبين التحليل الشكلي زيادة في سمك جدار النبيبات المنوية مع تقدم العمر وانخفاض في سمك الغشاء القاعدي وقطر النبيبي المنوي مع تقدم العمر. وأظهرت الحوصلة المنوية في الجاميع الثلاث 6 و12 و18 شهر فرط تنسج للظهارة الغدية. وسببت آفات غدة البروستات في المجاميع الثلاث فرط تنسج للظهارة والتي أمتدت كبروزات شبيهة بالأصابع مع وجود كميات متباينة من مواد غروية في تجويف الظهارة. نستنتج من هذه الدراسة أن تقدم العمر في الجرذان مرتبط بتغيرات نسجية وشكلية غير طبيعية في المجرى البولي التناسلي.

Introduction

Aging is a syndrome of changes or biological process accompanied by gradual deterioration of the physiological function and metabolic process. This multifactorial process which is affected by the sum of genetic, diet, social and environmental factors, encompasses various organs and tissues (1). Aging damage occurs to molecules (DNA, proteins, lipid) and to cell and organs (2). There are physiological and structural alterations with age in almost all organ systems often without concomitant changes in body weight and body mass index (3). In addition, there is good evidence that aging is the result of progressive decline in the proliferative capacity and life span of cells and the effects of continuous exposure to exogenous influences that result in the progressive accumulation of cellular and molecular damage (4,5). The common age - related diseases include renal failure, osteoarthritis, muscle atrophy, cerebral atrophy from loss of cortical neurons, cardiovascular disease and reproductive disease (6,7). Aging is associated with declined renal function represented by glomerulosclerosis, tubular atrophy, proteinuria, reduced ability for concentration, impairment of electrolyte and ion transport, alteration in hormonal function and reduced bladder capacity (8). Aging changes in the male reproductive system include moderate decline of testicular gametogenic and endocrine function (7). The prostate gland enlarges with age as some of the prostate tissue is replaced with scar like tissue and this case is called benign hyperplasia (9) and it may cause problems to urination as well as to ejaculation (10). Also increased systematic oxidative stress which related with age may be lead to prostate cancer (11). One of the most important risk factors in the development of aging is the changes in reproductive system which is closely related to changes in the urinary system. The aim of this study was to observe the histopathological changes in aging kidney, urinary bladder, testis,

seminal vesicle and prostate.

Materials and methods

The experimental study included thirty albino rats which were divided into three groups of equality distributed ($n = 10$), young adult (6 months old), adult (12 months old) and adult (18 months old) male rats that were obtained from the animal house in the College of Medicine, Mosul University. They were fed ad libitum and had free access to water. All samples which were obtained from the kidney and bladder, prostate, testis and seminal vesicles of rats were cut into 0.5 cm pieces fixed promptly in 10% buffered neutral formalin, followed by a dehydration using a series of graded ethanol in serial concentrations (50%, 70%, 95%, 100%), immersed in xylene for clearing, infiltrated with paraffin wax. Four micrometer thick paraffin sections were obtained by using rotary microtome (Bright, MIC) and stained by hematoxylin and eosin examination (H&E) (12). Photography of sections was done with digital camera. Microscopic measurements taken using Ocular meter lens on 400x. The procedure employed was to examine 5 representatives (glomerular diameter, bladder wall thickness, bladder muscle thickness, wall thickness of Seminiferous tubule, basement membrane thickness, Seminiferous tubule diameter) sections per animal, viewing 10 randomly chosen measures of each sections and calculating their mean.

Statistical analysis

All data were expressed as mean \pm standard error of mean ($M \pm SE$) and statistical analysis was carried out using statistically available software of statistical package for social science (SPSS) version. One way analysis of variance (ANOVA) was performed to test for significance followed by Duncan multiple range comparison tests for comparisons between the groups ($P < 0.05$) and 0.01 were considered statistically significant.

Results and discussion

Microscopic examination of sections prepared from the specimens of rat's kidney (6months old) revealed presence of congestion of renal capillaries , acute tubular necrosis and reduction or shrinkage of glomerular tuft (Fig1) .Sections of the kidney of the 12 month old rats showed shrinkage of glomerular tuft and degeneration in tubular epithelium (Fig 2). Sections of the kidney of the 18 month old rats showed similar lesions in addition to tubular dilation (Fig 3). The morphometric changes of glomerular diameter from the three groups of rats showed significant difference between different groups (6 months mean was $53.180 \mu\text{m} \pm 2.252$; 12 months, mean was $46.900 \mu\text{m} \pm 1.005$; 18 months, mean was $35.000 \mu\text{m} \pm 1.037$).

