Investigation of the formation of bisphenol S from the metabolism of selected bisphenol S derivatives

Background

The clearance and metabolism of bisphenol S (4,4'-BPS or BPS, CASRN 80-09-1) and six derivatives (Table 1) were investigated in hepatocytes from rats, mice and humans using high resolution liquid chromatography coupled with mass spectrometry (LC-MS) (Waidyanatha et al., 2018). For some of these BPS derivatives, clearance was rapid and metabolites were rapidly formed. For other derivatives, clearance was slow and metabolites were not formed or easily detected (Waidyanatha et al., 2018). This document summarizes additional information retrieved from the existing LC-MS data for the potential formation of BPS from BPS derivatives from the above two experiments conducted. One experiment was conducted with incubations at 10 µM BPS derivative and analyzed by high resolution LC-MS to identify metabolites formed. However, this approach is limited in sensitivity and may not detect low levels of formation of a metabolite. The second experiment was conducted to assess clearance of the parent, with incubations with 1 µM BPS derivative, and analysis by multiple reaction monitoring (MRM) using a triple quadrupole LC-MS. Multiple analytes were included in this method, with a calibration curve and MRM for assessing the clearance of 2,4'-BPS. Since 2,4'-BPS and BPS are isomeric, they give a fragmentation and similar response when determined by LC-MS, and re-examination of this data generated provided some insight into low levels of BPS formed from metabolism of BPS derivatives.

Methods

The formation of BPS as a metabolite of various BPS derivatives was investigated with available data from in vitro metabolism in hepatocytes using 10 µM BPS or derivative, and with the standard curve generated for 2,4'-BPS during clearance assay (1 µM).

During clearance investigations where analysis was conducted by tandem LC-MS, for all derivatives except for D90, MRMs for BPS or 2,4'- BPS were included in the analysis. Transitions monitored for quantitation were: BPS and 2,4'-BPS, 249.009 → 107.726; Bisphenol A (internal standard, IS), 226.993 → 211.887.

Results

For characterization of metabolites, incubations were conducted at 10 µM, and high-resolution LC-MS was used to identify metabolites formed. This approach has the advantage of collecting data for all masses during the evaluation and can be interrogated after the fact for metabolites not initially discovered. An example of the chromatograms of D8 and BPS in human hepatocytes is shown in Figure 1. Relative to the peak area for D8, BPS represented approximately 0.2%, suggesting that it is formed but at low levels. Detection of BPS as a metabolite in these incubations at low levels did occur, but did not provide quantitative information. The sensitivity of high-resolution analysis is limited compared with using MRMs with a triple quadrupole MS system.

Hence, the data collected from clearance incubations were used to look for the formation of BPS from derivatives where an MRM transition for BPS and 2,4'-BPS was included during the analysis using a triple quadrupole LC-MS/MS. The data showed a very low-level formation of BPS in some of the BPS derivatives. No BPS was detected in TGSA or BPS-MAE.

An example is shown for the formation of BPS in D8 incubations in Figure 2. The limit of quantitation (LOQ) for BPS was 1 ng/mL (0.004 µM), and much of the data generated in these incubations were at or below LOQ, in male and female mouse, and male and female rat. Only the male and female human hepatocytes generated BPS concentrations that rose above LOQ (Figure 2).

In Figure 3, a similar comparison is shown for BPS-MPE, which also had detectable BPS present during the time-course of the clearance incubations using LC-MS/MS analysis. The male and female rat hepatocytes had the highest levels of BPS detected. All of the other preparations, with the exception of female mouse at 30 min, were at or below the LOQ.

Figure 1. BPS in human hepatocytes incubated with 10 µM D8.

