Electronic Supplementary Information for:

Chip-based liquid phase microextraction combined with electrothermal vaporization-inductively coupled plasma mass spectrometry for trace metal analysis in cell samples

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Fabrication of microfluidic devices

Soft lithography and rapid prototyping with PDMS technology are employed for the fabrication of microfluidic devices. A transparent mask patterned with a high resolution laser printer was used to make a master on a silicon wafer with AZ 50 photoresist. Before PDMS casting, the master was exposed to trimethylchlorosilane vapor for 3 min to avoid the adhesion between PDMS and silicon wafer. GE RTV 615 (PDMS) component A and B were mixed at a ratio of 10:1 and cast on the master after air bubbles disappeared. After incubating at 75^{o}C for 3 h, the solidified PDMS was peeled off, drilled on demand, and exposed to oxygen plasma together with a clean glass. Then the PDMS microfluidic chip was prepared by bonding the treated PDMS with the glass.

Sample preparation

Two kinds of Certified Reference Materials, GBW07605 human hair and GBW09152 human serum, were employed for the validation of the accuracy of the proposed method. In addition, three kinds of real samples, human serum sample (Wuhan University Zhongnan Hospital, Wuhan, China), Jurkat cell sample and HepG2 cell sample, were analyzed. Details of the sample preparation are as follows.

The Certified Reference Materials and real human serum samples were weighed into PTFE digestion vessels. After adding 4 mL of HNO_3, the vessels were put on a electric hot plate (85ºC) for 3 h. The vessels were then placed on a turntable and subjected to microwave digestion. The microwave system was operated as follows: 5 min at 10 MPa and 150 ºC, 5 min at 18 MPa and 180 ºC, 5 min at 20 MPa and 200 ºC. After the microwave treatment, the vessels were cooled in air to room temperature, and then put on an electric hot plate (85ºC) again. When the samples were nearly dryness, the digest was transferred and diluted with highly pure deionized water. After the addition of an appropriate amount DDTC (concentration of 0.5% (m/v)) and adjusting of pH to 9.0, the samples were served to chip-based LPME-ETV-ICP-MS analysis.

In addition, Jurkat T cell (purchased from Wuhan University Center for Animal Experiment/A3-Lab, Wuhan, China) and HepG2 cell (kindly provided by Prof. Ying Zhu, College of Life Sciences, Wuhan University, Wuhan, China) samples were countered. After centrifugation, the supernatant was removed and the cells were washed with PBS twice. Then the amount of cells

was counted, and the density of HepG2 cells and Jurkat T cells were diluted to ~3.6×10^5/mL and ~2.2×10^6/mL, respectively. Then 1 mL of the cells dispersion were subjected to ultrasounication for 40 min. Next, a centrifuge was used to separate the cell sap and the deposit. The cell sap was transferred into a polyethylene tube, then an appropriate amount DDTC was added (concentration of 0.5% (m/v)) and the sample solution was adjusted to pH 9.0 with diluted NaOH solution and diluted to 2 mL with highly pure deionized water.

Determination of the procedure blanks

For the determination of procedure blanks, highly pure deionized water was used as the blank sample. After an appropriate amount DDTC was added (concentration of 0.5% (m/v)) and the pH was adjusted to 9.0, it was subjected to the chip-based LPME and the blank values were determined by ETV-ICP-MS.

Effect of flow rate

On a microfluidic chip, flow rate is a great influence factor on the system stability. If the flow rate is too high, it is hard to form stable laminar flow during the extraction. To examine the effect of flow rates, the organic phase flow rate was fixed at 2 μL min^{-1}, while the aqueous phase flow rates were varied over the range of 20~100 μL min^{-1}. As can be seen in Fig. S-1, the aqueous phase occupied more space in the central channel with the increase of the flow rate, while the laminar flow was stable throughout the examination. Then the effect of flow rate on the extraction of target metal ions was studied with the organic phase flow rate ranging from 1 μL min^{-1} to 4 μL min^{-1} by fixing the flow rate ratio of aqueous phase to organic phase at 40:1, and the results are shown in Fig. S-2. The extraction efficiency was not decreased obviously with increasing flow rate from 1 to 3 μL min^{-1}, and then decreased with further increasing the flow rate to 4 μL min^{-1}. Therefore, 2 μL min^{-1} was selected as the organic phase flow rate, and accordingly the aqueous phase flow rate was 80 μL min^{-1}. After extraction for 3.5 min, 7 μL of the organic solvent was collected and introduced into ETV-ICP-MS for subsequent analysis, and 280 μL of sample solution was consumed.

