BISPHENOL A CONCENTRATIONS IN WASTEWATER AND RIVER WATER SAMPLES FROM KENTUCKY AND GEORGIA, USA

Sanaa A[1], Sajwan K[2 *], Loganathan BG[1]

[1]Murray State University, 1201 Jesse D Jones Hall, Murray, KY 42071, USA; [2]Savannah State University, 3219 College Street, Savannah, GA 31404, USA.

Introduction

Bisphenol-A (BPA), 2,2`-bis-(4-hydroxyphenyl)propane ($C_{15}H_{16}O_2$) is one of the compounds of the Bisphenols family having two phenolic rings joined together through a bridging carbon [1] BPA was synthesized for the first time in 1891 by Dianin through condensation of acetone with phenol[2]. The industrial applications of BPA were discovered in the late of 1930s. Since the 1940, a large quantity of BPA has been produced and used as a plastic component. BPA was used in can linings by the 1960s. Currently, over 8 billion pounds are produced annually throughout the world. BPA is used in the manufacture of polycarbonate plastics, epoxy resins and flame retardants.[3,4] Epoxy and polystyrene resins are used in plastic coating, medical devices, food packaging, linings of beverage and food cans, optical lenses, baby bottles, and water containers. [5,6] Materials that contain this material are marked with the number 7 or the letters "PC" near the recycle symbol.[7] Furthermore, BPA is used in the final manufacturing process of protective coating, automotive lenses, protective window glazing, compact disks, building materials, thermal paper, paper coating, and electronic parts. It is also used in some polymers in dental sealants or composites.[7]

Due to its widespread use in industrial applications and many household products, Bisphenol A has been detected in wastewater treatment plant samples, lakes, rivers, the ocean, sediments, soil, the atmosphere, foodstuffs, beverages, paper products, indoor dust, as well as in human blood and urine samples.[7] BPA is most frequently reported in samples from municipal wastewater discharges and in leachate from landfills. BPA has attracted a lot of public attention due to its potential association with adverse health effects such as prostate cancer, obesity, diabetes, neurobehavioral, and reproductive problems.[8] This study was conducted to determine levels of Bisphenol-A in water samples collected from the local wastewater treatment plant, Bee Creek, and Clarks River in Kentucky (Figure 1). For comparison purposes, three wastewater treatment plant samples from Georgia were also analyzed.

Population demographics of Murray, Kentucky in 2014 and 2015 reveal that the Murray community population was 17,741 not including the Murray State University (about 10,500 students). The Murray WWTP is a small facility that processes about 5.2 million gallons per day. It receives wastewater from homes, small businesses, schools, hospitals, and Murray State University. Knowledge on the current levels of BPA in influent, effluent, upstream and downstream of Bee Creek, and Clarks River is essential in order to understand the contamination levels and behavior of BPA in these aquatic systems, prevent further contamination, and to protect living organisms in the western Kentucky watershed. Based on this knowledge of Murray and the persistent properties of BPA, it can be hypothesized that detectable levels of BPA may be found Western Kentucky waters and WWTP. The objectives of this study were to (i) identify and determine the levels of BPA in sewage and river water samples using GC-MS, (ii) determine the spatial variation in BPA levels in wastewater, Bee Creek River, and Clarks River and (iii) determine the sources of BPA to the Bee Creek, and Clarks River

Materials and Methods

Sampling sites including Murray Wastewater Treatment Plant (influent, effluent, upstream of the MWWTP of Bee Creek, and downstream of the MWWTP of Bee Creek), upstream Bee Creek at 4th Street, upstream Clarks River at Highway-94, and downstream Clarks River at the Squire Holland Rd bridge (Figure 2). In addition, samples of deionized water were collected in the laboratory and drinking water from the water fountain in the Chemistry building in order to study contamination profiles of Bisphenol-A (Figure 1). In addition, influent and effluent samples from the President Street, Cross Roads and Tybee WWTPs (from Georgia) were also analyzed.

