

Research Article



Urinary Bisphenol A (BPA) Level and Consumption of Water/ Soft Drinks / Food from Plastic Containers

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ABSTRACT

The objective of this study is to estimate Bisphenol A (BPA) level in human urine and correlate the level with extent and duration of consumption of food/ drink from plastic containers. 80 participants who fulfilled the selection criteria were recruited to the study after getting written informed consent. Information on the nature of plastics used, duration of use and demographic details were obtained from the participants. The first morning urine sample was collected from all of them and BPA was estimated using ELISA kit. Data were analyzed using t-test followed by one way ANOVA. All the urine samples were found to contain BPA and its level ranged from 2.79 to 19.45 ng/ml. The average (arithmetic) level was 7.36ng/ml (6.95ng/ml geomean). BPA was higher in males (7.72 ng/ml) than females (6.3 ng/ml). It significantly correlated with BMI. ($p = 0.05$ to 0.001). Homemakers (5.9 ng/ml) had the lowest BPA. Maximum level was seen in those who reused plastics as well as worked with BPA containing materials (9.01 ng/ml). The average level found in this study was higher than the level reported earlier (1.71 ng/ml geomean) in the Indian population. The average urinary BPA level was higher than the levels reported earlier. BPA was higher in males than females. Home makers had the lowest BPA. The BPA levels was maximum in those who re used plastics as well as worked with BPA containing materials.

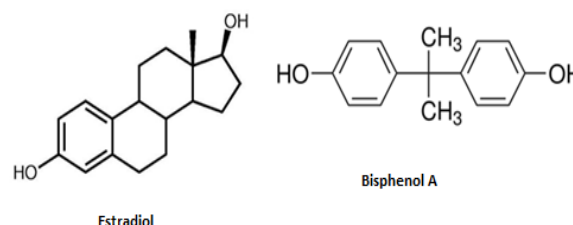
Keywords: Bisphenol A, BPA, polycarbonates, epoxy resins, Estradiol, estrogen related receptor, plastics, consumption.

INTRODUCTION

Bisphenol A (BPA) is one among the highest produced chemicals worldwide¹. BPA was first synthesized by acetone condensation of two phenol equivalents by Russian chemist, A P Dianin in 1891² and has been widely used in the manufacture of polycarbonate plastics and epoxy resins with which common consumer products such as infant feeding bottles, food or beverage packaging and other containers are produced³. BPA is also used in printing industry to produce bill receipts. Use of BPA containing plastics leads to exposure to BPA through food and drinking water stored in and consumed from these containers. Exposure can also occur through BPA containing dental sealants, dermal exposure to printed receipts and inhalation of household dusts^{4,5}. A recent study conducted in 2016 found that, urinary BPA level was higher in population living in close to e-waste recycling location than population living in rural area⁶.

BPA is completely absorbed from gastrointestinal tract on oral exposure and through skin from dermal exposure. First-pass metabolism of BPA occurs in the gastrointestinal tract and liver. BPA is rapidly bio transformed in the liver through glucuronidation to BPA-glucuronide and BPA sulphate and excreted completely in urine^{4,7}.

BPA mimics natural estrogen (Estradiol or E2) due to its phenol groups similar to that of estradiol. It is known as an "Endocrine disruptor" and it competitively inhibits oestrogen due to the structural similarity⁸.



BPA and Estradiol structure⁹

The toxicity of BPA in humans has been the subject of extensive research and is found to be linked with several disorders such as obesity¹⁰, thyroid dysfunction¹¹, liver damage¹², hypertension¹³, diabetes mellitus¹⁴ and prostate cancer¹⁵. A new pathway for BPA in body via a protein known as ERR γ (Oestrogen-Related Receptor) has been identified. It was observed that ERR γ was 1000 times more sensitive to BPA than to oestrogen receptors and ERR γ played an important role in metabolism and that BPA could be a cause of obesity and diabetes¹⁶.

