

*Research Article*

## Histological Study of Toxic Effects of Acrylamide on the Liver and Kidney of Adult Male Albino Rats

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### Abstract

**Background and objectives:** Acrylamide has become one of the important public health concerns as it was detected in widely consumed food items; for example, fried bread, potato chips, any carbohydrate-rich food products cooked at high temperatures, roasted coffee. Several health hazards were reported after acrylamide exposure in animal and human models. Many studies reported that acrylamide can induce toxicity to many body organs such as reproductive toxicity, genotoxicity and neurotoxicity. **Materials and methods:** Forty adult male albino rats were randomly divided into 2 groups (n=10 per group). **The control group (group I):** received only standard diet and drinking water for 6 weeks. **The acrylamide group (group II):** received acrylamide via in tragastric gavage route at dose of 20 mg/kg once daily for 6 weeks. Histological and examination of the liver and kidney was done. **Results:** There were marked structural changes in kidney and liver of group II in the form of congestion apoptosis and vacuolations if compared to the control group. **Conclusion:** Administration of acrylamide continuously for six weeks causes massive destruction in both liver and kidney.

**Key words:** Acrylamide, public health, body organs

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### Introduction

Acrylamide has become one of the important public health concerns as it was detected in widely consumed food items; for example, fried bread, potato chips, and any carbohydrate-rich food items cooked at high temperatures (higher than 200°C) (Erdemli et al., 2016).

It represents an industrial chemical used in the manufacturing of polyacrylamides that are commonly used in personal care and grooming products (e.g. lotions and cosmetics), wastewater treatment soil conditioning, and paper and textile industries. In addition to such industrial and laboratory uses, high levels of acrylamide were also detected in tobacco smoke (Abdullah, 2019).

Several health hazards were reported after acrylamide exposure in animal and human models. Many researches reported that acrylamide can induce reproductive toxicity, genotoxicity, hepatotoxicity and carcinogenicity in animal models (Rydberg et al., 2005). Similarly, the neurotoxicity of acrylamide was clearly demonstrated in human and the

immunotoxicity of acrylamide was recently gained attention by scientists (Wei et al., 2015).

The liver and kidney are very important organs and play many important functions in the human body so this study assesses the effect of the acrylamide on histology of the liver and kidney.

### Aim of the work

The aim of this study is to assess the toxic effects of acrylamide on the histology of liver and kidney of adult male albino rats.

### Material and methods

#### Animals:

This study was performed in the Histology and Cell Biology Department of the Faculty of Medicine, Minia University, Egypt. This work was carried on 20 adult male albino rats which were weighing about 150-250 g, of 6-8 weeks. Animals were obtained from the Study Animal House of Minia University Laboratory Animals Growing Center of the Faculty of Agriculture.

Rats were housed in clean plastic cages and fed a standard laboratory diet with free access to diet

and water. Rats were maintained at a laboratory temperature ranged from 25-30°C and exposed to 12 hours light and 12 hours dark cycle. Rats were acclimatized for 2 weeks before the experiment. All rules of animal care and treatment were carried out according to the local guidelines of the Ethical Committee of the Faculty of Medicine of Minia University.

#### Reagents:

1- **Acrylamide:** “(C<sub>3</sub>H<sub>5</sub>NO), >99.5% pure, catalog no. 004027” was purchased from Medico company, Egypt. It was dissolved in distilled water (Mahmood et al., 2015).

#### Experimental design:

The 20 adult male albino rats were randomly divided into 2 groups (n=10 per group) as follows:

**1- Group I (Control group):** They were received distilled water and standard diet for 6 weeks.

**2- Group II (Acrylamide group):** They were received acrylamide “dissolved in 0.2ml of distilled water” orally via intragastric gavage route at dose of 20 mg/kg once daily for six weeks (Kopanska et al., 2017).

After 6 weeks, all rats were anesthetized and euthanized by cervical decapitation in the morning hours between 8.30 h and 9.00 h, 24 h after the last treatment. Tissue samples of kidney and liver were immediately collected and cut into small pieces.

#### For light microscopic examination:

##### a) The Paraffin Technique (Suvarna et al., 2018):

The liver and kidney specimens were fixed in 10% neutral-buffered formalin for 2 days at room temperature. After appropriate fixation,

tissue samples were dehydrated in a graded alcohol concentrations, cleared in xylene and embedded in paraffin wax then cut by a microtome. 5µm sections were mounted on glass slides to be stained.

##### b) Staining with Hematoxylin and Eosin (H&E) (Suvarna et al., 2018):

Some sections, mounted on glass slides, were deparaffinized to be stained with H&E. They were placed in Hematoxylin stain for 7 minutes, washed well in running tap water, then put in eosin for 3 minutes and the excess stain was washed by water. The sections were dehydrated by alcohol, cleared by xylene and then mounted on glass slides to be viewed by the light microscopy for histological analysis.

