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Estimation of Salicylic Acid (aspirin) on Sperm Specification and Histopathological Effects in Testis of Male Albino Rats (*Rattus ruttus*)

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Abstract.

Introduction *Excessive and uncontrolled use of most sedatives by patients suffering from limited-range pain or chronic pain that considered a threat to the general health of the body and does not recover over time.*

Material and Methods: *Twenty-five white rats were used, the group was divided into five subgroups, each containing five rats for one months (main groups) after oral administration of 50, 100, 150 and 200 mg/kg/day. Each main groups dealing with the control group receiving normal physiological saline (five rats), the second group (five rats) given with 50 mg/kg/day of the drug, the third group (five rats) given with 100 mg/kg/day of the drug, the fourth group (five rats) was given 150 mg/kg/day of the drug and the fifth subgroup (five rats) administered with 200 mg/kg/day of the drug. The rats were slaughtered after the groups received the treatment for two months. in addition to semithin sections for light microscopic examination, followed by statistical studies.*

Results *showed widespread pathological effects such as the appearance of hypertrophic epithelium in the lumen and an apparent decrease in both the wall thickness of the seminiferous tubules and the number of their cell layers, in addition to a marked decrease in the number of spermatogonia in the scaly spermatogenic cells in the lumen of the seminiferous tubules. The intensity of virulence increases gradually with increasing concentration and prolonging exposure time*

Conclusion*: Aspirin can cause significant but long-term harm. so this attempt was to determine the effect of aspirin on many physiological criteria and several histological indicators in the testes of male rats*

Keywords*: NSAID, Aspirin, testes****.***

I. INTRODUCTION

Muscle stress resulting from life demands and psychological stress leads to an exacerbation of the flow of hormones as an immediate response to the imbalance in cellular balance. This imbalance appears in side effects, the most important of which are headaches, tension, and sometimes a rise in temperature accompanied by the use of sedatives and anti-anxiety drugs(Chu *et al*., 2024). One of the most important indicators of pain is the hormone prostaglandin. The prostaglandins, thromboxanes, and leukotrienes are among the highly active molecules in the eicosanoid's family (Tang et al., 2022). They have an impact on many different bodily functions, including inflammation. Aspirin consumption inhibits the synthesis of prostaglandins and thromboxane, causing both favorable and undesirable consequences (Prizment et al., 2020). The NSAIDs were created to function similarly to aspirin with less negative effects. There are several NSAIDs now on the market, and choosing which agent to use relies on a number of parameters. Surprisingly, current research indicates that certain NSAIDs could prevent the body from mending (Müller, 2019). Acetaminophen has several significant therapeutic applications despite not

being an NSAID (Pergolizzi et al., 2021). Having knowledge of these medications' actions and any potential side effects, the sports trainer can work with the team doctor to create an aspirin-free plan (Patterson et al., 2022).

EXPERIMENTAL ANIMALS

The own treaties will be dealing with 25 male albino rats (*Rattus rattus*) with weights between 200 and 230 g they were gotten from the University of Kufa's Faculty of Science's animal house. The rats are housed in 40 cm long by 23 cm wide by 20 cm high plastic cages with metal coverings that were made especially for them. The rats are kept in the proper laboratory settings, which include a temperature range of 20 to 25 degrees Celsius, a light/dark cycle of 10 to 14, a ventilation rate of 15 to 20 breaths per hour, and a relative humidity range of (30 – 70) %.

Drug Used

The treated groups with aspirin with 100 mg doses by using therapeutic doses and calculated for the weight of the rats. The drug has been dissolved it in a normal saline (N.S) for the purpose of preparing different doses of the compound and according to the groups mentioned in the design of this study (Barbaud et al., 2020). The selection of N. S as diluting liquid depending on the aspirin doses that are supposed to be given when doubling the volume of the initially prepared solution (HADI & ABOOD, 2022) as follow:

1- Group one (five rats) was administrated 1 ml of normal saline per day as control group.
2- Group two (five rats) was administrated 50 mg/kg taking 1 ml from prepared solution per day
3- Group three (five rats) was administrated 100 mg/kg taking 1ml from prepared solution per day.
4- Group four (five rats) was administrated 150 mg/kg taking 1ml from prepared solution per day
5- Group five (five rats) was administrated 200 mg/kg taking 1ml from prepared solution per day.

SACRIFICE OF THE ANIMALS

Animals has been scarified after end of experiment by mix of ketamine: Xylazine (90mg/kg:10mg/kg intra peritoneal), used ketamine 0.5ml and xylazine 0.1ml to each 250g of body weight for anesthesia the animals for all groups. Saved the testis in containers contain ten percent formalin (Imarah et al., 2022).

