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Histological Effects of Acrylamide on the Reproductive Organs of Male Rats

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Abstract: ***Acrylamide is a chemical compound which has been discovered in overcooked carbohydrate foods in recent years. Several studies have reported that acrylamide has toxic effects on different systems of the human and animal body. The present study was carried out to investigate its effect on the histology of the reproductive organs of male Sprague-Dawley rats. The study animals were divided into four groups with five animals in each group. Group one served as the control while groups two to four were administered oral acrylamide doses of 10 mg/kg, 15 mg/kg and 20 mg/kg respectively for a period of four weeks. The results obtained from the study showed that acrylamide administration generally induced abnormal histological changes on the reproductive organs of treated animals in a dose dependent manner. Acrylamide induced injurious effects on the reproductive organs of male Sprague-Dawley rats that may contribute to male reproductive organs dysfunctions.***

Keywords: Acrylamide, Sprague-Dawley rats, male reproductive organs, reproductive organ dysfunction

1. Introduction

Acrylamide is a chemical compound with the chemical formula C_3H_5NO. It is a white crystalline solid, odorless and soluble in water, ethanol, ether and chloroform. Acrylamide can also be called acrylic amide and its IUPAC name is prop-2-enamide. Acrylamide is a potential human carcinogen, as it has been categorized as a 'B2' carcinogen (US, EPA). Its non-thermal decomposition results in the formation of ammonia but thermal decomposition forms oxides of nitrogen, carbon monoxide and carbon dioxide. It decomposes where oxidizing agents, acids, bases, iron salts and iron are present. Acrylamide has been found to have several uses ranging from the medical, laboratory to the industrial were companies use it in the manufacture of polyacrylamide and in water treatment (Ran *et al.*, 2014). Since the discovery of acrylamide in overcooked carbohydrate foods in the year 2002 by an Eritrean scientist Eden Tareke in Sweden, it has raised serious concerns as per whether or not it sis able to elicit systemic toxicity in humans (Tareke *et al.*, 2002). Several studies have reported the toxic effect of acrylamide on the various systems of the body but few have discussed its effect on the histology of reproductive tissues. Acrylamide has been reported to have adverse effect on reproduction as shown by dominant lethal effects, distortion of testicular epithelia, and morphological sperm abnormalities (Dearfield *et al.*, 1988). Acrylamide is evidently formed during frying, grilling, roasting as well as baking carbohydrate-rich foods especially at temperatures that are above 120 °C. This happens through interactions of amino acids with the reducing sugar (Tareke *et al.*, 2002). Glycidamide is an epoxide metabolite of ACR and is believed to be responsible for the ACR genotoxicity in humans and animal models (Kruser and Flynn, 2011). This study was designed to investigate the effects of varied doses of acrylamide on the histological architecture of male reproductive tissues in Sprague-Dawley rats.

2. Materials and Methods

Chemical compound

A 98% acrylamide with a net quantity of 100g was purchased from Pyrex-IG Scientific Company, Benin City, Edo State Nigeria. The compound was stored in a cool dry place, until it was needed for use. Distilled water used as solvent to produce the stock solution was also purchased from Pyrex- IG Scientific Company.

Experimental animals

Adult male rats, having body weights between 120 to 220g were purchased from Igbinedion University animal husbandry. They were housed under natural light and dark cycles, and were given daily access to food and water. The animals were acclimatized for one week before the experiment was carried out.

Experimental design

Twenty animals were randomly placed into four groups with each group consisting of five rats each. Three out of the four groups were subjected to one month of oral administration of acrylamide while the first group was used as the control group. The dosage administered to each group is as follows: Group 1 = Control group, Group 2 = 10 mg/kg, Group 3 = 15 mg/kg, Group 4 = 20 mg/kg. The animals were weighed weekly and the weights of the individual animals per group were taken using a digital scale. After the period of one month the rats were sacrificed by cervical dislocation. Through an abdominal incision, the reproductive organs were exposed together with other organs of the body systems. The testes (right and left) and caudal epididymis were harvested and weighed using an electrical weighing balance. The testes and caudal epididymis were fixed in Bouin's fluid contained in universal containers.

