

## **Effect of paint fumes on histoarchitecture of the testes of adult male Wistar rats**

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### **Short Report**

#### **ABSTRACT**

**Aim:** To investigate the effect of paint fumes on the histoarchitecture of the testes.

**Methodology:** In this study, we reported sixteen (16) Wistar rats mainly male of an average weight of 180g. Four groups of four rats each were divided into groups A-D. The treated rats were exposed to paint fumes for 8 hours daily, group A for three weeks, B for four weeks and C for five weeks. Group D animals (control group) were exposed to air for 8 hours for five weeks. On the last day of exposure, animals were euthanized, sacrificed and thoraco-abdominal incision was performed after which one testis was received from each rats, preserved in 10% formal saline and further processed for histological study using Hematoxylin and eosin technique.

**Results:** The testes of the paint fumes exposed group shows necrotic cell death of the germ cells (spermatogonia) and reduced sperm cells in the central lumen. This is an indication of altered spermatogenesis.

**Conclusion:** Paint fumes that contain volatile organic compound cause the necrotic death of testicular germ cells in exposure dependent manner and there were reduced sperm cells in the lumen. This can lead to infertility.

**Keywords:** histoarchitecture, testes, spermatogonia, infertility.

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## Effet de la peinture sur l'histoarchitecture des testicules des mâles adultes Wistar rats

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### Rapport court

#### RÉSUMÉ

**Objectif:** étudier l'effet de la peinture sur l'histoarchitecture des testicules.

**Méthodes:** Seize adultes mâles Wistar rats pesant environ 180-200g ont été utilisés pour l'étude. Les rats ont été divisés en quatre groupes de A à D de quatre rats par groupe. Les traités étaient des rats exposés aux vapeurs de peinture pour 8 heures par jour, un groupe de trois semaines, B pour quatre semaines et C pendant cinq semaines. Groupe D animaux (groupe contrôle) ont été exposés à l'air pendant 8 heures pour une durée de cinq semaines. Les animaux ont été sacrifiés et les testicules ont été excisées et fixés à 10% formelle saline et traités pour étude histologique aide hématoxyline et d'éosine tache

**Résultats:** Les testicules des odeurs de peinture groupe exposé montre nécrotiques mort cellulaire des cellules germinales (spermatogonies) et réduit les spermatozoïdes dans le centre lumen. C'est une indication de modifier la spermatogenèse.

**Conclusion:** La peinture qui contiennent composés organiques volatils cause la mort nécrotiques testiculaire de cellules germinales dans l'exposition dépendantes et il y a eu une réduction les spermatozoïdes dans le lumen. Ceci peut entraîner l'infertilité.

**Mots-clés:** histoarchitecture, testicules, spermatogonies, stérilité.

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## INTRODUCTION

Paint is defined as any composition of either liquid or mastic which after application to a substrate in a thin layer is converted to a solid film (1). It is generally used to add colours and protect objects on which it is applied. It is applied to most things in our environment (e.g. house, car etc.) hence, we are exposed to it always.

Compounds at room temperature that have very high vapour pressure are commonly referred to as Volatile organic compounds (VOCs) (2). Some VOCs are naturally occurring (e.g. isoprene, terpenes etc) produced by plants while the anthropogenic VOCs are from man-made activities (3), which are mainly derived from paints (3) and coatings, oil refinery and industrial waste (e.g. benzene, Ethylbenzene, Xylenes) (3). Exposure to paint fumes containing VOCs either during painting or in newly painted environment has been associated with nausea (4), sensory irritation (5), respiratory distress, allergic and immune effects (6, 7, 8).

Benzene (example of VOC) has been found to be carcinogenic (9). Studies have shown that some VOCs cause liver damage and CNS disorder (10, 11). It has been previously reported that there were reversible encephalopathy, bone marrow suppression, biochemical and histological evidence of a hepato- cellular insult after exposure to paint fumes (12). Infertility is a common thing in this part of the world and many factors have been implicated as the causative reasons. 30% of infertility has been blamed on male and both reduced sperm production in the testes and abnormally functioning sperm are the most common etiology in cases of infertility in male (13). As the population boom, people get more exposed to paint fumes through different ways with little or no safety procedure, the present work investigated if paint fumes is deleterious to the male reproductive organ.

## MATERIALS AND METHOD

### Procurement of Paint

SeniorLux gloss paint manufactured by Prince and Princess industry (Ilorin,

Nigeria) was purchased from Taiwo Paint Store, Moraba Ilorin. The paint was purchased in February, 2012 with the expiry date put at January, 2014. The content of the paint was given as:

- Not manufactured with lead or mercury containing materials
- Alkyd Resin
- Extender Pigments
- Titanium Dioxide

The VOC's content was given as 380 g/L (3.17 lbs/gal) maximum.

