Supporting Information for

"**Isomer Specific Accumulation of PerfluorooctaneSulfonamide Ethanol-based Phosphate Diester to PerfluorooctaneSulfonate in Japanese Medaka(*Oryziaslatipes*)"**

Hui PENG, Shiyi Zhang, Jianxian SUN, Zhong ZHANG, John P. Giesy, and Jianying HU*

MOE Laboratory for Earth Surface Process, College of Urban and Environmental Sciences, Peking University, Beijing100871, China

Words 406

This supporting information provides tables and figures addressing (1)Multiple reaction monitoring (MRM) transitions of diSPAP and its metabolites; (2) Recoveries (n=3) and method detection limits (MDLs, ng/g wet weight (ww)) of perfluorinated compounds in fish samples; (3) Typical ^{19}F spectra of purfied diSPAP; (4) Typical chromatogram (a) and spectra (b) of purified diSPAP standard using UPLC-Q-TOF;(5) Typical chromatograms of isomers of EtFOSAA, PFOA, EtFOSA and PFOS using BEH fluoro-phenyl column; (6) Typical chromatograms of EtFOSE and B-FOSAA using BEH C18 column; (7) Typical UPLC-MS/MS chromatogram of diSPAP in culture water sample; (8) Typical chromatograms of EtFOSA in standard and exposed fish samples.

Purification of diSPAP by HPLC fractionation. HPLC fractionation was used to isolate diSPAP from technical product FC-807 which was obtained from Hubei Hengxin Company. Fractions were collected at 2-min interval from 65 min to 95 min, and then diSPAP in each fraction was quantified by use of UPLC-MS/MS after 10,000-fold dilution with methanol. Fractions which contained diSPAP were collected and combined, and then evaporated. Fractionation was conducted by use of a Waters HR C18 column (6 μm; 19 mm×300 mm) which was maintained at 40°C. The flow rate and the injection volume were 6 mL/min and 100 μL, respectively. Methanol(A) and ultrapure water (B) were used as mobile phases: 20% A was increased to 50% in 10 min, and then to100% in 100 minand kept for 5 min, followed by a decrease to initial conditions of 20% and held for 20 min to allow for equilibration. After purification, the diSPAP was characterized by NMR spectra (Figure S1). The purified diSPAP (5 mg/L) was also characterized using UPLC-Q-TOF with full scan range from m/z 100-2000. A single peak of diSPAP was clearly observed, and the intensities of other peaks were more than 50-folds lower than diSPAP (Figure S2). Finally, the potential impurities (perfluorooctanesulfonate (PFOS), perfluorooctane sulfonamide ethanols (FOSE), perfluorooctane sulfonamide ethanol (FOSE) based phosphate monoester (monoSPAP), perfluorooctane sulfonamide ethanol (FOSE) based phosphate triester (triSPAP), NEtFOSE, NMeFOSE, perfluorooctane sulfonamide (PFOSA), NEtFOSA, NMeFOSA, 2-(perfluorooctanesulfonamido) acetic acid (FOSAA), NEtFOSAA, and NMeFOSAA) were quantified using targeted monitoring potential contaminants, and none of these chemicals were detected in purified diSPAP (10 mg/L).

UPLC-Q-TOF Analysis. UPLC-Q-TOF was used to characterize the purification of

diSPAP. Separation of purified diSPAP was achieved with a Waters ACUITY UPLC BEH C18column (1.7 μm; 2.1 mm × 100 mm). Injection volume was 5 μl. Methanol (A) and 5 mM ammonium acetate (B) were used as the mobile phases. Initially 10% A was increased to 100% in 8 min and kept for 2 min, followed by a decrease to initial conditions of 10% A and held for 2 min to allow for equilibration. The flow rate was 0.2 mL/min. Mass spectrometry was performed using a Waters XEVO G2QToperated with an electrospray ionization sourcein a negative ion mode. Sodium formate was used for a mass calibration check with the mass range of *m/z* 100-2000, and leucine-enkephalin (MW=555.62 Da) was used as a lock mass. The instrument was set to acquire over the m/z range 100-2000 with scan time of 0.5 s, and data were collected in centroid mode.