Histological processes

The harvested testes and epididymis were taken through the following tissue processing procedures; Fixation using freshly prepared Bouin's fluid, Dehydration, Clearing, Impregnation, Embedding, Sectioning and Staining using routine haematoxylin and eosin staining method.

Statistical analysis

All statistical analysis of data obtained in the course of this study were carried out using the Statistical Package for

Social Sciences (SPSS version 20). The results are expressed as mean ± SEM. Statistical significance was determined by analysis of variance (ANOVA). Significant differences between groups were determined in ANOVA using Duncan post-hoc test at $p<0.05$.

3. Result

Effect of acrylamide administration on the body weight

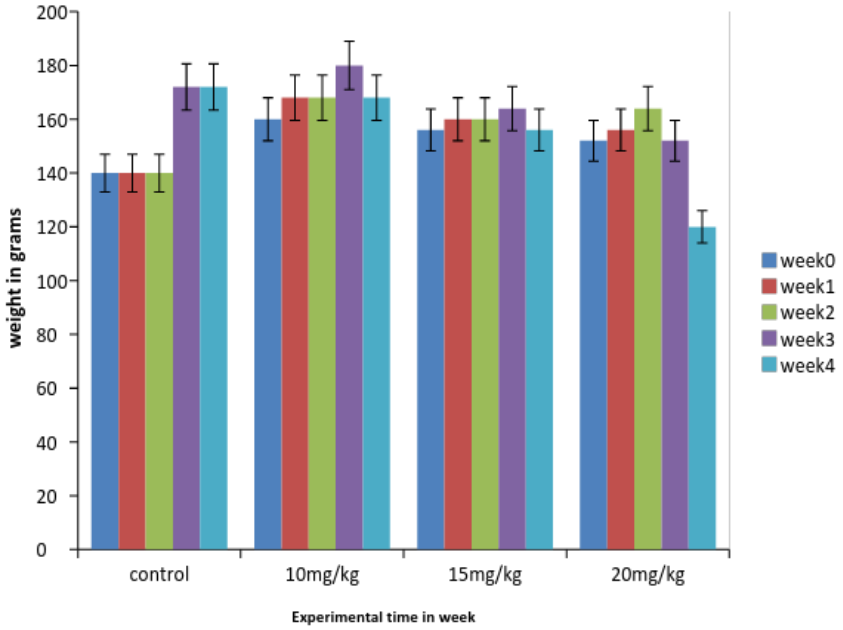

Figure 1: Effect of daily administration of varied doses of acrylamide (10mg/kg, 15mg/kg and 20mg/kg b. wt.) on the body weight of male Sprague-Dawley rats treated for four weeks.

The study animals treated with 10 mg/kg, 15 mg/kg and 20 mg/kg showed a significant increase ($p<0.05$) in body weight from week 0 to week 2, compared to the control group. During the 3rd week of treatment, there was no significant difference ($p>0.05$) in body weight across all the groups. But, in week 4, the study animals treated with 20 mg/kg showed a marked significant decrease ($p<0.05$) in body weight compared to the study animals treated with 10 mg/kg, 15 mg/kg and the control group.

