



Multi-residue Pesticide Analysis of Botanical Dietary Supplements using SPE Clean-up and GC-Triple Quadrupole MS/MS*

UCT Part Numbers:

ECPSACB256 (500 mg PSA, 250 mg GCB, 6 mL cartridge)

ECMSSC50CT-MP (4000mg MgSO₄, 1000mg NaCl)

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Summary

A screening method for the analysis of 310 pesticides, isomers of organohalogen, organophosphorus, organonitrogen and pyrethroid pesticide metabolites in a variety of dried botanical dietary supplements, spices, medicinal plants, herbals, teas, and phyto-medicines is described. Acetonitrile/water is added to the dried botanical along with anhydrous MgSO₄ and NaCl for extraction. This is followed by clean-up using a tandem SPE cartridge consisting of graphitized carbon black (GCB) and primary-secondary amine sorbent (PSA). Pesticides in the study were spiked at 10, 25, 100 and 500 µg/kg. Mean pesticide recoveries were 97%, 91%, 90% and 90%. Percent RSDs were 15%, 10%, 8%, and 6% respectively.

Some Pesticides Screened by this Method

Azoxystrobin	Chlorpyrifos	DDT
Diazinon	Dimethomorph	Hexachlorobenzene
Hexachlorocyclohexanes	methamidophos	Pentachloroaniline
Pentachloroanisole	Pentachlorobenzene	Pentachlorothioanisole
Quinoxifen	Quintozene	Tecnazene
Tetraconazole	Tetramethrin	

Prepare stock solutions of individual standards by dissolving 25–100 mg of pesticide in 25 mL of toluene.

Procedure

1. Botanical Preparation

- a) Add dry botanical powder (1.00 ± 0.02 g) to the 50 mL centrifuge tube
- b) Add 10 mL water and 10 mL extraction solvent (60 $\mu\text{g/L}$ of the internal standard, tris-(1,3-dichloroisopropyl)phosphate in acetonitrile)
- c) Shake vigorously to insure the botanical is completely wetted
- d) Allowed to stand for 15 minutes
- e) Add the contents of **ECMSSC50CT-MP** pouch to each centrifuge tube
- f) Shake vigorously after addition to disperse the salts
- g) Shake samples vigorously for 1 minute
 - a. Centrifuge at 4500 rpm (4200g) x 5 min

2. Solid-phase Clean-up

- a) Condition **ECPSACB256** cartridge(s) on a manifold using 3 x 6 mL acetone
- b) Do not let cartridge go to dryness after last acetone wash
- c) Insert 15 mL disposable centrifuge tubes in the vacuum manifold
- d) Add a layer of anhydrous sodium sulfate to the top of each cartridge
- e) Add a 1.25 mL aliquot of the extract to the cartridge
- f) Allow to percolate through the cartridge. Apply low vacuum if needed
- g) Rinse cartridge with 1 mL of acetone and continue to collect

3. Cartridge Elution

- a) Elute cartridge with 12 mL of 3:1 acetone:toluene
- b) Reduce extract to approximately 100 μL with a gentle N_2 stream in a water bath at 50-55 $^\circ\text{C}$
- c) Add 0.5 mL toluene, QC standards (50 μL of deuterated polycyclic aromatic hydrocarbons mixture, 500 $\mu\text{g/L}$), and 25 mg of magnesium sulfate
- d) Centrifuge at 3500 rpm x 5 min
- e) Divide the toluene extracts between two GC vials with 250 μL vial inserts keeping one vial as a reserve spare

4. GC-MS/MS Analysis

GC-MS/MS Parameters

(Equivalent equipment may be used)

GC: TRACE Ultra Gas Chromatograph
MS: TSQ Quantum triple quadrupole
Autosampler: TriPlus (Thermo Fisher Scientific)
Column: 30 m x 0.25 188 mm id HP-5MS fused silica capillary column (Agilent Technologies, Santa Clara, CA, USA)
Guard Column: deactivated 5 m x 0.25 mm I.D, Restek Corp., Bellefonte, PA
Oven Temperature: Program, initial 105° C for 3 min, 130° C/ @ 10° C/min, 200° C @ 4° C/min, 290° C @ 8° C/min. Hold 6 min.
Column Flow Rate: 1.4 mL/min He
Injector: PTV 100° C for 0.05 min, ramp 12° C/sec to 280° C
Autosampler: TriPlus Thermo Fisher Scientific
Auto-sampler Temperature: 10 °C
Injection Volume: 2.0 µL splitless mode
Injection Liner: 2 mm id x 120 mm open baffled fused silica deactivated
Ion Source & Transfer T: 250°C and 280°C, respectively
Electron Multiplier V: auto-tune approx. 1400 V
Ar Collision gas: 1.5 mTorr
Cycle Time: 0.5 sec
Q1 entrance mass width (FWHM): 0.7 amu.
Stock pesticide standards: Full scan 50-550 m/z