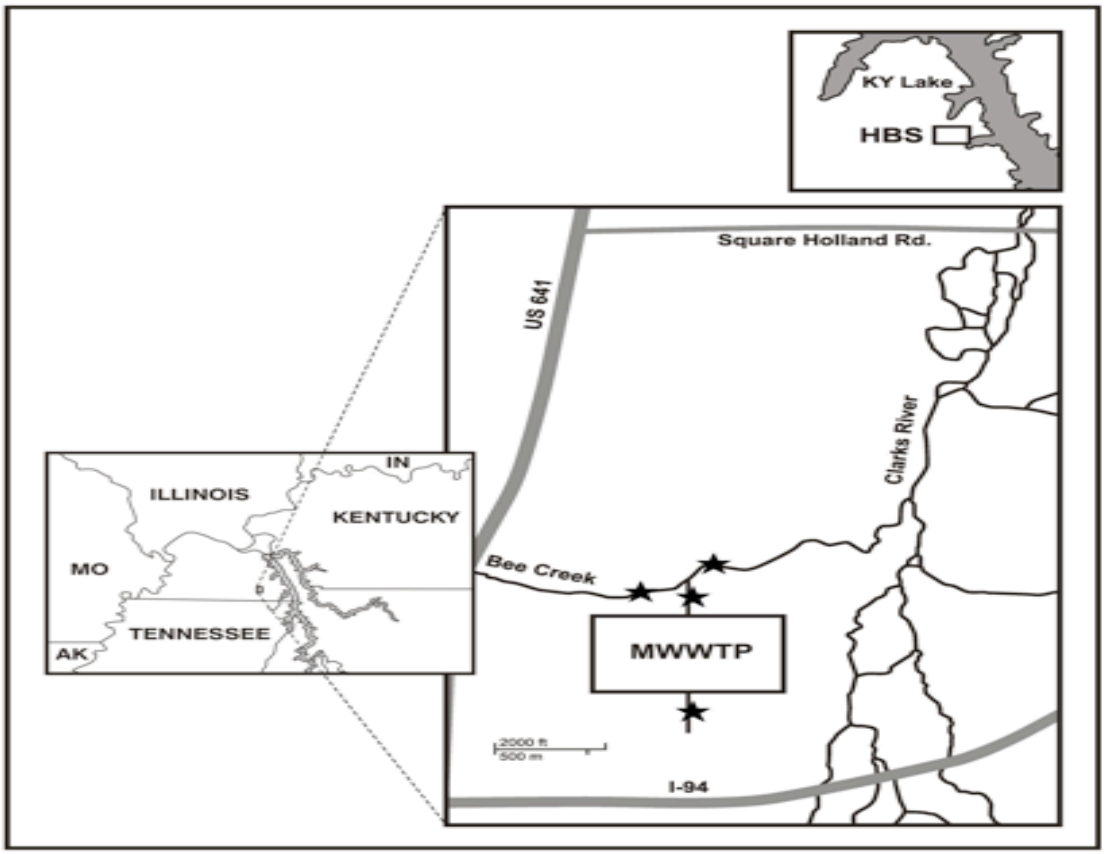

Figure 1. Map showing sampling locations (★). MWWTP: Murray Wastewater Treatment Plant.

About one liter of wastewater from MWWTP and the target samples from each site were filtered using the cellulose acetate filter with a pore size of 0.45µm (Millipore HAWP 04700) under vacuum. About 700 mL of sample was transferred to a pre-cleaned separatory funnels. 0.030 -0.034 g of NaCl and 4-5 drops of 50% HCl were added to the water samples. 50 mL of dichloromethane was then added. The samples were extracted by vigorously shaking for 10 min and the organic phase was collected in pre-cleaned Erlenmeyer flasks. Another 50 mL of dichloromethane was added and extracted for 10 minutes more, and the organic phases collected were combined for further analysis. The extract was passed through anhydrous sodium sulfate (Na2SO4) salt to remove water molecules adhering to the eluent.[9] The eluent was concentrated to 5 mL in a Kuderna-Danish (KD) concentrator then transferred to test tubes. A gentle stream of ultra-pure nitrogen gas (>99.9 %) was used to reduce the sample eluent volume close to dryness. The BPA was then derivatized by adding 70 µL of BSTFA [N,O-bis(trimethylsilyl)-trifluoroacetamide)], 20 µL of 1% TMCS (trimethylchlorosilane) and 10 µL of pyridine to the test tube and then placed in the oven at 70 ºC. for 15 minutes.[10] Once complete, the reaction mixture was blown to dryness under N2 gas and then 1 mL of hexane was added. Analysis was performed by GC-MS. Three characteristic ions namely m/z 372, m/z 357, and m/z 207 confirmed the presence of BPA. The primary molecular ion m/z 372 and the daughter ions (secondary ions) m/z 357, and m/z 207 were examined in each of the samples analyzed. BPA quantitation in samples were determined using peak area of TIC and the slope determined using the calibration standard.