Effect of acrylamide administration on organ weights

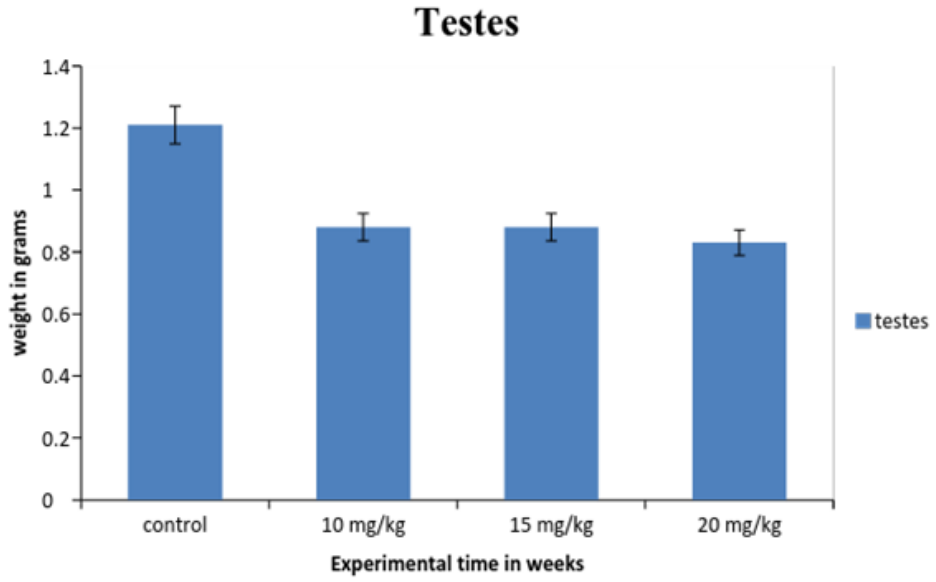

Figure 2: Effect of daily administration of varied doses of acrylamide (10mg/kg, 15mg/kg and 20mg/kg b. wt.) on the testicular weight of male Sprague-Dawley rats treated for four weeks

The study animals treated with 10 mg/kg, 15 mg/kg and 20 mg/kg showed a significant decrease ($p<0.05$) in the weight of their testes compared to the control in a dose dependent manner. Though, the study animals treated with 10 mg/kg

and 15 mg/kg decreased at the same rate as seen from the graph above.

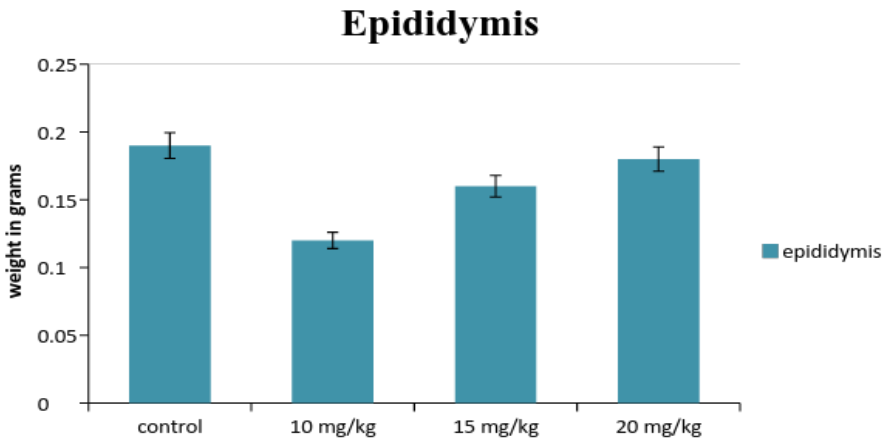

Figure 3: Effect of daily administration of varied doses of acrylamide (10mg/kg, 15mg/kg and 20mg/kg b. wt.) on the epiddymal weight of male Sprague-Dawley rats treated for four weeks

The study animals treated with 20 mg/kg, 15 mg/kg and 10 mg/kg showed a significant dose dependent decrease ($p<0.05$) in the weight of the epididymis compared to the control.

Effect of varied doses of acrylamide (10 mg/kg, 15 mg/kg and 20 mg/kg) on histology of reproductive organs of male Sprague-Dawley rats.

