Biological Forum – An International Journal ***(Special Issue 2015)*** **7**(2): 79-90(2015)

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

The effect of Bisphenol A on serum parameters and morphology of kidney's tissue

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(Received 28 June, 2015, Accepted 07 August, 2015)
(Published by Research Trend, Website: www.researchtrend.net)

ABSTRACT: Bisphenol A is chemical material that because of its positive characteristics is using in polycarbonate plastic and epoxy resins. BPA may be absorbed in the gastrointestinal tract after ingesting products packed in plastic containers. BPA is conjugated by glucuronic acid in bowel and liver and excreted in urine as BPA-glucuronide. Renal injury is one of side effects of exposure to BPA. In this study we used from 4 groups each of 10 rats in each group (control and three groups were receiving BPA at dose 10, 50 and 100?g/kg/day respectively). Rats given BPA by intra peritoneal injection for 15 days. The results showed that low doses of BPA including 10 and 50µg/kg/day have no significant effect on serum concentration of urea, but high dose of BPA namely100?g/kg/day has a significant effect on urea levels ($p<0.05$). The creatinine levels in serum increased accordingly dose depended of BPA in treated groups. Too histologic finding showed that only 100?g/kg/day dose of BPA caused renal lesions such as dilation and propagation of glomeruli and degeneration of epithelium of proximal tubule.

Key words: Bisphenol A, Kidney histology, Urea, Cereati

INTRODUCTION

Currently Bisphenol A (BPA), is a key monomer in plastics and epoxy resins, and used as a additive material on them. BPA adds rigidity, lucidity, lightweight, and resistance to temperature to these products. Polycarbonates are used in plastic containers commonly used in the food industry and at home, such as plastic bottles, lenses, and medical devices. BPA epoxy resins are used as coating in food and beverage cans. However, due to the potential impact on health, in Japan epoxy coating was replaced by a polyester film (Thomson, & Grounds, 2005; Willhite *et al*., 2008). BPA is a chemical switch in endocrine processes, and may impact reproduction, weight, and development. BPA acts like a hormone, altering cellular function at very low concentrations, with the maximum safe levels of 5mg/kg/day (Vandenberg *et al*., 2010). BPA may be absorbed in the gastrointestinal tract after ingesting products packed in plastic containers. Like intestinal phenols, BPA is conjugated by glucuronic acid in intestine and liver and excreted in urine as BPA-glucuronide (Dekant, & olkel, 2008). It acts as an estrogenic compound in both in vivo and in vitro researches.

Also there are some evidences from animal studies that shows changes in adult animals exposed to BPA resulting due to estrogenic activity (Lemos *et al*., 2010) or causing liver damage (Bindhumol *et al*., 2003; Tyl, 2008), thyroid disorders and diabetes mellitus type 2 (Richter *et al*., 2007), pancreatic damage (Ropero *et al*., 2008), and or obesity (Vasiliu *et al*., 2006). There are no studies linking serum BPA to disease in humans, so it is unknown whether there are cutoff levels that may be worrisome, but available studies have evaluated BPA exposure by assessing urinary BPA (Vandenberg *et al*., 2010). A little amount of BPA released from said products leads to human exposure. Thus, in humans, BPA is detected not only in serum and urine but also in the placenta and amniotic fluid. Exposure to BPA containers, such as bottles, for 7 days increases urinary BPA excretion in normal subjects from 1.2 mcg/g creatinine to 2 mcg/g (Carwile *et al*., 2011; Korkmaz *et al*., 2010). BPA shows potential acute, short-term, and subchronic toxicity (Tyl *et al*., 2002; Tyl 2008). Accordingly, the aim of the present study was to investigate the effect of the administration of different doses of BPA on the kidney histology and sera factors in adult male rats.

MATERIALS AND METHODS

Forty male wistar albino rats ranging 250-300 grams were purchased from the Experimental Animals, University of Medical Sciences, Tabriz, Iran. Handling of animals was in compliance with the Guidelines for the care and use of animals for scientific purposes. The animals were caged in a well-ventilated animal room with a 12:12 dark/light cycle and controlled temperature at 23 ± 2.C and all had free access to standard rodent diet and had free access to water supplied in glass bottles.