Rats have been anesthetized utter, the blood samples for hormonal analysis (FSH, LH, and testosterone) have been collected from the right ventricle of the animals via a syringe. The blood samples were collected into the gel tubes, also the plasma samples have been obtained by centrifuging the tube at 2–8C and at 1000g for 15 min. The hormonal analyses were performed by using the commercially suitable kits and according to the instructions of the manufacturer (Ali et al., (2022).

HISTOLOGICAL PREPARATIONS

All samples have been fixed after dissected from animals in containers contains 10% formalin (38%100ml formalin in 900ml tap water) to be ready for processing them in following steps in (Talib Dawod & Hassan Abood, 2022).

Resalt

Testis function test

Effect of Interactions of Oral administration of Different Concentrations of Aspirin on testosterone, LH and FSH (ng/ml) in Male Rats.

The results displayed a significant increase ($p<0.05$) in FSH levels (369±0.29, 523.00±0.32, 539.50±0.22, 634.2±0.29) ng/ml and LH levels (140.5±0.32, 157.35±0.22, 163.00±0.32, 167.1±0.32) ng/ml concentrations respectively, whereas; the testosterone levels showed a significant decrease ($p<0.05$) (2.48±0.29, 1.76±0.4 , 1.44±0.32 and 1.31± 3.4) ng/ml after oral administration of concentrations 50,100,150 and 200 mg/kg/day of aspirin compared with control group (363.5±0.29, 132.25±0.29 and 2.95±0.39) ng/ml,

Aspirin mg/kg/day	Testosterone (ng/ml) (Mean± S.E)	LH (ng/ml) (Mean± S.E)	FSH (ng/ml) (Mean± S.E)
Control	2.95±0.39 (E)	132.25±0.29 (A)	363.5±0.29(A)
T1/50	2.48±0.29 (D)	140.5±0.32 (B)	369±0.29(B)
T2/100	1.76±0.4 (C)	157.35±0.22 (C)	523.00±0.32(C)
T3/150	1.44±0.32 (B)	163.00±0.32 (D)	539.50±0.22(D)
T4/200	1.31± 3.4(A)	167.1±0.32 (E)	634.2±0.29(E)

HISTOLOGICAL STUDY

Histopathological alterations in the testis of male rats in various concentrations of aspirin (figures B to D). Figures B to D show mild to severe histopathological changes in the testis of male rats given varying doses of aspirin. such as severe hypertrophied epithelium projecting into the lumen and an apparent decrease in both the thickness of the seminiferous tubule wall and the number of its cell layers, as well as a marked reduction in the number of sperm counts in the lumen desquamated spermatogenic cells in the lumen of a seminiferous tubule.

Degeneration of germinal epithelia, duplication of Leydig cells, and expansion of intratubular space with high dosages of Aspirin Testicular amyloid consists of extracellular accumulation of homogeneous, eosinophilic, and amorphous material in the interstitiaium, which appears pinkish in color and appears to cause significant sloughing of testicular interstitial cells in the high dose treated rats, according to the histoarchitecture of the testes

The results of this study confirmed the occurrence of hypertrophy of the testicle and enlargement of the seminiferous tubules with a stop in sperm production. The testicular aging process is quite subjective, and several research have looked into the effects of nonsteroidal anti-inflammatory medicines (NSAIDs) in cellular and animal models of testicular aging.

