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Protective and ameliorative role of date palm pollen and vitamin C against gastric toxicity induced by bisphenol-A in male albino rat

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Abstract: This study evaluated the protective and ameliorative role of date palm pollen and vitamin C against bisphenol- A (BPA) induced gastric structure toxicity in rats. Control rats received only food and water. Second group administrated BPA (30 mg/kg b.wt.). Third group was given DPP (100 mg/kg b.wt.).Fourth group was given Vit. C (160 mg/kg b.wt.). Fifth group was given DPP 12 hrs before administration of DPP, while the sixth group was given BPA 12 hrs before administration of BPA. The seventh group was given Vit. C 12 hrs before administration of BPA, whereas the eighth group was given BPA 12 hrs before administration of vit. C. The ninth group was given DPP combined with Vit. C 12 hrs before administration of BPA.BPA induced histopathological changes in the stomach indicated by congestion at the vasculatureand edema of the submucosal layer of the non-glandular part, and congestion of the blood vessels at the muscular layer of the glandular part. However, administration of each DPP and vitamin C before and after BPA, or combined with each other before BPA, restored the normal histological structure of the stomach. So, this study revealed that DPP and Vit. C protected and ameliorated gastric structure against BPA toxicity via their antioxidant activities.
Keywords:Bisphenol-A, gastric tissue, date palm pollen, Vitamin C, male rats.

1Introduction

Bisphenol -A (BPA) has received a great attention due to its toxicity on the environment and biological system in the humans and the experimental animals. BPA is known as endocrine disrupting chemical. BPA is used globally in polycarbonate plastic industries and daily in products such as food packaging, infant feeding bottles and medical devices [1].

BPA can leak into the food from plastic container. So, humans are exposed to BPA mainly via gastrointestinal tract, and then distributed throughout the body via the blood circulation [2]. BPA is absorbed by the gastrointestinal tract, metabolized in the liver into BPA glucuronide then transported to the blood and tissue via endocrine system. It induces neurological toxicity in the wall of the gastrointestinal tract affecting stomach and intestine by decreasing cholinergic neurons in the all parts of the stomach wall [3]. The toxic effect of BPA takes place via induction of oxidative stress and inhibition of the anti-oxidant enzymes activities [4-5].

Date palm pollen (DPP) is a male reproductive cell of palm flower of *Phoenix dactylifera* [6].It is fine powder-like material produced by seed palm [7]. It has been used as a traditional herbal medicine for improving male and female fertility by ancient Egyptian and Chinese peoples [8]. It is a natural antioxidant polyphenol and flavonoids used for improving hepatorenal structure and function, hematological parameters and biological system against the toxicity of the environmental harmful materials [9]. Also, it has anti-inflammatory and anti-diarrheal activity [11-12].

Vitamin C is a water soluble natural antioxidant acts as free radical scavenger in the biological systems reducing the toxic environmental pollutants on the animals [13]. As an antioxidant, Vitamin C acts as hepatorenal protective agent against the toxicity of BPA in rats [14]. Also, it reserved the effect of BPA-induced oxidative stress in the testis of rats to the normal structure, via its antioxidant activity [15].

Moreover, as evidence for the dangers of BPA exposure grows, most studies have been focused to evaluate these dangers on male and female reproductive system with limited studies on the toxic effect of BPA on the gastric structure. Therefore, the current study was conducted to evaluate the toxic effect of BPA on the histological structure of the non-glandular and glandular parts of the stomach in male albino rats. Furthermore, we examined whether administration of each DPP and vitamin C before and after

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BPA could treat the histological changes induced by BPA.

2 Material and Methods

Chemical:

BPA and vitamin C used in the current study were purchased from LOBA Chemé Company (India). Date palm pollen grains, small oval gametocytes with Fine Park were collected from the farm of agriculture faculty, Sohag University, Egypt. The grains were separated from the park, washed, dried and blended. The powder was kept in the refregeder until use.

Experimental animals

Fifty-four adult male albino rats kept in the Animal house of Zoology Department, faculty of Science, Sohag University were used. The rats were obtained from the animal house of the faculty of Medicine, Assuit University, Egypt. The rats aged 3 months and weighing about 200±20 g. The animals were kept in stainless steel cages in a well ventilated room and were allowed to acclimatize two weeks to the environmental conditions (12:12 h light-dark photoperiod; temperature 24±3 oC). They were allowed free access to food and water ad libitum.

