Possible Protective Effect of Ascorbic Acid on the Histopathological Changes of Acrylamide on Jejunum of Adult Male Albino Rats

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Abstract

Introduction: Acrylamide (ACR) is a water soluble vinyl monomer that has multiple chemical and industrial applications. Several studies were used with liver, kidney, brain, erythrocyte and proved that (ACR) increase glutathione-s-transferase and causing oxidative stress. Jejunum is the most sensitive part of the intestine, and most of the absorption process occurred in it. Ascorbic acid is natural antioxidant that prevents the increased production of free radicals induced by oxidative damage to various cellular components.

Aim of the work: to study the effect of ACR on the jejunum of adult male albino rats and to evaluate the possible role of ascorbic acid as a protective agent.

Material and methods: A thirty albino rats were divided into 3 groups:

Group I: were served as untreated control group.

Group IIa received ACR 30 ml\kg b.w orally dailly for 28days

Group IIb: received both ACR 30 ml\kg b.w orally simultaneously with ascorbic acid 400ml \kg.b.w intraperitoneally daily for 28 day.

Results

Exposure to ACR led to histological changes in **Group IIa** in the form of widening and shortening of villi compared to control group sections. There was inflammatory cell infiltration in the lamina propria,

Group IIb showed an improvement in comparison with a groupIIa. This improvement include restoration of villous shape and length, less inflammatory cells infiltration

Conclusion: ACR led to histopathological changes in jejunum. These changes attenuated by simultaneous administration of ascorbic acid with ACR.

Introduction

ACR is a water soluble vinyl monomer that has multiple chemical and industrial applications (**Hammad et al., 2013)**. It has been used in the industrial sector, including clarifying drinking water, treating industrial waste waters, and in polyacrylamide gels in biotechnology laboratories since 1958 **(Doerge et al.,2005).** ACR is produced in starchy food that are baked, roasted or fried at high temperature **(Hammad et al., 2013)**.

Potentially toxic (ACR) is largely derived from heat induced reactions between the amino group of the free amino acid asparginase and the carbonyl group of glucose and fructose in cereals, potatoes and other plant-derived foods **(El-Mottaleb and Rashed,2008)**. Several studies were used with liver, kidney, brain, erythrocyte and proved that (ACR) increase glutathione-s-transferase and causing oxidative stress **(Mansour et al**., **2008)**.

The small intestine is the longest part of the gastrointestinal tract and extends from the pyloric orifice of the stomach to the ileocecal junction, jejunum is most sensitive part of the intestine, also most of the absorption process occurred in it **(Drake et al., 2007).**

Ascorbic acid, according to **Nayanatara et al**. **(2008)** is a natural

antioxidant that prevents the increased production of free radicals induced by oxidative damage to various cellular components.

Materials and methods:

Animal: A thirty albino rats (2-3 months old) were used in the present study. Their weights ranged from 200-250 grams. The rats were maintained in a suitable temperature in a well-ventilated animal house.

Materials: ACR and ascorbic acid were purchased from Sigma-Aldrich Chemical Company.

Methods: The rats were divided into 3 groups, 10 animals for each.

Group I: untreated control group.

Group IIa: received ACR 30 ml\kg b.w orally dailly for 28days

Group IIb: received both ACR 30 ml\kg b.w orally simultaneously with ascorbic acid 400ml \kg.b.w intraperitoneally daily for 28 day.

By the end of the experiment, the animals were anaesthetized by ether and sacrificed. The jejunum specimens were dissected and processed for light microscopic examination.

Jejunum specimens were fixed in 10% formalin for at least 24 hours, washed well in water, dehydrated in ascending grades of alcohol, cleared in xylene, impregnated in soft paraffin, and then embedded in hard paraffin. Sectioning was done by a microtome (Leica RM2035) at 5 micrometer, mounted on glass slides and stained with:

Haematoxylin and eosin stain (H&E) for general histological examination.

Results

I-Control group:

Jejunum wall was formed of four layers; mucosa, submucosa, musculosa and serosa .The mucosa was the layer facing the intestinal lumen showed finger & leaf like villi, each villus had a central core which was formed of loose connective tissue & numerous fibroblasts and lined with simple columnar epithelium (enterocytes) with goblet cells. Enterocytes were tall columnar cells with acidophilic cytoplasm and oval basally located basophilic vesicular nucleus with apical brush border. Goblet cells were unicellular glass shaped cells; its basal part was basophilic. Its apical part appeared faint .The lamina propria, forming the villus core. Between the villi, there were invaginations called intestinal gland or crypts of Lieberkühn which were simple tubular structures and lined by simple columnar epithelium. They extended from the bases of villi through the whole thickness of the lamina propria. Muscularis mucosa appeared as a thin layer of spindle shaped smooth muscle fibers circularly arranged with fusiform nuclei and acidophilic cytoplasm. The submucosa appeared as a loose connective tissue layer with comparatively large blood vessels. The Musculosa consisted of two layers of smooth muscle; the inner one was circular and formed of spindle shaped smooth muscle fibers with fusiform nuclei and the outer one was longitudinal and appeared as circular sections with centrally located rounded nuclei. The serosa was the most outer layer and was formed of with a thin layer of loose connective tissue covered with mesothelium **(Fig 1)**.