The histological sections from rat's urinary bladder (6month old) showed hyperplasia of the transitional epithelial lining the urinary bladder and it's extension as papillae into the lumen , hypertrophy of muscle fibers, desquamation of epithelium of the lining mucosa and accumulation of exudate in lumen of bladder (Fig 4) . In sections of urinary bladder of the 12 month old rats showed thickening of the mucosa, hyalinization of muscle fibers in muscular layer, and desquamation of epithelial mucosa into lumen (Fig 5). Sections of the urinary bladder of the 18 month old rats showed changes similar to those seen in 6 and 12 months old rats in addition to hyperplasia of the transitional epithelial mucosa (Fig 6). The wall thickness of bladder from three group showed significant difference between 6 and 12,18 months of old rats and not found between 12 and 18months old rats (6 months, mean was $4.12\mu\text{m} \pm 0.485$; 12 months, mean was $12.52\mu\text{m} \pm 1.830$; 18 months, mean was $12.78\mu\text{m} \pm 1.125$). Bladder muscle did not show significant difference between three groups of rats (6 months' mean was $50.6 \mu\text{m} \pm 6.713$; 12 months mean was $61.00\mu\text{m} \pm 5.079$; 18 months, mean was $68.76\mu\text{m} \pm 2.211$).

The sections which prepared from the testis of the 6 month old rats showed congestion of blood vessels in the interstitium , loss of sperms from the lumen of some seminiferous tubules , in addition to degeneration of spermatocytes and spermatozoa (Fig 7).Testis of the 12 months old rats exhibited severe vacuolar degeneration of the spermatocytes and lack of spermatozoa and hyperplasia of Leydig cells (Fig 8). In sections of the testis of the 18 months old rats there were degeneration of spermatocytes thickening of interstitium and hyperplasia of Leydig cells(Fig 9).Sections of the seminal vesicles of the 6,12 and 18 months old rats exhibited hyperplasia of glandular epithelial lining the tubular vesicular glands (Fig 10,11). Seminiferous tubule wall thickness showed significant difference between 6 and 12,18 months, and there was no significant difference between 12 and 18months old rats (6 months, mean was $3.46\mu\text{m} \pm 0.150$; 12 months $9.88\mu\text{m} \pm 0.418$; 18 months, mean was $11.00 \mu\text{m} \pm 0.443$).The thickness of basement membrane of the Seminiferous tubule wall showed significant difference, in which the tubule wall thickness decreased with age, (6 months, mean was $3.660\mu\text{m} \pm 0.172$; 12 months, mean was $0.880 \mu\text{m} \pm 0.097$; 18 months, mean was $0.540\mu\text{m} \pm 0.067$). Seminiferous tubule diameter showed significant difference between 12 and 18 months of old rats .(6 months, mean was $26.01 \mu\text{m} \pm 0.811$; 12 months , mean was $37.120 \mu\text{m} \pm 1.085$; 18 months , mean was $31.830 \mu\text{m} \pm 1.522$). Sections of the prostate gland of the 6 months old rats exhibited hyperplasia of epithelial lining the wall of acinior alveoli and the presence of variable amounts of colloid substances (Fig 12). The prostate gland of the 12 month old rats exhibited hyperplasia in epithelial lining the wall of alveoli which extended as finger- like projections (Fig 13). The prostate gland of the 18 month old rats showed changes similar to those seen in the 12 month old rats (Fig 14). In the present study lesions that occurred in the kidney of aging rats included shrinkage of the glomeruli, acute tubular necrosis, and tubular

dilatation. So for these results the morphometric changes of the glomerular diameter will demonstrate decreases with advanced age (12 and 18 months groups), These findings are in accordance with those of other findings (13,14,15). Other researchers found that there is decrease in body physiology, reproductive activity and renal mass and an increase in disordered inflammation and incidence of sclerotic glomeruli with advanced age in human (16,17). Histopathology of the urinary bladder in rats of the three age groups (6,12 and 18 months) revealed papillary hyperplasia of the transitional epithelium, hypertrophy and hyalinization of the muscular layer, desquamation of the lining epithelium, and cystitis. Thickness of the bladder wall revealed significant changes between 6 months and 12,18 month groups in which bladder wall thickness increases with age, while morphometric analysis did not show any changes with age in the bladder muscle thickness and bladder function, Similar thickening of the aged rat bladder has also been reported by others (18,19,20). While (21) are revealed that the alteration of genes which associated with aging may lead to the functional deterioration in mice bladder. In the testis of the aged rat the changes that were seen in this study included degeneration of the spermatocytes and spermatozoa, lack of spermatozoa, and hyperplasia of the interstitial cells, also in this study the wall thickness of Seminiferous tubule showed significant differences between three groups in which increases in tubular wall with increases of age which must be due to pathological changes as seen in fig 8,9, also our results indicate that basement membrane thickness is decreased with age. These changes agreed with (22,23) who reported decrease of basement membrane (seminiferous tubules) thickness and an increase in the lamina propria with aging in rats. Morphological studies showed by (24) revealed that Leydig cells were more abundant in the testis interstitium at 6 and 24 months when compared to 3 months. Other changes that have been

