

1 **Investigating the effects of PFOA accumulation and depuration on**
2 **specific phospholipids in zebrafish through imaging mass**
3 **spectrometry**

4 *Qiuyue Shi^a, Zhengfen Wan^a, Shaoyong Lu^{*a}, Cheng Fang^{b,c}, Changzhou Yan^d, Xian Zhang^{*d}*

5 *a. State Key Laboratory of Environmental Criteria and Risk Assessment, National*
6 *Engineering Laboratory for Lake Pollution Control and Ecological Restoration,*
7 *State Environmental Protection Key Laboratory for Lake Pollution Control,*
8 *Chinese Research Academy of Environmental Sciences, Beijing 100012, China*

9 *b. Global Centre for Environmental Remediation, University of Newcastle,*
10 *Callaghan, NSW 2308, Australia*

11 *c. Cooperative Research Centre for Contamination Assessment and Remediation of*
12 *the Environment, University of Newcastle, Callaghan, NSW 2308, Australia*

13 *d. Key Laboratory of Urban Environment and Health, Institute of Urban*
14 *Environment, Chinese Academy of Sciences, Xiamen 361021, China*

15

16

17

18

19

20

21

22 Table S1. Phospholipid molecular species preliminary identified from LIPID

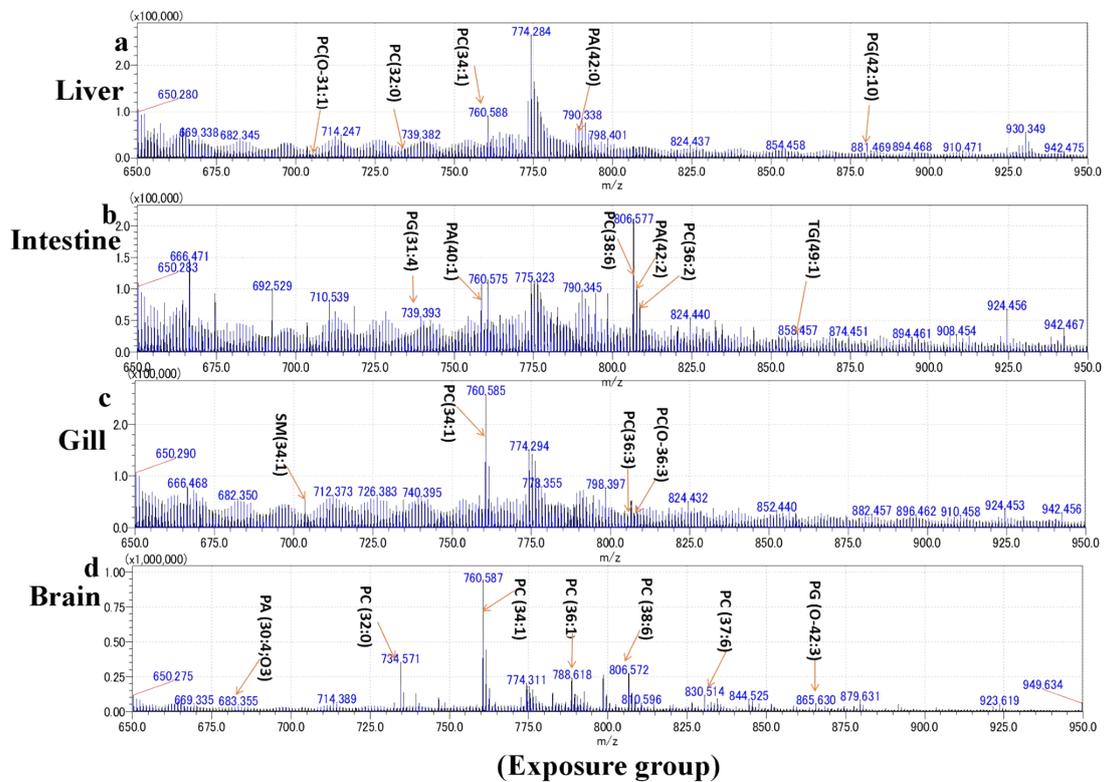
23 MAPS lipid database and the functions of difference phospholipids.