### Animals

Sixteen adult male Wistar rats were purchased from Adedeji Ventures, Tanke Ilorin. The rats were housed in the Anatomy Department animal holdings and were kept in standard cages. The rats were fed with Bendel feeds (Ewu, Nigeria) and distilled water *ad libitum*. They were exposed to an environment of 12-hour light/dark cycle at  $24^{\circ}\text{C} \pm 2^{\circ}\text{C}$ .

### Animal treatment

Using random selection method, the animals were assigned into four groups A-D (4 animals each). The animals were exposed to 20ml paint fumes for 8 hours daily in the following order:

Group A: three weeks

Group B: four weeks

Group C: five weeks

While group D were exposed to fresh air for five weeks serving as the control.

### Method of exposure

The rats were put in a box (75cm X 50cm X 30cm), hole was bored on the cover of the box to allow little ventilation for breathing of animals. 20ml of gloss paint was put in a small container and hung at the upper part of the box. The container is not closed to allow escape of the fume into the box. A fume chamber is the ideal tool for the experiment but the researchers could not get one at the time of the experiment. All animals used for this study were treated in accordance with the ethics and guidelines of the Institutional Animal Care and Use Committee (IUCAC) of University of Ilorin.

### Animal sacrifice and fixation

24 hours after the last exposure, all animals were euthanized and their testes were cut, trimmed of fat, preserved in 10% formal saline and further processed for histological study using Hematoxylin and Eosin staining.

### RESULTS

Spermatogonia disruptions and vacuolations were observed from the histological section of testes of exposed animals as compared to the control group in exposure dependent manner. There was also spermatozoa reduction in the lumen of the testes in exposure dependent manner as compared with that of control animals.

The primary spermatocytes of the paint fumes exposed rats show a disrupted plasma membrane and rarefied cytoplasm although with normal state of nuclei suggesting that the cells are in a necrotic state. This is different from what is seen in the control where the nuclei, plasma membrane and cytoplasm were intact.

### DISCUSSION

After life, procreation is next. About one out of every ten couple has been reported to be infertile and the number keeps increasing every year. Numerous factors cause or aids infertility and many of the cause are just been discovered (13). In this project work, we report the effect of paint fumes that contain VOCs on the histoarchitecture of the testes using Hematoxylin and eosin stain.

From the histological technique, it was found that exposure of Wistar to paint fumes for 3, 4 and 5 weeks shows progression of testicular lesion that is the characteristics of an acute toxic insults. The toxic effect of the paint fumes may be attributed to the inclusion of VOC's in paint which are the chemicals that humans are allergic to.

This study has shown that there is testicular disruption following exposure to paint fumes; this may be due to Styrene, a known volatile organic compound which is major compound used in gloss paint. Styrene has been reported to reduce testicular enzymes, decrease sperm production and

testicular degeneration at high. (14) This is similar to what was seen in the paint fumes exposed rats especially those exposed for 5 weeks.

Paint thinner is another component of gloss paint (the type of paint used in this experiment) which contains 66% toluene, has been found to inhibit both the secretion and synthesis of testosterone in a reversible (15) Which can also inhibit sperm production and agenesis of spermatogonium. This is similar to what is obtained in the rats exposed to paint fumes where disruption in spermatogonium was seen.

The exact mechanism by which paint fumes containing volatile organic compounds affect the testes is still unclear, presently our laboratory is working on the mechanism by which paint fumes affect the structure and functions of the testes.

### CONCLUSION

The result has shown that paint fumes (VOCs) have adverse effect on the testes causing disruptions to the spermatogonia. Long term exposure reduces the number of sperm cells; this can be inferred that long term exposure could lead to infertility.

