

Supplementary Data

This supplementary data is a part of paper entitled "Development of an Analytical Method for Kasugamycin Residue in Herbal Medicine, *Achyranthes japonica* Nakai".

Table S1. Recovery rate (%) according to the difference in extraction solvents

Compound	Recovery rate (%) ¹⁾		
	Methanol	Methanol:water (50:50, v/v)	Water
Kasugamycin	58.4 ± 2.6	90.2 ± 2.1	96.3 ± 2.7

¹⁾ Triplicate recovery average ± RSD

Table S2. Recovery rate (%) by loading volume of SCX SPE cartridge after matrix application

Loading volume	Recovery rate (%)				Total
	Loading	Water 10 mL ¹	Methanol 10 mL ¹	5% NH ₄ OH in methanol 10 mL ²	
1 mL	-	-	-	95.5	95.5
2 mL	-	-	-	101.2	101.2
3 mL	-	-	-	97.4	97.4
4 mL	-	24.5	-	69.5	94.0

¹⁾ Washing

²⁾ Elution

"-" means that the target compound was not eluted from the column and therefore no recovery

Table S3. LC/MS/MS calibration curve of working solution at various concentrations range

	STD #1	STD #2	STD #3	STD #4	STD #5	STD #6
Concentration (µg/mL) at volume 500 µL	0.005	0.008	0.01	0.05	0.08	0.1
Matrix (µL)	500	500	500	500	500	500

Table S4. Recovery rate and MLOQ for chlorantraniliprole in *Achyranthes japonica* Nakai root

Herbal medicine	Fortification (mg/L)	Recovery (%) ^{a)}	C.V. (%) ^{b)}	MLOQ (mg/kg)
<i>Achyranthes japonica</i>	0.04	97.2	8.8	0.04
Nakai	0.4	86.3	2.8	