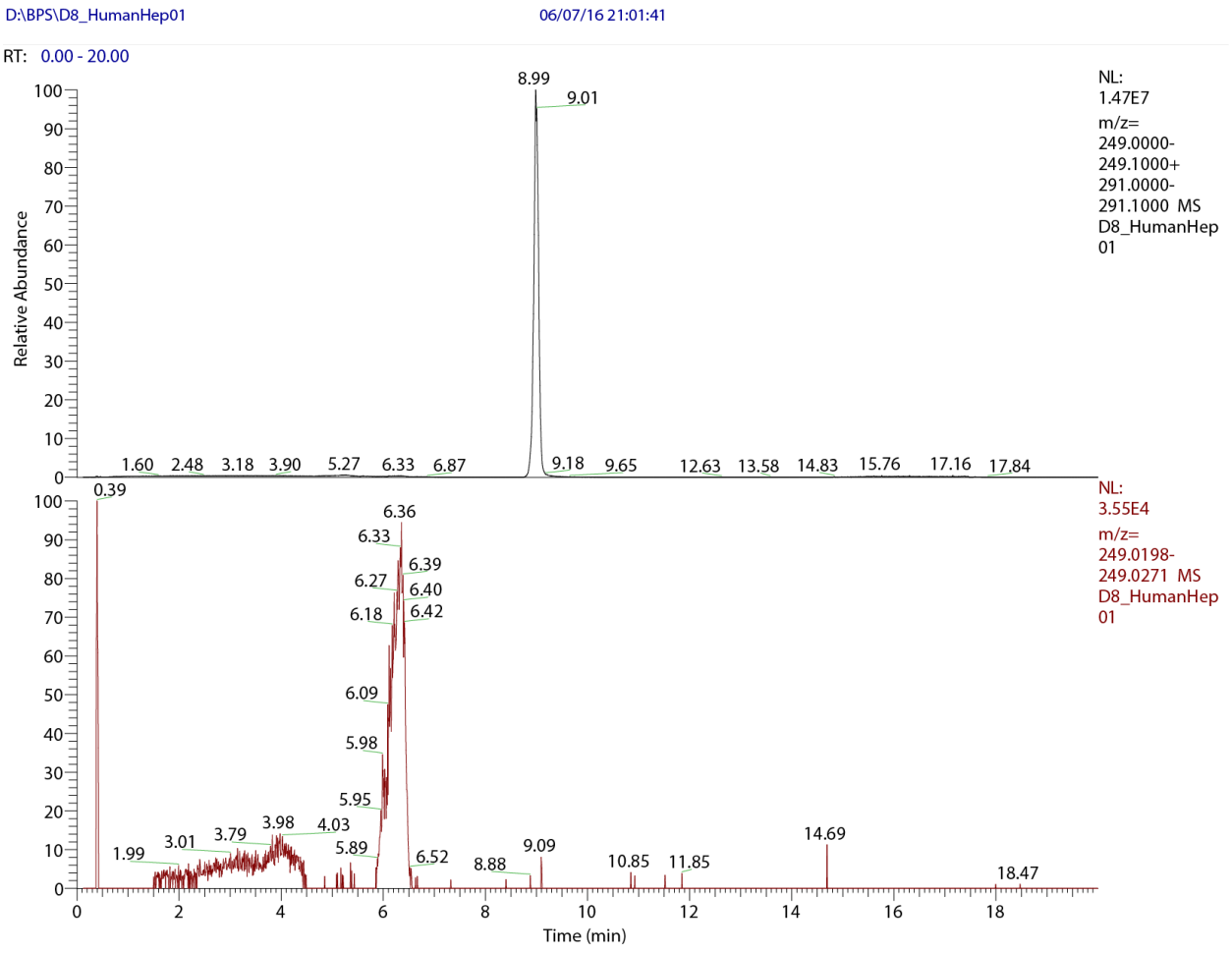


Figure 2. Formation of BPS in male and female human, mouse and rat hepatocytes incubated with 1µM D8.