reported by others investigators include narrowing of tubular diameter, thickening of the basement membrane, reduced in the number of sertoli and spermatogenic cells, and the appearance of multinucleated giant cells (25,26). An important role of basement membrane is maintaining the integrity of tissues (26). Therefore, alteration of basement membrane structure has been associated with severe functional impairment of the testis in several conditions, including vasectomy, autoimmune orchitis, cryptorchidism and following x-irradiation (27). In histological study of spermatogenesis in 30 men who died of trauma or myocardial infraction, spermatogenesis, as defined by spermatid nuclei, decreased with increasing age (27). Furthermore, the age-associated imbalances between proliferation and programmed cell death lead to defected in spermatogenesis and infertility (28) In the prostate of the aging rats included papillary hyperplasia of the lining epithelium and the presence of variable amounts of colloid substances. These changes are agreement with (29).

Conclusion

This study concludes that the variable histological and morphological changes of urogenital tract is related with aging in the rats. Our observation allows us to advance the histomorphometric changes which effects on the capacity of kidney, bladder, testis, seminal vesicle and prostate to respond to a variety of physiologic and pathologic stresses and then effects on function of these organs. Severe changes began around 18 months although the histomorphometrically changes in these organs was detectable at 12 months. thus these changes may explain that the one of the important factors in the development of aging is the changes in reproductive system which is related to changes in the urinary system.

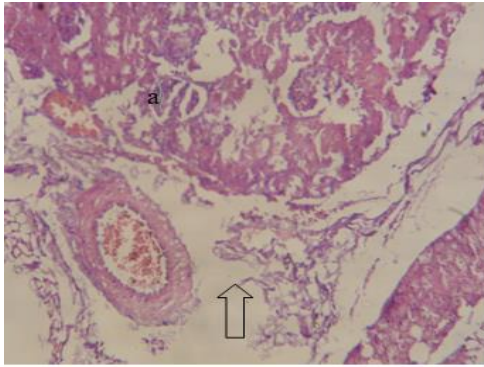


Fig1. Section of a rat kidney at 6 month of age showing shrinkage in glomerular tuft (a), and increase in interstitial space (arrows). H&E.106105 X

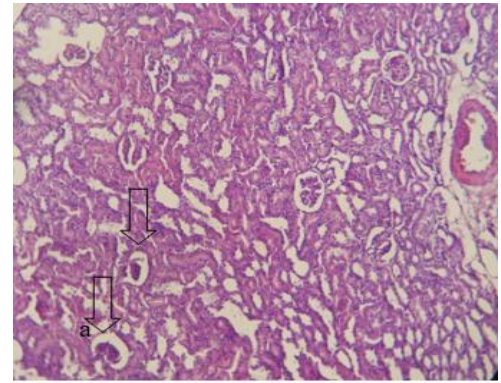


Fig 2. Section of a rat kidney at 12 month of age showing shrinkage in glomeruli (arrows). H&E. 105 X

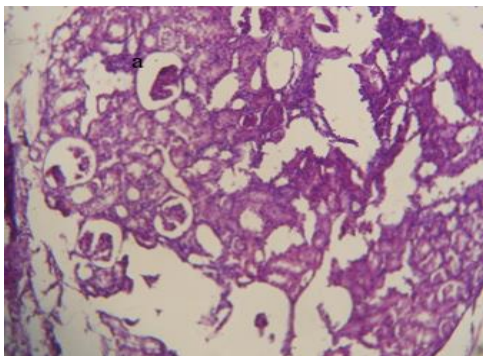


Fig 3. Section of a rat kidney at 18 month of age showing tubular dilatation as above. H&E. 105 X