Considering the health risks involved with use of BPA containing materials and the limited studies conducted in India on BPA, the present study was planned to assess the urinary level of BPA in healthy individuals and its association with the extent of use of plastics containing BPA.

MATERIALS AND METHODS

The study was started after obtaining Institutional Human Ethics Committee approval (IHEC/03/04sep 2015/Desp.no.135/25.09.15).



Subjects selection

A total of 150 prospective participants were interviewed from among the healthy subjects who accompanied the patients in a tertiary care hospital. Among them 80 participants were recruited based on the following selection criteria after getting their informed consent.

Inclusion criteria

- Both male & female healthy subjects who were using plastic bottles/containers for drinking water/soft drinks/food for more than 1 year.
- Age between 18 and 60 years.
- Subjects willing to take part in the study

Exclusion criteria:

- Those who use /drink from non plastic containers
- Pregnant women & lactating mothers

A standardized questionnaire was given to each participant and sufficient time was given to them to read. The questionnaire allowed specifying demographic data, physical parameters including height, weight, BMI, occupation, duration of plastic use and reuse of plastic containers. Explanation to the questionnaire was given to needy participants. After getting informed consent from the participants, they were requested to provide 10ml of first morning urine sample in BPA free sterile container. The collected urine samples were stored at -80°C. All the samples were subjected to centrifugation at 2000rpm for 20 min to remove precipitation.

Assay was performed according to manufacturer's protocol.

The results were analyzed based on mean BPA in different groups of the study population and association between BPA level and extent of use of plastic materials.

Statistical analysis

Values were expressed as mean \pm standard deviation, analysis was done by student t-test followed by one way ANOVA, $P < 0.05$ was considered significant.

RESULTS

Among the 80 participants who completed the study, there were 58 males and 22 females. Their age ranged from 18 to 60 years (Fig. 1).

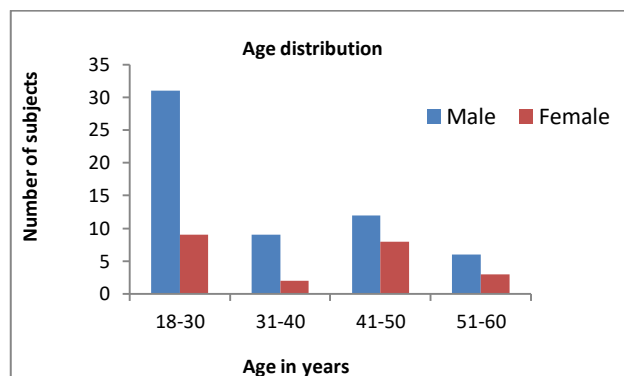


Figure 1: Age distribution

The category of participants included college students (group I), homemakers (group II), and those who worked in corporate and public sector organizations like banks, IT industry and private limited companies (group III). Group IV had participants who were doing their own small business such as selling stationeries, plumbing, running small hotels and working as billers in super markets and ATMs (fig.2).

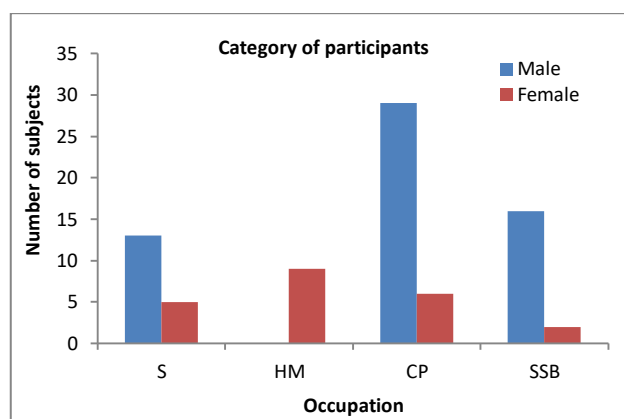


Figure 2: Category of participants

Note: S- Students, HM- Home makers, CP- Corporate and public, SSB- Small scale business

Their BMI ranged from 18.6 to 35.7. The BMI was in the range of overweight and not obesity (25-30) in group II to IV (fig.3).