Expermental design:

BPA, DPP and vitamin C were suspended in dist. Water for oral administration in accordance with the body weighs. The treated groups consisted of nine groups with six rats each, as follows:

- Group 1 (control): was provided with food and water only daily.
- Group 2 (BPA-treated rats): 30 mg/kg b.wt/day administrated daily.
- Group 3 (DPP-treated rats): 100 mg/kg b.wt/day administrated daily.
- Group 4 (Vit. C-treated rats): 160 mg/kg b.wt/day administrated daily.
- Group 5 (DPP+BPA-treated rats): DPP was administrated 12 hrs before administration of BPA, daily.
- Group 6 (BPA+DPP-treated group): BPAwas administrated 12 hrs before administration of DPP, daily.
- Group 7 (Vit. C+BPA-treated group): Vit. C were administrated 12 hrs before administration of BPA, daily.
- Group 8 (BPA + Vit. C-treated group): BPA was administrated 12 hrs before administration of Vit. C, daily.
- Group9 (DPP combined with Vit. C+ BPA-treated group): DPP combined with Vit, C was administrated 12 hrs before administration of BPA, daily.

The experiment was conducted for two months.

Histological examination:

The rats were fasted overnight, anesthetized using light diethyl ether, then scarified and stomach was removed, washed in normal saline (0.9 % NaCl) to remove the blood and the connective tissue, and blotted on dry filter paper, then fixed in neutral formaldehyde for histological examination. Gastric tissues were processed and embedded in paraffin wax and sections were made of about 3-5 µm. After staining with hematoxyline and eosin, slides were examined under the microscope (Olympus, Japan) for histological alternations and photographed.

3 Results

All animals were observed daily for mortality and general conditions during the period of the study. No mortality or clinical signs for BPA toxicities wereobserved.

Light microscopic examination of H&E stained sections demonstrated that the non-glandular and glandular parts of the control group showed normal histological structures (Fig. 1; Fig. 2).

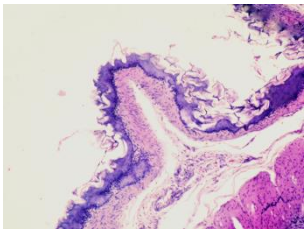

Fig. 1. Photomicrograph of from non glandular stomach section from rats of control group (G1) showing normal appearance. H&E (10x40).

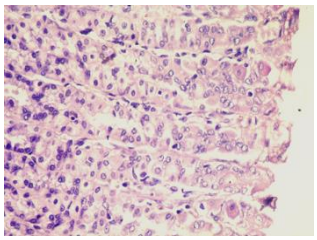

Fig. 2. Photomicrograph of glandular stomach section from rat ofcontrol group (G1) showing normal histological structure of gastric mucosa. H&E (10x40).

The gastric section of the non-glandular stomach part which was treated with BPA (G_2) showed congestion of vasculature (arrows) and edema(arrow heads) at the submucosal layer (Fig. 3A), and that of the glandular part showed congestion of blood vessels(arrows) at the mucosal layer (Fig. 3B). The gastric section from DPP-treated rats (G 3) showed normal non-glandular part of the stomach (Fig. 4 A) like that of the control. Also, the non-glandular sections of the stomach from Vit.C-treated rats (G4) showed normal histological appearance similar to that of the contro (Fig 4B)

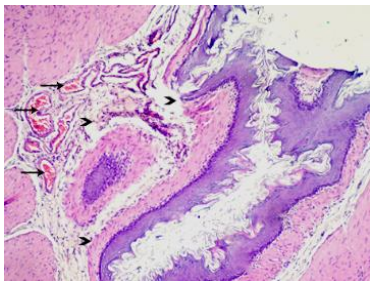

Fig. 3A. Photomicrograph of Non glandular stomach section from BPA.-treated group (G2) showing congestion of the vasculature (arrows), and edema (arrowheads) at the submucosal layer. H&E(10x10).

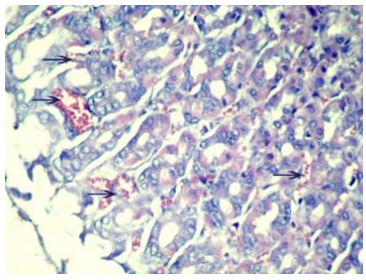

Fig. 3B. Photomicrograph of glandular stomach section from BPA.-treated group(G2)showing congestion of the blood vessels at the mucosal layer (arrows)H&E (10x40).

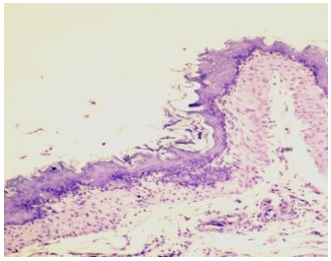

Fig. 4A.Photomicrograph of Non glandular stomach section from rats of DPP-treated group (G3) showing normal appearance.H&E(10x40).