To investigate the toxicity of BPA, a total of 40 rats were randomly divided into 4 groups consisting of 10 rats. Different groups of rats were administered freshly prepared BPA (Sigma-Aldrich, USA) was dissolved in 0.5 ml of olive oil via intra peritoneal injection at specific concentrations 10- 50 and 100µg/kg/day with 0.5 ml olive oil. The animals were treated via IP injection once daily 10- 50 and 100µg/kg/day. Group I was administered daily 1 µl/kg/day body weight (BW) of pure olive oil. Group II was administered daily 10µg/kg/day BW of BPA. Group III was administered daily 50µg/kg/day of BPA and group IV was administered daily 100µg/kg/day BW of BPA. At 24 hours after receiving the last dose, animals were anesthetized with intra peritoneal injection of 100 µg/kg body weight ketamine and 5 mg/kg body weight xylazine (Atalay and Laaksonen, 2004), and blood samples were obtained from the retro orbital sinus of the eye. Sera were separated for measurement of urea & creatinine. Immediately after blood samples were collected, animals were then sacrificed after exposure to ether and their kidneys were rapidly excised and part of it was immediately put at formalin 10% for histological studies. All data collected were analyzed using statistical package for social sciences (SPSS version 19 for windows) and comparisons were made using the one way ANOVA and regression statistics.

RESULTS

The results of this study indicated that low doses of BPA including 10 and 50 µg/kg/day have no significant effect on serum concentration of urea ($p>0.05$) in compared with control group. In contrast high dose of BPA consisted of 100µg/kg/day has a significant effect on urea concentration ($p<0.05$) (Table 1, Fig. 1A).

Table 1: The effect of IP injection of BPA at three doses on serum parameters in rats.

Group	Urea	Creatinine
Control	44.62 ± 0.35	0.47 ± 0.002
R BPA10 µg/kg/day	45.12 ± 0.29	0.51 ± 0.006
R BPA 50 µg/kg/day	45.57 ± 0.52	0.55 ± 0.014
R BPA100 µg/kg/day	54.72 ± 1.44	0.66 ± 0.014

Data are represented as mean ± SEM. Significantly different compared with the control value at $p<0.05$.

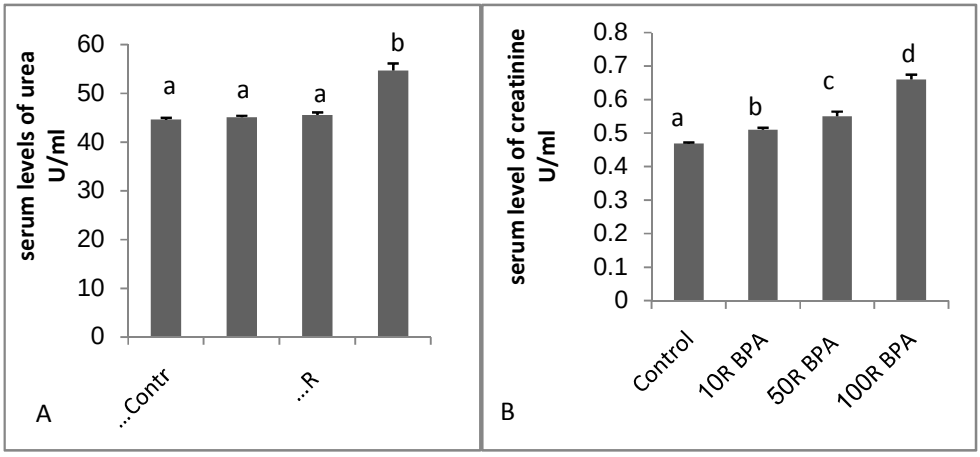

Fig. 1. Effect of BPA at three doses on serum parameters (urea and creatinine) levels in male rats. BPA was administered in rats via intraperitoneal injection for 15 days. Each bar represents the values as mean ±SEM (n=10). Abbreviation: RBPA10, receiving BPA at dose of 10µg/kg/day.

Creatinine is metabolic product of skeletal muscle creatinine. It is used to measure GFR. Excretion of creatinine in urine is equal to filtered of creatinine by glomerulus per minute time. Therefore creatinine clearance is a means to determine of GF. The results of our study showed that creatinine concentration in serum increased accordingly dose depended of BPA. As each three doses of BPA have significant effect on its concentration ($p<0.05$) in compared to control group.