**Result:** The cytoplasm stained light red while the nuclei stained blue.

#### Results

##### I-Histological results:

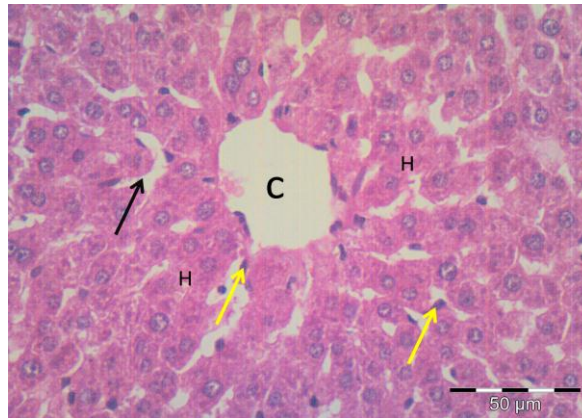
##### ➤ Hematoxylin and Eosin (H&E) stain results of liver tissues:

##### 1- Control group (group I):

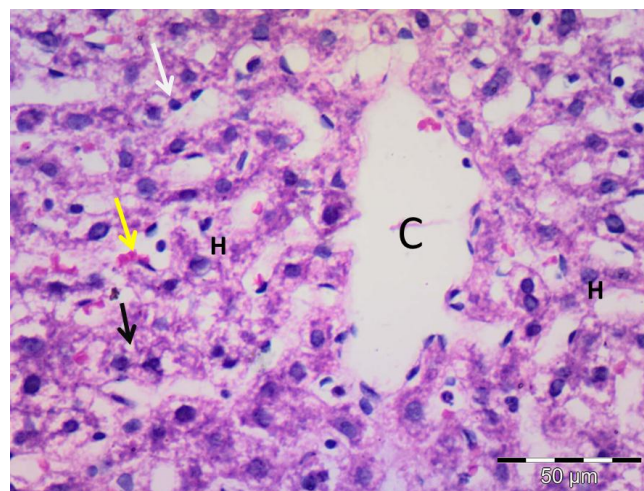
There was preserved histological architecture of the classic hepatic lobule in the form of central vein surrounded by cords of hepatocytes with round vesicular nuclei and prominent nucleoli. Blood sinusoids were present between cords of hepatocytes. They were lined with kupffer cells (**fig. 1**).

##### 2- Acrylamide group (group II):

There was disturbed histological architecture of liver. There was dilatation of the central vein. Hepatocytes appeared degenerated with vacuolated cytoplasm and pyknotic nuclei. Blood sinusoids appeared dilated and congested with blood in their lumen (**fig. 2**).



**Figure (1):** A photomicrograph of the rat liver tissue of the group I showing central vein (C) surrounded by cords of hepatocytes (H) with round vesicular nuclei and prominent nucleoli. Notice blood sinusoids (black arrow) are lined with Kupffer cells (yellow arrows). (H&E x400, scale bar=50μm).



**Figure (2):** A photomicrograph of the rat liver tissue of the group II showing and dilatation of the central vein (C). Hepatocytes (H) are degenerated as there are vacuolated cytoplasm (black arrow) and pyknotic nuclei (white arrow). Notice blood sinusoids are dilated and congested (yellow arrow). (H&E x400, scale bar=50μm).

➤ **Hematoxylin and Eosin (H&E) stain results of kidney tissues:**

**1- Control group (group I):**

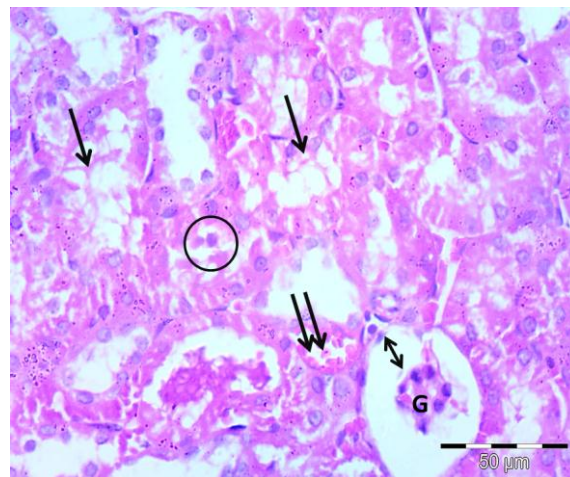
There was preserved histological architecture of kidney. Malpighian renal corpuscles showed glomerulus surrounded by Bowman's space. Proximal convoluted tubules were lined with high cuboidal cells with rounded basal nuclei. Distal convoluted tubules were lined with cubical cells with rounded central nuclei (Fig. 3).

**2- Acrylamide group (group II):**

There was disturbed histological architecture of kidney that was in the form of marked degeneration of the glomerulus and widening of the Bowman's space. There was degeneration of tubular lining cells with desquamation of some tubular cells in their lumina was observed. There was interstitial hemorrhage (Fig. 4).