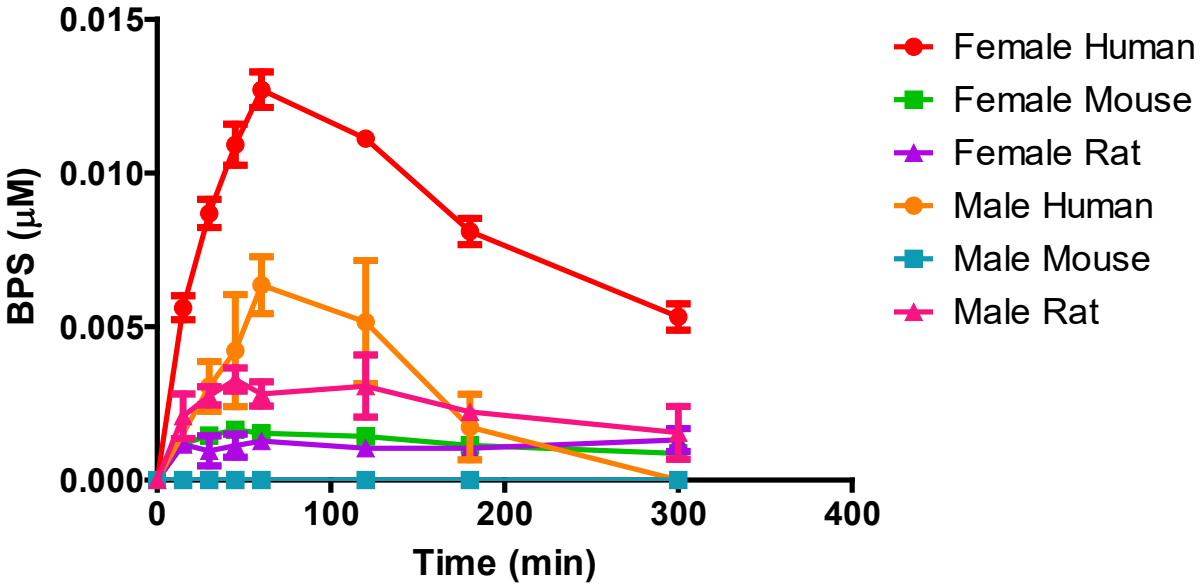


Figure 3. Formation of BPS in male and female human, mouse and rat hepatocytes incubated with 1µM BPS-MPE.

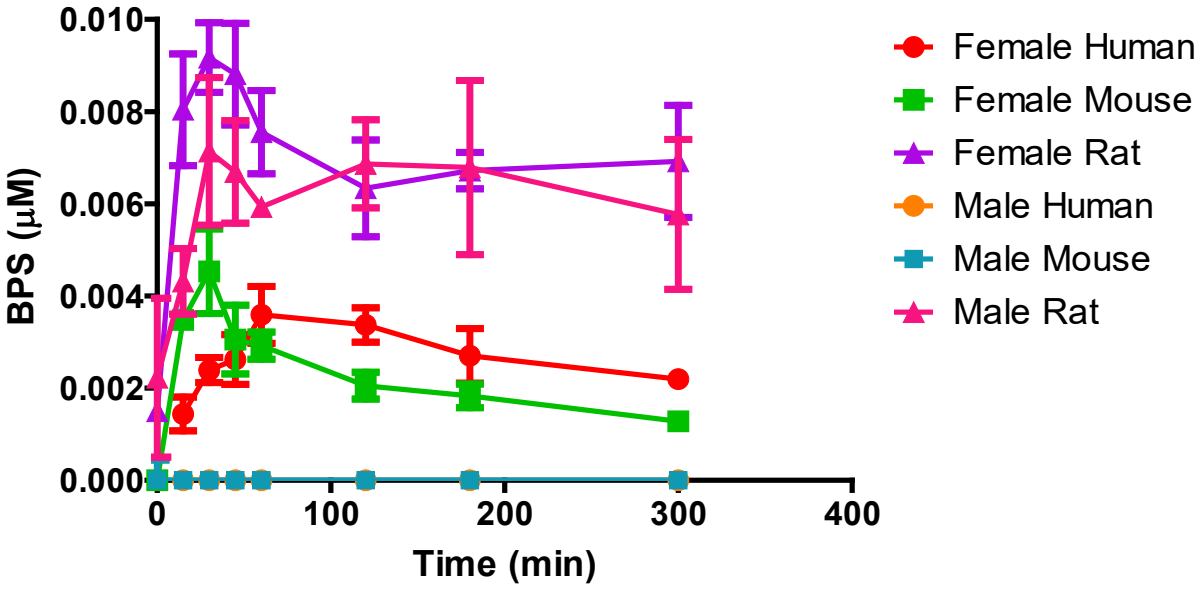


Table 1. Bisphenol S and derivatives used in the study (Waidyanatha et al., 2018)