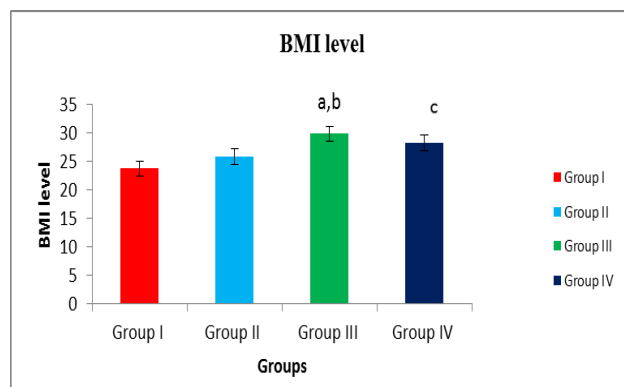


Figure 3: BMI level in groups

All the participants expressed that they had been using plastics since childhood. 85% (68 out of 80) of the

participants had been drinking water from plastic bottles as well as eating food from plastic containers. 49 % (39 out of 80) had often reused plastics. 14 participants were working with BPA containing materials (PVC pipes, lamination materials and printed receipts) (Table 1).

Table 1: BPA exposure and BPA level

Sl. no	Source of BPA exposure	BPA Level
1	Drinking water and eating food from plastic containers	8.23 ± 2.82
2	Reuse of plastic bottles for drinking	9.23 ± 2.53
3	Printed receipt handling	9.11 ± 0.43
4	Occupation related to BPA	9.08 ± 0.79

The BPA level was found to range from 2.79 to 19.45ng/ml. The mean BPA in males was 7.72 and in females 6.3ng/ml and the difference between male and female was statistically significant ($p=0.02$). There was a significant correlation between BPA and BMI in all groups except group II ($p < 0.05$).

The average BPA in group I was 6.2, in group II, 5.9, in group III, 8.5 and in group IV, 7.2. When the difference among the groups was statistically analyzed, significant difference was observed between group III and I as well as group III and II ($p=0.001$) (fig.4).

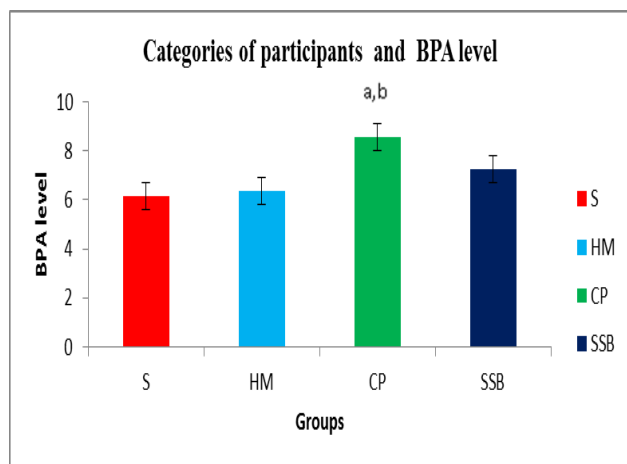


Figure 4: Categories of participants and BPA level.

Note: S- Students, HM- Home makers, CP- Corporate and public, SSB- Small scale business.

The average BPA in the age group between 18 to 30 years was 7.64, in age group 31 to 40 years was 7.42, in group 41 to 50 years, 7.04 and in 51 to 60 years 6.48. When BPA level was compared among the different age groups, the difference was not found to be statistically significant.

Seven males in the study population had occupation related to plastics and their average BPA was 9. Among 39 subjects who reused plastic containers, the minimum BPA level observed was 4.91ng/ml as against the minimum BPA value in the study population as a whole (2.79ng/ml). The mean BPA among the re users was found to be

statistically higher, 9.01 ± 2.57 whereas among the non re users it was 5.73 ± 1.33 (fig.5).