Results and Discussion

Table 1 shows bisphenol A concentrations found in various samples analyzed. Among various samples analyzed Murray wastewater treatment plant influent samples exhibited relatively higher concentrations of bisphenol A than other samples. Deionized water and drinking water samples revealed concentrations below the detection limits (<2.3 ng/L). Bisphenol A was barely detected or non-detectable in the upstream of wastewater treatment plant samples in Kentucky and most samples from Georgia (Table 1).

Table 1**.** BPA Concentrations (μg/L) in water samples collected from various locations in Murray, Kentucky and Georgia.

Sample Location	**Survey No.1**	**Survey No.2**
Blank (DI water)	BDL(<2.3ng/L)	Blank (DI water)
Influent (MWWTP)	5.27	2
Influent-duplicate	NA	2.5
Effluent (MWWTP)	1.4	BDL(<2.3ng/L)
Downstream Bee Creek	3.14	BDL(<2.3ng/L)
Upstream Bee Creek	1.71	0.81
Bee Creek at 4th Street	1.99	NA
Clarks River at Squire Holland Rd.	1.81	NA
Clarks River at Highway-94 site	1.65	NA
Drinking Water (Chemistry Building)	BDL(<2.3ng/L)	NA
President Street, Savannah, GA Influent Effluent	 0.7 BDL (<2.3ng/L)	NA
Cross Street, Savannah, GA Influent Effluent	 BDL (<2.3ng/L) BDL (<2.3ng/L)	NA
Tybee Island., Savannah, GA Influent Effluent	 2.32 BDL(<2.3ng/L)	NA

This preliminary study provides an assessment of Bisphenol-A contamination in the Murray Wastewater Treatment Plant, Bee Creek, and Clarks River samples in Kentucky and WWTP samples from Savannah, Georgia. Detectable amount of Bisphenol-A was found in the Murray Wastewater Treatment Plant, Bee Creek, and Clarks River samples as well as WWTP samples from Georgia.. Among the various samples analyzed, Murray Wastewater Treatment Plant influent samples contained relatively higher concentration of Bisphenol-A than other samples. Measurable amount of BPA in effluent waters indicate that the wastewater treatment process did not remove BPA completely during the process. Therefore, Murray Wastewater Treatment plant effluent waters seem to be a minor source of BPA to Bee Creek River and Clarks River. Further studies with more number of samples are needed to confirm the above results as well as to determine environmental distribution, behavior and fate of bisphenol A in western Kentucky watershed and in Savannah, Georgia, USA.

References

1. Olea, N., Rivas, A., and Leclercq, G. (2002) *The Journal of Steroid Biochemistry and Molecular Biology*. 82, 45-53.
2. Dianin, R. (1891) Bisphenol A. Zhurnal russkogo fiziko-khimicheskogo obshchestva. 23. pp.492.
3. Vandenberg, L. N., Colborn, T., Hayes, T. B., Heindel, J. J., Jacobs, D. R., Jr., Lee, D. H., Shioda, T., Soto, A. M., vom Saal, F. S., Welshons, W. V., Zoeller, R. T., and Myers, J. P. (2012) *Endocr. Rev.* 33, 378−455.
4. Huang, Y. Q., Wong, C. K., Zheng, J. S., Bouwman, H., Barra, R., Wahlström, B., Neretin, L., and Wong, M. H. (2012) *Environ. Int.* 42, 91−99.
5. Weisbons, W.V., Nagel, S.C., and van Seal, P.S. Large effects from small exposures: III Endocrine mechanisms mediating effects of bisphenol A at levels of human exposure.
6. Padmanaban, V., Siefert, K., Ransom, S., Jonhnson, T., Pinkerton, J., Anderson, I., Tao, L., and Kannaa, K. (2008) *J. Perinatol*. 28, 258-263.
7. Carlisle, J.;, Chan, D., Golub, M., Henkel, S., Painter, P., and Wu, KL. (2009) Toxicological Profile for Bisphenol A. Integrated Risk Assessment Branch office of Environmental Health Hazard Assessment. California Protection Agency. pp51.
8. Staples C. A., Dorn P. B., Klecka G. M., O'Block S. T., and Harris L. R. (1998) *Chemosphere* 36, 2149-73.
9. Del Olmo, M., Gonz6lez-Casado, A., Navas, N.A. , and Vilchez, J.L. (1997) *Analytica Chimica Acta.* 346, 87-92.
10. Ralph, M., and Pamela, S. (2011) *J. Chem.Educ.* 88, 1130-1132.