Pesticides in Fatty Matrices Extraction

UCT Part Numbers:
ECPSAC1856 (500 mg endcapped C18, 500 PSA, 6 mL cartridge)
CUMPSC18CT (150 mg $MgSO_4$, 50 mg PSA and 50 mg C18 in a 2 mL centrifuge tube)
ECMAG00D (500 g organic free $MgSO_4$ anhydrous)
ECNACL05K (5 kg NaCl)

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Procedure

1. Sample Preparation

a) Weigh 20.0 ± 0.10 grams (g) of homogenized sample into a 250 mL plastic centrifuge bottle, tared on a balance capable of weighing to 0.01 grams
b) Fortify each sample with process control spiking (PCS) solution
c) Add 50 mL of ethyl acetate (EtOAc) to each tube
d) Fortify each sample with internal standard (ISTD) spiking solution
e) Reduce sample material particle size by using a high speed disperser for approximately 1 minute
f) Add 2 g of anhydrous $MgSO_4$ **(ECMAG00D)** and 0.5 g anhydrous NaCl **(ECNACL05K)**

Note: Carefully add the reagents to the tube to avoid contaminating the threads or rims of the tubes otherwise leaks may result

g) Seal the tube and shake vigorously for approximately 1 minute either mechanically or by hand. Make sure the solvent interacts well with the entire sample and that crystalline agglomerates are broken up
h) Cool the sample in a -20 ºC freezer for approximately 30 minutes
i) Centrifuge at 10,000 RCF for 5 minutes
j) Decant at least 50 mL of the EtOAc layer into a 50 mL glass graduated centrifuge tube using a funnel and filter paper. Allow the extract to come to room temperature and adjust the volume with EtOAc to 50 mL using a Pasteur pipette
k) Concentrate the extract under a stream of nitrogen with a 70º C water bath until the volume remains constant (this will be ~ 3 mL and will take about 1 hour)

l) Dilute to 20 mL with acetonitrile (MeCN) and cap with a glass stopper, vortex for 1 minute

m) Freeze at -70 ºC for 30 minutes

n) Centrifuge the extract while frozen for 3 minutes (The MeCN will thaw during centrifugation)

o) Directly after centrifugation in step n), filter > 15 mL of the MeCN layer of the extract with a 0.45 µm syringe filter into a 15 mL glass centrifuge tube

p) Allow the extract to come to room temp, adjust the volume to 15 mL, and concentrate to 2.25 mL under a stream of nitrogen with a 70 ºC water bath

2. LC-MS/MS Analysis

a) Transfer 1 mL of extract to a 2 mL mini-centrifuge tube **CUMPSC18CT**

b) Vortex for 1 minute and centrifuge

c) Transfer to auto sampler vial. Sample is now ready for analysis

3. GC Analysis

a) For GC analyses, use the dual layer cartridge **ECPSAC1856**

b) Add approximately 0.75 – 0.80 grams (~ 0.6 cm = 0.25 inches) of anhydrous $MgSO_4$ added to the top of the cartridge

c) Condition the SPE cartridge by adding one cartridge volume (4.0 mL) of MeCN using a UCT positive pressure SPE manifold

d) Elute to waste

e) Place a labeled 15 mL graduated disposable plastic centrifuge tube below the cartridge in the positive pressure SPE manifold

f) Quantitatively transfer 1 mL of the sample extract from step 15 to the SPE cartridge

g) Elute SPE cartridge in a dropwise manner (Regulated Flow Pressure = 35 psi) into a labeled 15 mL graduated glass centrifuge tube using MeCN

h) Collect the eluate while washing the SPE cartridge **three times** with **4 mL of eluant**.

i) After the last 4 mL portion of eluant has passed through the cartridge move the switch of the positive pressure SPE manifold from “Regulated Flow” to “Full Flow/Dry” to dry the SPE cartridge for approximately 1 minute

j) Using an N-Evap (or equivalent) with the water bath set at 50°C and N_2 flow set at <10 liters per minute (LPM) (typical setting is 2 – 6 LPM), evaporate the sample to approximately 0.5 mL

k) Add 3 mL of toluene to the centrifuge tube containing the sample

l) Evaporate again to < 0.5 mL. (This is to insure all other solvents have been removed from the sample.)

m) Bring the volume to 1.0 mL with toluene and vortex to mix solvent into sample

n) Analyze by GCMS-EI and GCMS-NCI