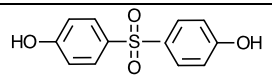
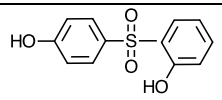
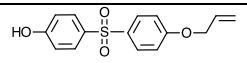
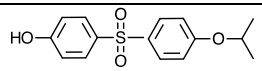
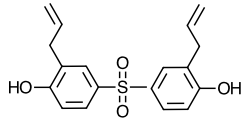
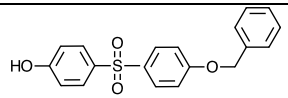
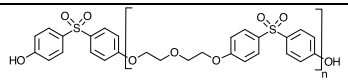
Chemical (Abbreviation)	CASRN	Structure	Molecular Weight	Vendor (Lot No.)	Vendor Purity (%)
Bisphenol S (BPS)	80-09-1		250.27	Sigma-Aldrich (St Louis, MO) (MKBM0826V)	99.3
2,4'-Bisphenol S (2,4'-BPS)	5397-34-2		250.27	TRC Inc. (Ontario, Canada) (1-ATH-73-4)	98
Bis(4-hydroxyphenyl)sulfonylphenyl (BPS-MAE)	97042-18-7		290.3	AV Square Chem Inc. (Monmouth Junction, NJ) (AVSC20142472A)	96.6
4-Hydroxy-4'isopropoxydiphenylsulfone (D8)	95235-30-6		292.35	Santa Cruz Biotechnology (Dallas, TX) (60314)	99.5
4,4'Sulfonylbis[2-(2-propenyl)]phenol (TGSA)	41481-66-7		330.4	AV Square Chem Inc. (Monmouth Junction, NJ) (AVSC20142472Bl)	98.6
4-Benzyloxyphenyl-4-hydroxyphenyl sulfone (BPS-MPE)	63134-33-8		330.39	TCI (Tokyo, Japan) (B06BA)	99.4
[a]Bis(2-chloroethyl)ether-4,4"-dihydroxydiphenyl sulfone copolymer (D90)	191680-83-8		570.12	AV Square Chem Inc. (Monmouth Junction, NJ) (104/SPL-842-01/054)	95.6

Table 2. Regression equations derived from 2.4'-BPS calibration curves used for quantitation of BPS

	Species	Sex	Regression	Weighting	R^2	Curve Equation
2,4'-BPS	Mouse	Male	Linear	1/x*x	0.9957	y = 0.146x + 0.907
2,4'-BPS	Mouse	Female	Linear	1/x*x	0.9965	y = 0.14x + -0.0216
2,4'-BPS	Rat	Male	Linear	1/x*x	0.9934	y = 0.839x + 0.145
2,4'-BPS	Rat	Female	Linear	1/x*x	0.9965	y = 0.107x + -0.00297
2,4'-BPS	Human	Male	Linear	1/x*x	0.9950	y = 0.178x + 0.362
2,4'-BPS	Human	Female	Linear	1/x*x	0.9965	y = 0.14x + -0.0172

References

Waidyanatha, S., Black, S.R., Snyder, R.W., Yueh, Y.L., Sutherland, V., Patel, P.R., Watson, S.L., Fennell, T.R., 2018. Disposition and metabolism of the bisphenol analogue, bisphenol S, in Harlan Sprague Dawley rats and B6C3F1/N mice and in vitro in hepatocytes from rats, mice, and humans. Toxicol Appl Pharmacol 351, 32-45. 10.1016/j.taap.2018.05.008