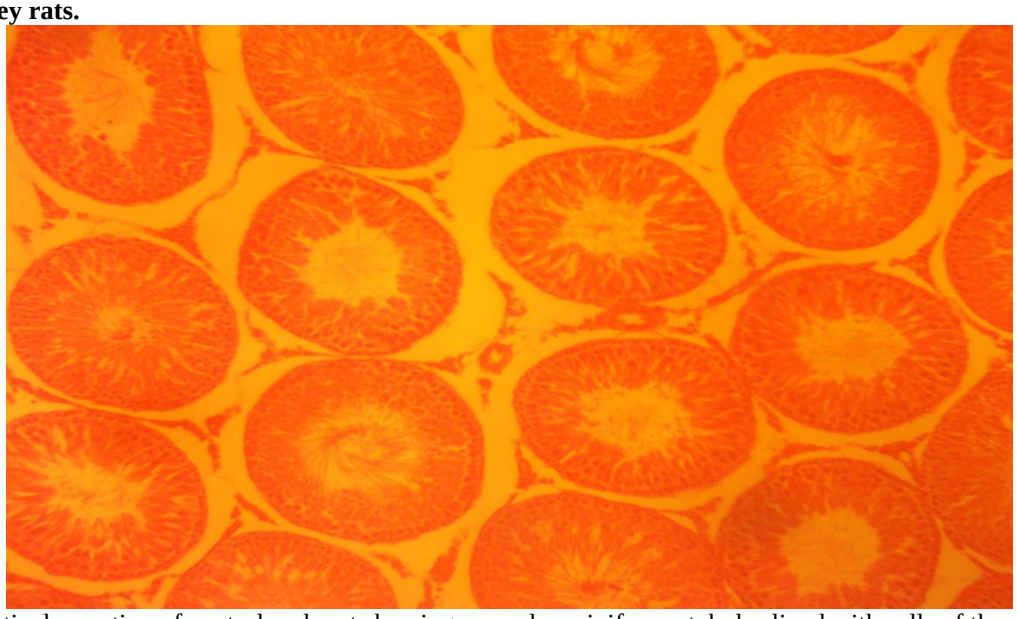

Figure 4: Testicular section of control male rat showing normal seminiferous tubules lined with cells of the spermatogenic series and separated by scanty stroma. (X100 magnification)