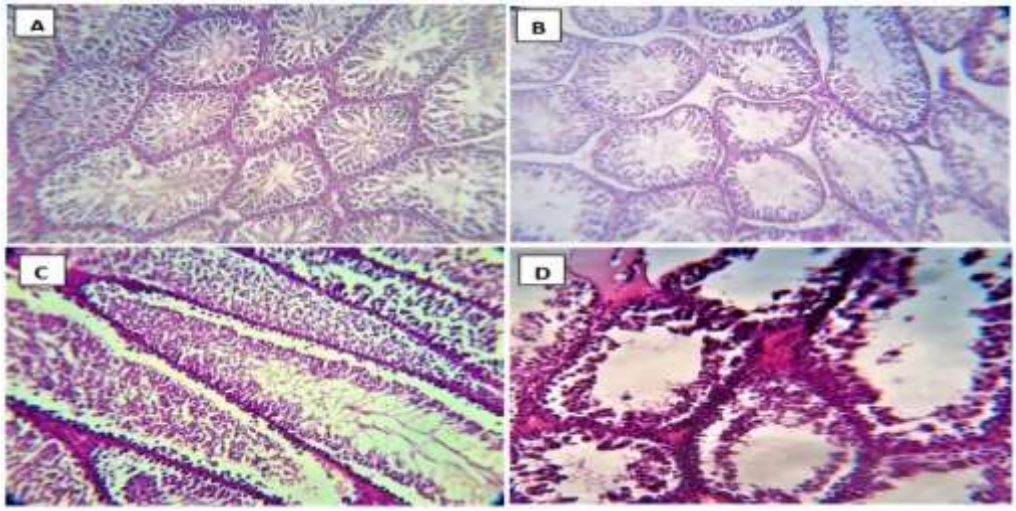

FIGURE 1. A: control, B: shows decrease in both thickness of the seminiferous tubule wall and number of its cell layers with apparent marked reduction of the sperms counts in the lumen and Leydig cells duplicating, C: shows homogenous acidophilic material and D: shows the desquamated spermatogenic cells in the lumen of a seminiferous tubule, seminiferous tubules lined by only one or two cell layers. (H&E stain, 100x).

III. DISCUSSION

The results of the current study showed that aspirin administration at 50, 100, 150, and 200 mg/kg daily for one month had a detrimental effect on the histologic structure of the rat testis as well as the hormonal indicators of sexual status, such as testosterone hormone, luteinizing hormone, and follicle-stimulating hormone. Additionally, there were obvious differences between the testis of the treated and control rats when different testis sections were examined under a light microscope. The testis of the aspirin-treated groups showed anomalies and histological changes, including the aspirin-treated groups' tested testes displayed abnormalities and histological

changes, such as severely hypertrophied epithelium projecting into the lumen and an apparent reduction in the thickness of the seminiferous tubule wall and the number of its cell layers, with an apparent marked reduction.

This study has demonstrated that, whereas rats' plasma levels of steadily decreased over the study period, levels of LH only marginally increased, which the researchers believe is likely due to cyclic hypothalamic release of LHRH (Naji et al., 2022; Álvarez-Maestro et al., (2021).

According to the Pituitary LH release is under feed-back control being controlled by plasma level of T, an increase in both LH and FSH is generated concurrently with a drop in plasma level of Testosterone hormone after 30 days of aspirin treatment (Cohen et al., 2020).

Ascorbic acid in gonads has shown to be a sensitive index of the release of LH from the pituitary We have indeed found a corresponding increase in plasma LH with decrease in testicular acid (Stukenborg et al., 2021). Our present data indicate that the effect of aspirin is impact primarily on testosterone level and whatever effect is produced on the LH level is secondary to it (Sharpe, 2020).

With high aspirin dosages, Leydig cells multiplied and the intratubular gap grew larger. Germinal epithelia also degenerated. Testicular amyloid is specifically demonstrated by the histoarchitecture of the testes in high dose treated rats. This amyloid is composed of extracellular accumulation of homogeneous, eosinophilic, and amorphous material in the interstiaium, which appears pinkish in color and appears to significantly slough testicular interstitial cells (Leydig) (Boizet-Bonhoure et al., 2022).

The findings of this investigation supported the existence of testicular hypertrophy and expansion of the seminiferous tubules in conjunction with a cessation of sperm production (Philibert et al., 2023). The loss in testicular structure and the aging process of the testis are both fairly related. Nonsteroidal anti-inflammatory medicines (NSAIDs) have been the subject of various research looking into how they affect cellular and animal models of testicular aging (Hallak et al., 2020; HADI, W. H., & ABOOD, A. H. (2022).

Aged Leydig cells produced much more testosterone when they were incubated with the COX2 inhibitor NS398. Additionally, blood testosterone levels and testicular StAR expression levels, which are necessary for testosterone biosynthesis and male fertility, were higher in rats given the COX2 inhibitor DFU [5,5-dimethyl-3-(3-fluorophenyl)-4-(4-methylsulphonyl) phenyl-2(5H)-furanone] than they were in rats given no DFU (Matzkin, et al., 2021; Blecharz-Klin et al., 2022; Hadi, W. H., & Abood, A. H. (2023, December).).

Although the effect of an analgesic like aspirin on testes is currently being studied in rats, it is disheartening that expected of causing multiple endocrine disturbances and testicular peritubular cells with this analgesic. These results were expected given that testes and aged Leydig cells express higher levels of COX2 (Mirabito Colafella et al., 2020; Schjerning et al., 2020).

IV. Conclusion

To conclude aspirin is a toxic agent which has deeply physiological and histological changes in testis on male rats when getting it orally permanently. The data proofed that the drug aspirin has been cuased a clear defect on severe hypertrophied epithelium projecting in to the lumen and apparent decrease in both thickness of the seminiferous tubule wall and number of its cell layers with apparent marked reduction of the sperms counts in the lumen desquamated spermatogenic cells in the lumen of a seminiferous tubule.

Reducing the use, the drugs without medical consultation or their abuse when necessary and increasing the proportion of drinking water accompanied by the drug.

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