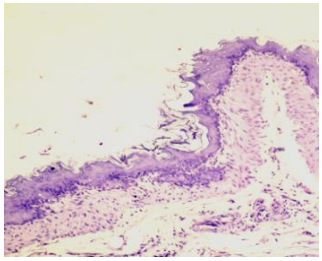

Fig. 4B.Photomicrograph of Non glandular stomach section from rats of Vit. C-treated group (G4) showing normal appearance.H&E(10x40).

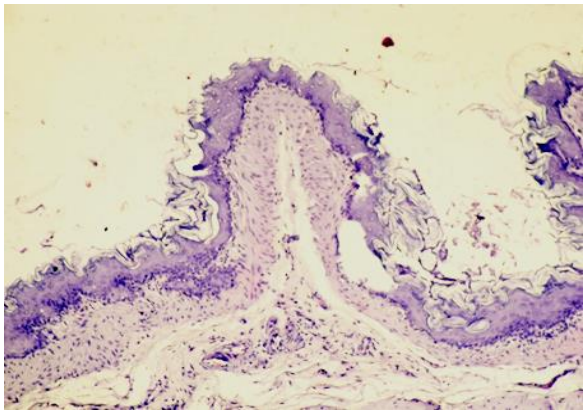

Fig. 5. Photomicrograph of nonglandular stomach section of rats from DPP + BPA.-treated group (G5)showingmore or less normal histological apperance. H&E (10x10)

The photomicrograph of the non-glandular gastric section of rats which was administrated DPP 12 hrs before administration of BPA (G_5) `showed normal histolological structure (Fig5), and that of glandular gastric section which was treated with DPP 12 hrs before treatment with BPA revealed more or less normal histological appearance (Fig. 6A),while the glandular part of the stomach which was treated with BPA 12hrs before DPP showed congestion of blood vessels(arrows) at gastric mucosa(Fig. 6B).

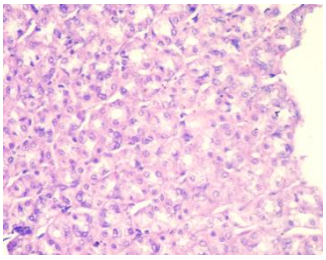

Fig. 6A. Photomicrograph of Glandular stomach section of rats from DPP +BPA group(G5) showing normal histolgicalstrucutre of the gastric mucosa H&E (10x40).

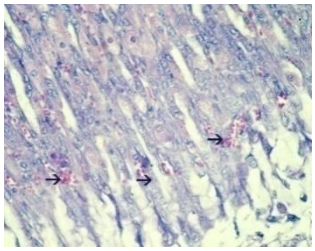

Fig. 6 B. Photomicrograph of glandular stomachsection of rats from BPA + DPP-treated group(G6) showing congestion of the blood vessels at the gastric mucosa (arrows). H&E (10x40).

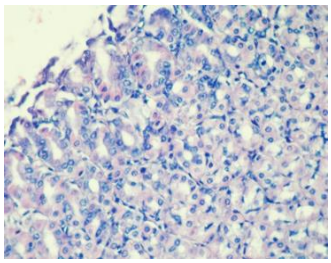

Fig. 7A. Photomicrograph of glandular stomach section of rats from Vit. C + BPA. Treated group(G7)showing normal histological apperance of the gastric mucosa.H&E (10x40).

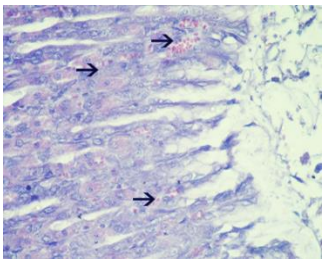

Fig. 7B. Photomicrograph of glandular stomachsection of rats from BPA + Vit.C-treated group(G8) showing sligth congestion of the blood vessel at the gastric mucosa (arrows). H&E (10x40).

The photomicrograph of glandular part of gastric section from the seventh group (G7) of rats which was administrated Vit. C 12 hrs before administration of BPA showed normal histological appearance of the gastric mucosa (7A). But, the gastric section of the glandular part of the stomach from the eight th group of rats which was treated with BPA 12 hrs before treatment with Vit.C

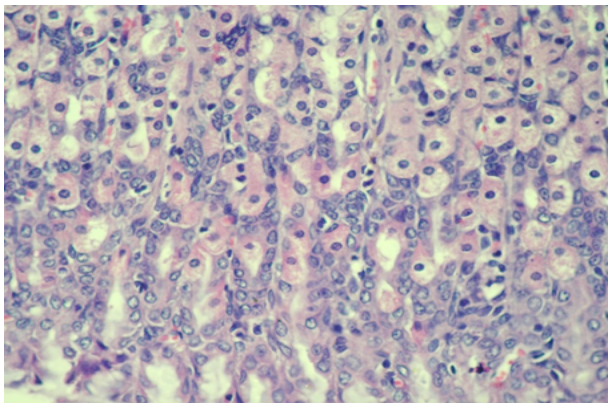

Fig. 8. Photomicrograph of glandular stomach section of rats from BPA + DPP + Vit. C-treated group(G9)showing normal apperance of the gastric mucosa H&E (10x40).