SUPPORTING INFORMATIONTABLES1. Multiple Reaction Monitoring (MRM) Transitions of diSPAP and Its Metabolites.

Compound	Acronym	Parent Ion	Daughter Ion	Cone Voltage	Collision Energy
perfluorooctanesulfonamide ethanol (FOSE)based phosphate diester	diSPAP	1203.5	169 526	70	68 48
2-(perfluorooctanesulfonamido) acetic acid	FOSAA	556	498 83	45	24 38
2-(N-methylperfluorooctanesulfonamide) acetic acid	NMeFOSAA	570	419 83	36	22 28
2-(N-ethylperfluorooctanesulfonamido) aceticacid	NEtFOSAA	584	419 83	32	22 22
perfluorooctane sulfonamide	PFOSA	498	78 99	42	34 30
2-N-methylperfluorooctane sulfonamide	NMeFOSA	512	169 219	46	24 22
2-N-ethylperfluorooctane sulfonamide	NEtFOSA	526	169 219	42	30 30
perfluorooctanesulfonamide ethanol	FOSE	602	59	35	30
2-N-methyl perfluorooctanesulfonamide ethanol	NMeFOSE	616	59	35	30
2-N-ethyl perfluorooctanesulfonamide ethanol	NEtFOSE	630	59	35	30

SUPPORTING INFORMATION TALBE S2. Recoveries (n=3) and Method Detection Limits (MDLs, ng/g wet weight (ww)) of Perfluorinated Compounds in Fish Samples.

	Recoveries (%)	MDLs (ng/g ww)
diSPAP	97±4.9%	29
PFOS	87±4.0%	2.0
PFOSA	88±11%	3.9
NMeFOSA	82±13%	2.6
NEtFOSA	91±2.7%	0.9
FOSAA	102±12%	0.4
NMeFOSAA	91±7.7%	1.5
NEtFOSAA	89±11%	2.6
FOSE	95±6.5%	19
NMeFOSE	89±13%	6.3
NEtFOSE	89±9.4%	5.6
$^{13}C_4$-PFOS	92±5%	-
d_5-NEtFOSA	85±7%	-
d_5-NEtFOSAA	98±4%	-

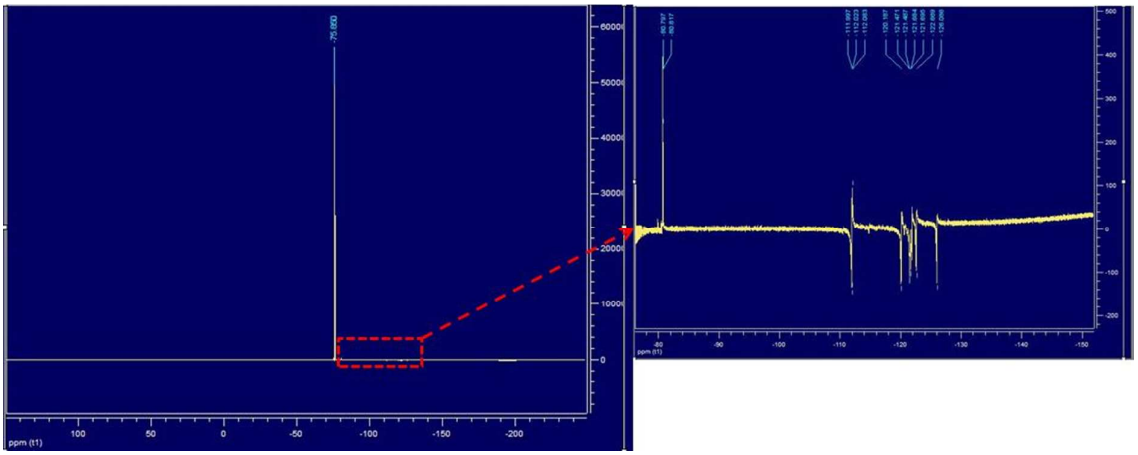
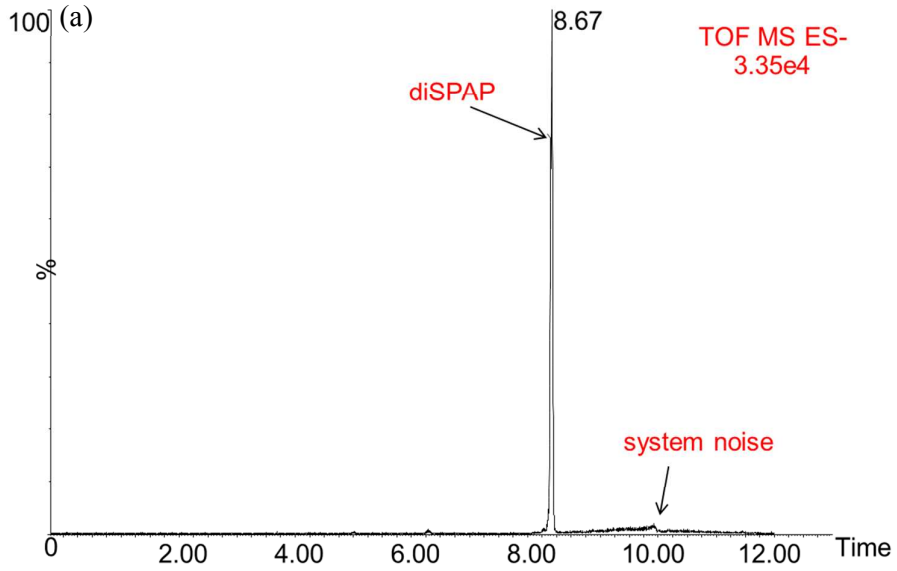