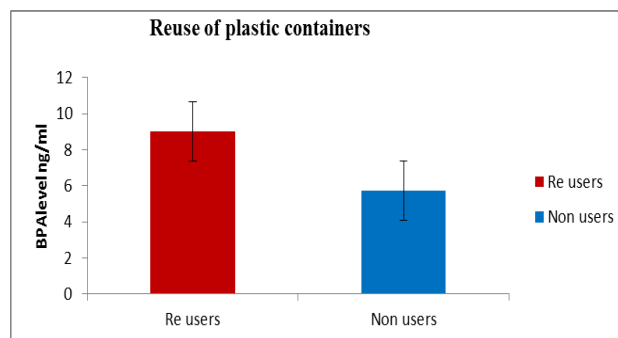


Figure 5: Reuse of plastic containers

DISCUSSION

The present study was undertaken with the objective of finding out the presence of BPA in humans and correlation of BPA level with the extent of use of plastics. As BPA is completely eliminated in urine and testing urine is non-interventional, urine samples were used for testing BPA. 80 participants who met the selection criteria were included and their urine samples were tested. All the urine samples were found to contain BPA and the level ranged from 2.79 to 19.45ng/ml and the average was 7.36ng/ml urine.

The BPA level was higher in males than females (7.4 vs 6.7). The gender difference observed could be due to the difference in the extent of use of plastics. Among the females 9 were home makers and they had a mean BPA level of 5.96 which was the lowest among all the groups. Home makers can consume water/food in household utensils rather than plastic containers which would have contributed to the lowest level of BPA in them.

Increased risk of low birth weight has been associated with prenatal exposure to BPA. Children who had higher level of BPA in urine were found to be at risk for development of Attention deficit hyperactivity disorder (ADHD) and its prevalence was higher in boys than girls¹⁷⁻¹⁸. Similarly prenatal exposure to BPA was also reported to be related to behavioural problems in school going boys than girls¹⁹.

The average BPA (geomean) found in urine among U.S citizens in 2005 was 2.2ng/ml in males and 1.8ng/ml in females²⁰. However these values were expressed in geomean. In our study, the geomean value was also found to be higher, 7.34ng/ml in males and 6.01ng/ml in females.

Gender difference in the level of BPA metabolites (BPA glucuronide and BPA sulfate) was also observed in 30 Korean subjects and males had a higher BPA glucuronide level²¹.

There was an age related decrease from 7.64 to 6.48 in BPA level in the age groups from 18 -30 to 51-60 years (table 2) and the difference in different age groups was not found to be statistically significant.

Table 2: Age and BPA Level

Groups	BPA level
18- 30 years	7.64 \pm 2.56
31- 40 years	7.42 \pm 1.84
41 - 50 years	7.04 \pm 3.19
51 – 60 years	6.48 \pm 1.92

This could be due to the lack of difference in the duration of use of plastic containers among the groups. However the lower level observed in the age group from 51 to 60 years might be because of restricted use of plastic containers among them compared to the younger age group.

In another study, the lowest level was 0.1ng/ml and the highest 30.1ng/ml and the highest level was found in the age group less than 19 yrs. The authors did not find any difference in concentration between males and females²². This again may be attributed to the increased use of plastic containers by the youngsters.

The use of polycarbonate bottles was found to increase urinary BPA concentration in a one week study conducted among 77 college students in Harvard Medical School²³.

U.S. National Toxicology Program panel reported that BPA affected behaviour and neural systems in children. Their study results indicated that most of the feeding bottles available in Indian market contained Bisphenol A irrespective of the brands. Traces of BPA were detected in all the samples collected from Bhopal and Baripada, in India. BPA was also found in the branded samples. Out of the fourteen baby feeding bottle samples analyzed 78.5% samples contained BPA. The maximum concentration of BPA was found to be 9.8ppm. Average concentration of BPA was found to be 1.68ppm. In 50% of BPA-free samples BPA had been detected above threshold limit of 0.6 ppm. In a bottle marked as "0 % BPA", BPA was detected²⁴.

A significant increase in BPA level was detected in cashiers handling thermal papers than other workers²⁵.

