

## Supplementary materials

# Determination of selenomethionine, selenocystine and methylselenocysteine in egg sample by high performance liquid chromatography - inductively coupled plasma mass spectrometry

Yue Zhao <sup>1,2</sup>, Min Wang <sup>1,2,\*</sup>, Mengrui Yang <sup>1,2</sup>, Jian Zhou <sup>1,2</sup> and Tongtong Wang <sup>1,2</sup>

<sup>1</sup> Institute of Quality Standards and Testing Technology for Agro-Products, Chinese Academy of Agricultural Sciences, Beijing 100081, P.R. China;

<sup>2</sup> Key Laboratory of Agro-Product Safety and Quality, Ministry of Agriculture and Rural Affairs, Beijing 100081, P.R. China.

zhaoyuecaas@163.com (Y.Z.); yangmengrui2014@163.com (M.Y.); zhoujian\_8382@163.com (J.Z.); wangtong123@126.com (T. W.)

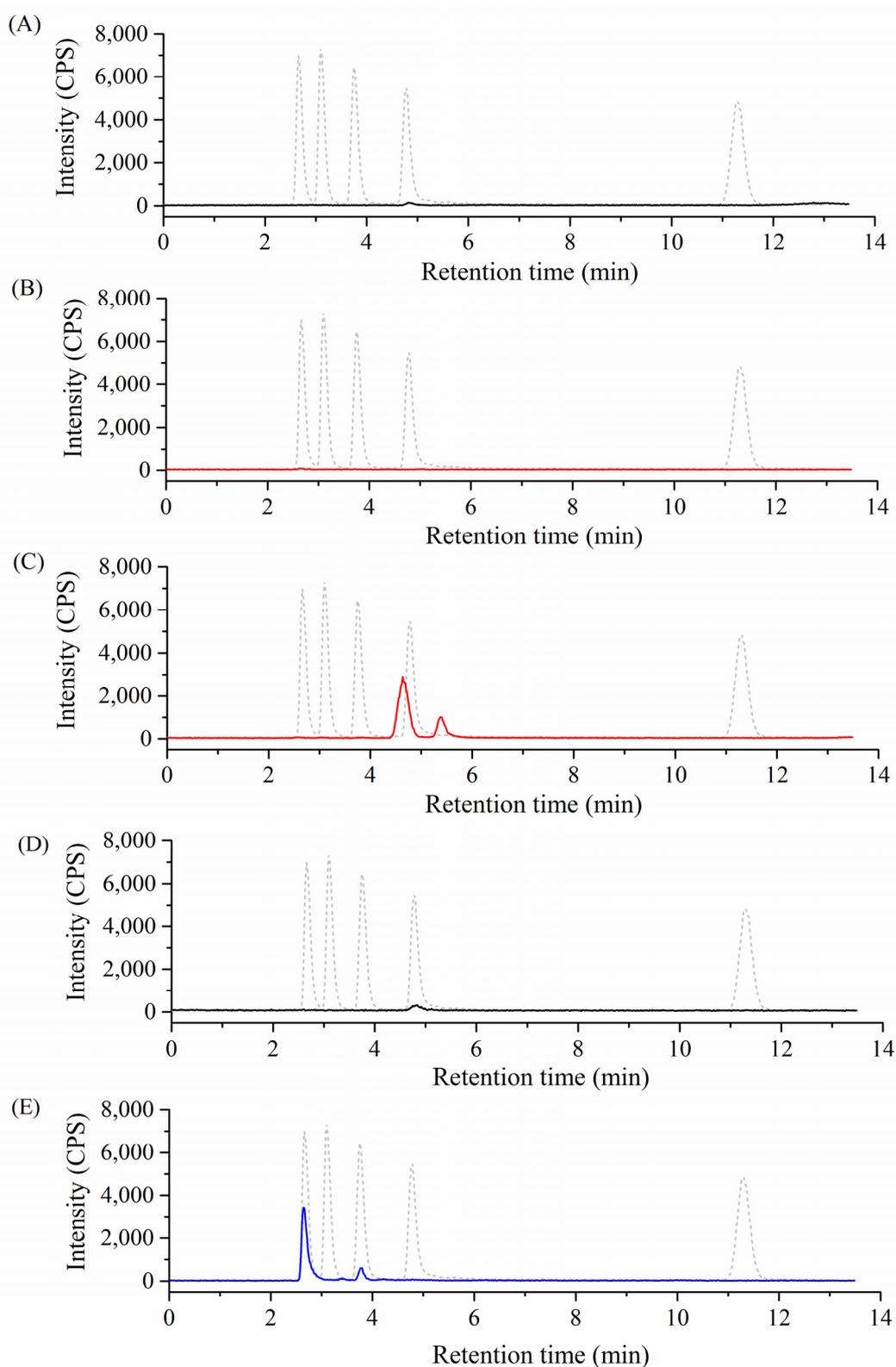
\*Corresponding author:

E-mail: wangmin@caas.cn

**Figure S1** HPLC–ICP–MS chromatograms by Agilent SB-Aq column of egg sample extracted by different extraction procedures: (A) Extraction with ultrapure water; (B) Extraction with HCl (0.1 M); (C) Extraction with NaOH (0.1 M); (D) Extraction with 20 mM ammonium acetate solution containing 5% of methanol (v/v); (E) Hydrolysis with protease XIV. Gray short dash line represents the HPLC–ICP–MS chromatogram of a mixture of Se species at 50  $\mu\text{g L}^{-1}$  (as Se) in solution on Agilent SB-Aq column.