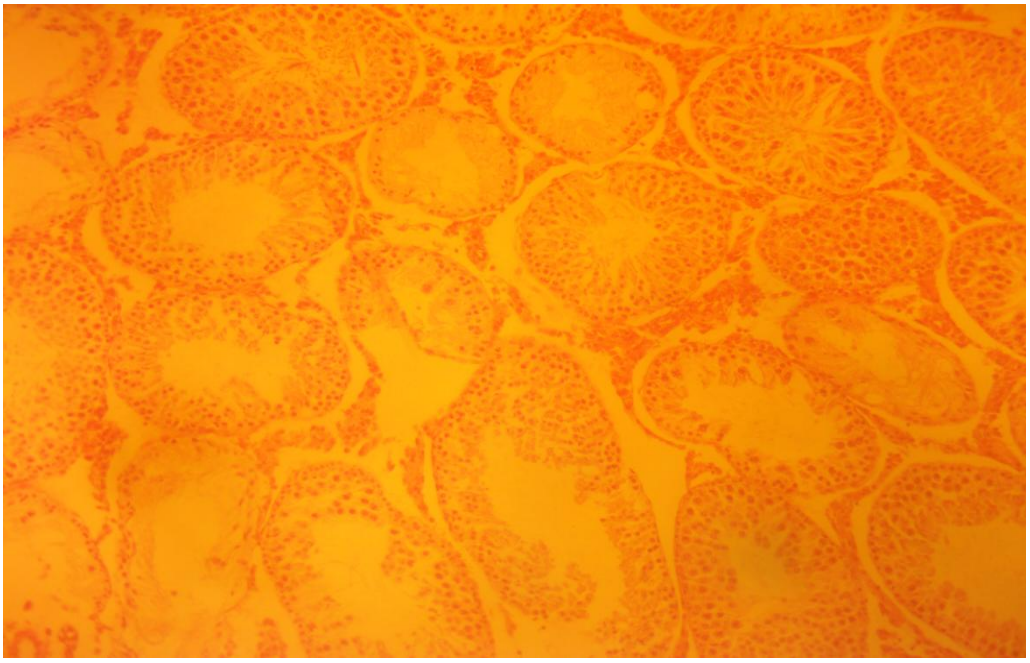

Figure 5: Testicular section of male rat administered 10 mg/kg b. wt. of acrylamide showing abnormal seminiferous tubules lined with degenerating cells of the spermatogenic series, partly separated by abnormal stroma and several tubules lacking spermatozoa. (X100 magnification)

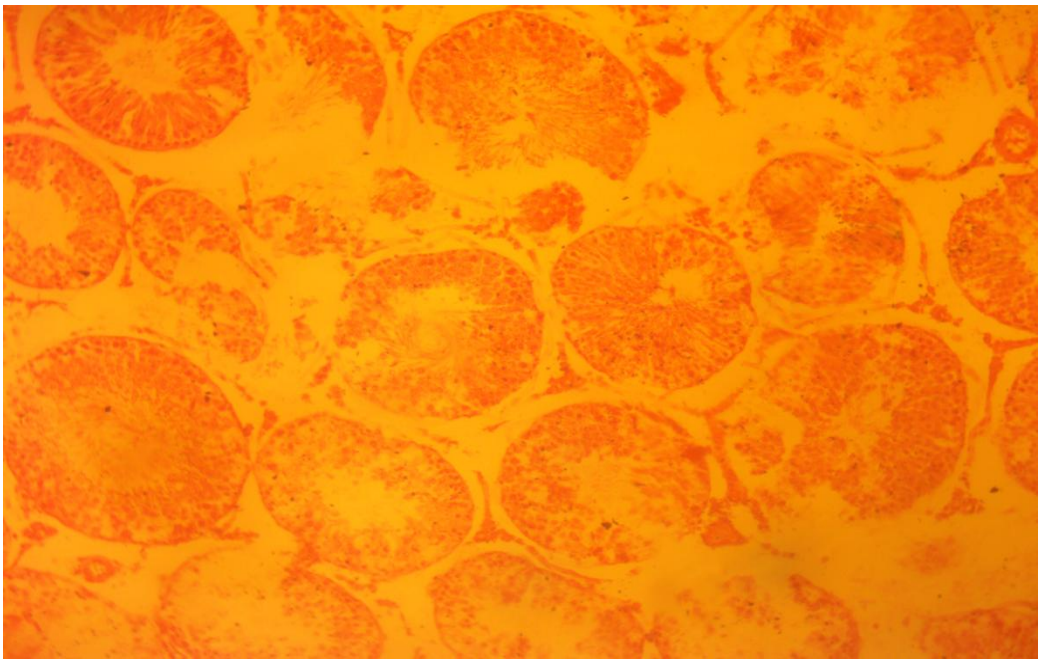

Figure 5: Testicular section of male rat administered 15 mg/kg b. wt. of acrylamide showing abnormal seminiferous tubules lined with degenerating cells of the spermatogenic series and partly separated by abnormal stroma. (X100 magnification)