A. Histological analysis

The tests of the BPA-treated animals exhibited morphological changes compared to the control group. BPA at the high dose 100 µg/kg/day cause dilation Bowman's space and hypercelluarity of glomerulus (Fig. 2 A). Also we observed that BPA induced degeneration of epithelial of proximal tube (Fig 2 B).

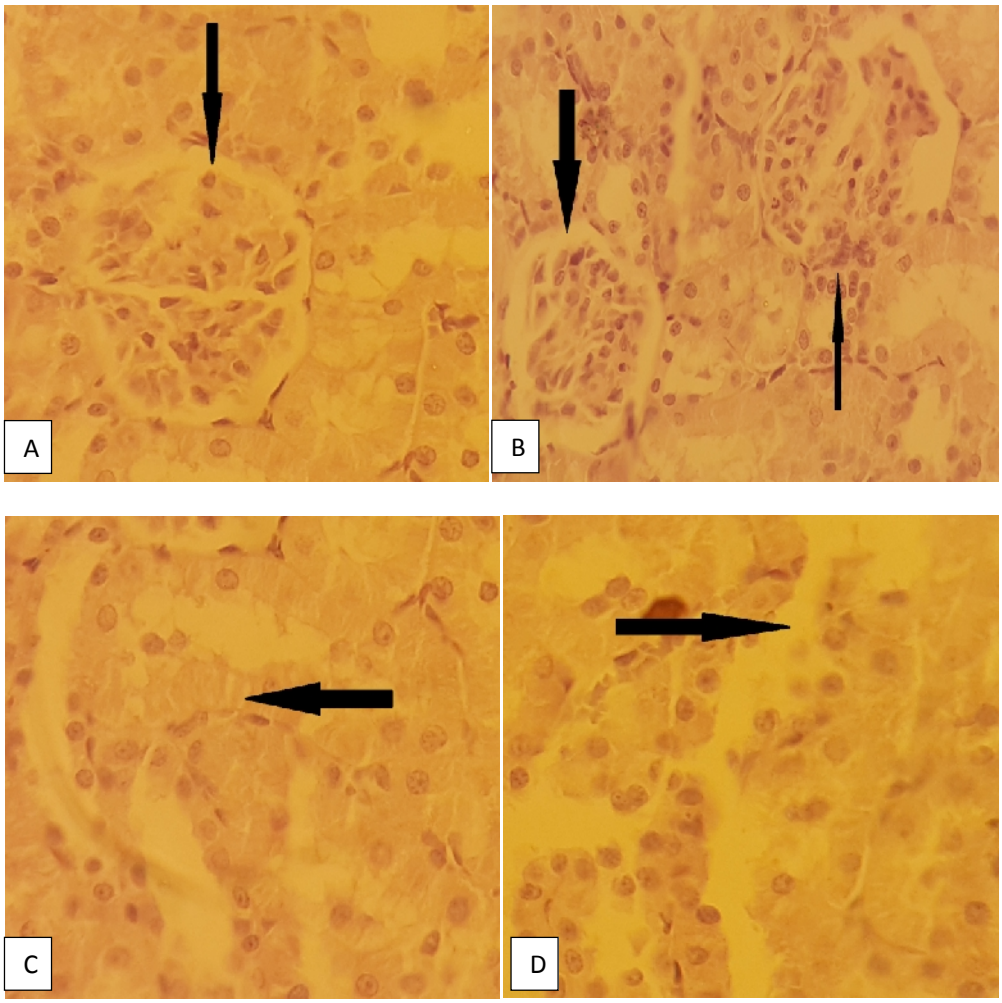

Fig. 2. Photomicrographs of a rat kidney tissue from receiving group BPA at dose of 100 (µg/kg/day). (A, B) Injured kidney tissue, the arrows showed glomerulus given to the dilation of Bowman's space and hypercelluarity (stained by hematoxylin - eosin, magnification × 400). (C, D)Tissue of kidney in group receiving 100 (µg/kg/day) dose of BPA, arrows showed degeneration of proximal tubule epithelium and symptoms of cell's nucleus piknosis (H & E, × 400).

DISCUSSION

BPA is an endocrine disorderly chemical released in environment, so most studies are focused on its effect on reproductive organs (Willhite *et al*., 2008; Haavisto *et al*., 2003). Due to limited information considering the toxic effect of different concentrations on kidney, our study used four different concentrations for evaluating the toxic effect of BPA on male Wister Albino rat. Kidney function testes including urea and creatinine evaluated the presence of kidney damage or disease.