FIGURE S1. Typical ^{19}F spectra of purified diSPAP.



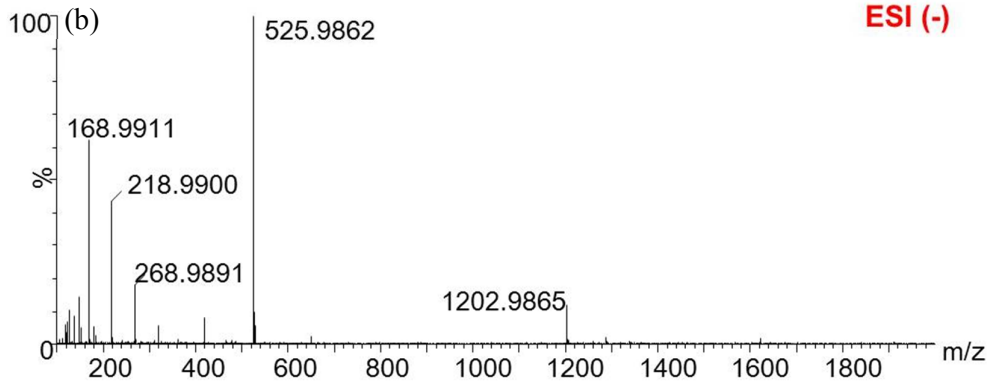


FIGURE S2. Typical chromatogram (a) and spectra (b) of purified diSPAP standard using UPLC-Q-TOF.