Showed slight congestion of the blood vessels(arrows) at the gastric mucosa (Fig. 7B).

The photomicrograph of the glandular gastric section of rats which was administrated with combined DPP and Vit. C 12 hrs before administration of BPA (G_8) revealed normal appearance of the gastric mucosa (Fig.8).

4 Discussion

BPA is globally used as a major component of manufacturing plastics, leading to much consideration about its health effect on humans. The size and structure of BPA are like estradiol, acts on estrogen receptors, disrupting the endocrine function. Many studies have been performed on BPA exposure, indicating its toxic effect on the hepatorenal structure, and male and female reproductive systems [16-17].

BPA may be absorbed in the gastrointestinal tract, conjugated by glucoronic acid in the intestine and liver, and

then excreted in the urine within 24 h [16]. The effect of BPA on the gastric structure was not sufficiently studied. Also, the use of the natural products may be good alternative with minimal side effects instead of the synthetic drugs. For many years, traditional herbal plants, such as DPP and Vit. C had been used as nutrient dense food source for the treatment of many diseases.

Our data revealed that BPA administration induced histopathological changes in the gastric structure indicated by congestion of vasculature and edema at submucosal layer of the non-glandular layer of the stomach, and congestion of the blood vessels at the mucosal layer of the glandular part. These results are in agreement with Ismail and El-Meligy [18] who mentioned that administration of BPA to male albino rats resulted in dilated congested blood vessels and submucosal edema at the glandular part of the stomach. It has been stated that edema with BPA administration may be related to the release of histamine following gastric damage which led to vasodilatation and an increase in the capillary permeability, and intestinal fluid [19]. It has been reported that dilated congested blood vessels may be BPA induced severe vascular congestion in the cardiac muscle and lung [20].

It has been reported that administration of BPA to rats with different doses decreases plasma levels of antioxidant enzymes such as SOD and CAT [21]. Free radical plays important roles in toxic-chemical-induced cellular damage that results in the cellular injury and damage of gastric mucosa [22]. So, the hitsopathological changes induced by BPPA on the non-glandular and glandular parts of the stomach can be explained by the increasing of free radicals induced by BPA, consequently upon resulting in the damage of the gastric structure. This assumption was supported by increasing LPO, SOD and NO on oral administration of BPA to rats (El-*sayed et al*., unpublished data). So, it can be concluded that BPA may be induced oxidative stress resulting in the production of free radicals species, leading to damage in the non-glandular and glandular parts of the stomach.

Oral administration of each DPP and Vit. C alone did not change the normal histological structure of the stomach. On the other hand, the oral administration of DPP 12hrs before BPA completely restored the normal histological structure of the non-glandular and glandular parts of the stomach. Also, the oraladministration of Vit.C 12 hrs before BPA, completely restorored the normal histological structure of the glandular portion of the stomach. However, with each DPP and vit. C administration 12 hrs after BPA, a partial restoration of the normal histological structure of the glandular part of the stomach was observed. But, the oral administration of combined DPP withVit. C 12 hrs before BPA, completely restoration of the glandular part of the stomach was noted. So, it can be concluded that each DPP and vit. C had potential protective impact against the toxicity of BPA on the gastric tissue in male albino rats. The observed restoration of the gastric histological structure with DPP and Vit. C administration before BPA may be attributed to their antioxidant activities. Also, it was indicated that oral administration of DPP and Vit. C before BPA caused a highly significant decrease in the serum LPO activity and a highly significant increase in the serum SOD activity (El-sayed*et al*., unpublished data). Thus, it can be stated that the potential protective effect of DPP and Vit. C against the toxicity of BPA on the gastric tissue may be related to their antioxidant activities which prevent the production of free radical species via stabilizing the membrane integrity of the gastric layers.

5 Conclusion

BPA has toxic effects on the non-glandular and glandular portions in the stomach of male albino rats via congestion of the vasculature and edema at the submucosal layer of the non-glandular part of the stomach, and congestion of the blood vessels at the mucosal layer of the glandular part of the stomach.However, administration of DPP and Vit. C before BPA prevented gastric damage via their antioxidant activities. Therefore ,it can be recommended that the extensive use of plastic products containing BPA must be avoided. Also, DPP and Vit. C can be used as altenative products instead of the synthetic drugs to protect workers in the plastic factories. Moreover, further studies should be performed to declare the mechanism of DPP and Vit. C against the toxicity of BPA on the gastric tissues.

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