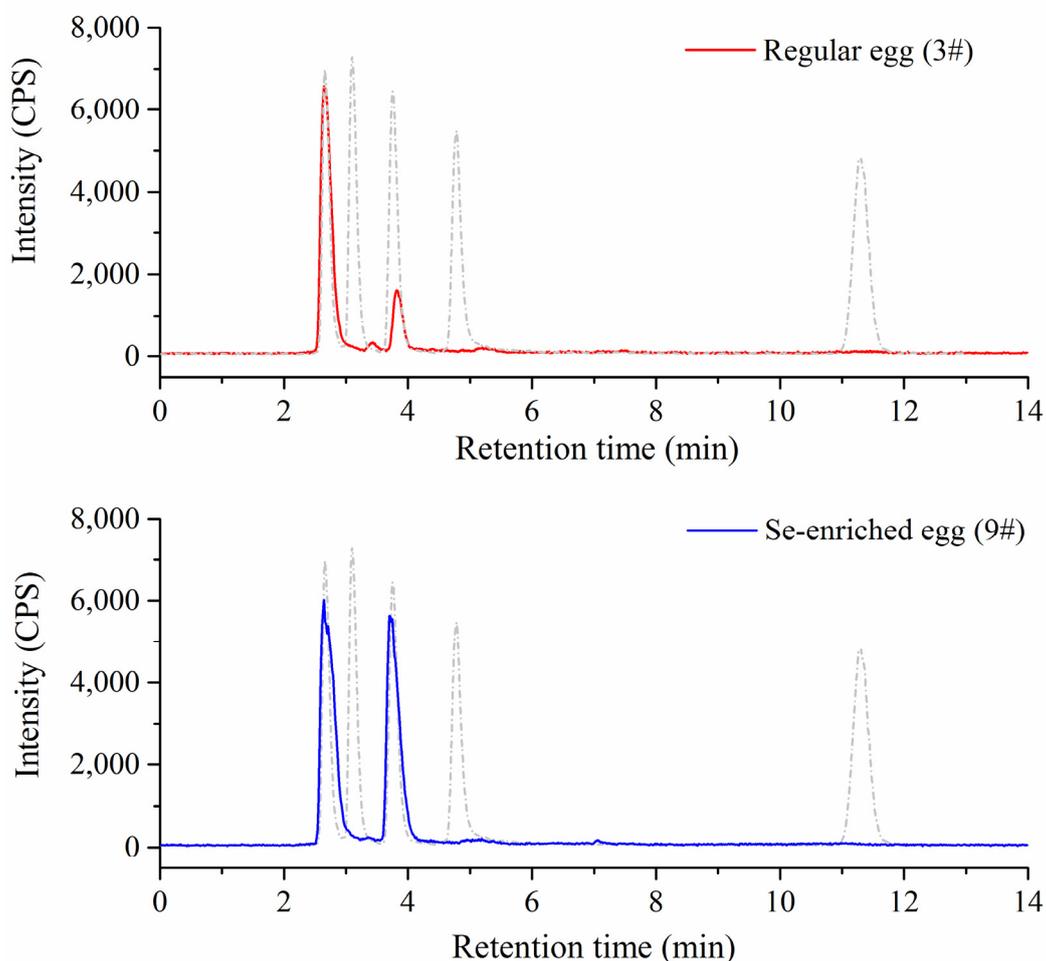
**Figure S2** HPLC–ICP–MS chromatograms by Agilent SB-Aq column of regular egg (red line) and Se-enriched egg (blue line). Gray short dash line represents the HPLC–ICP–MS chromatogram of a mixture of Se species at 50  $\mu\text{g L}^{-1}$  (as Se) in solution on Agilent SB-Aq column.

**Table S1.** Analytical performances of five Se species.



**Figure S1.** HPLC–ICP–MS chromatograms by Agilent SB-Aq column of egg sample extracted by different extraction procedures: (A) Extraction with ultrapure water; (B) Extraction with HCl (0.1 M); (C) Extraction with NaOH (0.1 M); (D) Extraction with 20 mM ammonium acetate solution containing 5% of methanol (v/v); (E) Hydrolysis with protease

XIV. Gray short dash line represents the HPLC–ICP–MS chromatogram of a mixture of Se species at 50 µg L<sup>-1</sup> (as Se) in solution on Agilent SB-Aq column.



**Figure S2.** HPLC–ICP–MS chromatograms by Agilent SB-Aq column of regular egg (red line) and Se-enriched egg (blue line). Gray short dash line represents the HPLC–ICP–MS chromatogram of a mixture of Se species at 50 µg L<sup>-1</sup> (as Se) in solution on Agilent SB-Aq column.

**Table S1.** Analytical performances of five Se species.

Se species	RT (min) <sup>a</sup>	Regression equation <sup>b</sup>	Liner range (µg L <sup>-1</sup> )	Correlation coefficient ( <i>r</i> )	LOD (µg L <sup>-1</sup> )	LOQ (µg L <sup>-1</sup> )
SeCys <sub>2</sub>	2.67 ± 0.01	$y = 1201.2x - 622.1$	2–100	0.9996	0.56	1.86
MeSeCys	3.11 ± 0.01	$y = 1181.4x + 988.66$	2–100	0.9997	0.49	1.62
SeMet	3.77 ± 0.02	$y = 1205.5x + 456.8$	2–100	0.9997	0.55	1.82
Se (IV)	4.83 ± 0.04	$y = 1149.1x - 142.2$	3–100	0.9999	0.71	2.37
Se (VI)	11.46 ± 0.08	$y = 1662.6x + 746.1$	2–100	0.9998	0.59	1.98

<sup>a</sup> Retention time (RT) is expressed as the mean ± SD of 60 replicates.

---

<sup>b</sup> Standard curves were constructed with six points ( $n = 6$ ) from seven replicates;  $x$  and  $y$  in the equations are the concentrations and corresponding peak areas, respectively.