^{a)} Mean values of triplicate samples with standard deviations

^{b)} CV (coefficient of variation, %) = standard deviation/average × 100

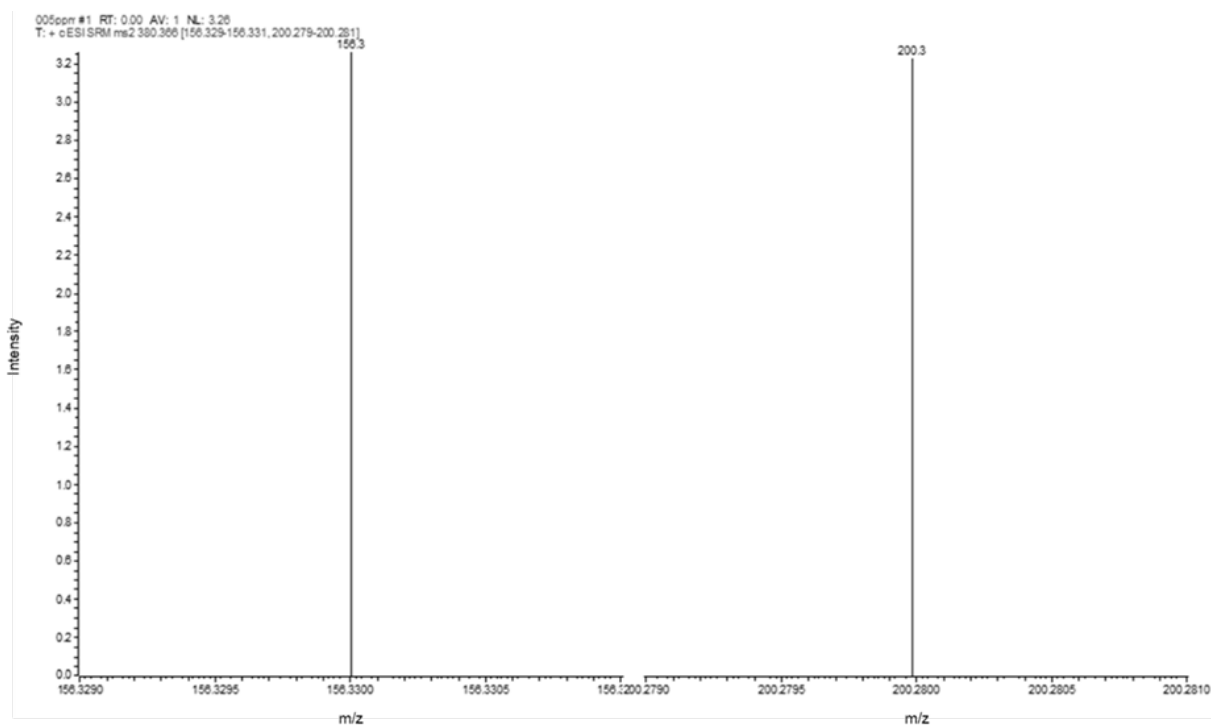


Fig S1. The MS-MS iron scan and fragmentation of kasugamycin

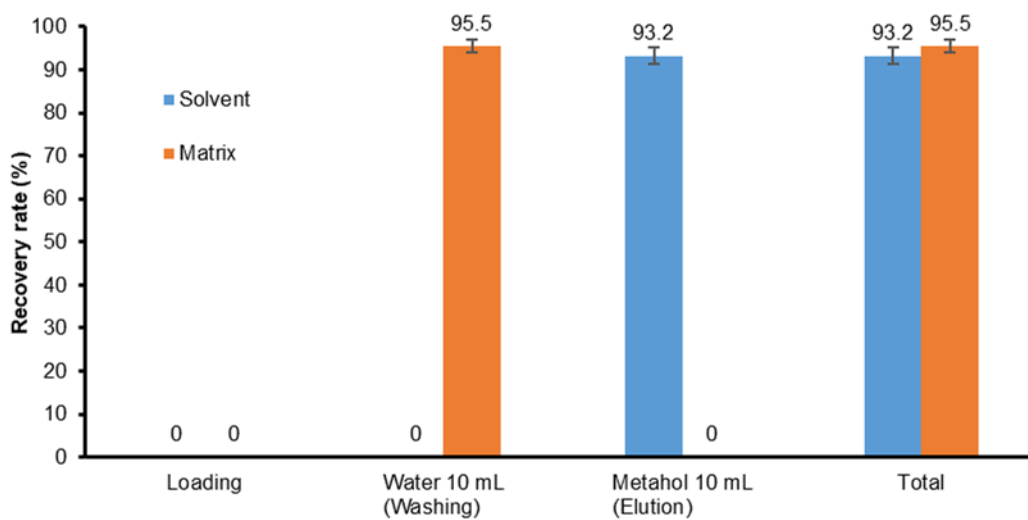


Fig S2. Recovery rate (%) elution purified by silica SPE cartridge

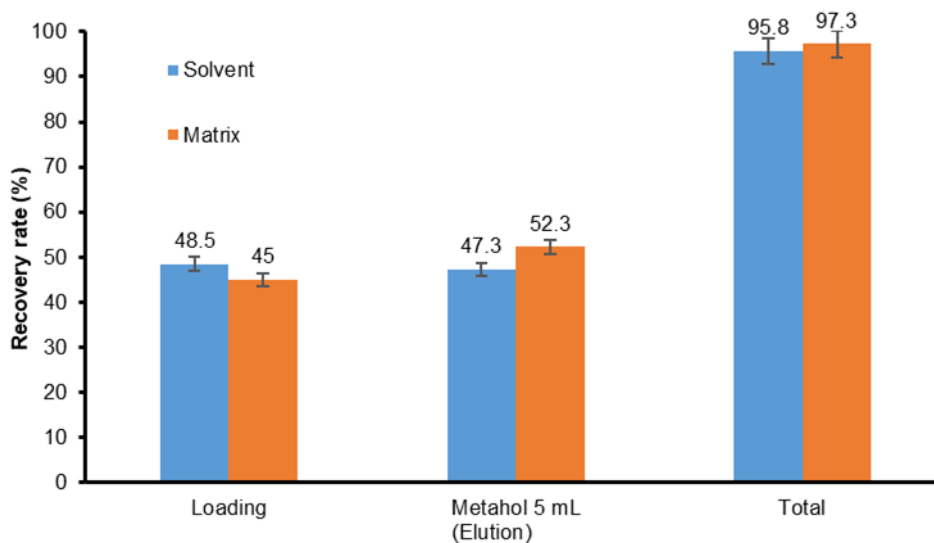


Fig S3. Recovery rate (%) of elution purified by HLB SPE cartridge

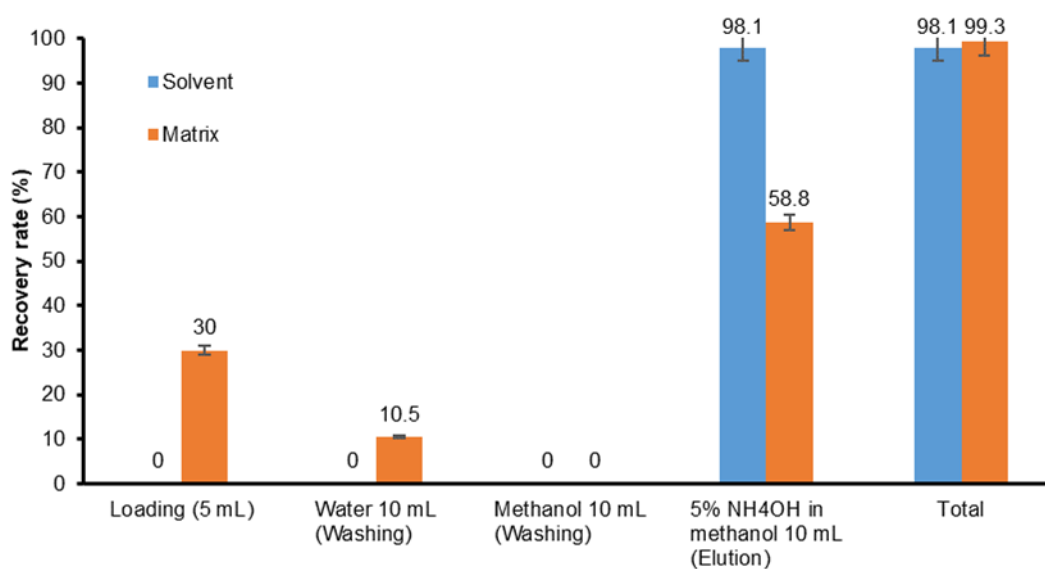


Fig S4. Recovery rate (%) of elution purified by SCX SPE cartridge