Experimental <i>m/z</i>	Theoretical <i>m/z</i>	Delta	Assignment	Functions of the PL
704.5793/704.5795	704.5589	0.0204/0.0206	PC (O-31:1)	
734.5697	734.5694	0.0003	PC (32:0)	Major constituents
760.5871-	760.5851	0.0023	PC (34:1)	of cell membranes
760.5874/ 760.5857				and lipid signaling
774.6038	774.6007	0.0031	PC (35:1)	
788.6161 /788.6061	788.6164	0.0003	PC (36:1)	
806.5680/ 806.5597	806.5670	0.0010/0.0073	PC (36:3)	
808.5863	808.5851	0.0012	PC (38:5)	
806.5759	806.5694	0.0065	PC (38:6)	
808.5831	808.5827	0.0004	PC (36:2)	
808.5738	808.5617	0.0121	PC (O-36:3)	
829.6486	829.6429	0.0057	PC (38:3)	
762.5947/762.5945/ 762.5940	762.6007	0.0060/0.0062/ 0.0130	PC (34:0)	
792.5622	792.5538	0.0084	PC (37:6)	

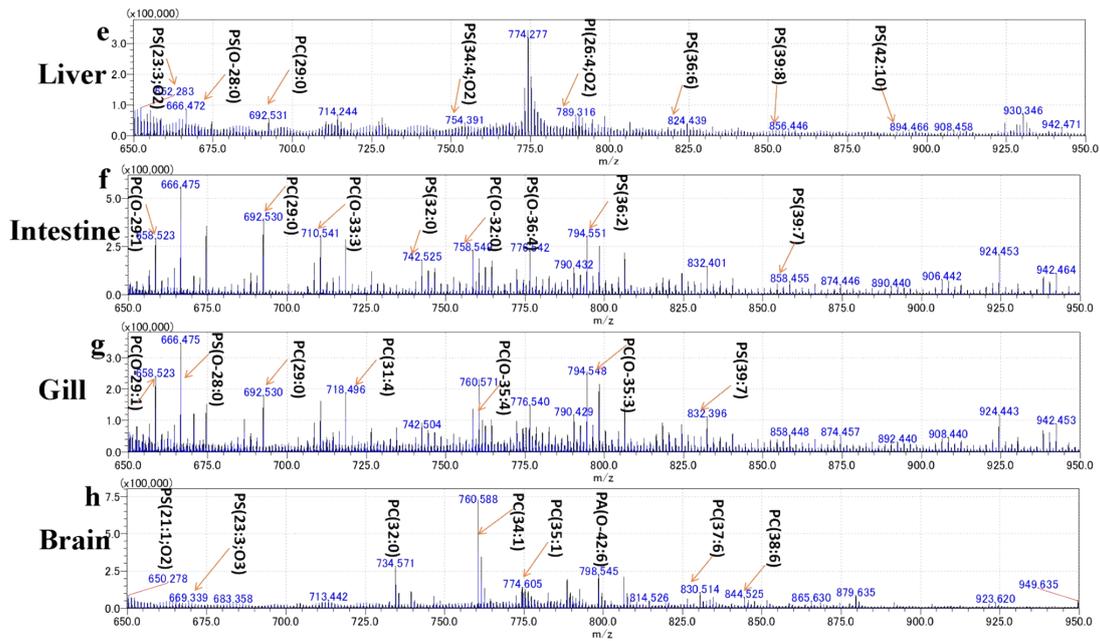
820.5903	820.5851	0.0052	PC (39:6)	
832.5894	832.5851	0.0043	PC (40:7)	
834.6020	834.6007	0.0013	PC (40:6)	
786.5873	786.5773	0.0100	PC (O-34:0)	
784.5631	784.5617	0.014	PC (O-34:1)	
789.6192	789.6368	0.0176	PA (42:0)	Precursor for the
807.5791/807.5792/	807.5874	0.0083/0.0082/	PA (42:2)	biosynthesis of
807.5786		0.0088		many other lipids
785.5873	785.5821	0.0052	PA (O-40:0)	and lipid signaling
759.5750	759.5898	0.0148	PA (40:1)	
761.5913/761.5916	761.6055	0.0142/0.0139	PA (40:0)	
703.5748	703.5636	0.0122	PA (O-37:1)	
775.5974	775.5847	0.0127	PG (O-37:2)	DNA replication,
801.6165/	801.5980	0.0185/0.0148	PG (O-37:0)	modification of
801.6128				cellular lipoprotein
773.5823	773.5691	0.0132	PG(0-37:3)	and cell death
857.6907	857.6995	0.0088	TG (49:1)	Supply and store
703.5958	801.5980	0.0111	TG (39:0)	energy
735.5754	735.5898	0.0144	DG (43:5)	Reduce visceral fat, inhibit weight gain, reduce blood fat
703.5760/703.5758/	703.5748	0.0012/0.0010/	SM(34:1; O2)	Membrane