There is not complete agreement over which transitions for a given pesticide are optimal for foods or dietary supplements. Reference information on SRM transitions for these analytes is provided in references.¹⁻⁴

Representative Recoveries (RSD) and Percent LOQ's in Each Botanical Matrix

Representative Recoveries (mean, n = 4) ± percent relative standard deviation (RSD) for pesticides by botanical, at 10 and 500 µg/kg and the number not detected (ND) at each fortification concentration

Botanical		10 µg/kg	ND	500 µg/kg	ND
Astragalus	<i>Astragalus membranaceus</i>	94 ±13	68	92 ±3	15
Bitter Orange Peel	<i>Citrus aurantium</i>	112 ±15	63	90 ±5	23
Black Cohosh Root	<i>Cimicifuga racemosa</i>	84 ±11	39	82 ±4	14
Chamomile	<i>Matricaria chamomilla</i>	87 ±11	68	91 ±4	29
Cinnamon	<i>Cinnamon verum</i>	63 ±26	149	101 ±7	9
Comfrey Root	<i>Symphytum officinale</i>	89 ±18	69	83 ±10	15
Dong Quai	<i>Angelica sinensis</i>	107 ±19	156	97 ±8	16
Echinacea	<i>Echinacea purpurea</i>	97 ±16	61	101 ±8	11
Fenugreek	<i>Trigonella foenum</i>	99 ±14	82	81 ±7	11
Garlic	<i>Allium sativum</i>	98 ±18	78	87 ±6	15
Ginger	<i>Zingiber</i>	103 ±14	211	104 ±6	59
Ginkgo Biloba	<i>Ginkgo biloba</i>	99 ±16	89	80 ±7	14
Ginseng	<i>Panax quinquefolius</i>	88 ±11	64	86 ±6	8
Green Tea		91 ±13	43	79 ±6	11
Hoodia	<i>Hoodia gordonii</i>	104 ±19	94	93 ±5	20
Hops	<i>Humulus lupulus</i>	111 ±10	233	102 ±6	53
Jasmine	<i>Jasminum odoratissimum</i>	100 ±14	65	84 ±4	10
Kava Kava	<i>Piper methysticum</i>	111 ±10	164	100 ±4	59
Licorice Root	<i>Glycyrrhiza glabra</i>	93 ±14	43	87 ±6	15
Milk Thistle	<i>Silybum marianum</i>	90 ±13	73	77 ±10	17
Psyllium	<i>Plantago psyllium</i>	99 ±11	39	95 ±4	16
Saw Palmetto	<i>Serenoa serrulata</i>	103 ±13	111	98 ±7	13
St. John's Wort	<i>Hypericum perforatum</i>	93 ±10	100	83 ±6	16
Valerian Root	<i>Valeriana wallichii</i>	101 ±19	68	94 ±10	13

* Adapted from, Douglas G. Hayward, Jon W. Wong, Feng Shi, Kai Zhang, Nathaniel S. Lee, Alex L. DiBenedetto, & Mathew J. Hengel. "Multi-residue Pesticide Analysis of Botanical Dietary Supplements using Salt-out Acetonitrile Extraction, Solid-phase extraction clean-up column and Gas Chromatography-Triple Quadrupole Mass Spectrometry" DOI: 0.1021/ac400481w

References

- 1) Pang, G. F.; Fan, C. L.; Liu, Y. M.; Cao, Y. Z.; Zhang, J. J.; Li, X. M.; Li, Z. Y.; Wu, Y. P.; Guo, T. T. J. AOAC Int. 2006, 89, 740–771
- 2) Walorczyk, S.; Gnuosowski, B. J. Chromatogr. A. 2006, 1128(1-2), 236-243
- 3) Okihashi, M.; Takatori, S.; Kitagawa, Y.; Tanaka, Y. J. AOAC Int. 2007, 90(4), 1165-1179
- 4) Frenich, A. G.; González-Rodríguez, M. J.; Arrebola, F. J.; Vidal, J. L. M. Anal. Chem. 2005, 77, 4640-4648