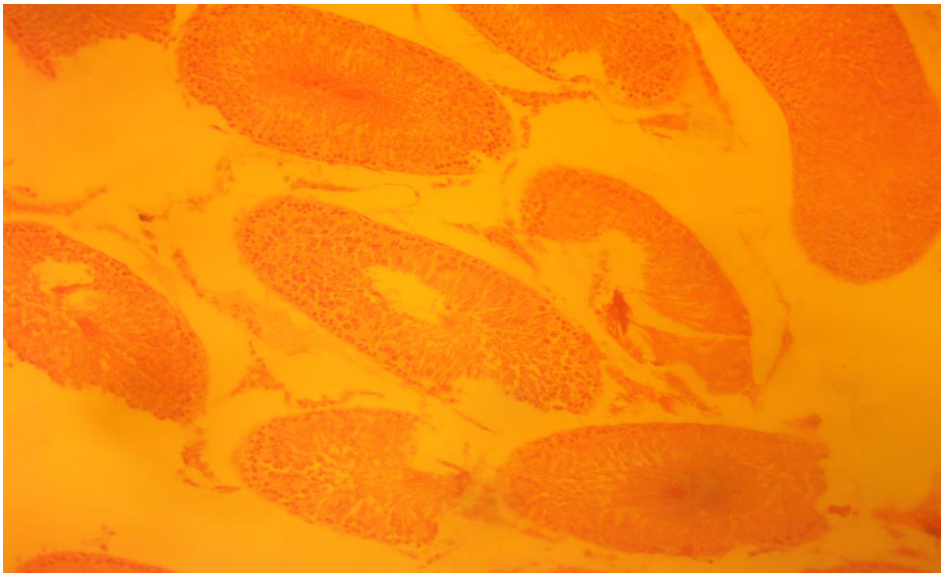

Figure 6: Testicular section of male rat administered 20 mg/kg b. wt. of acrylamide showing abnormal seminiferous tubules lined with degenerating cells of the spermatogenic series and partly separated by abnormal stroma. (X100 magnification)

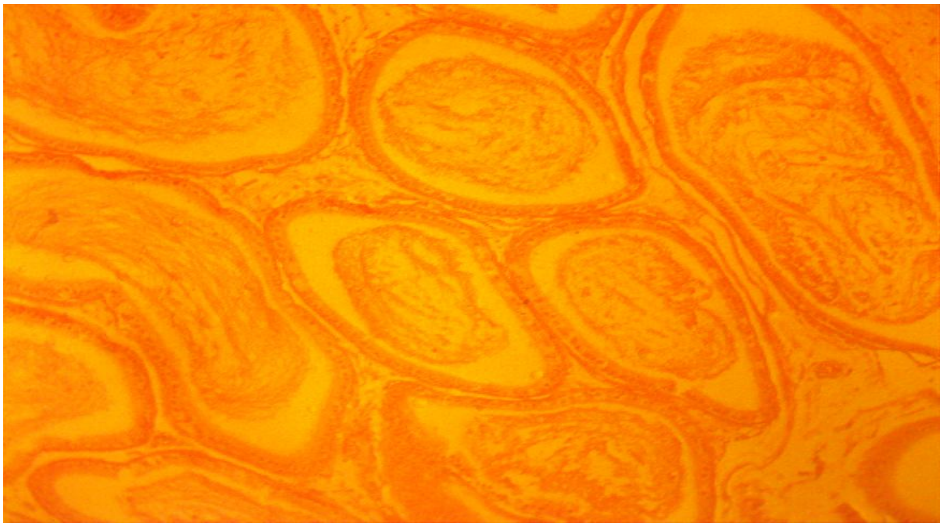

Figure 7: Epididymis section of control male rats showing normal epididymal tubules containing mature sperm cells. (X100 magnification)