**Figure (3):** A photomicrograph of the rat renal tissue of the group I showing Malpighian renal corpuscle containing glomerulus (G) surrounded by Bowman's space (double head arrow). Proximal convoluted tubules (P) are lined with high cuboidal cells having rounded basal nuclei. Distal convoluted tubules (D) are lined with cubical cells having rounded central nuclei. (H&E x 400, scale bar = 50  $\mu$ m).



**Figure (4):** A photomicrograph of the rat renal tissue of the group II showing degeneration of the glomerulus (G), widening of Bowman's space (double head arrow), degeneration of renal tubules (black arrows), with desquamation of some tubular cells in their lumina (black circle). Notice the interstitial hemorrhage (double arrows). (H&E x 400, scale bar = 50  $\mu$ m).

### Discussion

Acrylamide has become one of the important public health concerns as it was detected in widely consumed food items; for example, fried bread, potato chips, any carbohydrate-rich food items cooked at high temperatures, roasted coffee and cosmetics. Several health hazards were reported after acrylamide exposure in animal and human models (Sharma et al., 2013).

Many studies reported that acrylamide can induce toxic effect to many body organs such as reproductive toxicity, genotoxicity and neurotoxicity (Pennisi et al., 2013).

Acrylamide (>99.5% pure) was dissolved in distilled water and orally administered to rats via intragastric gavage route (Mahmoud et al.,

2015). Twenty adult male albino rats (weighting about 150-250g) were used in this study. Animals were divided into four groups, 10 animals for each group. Group I is the control group. Group II (acrylamide group) was received acrylamide at dose of 20 mg/kg once daily for six weeks. The dose was established by Elhelaly et al., (2019) as it causes significant damage to liver and kidney of rats while higher doses cause death of rats. The duration was selected previously by AL-Mosaibih, (2013) who found that this duration was enough to cause toxicity.

By using hematoxylin and eosin the histological results, there were preserved histological features of liver tissue sections of the control group (group I). These changes were in the form of apoptosis of hepatocytes. These results were in accordance with Ghorbel et al., (2015) who found degenerative changes such as apoptosis of hepatocytes.

These changes were explained by Kovac et al., (2015) who stated that acrylamide causes decrease in the reduced glutathione (GSH). Reduction in GSH leads to mitochondrial dysfunction and formation of reactive oxygen species (ROS). They cause to significant damage to cellular structures what is called the oxidative stress. Elhelaly et al., (2019) added this toxicity may cause damage to DNA and RNA, oxidative deactivation of certain enzymes, and the oxidation of polyunsaturated fatty acids into lipid peroxyl radicals and lipid hydroxides that cause affection of the balance between cell survival and death.

In addition, ROS disturb the vascular function.

This leads to more cellular damage and apoptosis (DeWitt & Prough, 2009). According to Raju et al. (2015) the acrylamide decreases the level of high density lipoprotein (HDL). Low HDL is associated with narrowing and blockage of the blood vessels leading to deleterious effect on them. Acrylamide also produces local toxic effects on the liver and kidney as they are the sites of its metabolism (Mahmoud et al., 2015).

Also, there was vacuolated degeneration of hepatocytes. This was explained by Roodi et al., (2018) who considered the vacuolation of

hepatocytes as ballooning degeneration and explained it as a type of cellular defense mechanism against toxins because these vacuoles collect the harmful elements and prevent them from affection of the biological activities of hepatocytes. As well, free radicals attack polyunsaturated fatty acids of plasma membranes leading to their degradation resulting in vacuolation.

Another observed finding was the vascular congestion which is previously mentioned by Raju et al., (2015). They explained that the vascular congestion was caused by the inflammatory response resulted from ROS.

Blood sinusoids were dilated as found by Elhelaly et al., (2019) who explained that ROS produced by acrylamide causes inflammation which in turn produce dilatation of blood vessels to increase the blood flow to the injured area

Kidney tissue sections of the control group showed preserved histological architecture. Kidney tissue sections of group II showed degeneration of the renal glomeruli, degeneration of tubular lining cells and interstitial hemorrhage. These results are in agreement with Erdemli et al., (2019) who found, on their study on the effect of acrylamide and vitamin E on kidney of rats, that acrylamide led to glomerular collapse, tubular degeneration and interstitial hemorrhage. They explained that acrylamide moves the oxidant-antioxidant balance to favor oxidants causing oxidative stress.

### Conclusion

From this study, it is concluded that acrylamide causes degenerative changes in the liver in the form of vacuolations in the cytoplasm of hepatocytes, apoptosis and congestion of the central vein. Regarding kidney, it causes degeneration of renal glomeruli and renal tubules and interstitial hemorrhage.

### Recommendations

We recommend avoid usage of products containing acrylamide and avoid cooking of potato and grain-based foods at high temperature for long time. We should use acrylamide mitigation strategies to minimize exposure to it as it causes many health hazards.

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