703.5763		0.0015		components,
821.5799	821.5933	0.0134	SM (40:3;O2)	cholesterol
				interactions &
				homeostasis
807.5788	807.5746	0.0042	PI (O-34:0)	Transfer protein and
				cellular nanoreactor
				for lipid signaling

24 Note: The mass accuracy of the instrument is less than 10 ppm. All phospholipid molecular
 25 species preliminary identified through LIPID MAPS lipid database under mass tolerance \pm
 26 0.05 m/z. To increase confidence in the identification, some MS/MS data of high abundance
 27 phospholipids were generated.



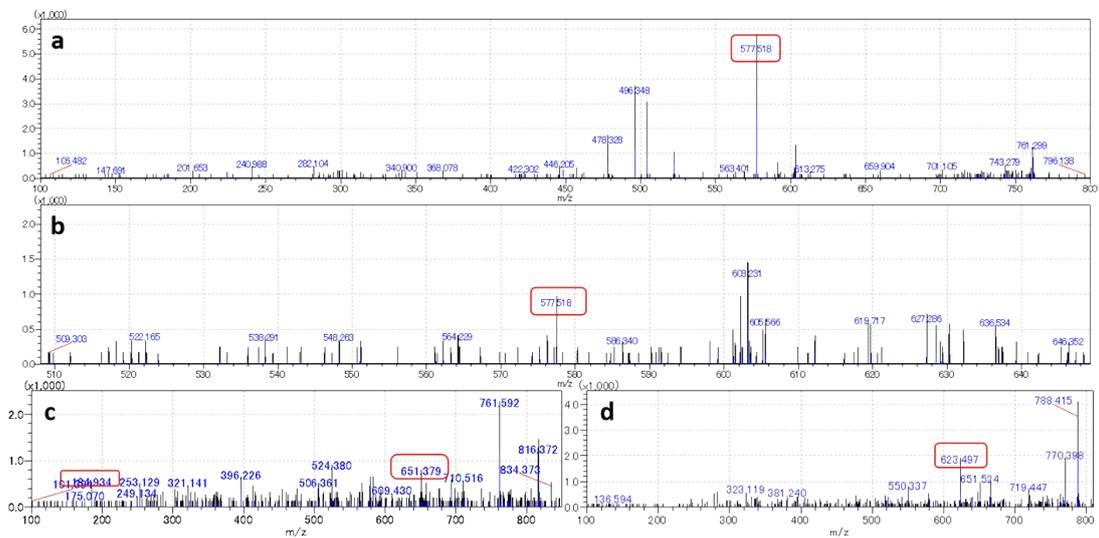
28



(Control group)

29

30 Figure S1: MALDI-TOF mass spectra from zebrafish tissues exposed to PFOA
 31 solution in the positive ion mode (a-d: exposure group; e-h: control group). The mass
 32 spectrum was from an average over the tissue area.