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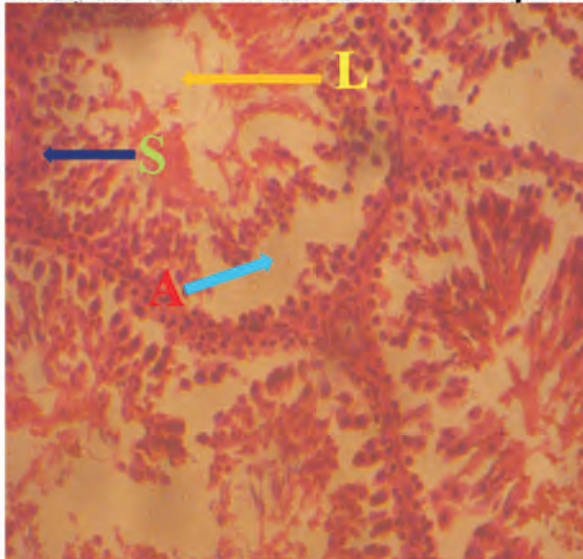
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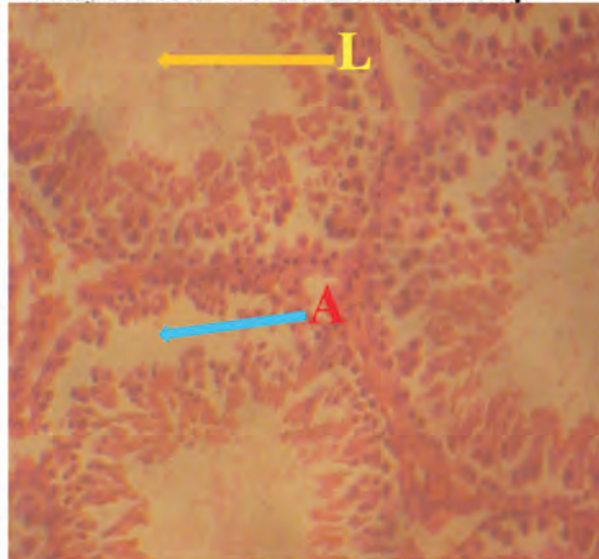


## HISTOLOGY RESULTS

**Plate 1: photomicrograph of testes of rats exposed to 20mL of paint 8 hours daily for three weeks. H&E x10 Obj.**

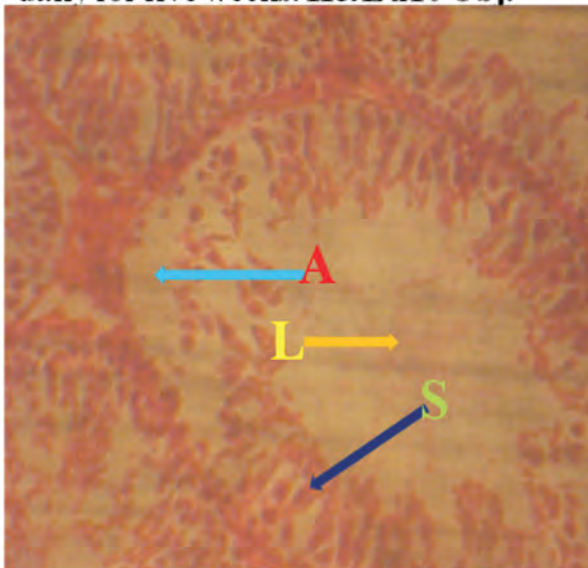


**Plate 2: photomicrograph of testes of rats exposed to 20mL of paint 8 hours daily for four weeks. H&E x10 Obj.**

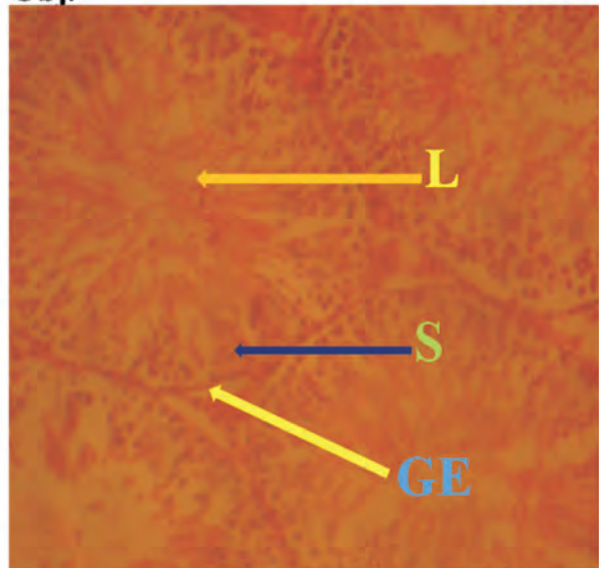


L: lumen, S: spermatogonium, A: area of cell disruption

**Plate 3: photomicrograph of testes of rats exposed to 20mL of paint 8 hours daily for five weeks. H&E x10 Obj.**



**Plate 4: photomicrograph of testes of rats exposed to air (control). H&E x10 Obj.**



A: area of cell death, L: lumen, S: spermatogonium, GE: germinal epithelium