Dual extraction technique combined with HPLC-ICP-MS for the speciation of seleno-amino acids in rice and yeast samples

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**Fig. S1** Effect of extraction time on the adsorption efficiency of seleno-amino acids by MSPE

Conditions: $C_{\text{SeCys2, MeSeCys, SeMet, SeEt}} = 25 \text{ ng mL}^{-1}$; sample volume, 10 mL; sample pH, 7.0

**Fig. S2** Effect of ethylenediamine concentration on the desorption of target seleno-amino acids

Conditions: $C_{\text{SeCys2, MeSeCys, SeMet, SeEt}} = 25 \text{ ng mL}^{-1}$; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; desorption time, 10 min; desorption volume, 1 mL; desorption time, 10 min

**Fig. S3** Effect of desorption time on the desorption of target seleno-amino acids by stirring mode

Conditions: $C_{\text{SeCys2, MeSeCys, SeMet, SeEt}} = 25 \text{ ng mL}^{-1}$; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; eluent, 1 mL 0.1 mol L$^{-1}$ ethylenediamine

**Fig. S4** Chromatography of seleno-amino acids in the 1$^{\text{st}}$ and 2$^{\text{nd}}$ desorption solvent

Conditions: $C_{\text{SeCys2, MeSeCys, SeMet, SeEt}} = 25 \text{ ng mL}^{-1}$; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; desorption time, 5 min; eluent, 1 mL 0.1 mol L$^{-1}$ ethylenediamine

**Fig. S5** Effect of extraction solvent on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys2}} = 2.5 \text{ ng mL}^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}} = 10 \text{ ng mL}^{-1}$; carrier concentration, 20% (v/v); acceptor solution, 0.6 mol L$^{-1}$ NaNO$_3$; stirring rate, 1100 rpm; extraction time, 30 min

**Fig. S6** Effect of sample pH on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys2}} = 2.5 \text{ ng mL}^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}} = 10 \text{ ng mL}^{-1}$; extraction phase: 20% (v/v) [MTOA]$^+ [\text{Cl}]^- $ in toluene; acceptor phase, 0.3 mol L$^{-1}$ NaNO$_3$; stirring rate, 1000 rpm; extraction time, 20 min

**Fig. S7** Effect of stirring speed on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys2}} = 2.5 \text{ ng mL}^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}} = 10 \text{ ng mL}^{-1}$; extraction phase: 15% (v/v) [MTOA]$^+ [\text{Cl}]^- $ in toluene; acceptor phase, 0.6 mol L$^{-1}$ NaNO$_3$; extraction time, 30 min

**Fig. S8** Effect of extraction time on the extraction of target seleno-amino acids

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Conditions: $C_{\text{SeCys}}=2.5 \text{ ng mL}^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}}=10 \text{ ng mL}^{-1}$; extraction phase: 15% (v/v) $[\text{MTOA}]^+\text{[Cl]}^-$ in toluene; acceptor phase, 0.6 mol L$^{-1}$ NaNO$_3$; stirring speed, 1100 rpm
Fig. S1 Effect of extraction time on the adsorption efficiency of seleno-amino acids by MSPE

Conditions: $C_{\text{SeCys}2, \text{MeSeCys}, \text{SeMet}, \text{SeEt}}=25 \text{ ng mL}^{-1}$; sample volume, 10 mL; sample pH, 7.0
**Fig. S2** Effect of ethylenediamine concentration on the desorption of target seleno-amino acids

Conditions: C₅SeCys₂, MeSeCys, SeMet, SeEt=25 ng mL⁻¹; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; desorption time, 10 min; desorption volume, 1 mL; desorption time, 10 min
**Fig. S3** Effect of desorption time on the desorption of target seleno-amino acids by stirring mode

Conditions: C$_2$SeCys, MeSeCys, SeMet, SeEt=25 ng mL$^{-1}$; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; eluent, 1 mL 0.1 mol L$^{-1}$ ethylenediamine
Fig. S4 Chromatography of seleno-amino acids in the 1\textsuperscript{st} and 2\textsuperscript{nd} desorption solvent

Conditions: C\textsubscript{SeCys2}, MeSeCys, SeMet, SeEt=25 ng mL\textsuperscript{-1}; sample volume, 10 mL; sample pH, 7.0; extraction time, 10 min; desorption time, 5 min; eluent, 1 mL 0.1 mol L\textsuperscript{-1} ethylenediamine
Fig. S5 Effect of extraction solvent on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys}}=2.5$ ng mL$^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}}=10$ ng mL$^{-1}$; carrier concentration, 20% (v/v); acceptor solution, 0.6 mol L$^{-1}$ NaNO$_3$; stirring rate, 1100 rpm; extraction time, 30 min
**Fig. S6** Effect of sample pH on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys2}, \text{MeSeCys}, \text{SeMet}, \text{SeEt}} = 10 \text{ ng mL}^{-1}$; extraction phase: 20% (v/v) [MTOA]$^+\cdot$[Cl]$^-$ in toluene; acceptor phase, 0.3 mol L$^{-1}$ NaNO$_3$; stirring rate, 1000 rpm; extraction time, 20 min
Fig. S7 Effect of stirring speed on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys2}} = 2.5$ ng mL$^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}} = 10$ ng mL$^{-1}$; extraction phase: 15% (v/v) [MTOA]$^+$/[Cl$^-$] in toluene; acceptor phase, 0.6 mol L$^{-1}$ NaNO$_3$; extraction time, 30 min
Fig. S8 Effect of extraction time on the extraction of target seleno-amino acids

Conditions: $C_{\text{SeCys}_2} = 2.5$ ng mL$^{-1}$; $C_{\text{MeSeCys, SeMet, SeEt}} = 10$ ng mL$^{-1}$; extraction phase: 15% (v/v) [MTOA]$^+$(Cl)$^-$ in toluene; acceptor phase, 0.6 mol L$^{-1}$ NaNO$_3$; stirring speed, 1100 rpm