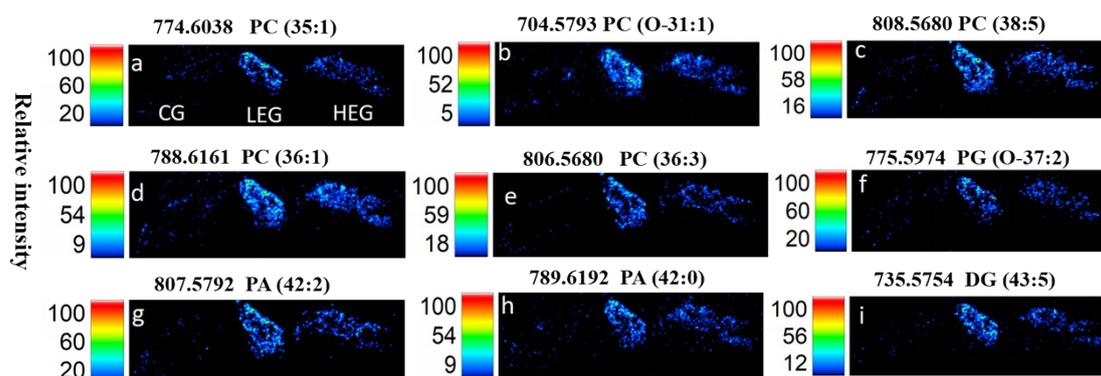
33

34 Figure S2: MALDI-MS/MS spectra of peaks at (a) m/z 760.585 acquired from PC
 35 (32:1) standard; (b) 760.587, (c) 834.602, and (d) 806.568 acquired from a zebrafish
 36 tissue. The red circle shows fragmented ion peaks from the neutral loss of
 37 PHOSPHOCHOLINE (NL183Da).

38 Table S2. Phospholipid molecular species identified from MS/MS experiment directly
 39 performed on the zebrafish tissue in positive ion mode.

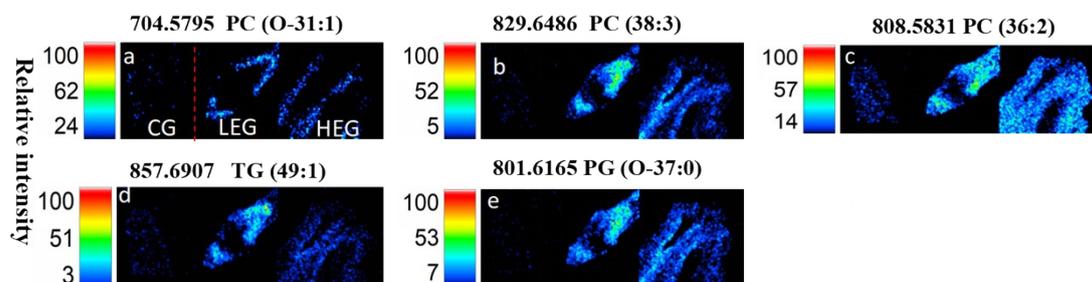
Experimental m/z	Theoretical m/z	Assignment	Structurally specific CID product ions (m/z)	
			Phosphocholine	Neutral loss of PHOSPHOCHOLINE(NL183Da)
760.5874	760.5851	PC (36:1)	184	577
834.6020	834.6007	PC (40:6)	184	651
806.5680	806.5670	PC (36:3)	184	623

40



41

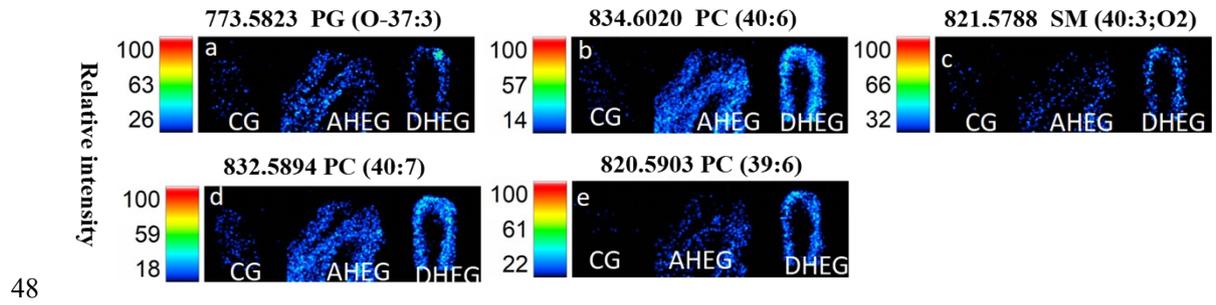
42 Figure S3. Visual distribution of phospholipids in zebrafish liver after PFOA exposure
 43 and control group, a single *m/z* value was indicated for each images. (CG: control
 44 group; LEG: low-exposure group; HEG: high-exposure group).



45

46 Figure S4. Visual distribution of phospholipids in zebrafish intestine after PFOA

47 exposure and control group, a single m/z value was indicated for each images.



49 Figure S5. IMS of zebrafish intestine after PFOA exposure and control group during
50 accumulation and depuration phase in the high-exposure group.