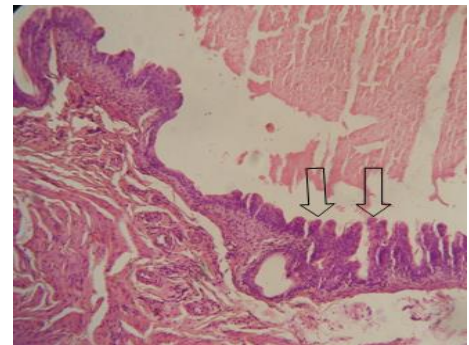


Fig 4. Section of rat urinary bladder at 6 month of age showing hyperplasia of the transitional epithelial lining the urinary bladder and it's extension as papillae into the lumen (arrows). H&E. 105 X

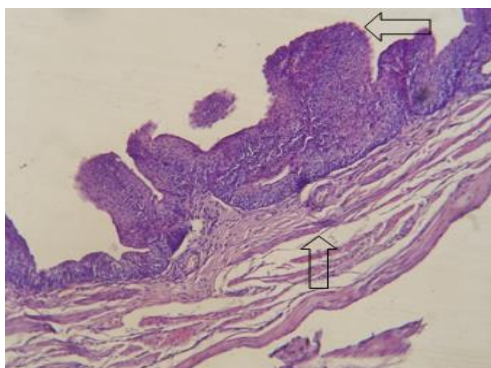


Fig 5. Section of rat urinary bladder at 12 month of age showing irregular thickening of the mucosa and hyalinization of muscle fibers in muscular layer (arrows). H&E 105 X

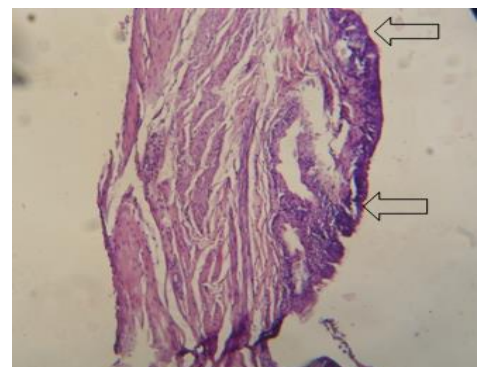


Fig 6. Section of rat urinary bladder at 18 month of age showing hyperplasia of epithelial lining (arrows). H&E. 105 X

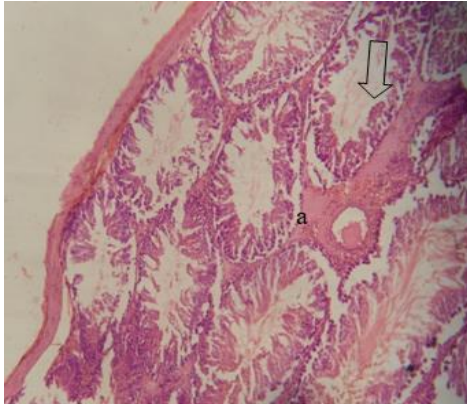


Fig 7. Section of a rat testis at 6 months of age showing congestion of blood vessel in the interstitium (a) and degeneration of the spermatocytes and spermatozoa (arrows). H&E.

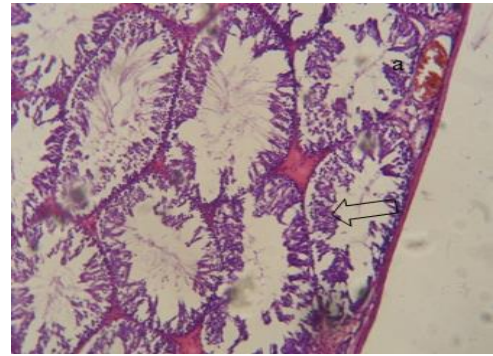


Fig 8. Section of a rat testis at 12 month of age showing congestion (a) vacuolar degeneration of the spermatocytes and hyperplasia of Leydig cell (b) and lack of spermatozoa (arrows). H&E. 105 X

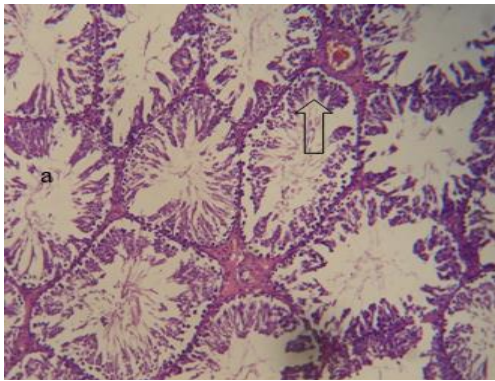


Fig 9. Section of a rat testis at 18 month of age showing thickening of intrstium associated with hyperplasia of Leydig cells (a) and degeneration of the spermatocytes (arrows). H&E. 105 X