The reuse of plastics has correlated with elevated the BPA level in our study and the highest level was observed in printed bill receipt handlers and this was not reported earlier.

Both BPA and BPS were estimated in thermal papers obtained from super markets, bus station and other similar places and was compared with that of papers handled in general population in Brazil and found that the median intake levels of BPA and BPS in the study population was several folds higher, 71 vs 1.42 μ g/d. Such a high level of intake will certainly lead to higher blood level of BPA and its consequent adverse effects. The home makers had the lowest level of BPA. This again confirms the relationship between the extent and duration of plastic use and BPA level. This study has

shown that BPA level is influenced by use, reuse and handling of BPA containing material²⁶.

As discussed earlier, the mean urinary BPA level found in 77 college students after consumption of beverages from polycarbonate bottles for the duration of one week was 2.6 μ g/L and it equals to 2.6ng/ml²³. Even exposure to one week had resulted in a detectable level. The mean BPA level in our study was much higher, 7.36ng/ml. This could be due to the limited period of exposure in the previous study and the long duration of exposure (more than 1year) in our study.

Exposure to BPA is also linked to the increased incidence of polycystic ovarian syndrome (PCOS), ovarian and prostate cancers. BPA level was higher among those with PCOS²⁷. BPA level is also suggested to be a prognostic marker in prostate cancer¹⁵.

BPA and other endocrine disruptor like phthalate can affect signaling pathways involved in glucose metabolism and body weight at the cellular level and that the reports linking BPA and obesity were inconsistent¹⁴. However a positive association between BPA exposure and body fat/obesity was also observed²⁸. In the present study BMI directly correlated with BPA, in support of this observation.

BPA had been detected in newborns also. The plastic medical devices used in neonatal intensive care unit (ICU) were found to contribute to BPA exposure in premature infants. The authors of this study had observed that infants on whom 4 or more medical devices were used over a period of 3 days had significantly higher median urinary BPA level (36.6 μ g/L) than infants who needed 3 or less than 3 devices (13.9 μ g/L)²⁹.

BPA was found in the cord blood of newborns as well as in the mothers of the newborn (2.5ng/ml in the mothers and 0.5ng/ml in the cord blood)³⁰. This finding indicates the placental transfer of BPA as well as caution one has to take in using plastic materials during antenatal period.

The subjects who worked with BPA containing materials like PVC pipes, polycarbonate bottle manufacturing units, printed receipt handling and those who re used plastic bottles had the maximum levels of urinary BPA in our study. A study in Taiwan estimated an increased BPA concentration in water stored for a week in PVC pipe than stainless steel pipe³¹. Leaching of BPA from polycarbonate bottle occurs during cleaning in a dish washer and sterilization with boiling water³². Another study on baby feeding bottles identified that temperature was a crucial factor in leaching of BPA into water stored in polycarbonate bottles³³.

The concentration of urinary BPA estimated in nine different countries is given in table 3.

Table 3: BPA levels among Asian countries

Sl no	Population	BPA level (ng/ml)	Year	Our study
1.	Canada	GM-1.16	201234	AM-7.36ng/ml GM-6.95ng/ml
2.	USA	GM-2.4		
3.	Kuwait	GM-3.05		
4.	Korea	GM-2.17	201122	
5.	India	GM-1.71		
6.	Vietnam	GM-1.18		
7.	China	GM-1.10		
8.	Malaysia	GM-1.06		
9.	Japan	GM-0.95		

AM-Arithmetic mean; GM-Geometric mean

Compared to all the previous reports, a higher BPA was observed in the current study. This may be due to the change in food habits like eating fast foods which are served in plastic containers, changes in quality of life or lack of awareness of BPA.

CONCLUSION

We can conclude from our study that, all the participants had BPA in their urine samples and the average level was much higher (7.36ng/ml) than the levels reported earlier. BPA was higher in males than in females and home makers had the lowest BPA. The BPA levels were maximum in those who reused plastics as well as those worked with BPA containing materials. Considering the negative health effects of BPA on various systems and organs, plastic use in food industry and cooking can be avoided.

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