EBPI Technical Document 1.3.3

Solvent Exchange Technique for Volatile Compounds in Organic Extracts

1.0 Purpose

This document aims to provide guidance for solvent exchange procedures used when volatile contaminants are extracted in a solvent that is incompatible with a bioassay from EBPI. Usually the organic solvents will need to be exchanged with DMSO or methanol to facilitate running them with the bacterial-based genotoxicity and mutagenicity products. EBPI recommends that clients consult literature procedures as this is only meant as a guide and must be adapted to fit individual laboratory setups.

2.0 Procedure

2.1 Perform extraction procedure in desired solvents by sonication, shaking or soxhlet procedures. Solvent of choice will be determined by the nature of the target contaminants but most commonly, acetone, DCM and chloroform are used.

2.2 Combine the collected extracts into a round bottom flask of appropriate size.

2.3 Evaporate (approximately 90%) of the total solvent volume under vacuum on a rotary evaporator (rotovap) gently (at low heat). Reduce the volume to between 1-5 mL depending on the original volume of extract.

2.4 Remove the flask from the rotovap and further concentrate the sample by evaporation under a gentle stream of nitrogen gas. Reduce the volume to approximately 1.0 mL. Hold the temperature steady between 30 and 40 °C during this step using a water bath.

2.5 Add a 2.0 mL aliquot of the exchange solvent can be directly to the remaining primary extract in the same reaction flask. Ensure the exchange solvent is added in excess.

2.7 Place this mixture under nitrogen gas in a water bath (40 °C) for 30 min to facilitate solvent exchange. Nitrogen should be bubbled through the solution to encourage evaporation of the extraction solvent

2.8 Once the final volume is reduce to approximately 2.0 mL, repeat the evaporation procedure once more by adding another 2.0 mL aliquot of exchange solvent to the vessel.

2.9 Place the flask under nitrogen gas in a water bath (40 °C) for 30 min again to facilitate solvent exchange. Nitrogen should be bubbled through the solution to encourage evaporation of the extraction solvent. Leave the vessel for an additional 15 minutes under nitrogen to complete evaporation

2.10 Combine the final exchange solvent aliquots and bring the final volume to 5.0 mL using the exchange solvent.

2.11 Check amounts of residual extraction solvent in the final solution by HPLC if cytotoxicity is observed in the assay

The final solution can now be used in the Bioassay. Ensure that you have calculated the concentration effect as this may become important when using toxicity equivalency quotients for your result interpretation. If you have any further questions, contact a representative at EBPI
www.biotoxicity.com