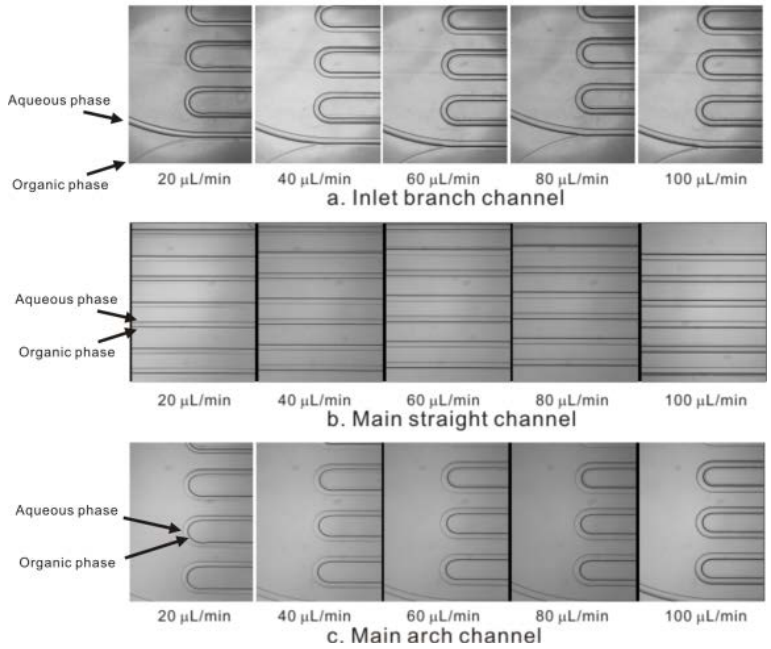

Fig. S1 Visible light images of the extraction of complex from aqueous phase to the organic phase.

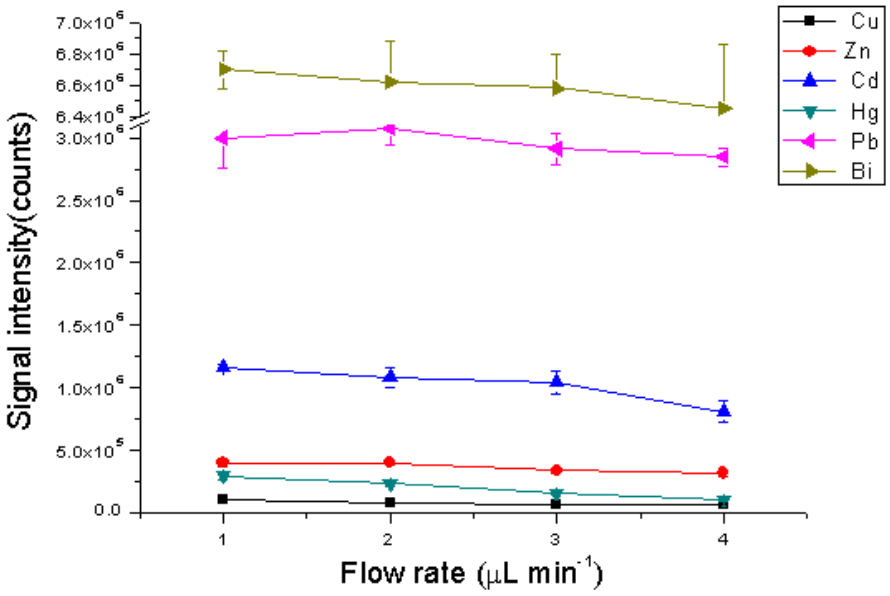

Fig. S2 Influence of flow rate on signal intensity in chip-based LPME.

Table S1 Influence of coexisting ions

Coexisting ions	Tolerance concentration (μg mL^{-1})					
	Cu	Zn	Cd	Hg	Pb	Bi
K^+	5000	10000	10000	5000	10000	10000
Na^+	5000	5000	10000	5000	10000	1000
Ca^{2+}	5000	1000	5000	5000	1000	5000
Mg^{2+}	1000	1000	1000	1000	1000	500
Fe^{3+}	0.5	0.5	0.2	1	1	1
Al^{3+}	1	1	1	1	1	1
Cl^-	8000	16000	16000	8000	16000	16000
NO_3^-	8000	8000	16000	8000	16000	1600
SO_4^{2-}	5000	2000	5000	5000	2000	2000

Table S2 Preparation reproducibility

Analytes	Chip to chip RSD (%,n=4, c=5 ng mL^{-1})	Batch to batch RSD (%,n=5, c=5 ng mL^{-1})
Cu	6.37	9.35
Zn	5.46	7.56
Cd	3.18	4.82
Hg	4.74	6.59
Pb	7.35	6.74
Bi	5.46	7.46

Table S3 Comparison of LODs found in literatures following different analytical approaches.

Methods	LOD (pg mL^{-1})						Time (min)	Volume (mL)	Refs.
	Cu	Zn	Cd	Hg	Pb	Bi			
Chip-based LPME-ETV-ICP-MS	89.3	59.8	7.5	21.6	19.2	6.6	3.5	0.28	This work
micro-fluidic sample introduction system-ICP-MS	-	-	19.8	-	10.4	-	1.3	0.4μL	[7]
HF-LPME-ETV-ICP-MS	12.4	28.7	4.5	3.3	4.8	1.6	15	2.5	[20]
SPE-FI-ICP-MS	30	260	4	-	20	-	13.5	1.8	[22]
MNPs-SPE-ICP-OES	-	200	300	-	800	-	5	50	[23]
MNPs-ICP-MS	600	600	-	-	60	-	5	15	[24]

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