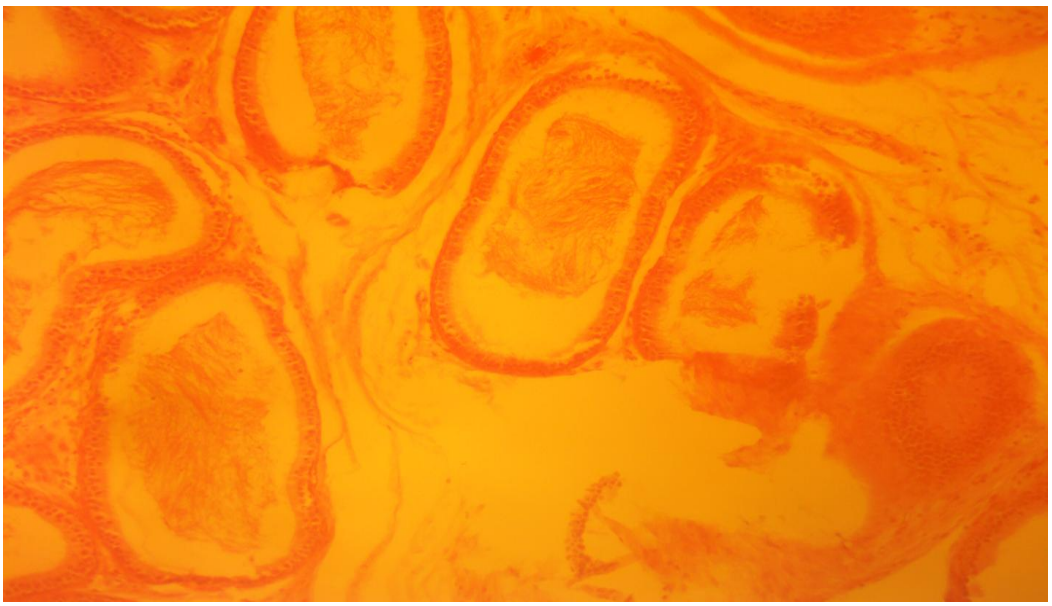

Figure 8: Epididymis section of male rats administered 10mg/kg b. wt. of acrylamide showing mild degeneration of epididymal tubules. (X100 magnification)

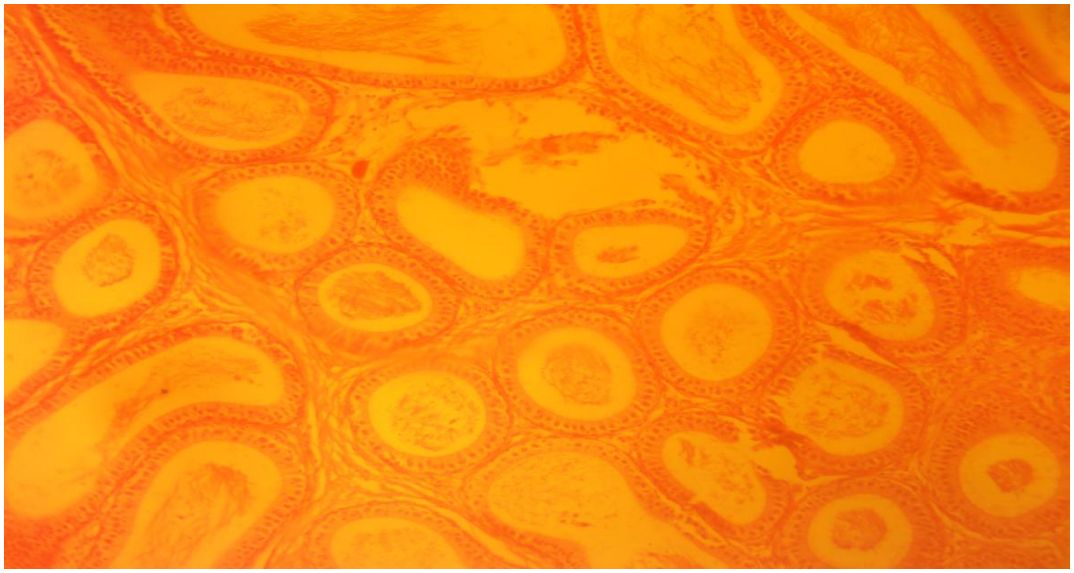

Figure 9: Epididymis section of male rats administered 15mg/kg b. wt. of acrylamide showing mild degeneration of epididymal tubules and reduction of luminal sperm population. (X100 magnification)