Data presented in our study demonstrate that high dose of BPA 100µg/kg/ day significantly increased the urea and creatinine levels in treated group than control group. BPA levels have been measured in human fluids and tissues in many developed countries of the world. A general consensus has been accepted that BPA can be detected in the majority of individuals in these countries. Urinary excretion of triclosan, and possibly BPA, decreased with decreasing renal function. The associations might differ by age or sex (You *et al*., 2011). BPA is eliminated by the kidney, and increased blood levels have been observed in CKD. Serum BPA is virtually undetectable when renal function is normal. Mean BPA values in patients with CKD were reported to be 0.23 ng/mL. Higher BPA values were reported in patients dialyzed with first generation polysulfone membranes (4.83 ± 1.94 ng/mL) and increased postdialysis (6.62 ± 3.09 ng/mL), while lower values were found in patients using second generation polysulfones (Murakami *et al*., 2007).

As mentioned, BPA is an estrogenic endocrine disruptor molecule of phenolic structure used in plastics, which has renal elimination, and builds up when the glomerular filtration rate decreases. Hemodialysis patients may be exposed to BPA from the environment and also from hemodialysis filters. However, to date no adverse effects have been linked to BPA in dialysis patients (González-Parra *et al*., 2013).

In the kidney, non-significant changes in lipid peroxidation and GSH levels and in the activity of catalase occurred after the daily oral administration of BPA at the two dose levels. The only significant change obtained was an increase in GST activity after 6 weeks of oral administration of 25 mg/kg of BPA (Mourad, & Khadrawy, 2012).

The present data showed non-significant changes in oxidative stress parameters in the kidney due to BPA treatment. However, serum uric acid increased significantly after 6 weeks of the daily oral administration of BPA at both the high (25 mg/kg) and low (10 mg/kg) dose levels. In the light of the present results, this increase in uric acid could not be attributed to impairment in kidney function (Mourad, & Khadrawy, 2012).

The study that carried out by Reimschuessel, 2001 on fish showed that kidney, liver, brain and gills are the most vulnerable organs of a fish exposed to the medium containing any type of toxicant. They reported that after exposure to a variety of renal toxicants, effects in many fish species at all life stages, including rainbow trout, goldfish, tilipia and Zebrafish. The renal tubules are particularly sensitive to toxic influences, in part because they have high oxygen consumption and vulnerable enzyme systems, and in part because they have complicated transport mechanism that may be used for transport of toxins and may be damaged by such toxins. Also the tubules come in contact with toxic chemicals during their excretion and elimination by the kidneys (Tisher *et al*., 1989).

Many chemicals had a direct nephrotoxic action and excreted their effects principally on the proximal convoluted tubules. The presence of necrosis may be related to the depletion of ATP, which finally leads to the death of the cells (Shimizu *et al*., 1996). Bhattacharya *et al*., (2008); Zha *et al*., (2008) showed that NP (Nonylphenol) (at concentration from 37 to 150 µg/l) caused a modification of the structure and function of the kidney and other organs such as the liver and gills (Bhattacharya *et al*., 2008; Zha *et al*.,2008). The organization of the urinary tubules and seminiferous tubules of the kidney was found altered (Roig *et al*., 2014). Tan *et al*., (2003) observed that BPA also caused the enlargement of the kidney and hydronephrosis. Jyothi *et al*., (2009) observed significant increases values in the concentration of serum creatinine when exposed with cyclophosphamide in rats on kidney. Okolie and Osagie (1999) has observed that serum urea and creatinine were significantly higher in the cyanide group relative to controls. Zurovsky and Haber (1995) have reported that sodium nitrate treatment, urea and creatinine were increased in the serum but decreased in the kidney, suggesting an impairment of kidney functions. In this study we observed that distribution of BPA at dose 100 µg/kg/day for 15 days caused significant increasing in serum urea and cereatinine levels. In contrast with our finding in study of carried out by Murmu and Shrivastava, 2014 showed that BPA insignificantly increased the creatinine level after 15, 30 and 60 days as compared to control. They indicated that supplementation of vitamin-C along with bisphenol-A showed recovery in renal values, renal reaction after 15, 30 and 60 days as compared to control group. So, this indicated that vitamin-C works as antidote against bisphenol-A toxicity in Cirrhinus mrigala (Murmu, & Shrivastava, 2014).

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