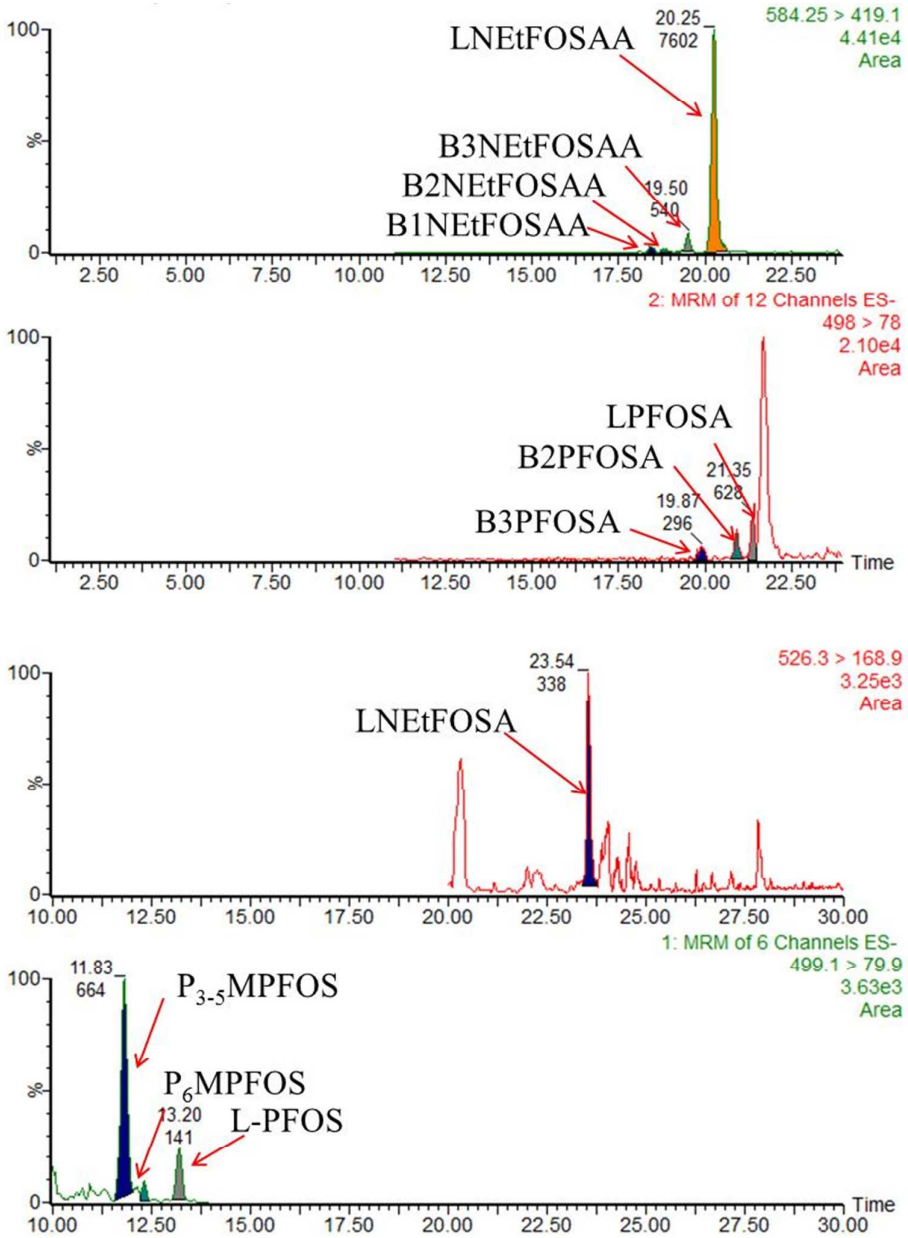


FIGURE S3. Typical UPLC-MS/MS chromatograms of isomers of EtFOSAA, PFOSA, EtFOSA and PFOS using BEH fluoro-phenyl column.