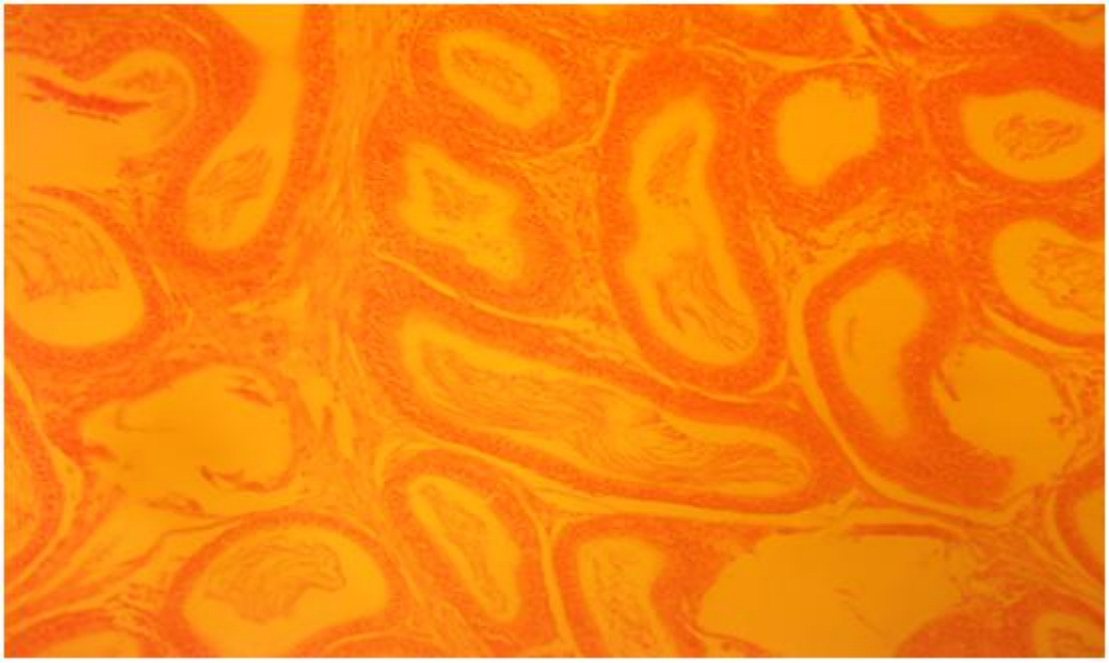

Figure 10: Epididymis section of male rats administered 20mg/kg b. wt. of acrylamide showing focal degeneration of epididymal tubules and reduction of luminal sperm population. (X100 magnification)

4. Discussion

This work examined the effect of acrylamide on the histology of the reproductive tissues of male Sprague-Dawley rats. It is a general perception that alteration in body weight is a pointer to impairment in the normal functioning of organisms and is also useful in ascertaining the toxicity of substances (Grance *et al.*, 2008). One of the most sensitive and effective drug toxicity indicators is the organ weight which often times is altered before changes in appearance, form and shape becomes affected and evident (Ying-Piao *et al.*, 2013).

Administration of varied doses of acrylamide to mature male Sprague-Dawley rats caused general distortion in the histoachitecture of the reproductive tissues of the animal models in a dose dependent manner relative to the control. The seminiferous tubules of the testes showed marked depletion of the spermatogenic series in the treated animals, while the epididymal tubules also showed focal areas of cell degeneration. These defects seen in this study could be due to alterations in the normal reproductive hormonal levels and is a clear indication that acrylamide may have the ability to impair the process of spermatogenesis and also may have the propensity to cause low sperm count (Zenick *et al.*, 1986), which is a potent pointer to reduced fertility and/or infertility.

In conclusion, acrylamide caused deleterious effects on the histoarchitecture of the animal models used in this study, and whilst the recorded effect cannot be immediately extrapolated to humans, consumption of overcooked/over processed starchy food should be avoided. Further studies to ascertain which exact carbohydrates food accumulate acrylamide and at what level of cooking are recommended.

5. Acknowledgement

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