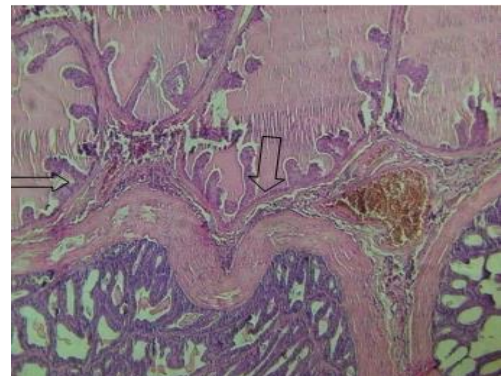


Fig 10. Section of a rat seminal vesicle at 6 month of age showing hyperplasia in epithelial lining (arrows) . H&E. 350 X

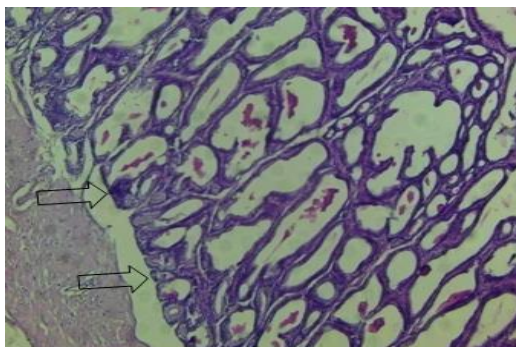


Fig 11. Section of a rat seminal vesicle at 6 month of age showing hyperplasia in epithelial lining (arrows). H&E. 105 X

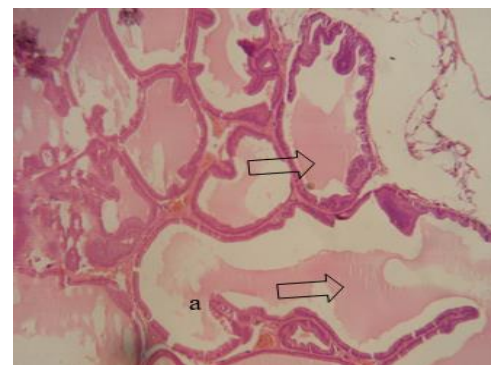


Fig 12. Section of a rat prostat at 6 month of age showing hyperplasia of the epithelial lining the alveoli (a) and the presence of variable amounts of colloid substances (arrows). H&E.

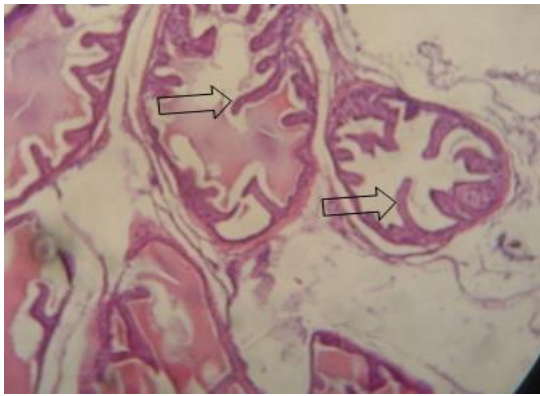
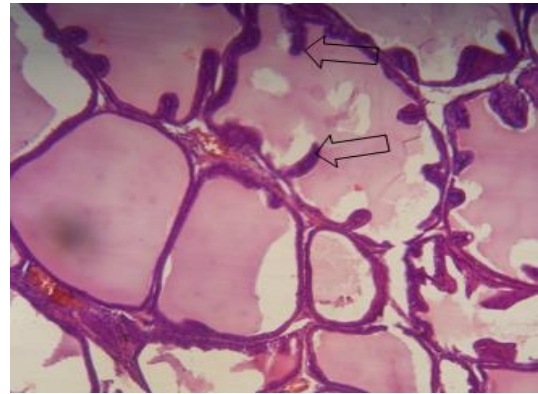


Fig 13. Section of a rat prostate at 12 month of age showing hyperplasia in epithelial lining the alveoli which extended as finger – like projections (arrows). H&E. 105X



8.

Fig 14. Section of a rat prostate at 18 month of age showing hyperplasia of epithelial lining the alveoli which extended as finger –like projections into lumen (arrows). H&E. 105 X

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