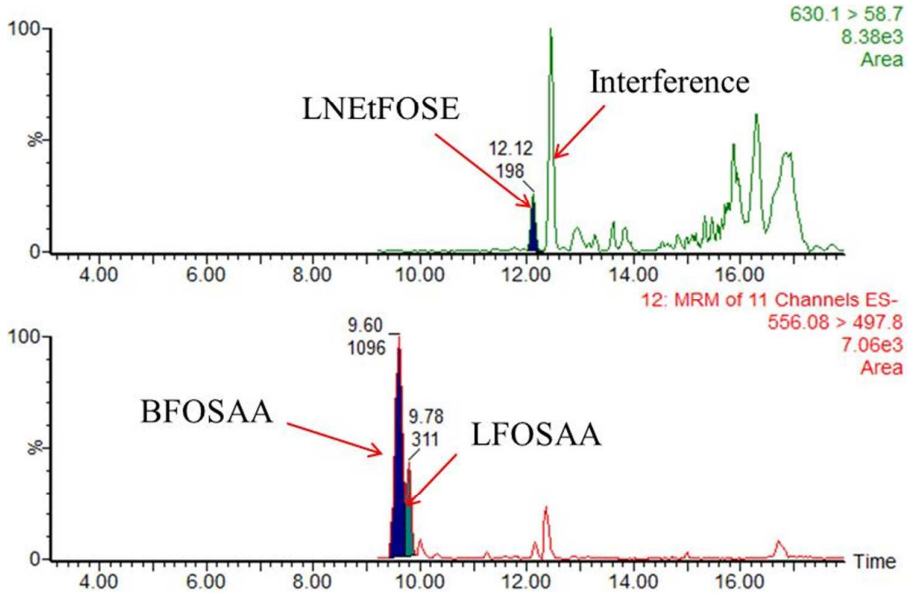


FIGURE S4. Typical UPLC-MS/MS chromatograms of EtFOSE (branched isomers of EtFOSE were not detected) and FOSAA in exposed fish sample using BEH C18 column.

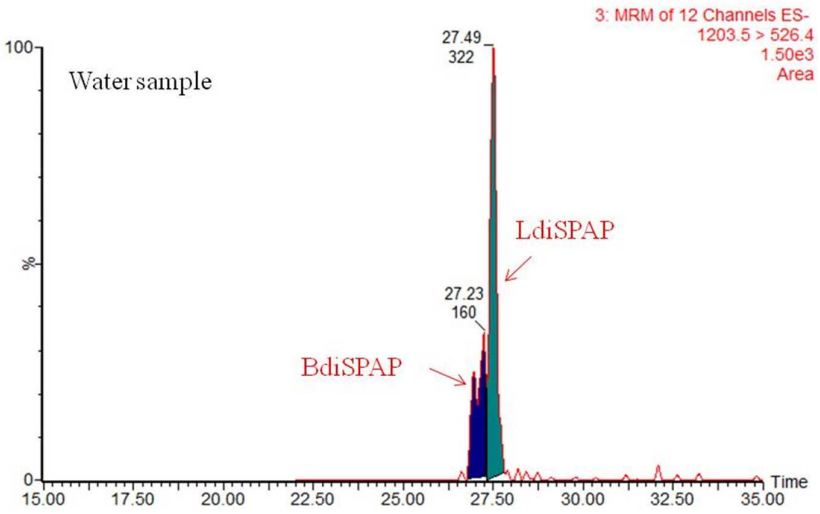


FIGURE S5. Typical UPLC-MS/MS chromatogram of diSPAP in culture water sample.

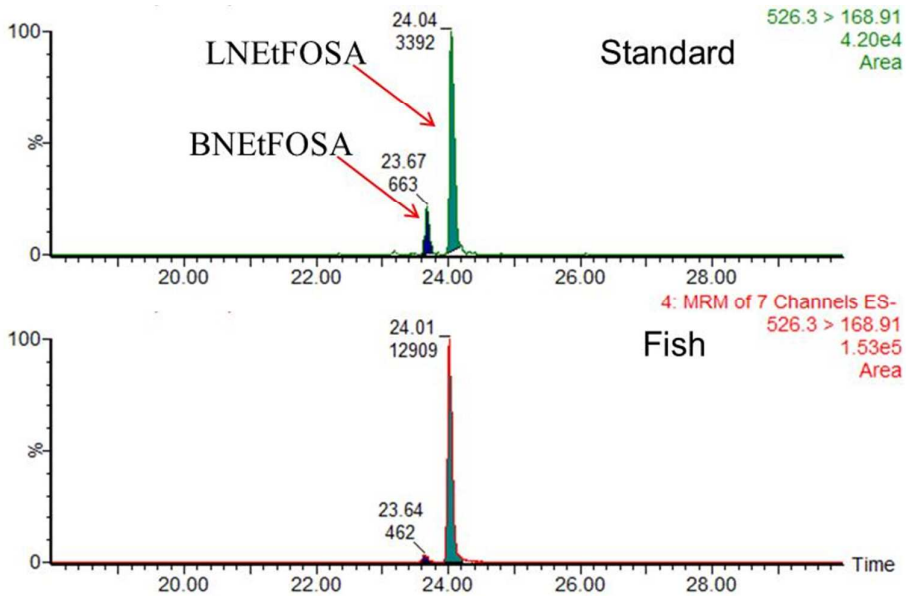


FIGURE S6. Typical UPLC-MS/MS chromatograms of EtFOSA in standard and exposed fish samples.