Group IIa: Examination of this group showed widening and shortening of villi compared to control group sections. There were inflammatory cell infiltrations in lamina propria (**Fig 2)**.

Group IIb: Examination of this group showed restoration of villous shape to some extent compared to group IIa. The inflammatory cell infiltrations were less than group IIa **(Fig 3)**.

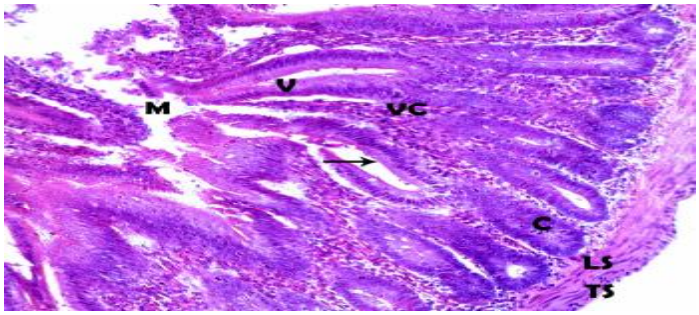

Fig. 1) : A photomicrograph of a transverse section of a jejunum of adult control animal (V), villus core (VC) ,brush border of enterocytes (arrow) ,the intestinal showing; villi glands (C),circular muscles (LS) longitudinal muscle (TS).
(H&EX200)

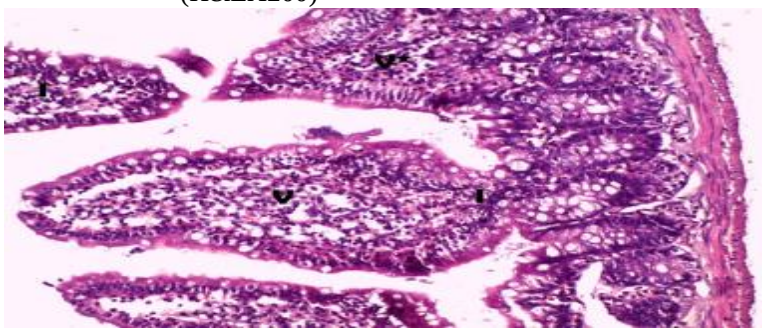

Fig.2): A photomicrograph of a transverse section of a jejunum **(group IIa),** showing shortening **(V*)** and broadening of villi (**V**) slight inflammatory cell infiltrations(**I**).
(H&E.X200)

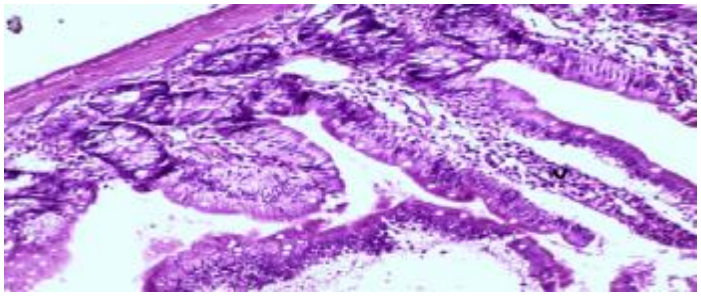

Fig.3): A photomicrograph of a transverse section of a jejunum **(group IIb**), showing the intestinal villi with some villi height restoration **(v),** and less inflammatory cell infiltrations compared to group IIa.
(**H&E. X200)**

Discussion

The present work revealed that ACR induced histological alteration in the jejunum of adult male albino rat.

These changes were in the form of widening and shortening of villi compared to control group sections .There were inflammatory cell infiltration in the lamina proria.

One of the prominent findings in the treated group with ACR was in the form of shortening and broadening of jejunal villi.

This agree with **Dobrowolski and his colleagues (2012**) who found that ACR altered morphology and histology of the small intestinal wall, decrease musculosa and submucosal thicknesses, villus length, crypt depth, crypt number, and the small intestinal absorptive surface. These results

coincide with the results of **Tomaszewska et al. (2014)** who found that ACR increased the number of damaged villi in the duodenum and jejunum. Furthermore, **Lee and his colleagues (2005)** found that ACR caused oxidative stress by inducing generation of reactive oxygen species (ROS), that reduce the antioxidant defense systems of the cells by depleting non-enzymatic antioxidant systems (vitamins and glutathione) and increasing susceptibility of cells to oxidative attack by altering the membrane integrity and fatty acid composition.

In the present study, we observed inflammatory cellular infiltrations in the lamina propria of mucosa and submucosa. These results are in accordance with the results of **Hammad et al. (2013)**. **Naruszewicz and his colleagues (2009)** found that chronic ingestion of dietary ACR might induce oxidative stress in humans through leukocyte activation and increased production of reactive oxygen radicals causing chronic inflammation.

In the present work, group of rats administrated ascorbic acid with ACR showed a partial improvement in comparison with a group of rats received ACR only. This improvement includes restoration of villous architecture, less inflammatory cell infiltration. These results are in agreement with the previous studies of **EL-Kenawy et al. (2013)** who reported that ascorbic acid led to improvement in the histopathological degeneration in tissues by toxic agents as Vitamin C has been established as an antioxidant which removes free radicals produced in the body and scavenge superoxide, hydrogen peroxide, and hydroxyl radicals.

Conclusion: ACR led to histopathological changes in jejunum. These changes attenuated by simultaneous administration of ascorbic acid with ACR.

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