Extraction of MI (methylisothiazolinone) from Sun Lotion Using ISOLUTE® SLE+ Prior to GC/MS Analysis

Introduction

Methylisothiazolinone (MI) is an antimicrobial preservative that can potentially be used in a variety of personal care products. However a percentage of the population is at risk from contact dermatitis when exposed to this compound at sufficient concentrations. From July 2015, the European Commission will adopt a ban of MI as an ingredient in leave-on cosmetic products.

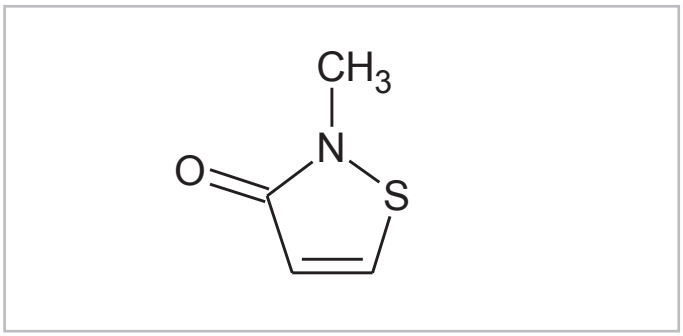

Figure 1. Structure of MI

This application note describes the extraction of methylisothiazolinone from sun lotion, prior to GC/MS analysis. **Figure 1** shows the structure of MI.

ISOLUTE® SLE+ (Supported Liquid Extraction) columns offer an efficient alternative to traditional liquid-liquid extraction (LLE) for bioanalytical sample preparation, providing high analyte recoveries, no emulsion formation, and significantly reduced sample preparation.

This application note describes an effective and efficient ISOLUTE SLE+ protocol optimized for the 1 mL sample capacity column format.

Sample Preparation Procedure

Format:	**ISOLUTE® SLE+ 1 mL Sample Volume Columns, Part Number 820-0140-C**
Sample Pre-treatment:	Measure 100 mg of sun lotion into a 2 mL vial. Add 1.5 mL of 50/50 methanol/sodium chloride (1M (aq)) and vortex mix for at least 10 seconds. Centrifuge at 17,000 G for 10 minutes and decant 1 mL into a glass tube.
Sample Loading:	Load 500 µL of the decanted solution onto the column and apply a pulse of vacuum or positive pressure (3–5 seconds) to initiate flow. Allow the sample to absorb for 5 minutes.
Analyte Extraction:	Apply hexane/ethyl acetate (1:1, v/v, 2 mL) and allow to flow under gravity for 5 minutes. Repeat this procedure two more times for a total of 6 mL of elution solvent. Apply vacuum or positive pressure (5–10 seconds) to pull through any remaining extraction solvent.
Post Elution and Reconstitution:	Before evaporation, add 200 mM HCl in isopropanol (100 µL) to each collection tube. This will stabilize the analyte and minimize losses during evaporation. Dry the extract in a stream of air or nitrogen using a SPE Dry (30 °C, 20 to 40 L/min) or TurboVap® (1.0 bar at 30 °C) for 15–20 minutes. Take care not to over-dry the samples. Upon dryness, reconstitute with 200 µL ethyl acetate.

Biotage®

GC Conditions

Instrument:	Agilent 7890A with QuickSwap
Column:	Agilent J&W DB-5, 30 m x 0.25 mm ID x 0.25 µm
Carrier:	Helium 1.2 mL/min (constant flow)
Inlet:	250 °C, Split, (ratio 40:1)
Injection:	1 µL
Wash Solvents	Hexane, Ethyl acetate
Oven:	Initial temperature 60 °C
	Ramp 10 °C/min to 110 °C
	Ramp 100 °C/min to 325 °C, hold for 2:00
Transfer Line:	280 °C

MS Conditions

Instrument:	Agilent 5975C
Source:	230 °C
Quadrupole:	150 °C
MSD Mode:	SIM

SIM Parameters

Table 1. Ions acquired in the Selected Ion Monitoring (SIM) mode

SIM Group	Analyte	Target (Quant) Ion	1st Qual Ion	2nd Qual Ion
1	MI	115	87	58

Results

Sun lotion (100 mg) was spiked with 20 µg of MI to evaluate extraction recovery and results were compared with samples fortified with one third of this spike, reflecting the one third supernatant load. The percentage of MI recovered after extraction was measured to be 99.6% with an RSD value of 9.5%

Calibration Curves

A calibration curve of MI concentrations was extracted with values of 50, 100, 200, 375, 400 and 750 ng/mg. The 375 ng/mg value is the maximum concentration allowed in rinse-off commercial personal care products in Europe, based on the European Commission limit of 0.0015% amount of MCI:MI in the ratio of 3:1. The curve is demonstrated below in **Figure 2**. The coefficient of determination (r^2) is 0.9953.

Solution Preparation

1. 200 mM HCl in IPA: add 200 µL concentrated hydrochloric acid to 11.8 mL isopropanol (HPLC grade). The concentration of hydrochloric acid stock is commercially available ~12M.

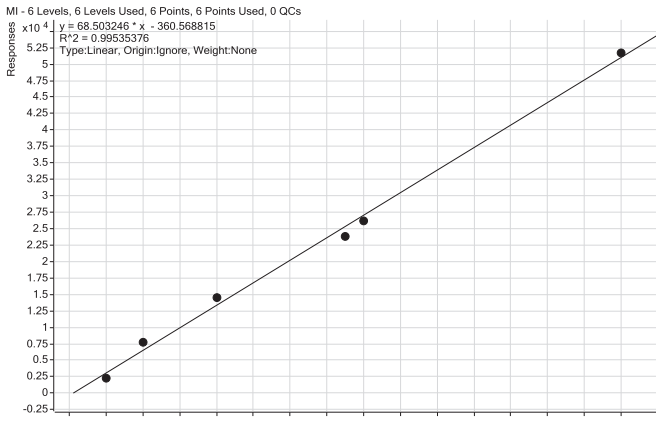

Figure 1. Calibration Curve for MI using ISOLUTE® SLE+ 1 mL columns

Ordering Information

Part Number	Description	Quantity
820-0140-C	ISOLUTE SLE+ 1 mL Supported Liquid Extraction Column	30
PPM-48	Biotage® PRESSURE+ 48 Positive Pressure Manifold for Columns	1
SD-9600-DHS-EU	Biotage® SPE Dry Sample Concentrator System 220/240 V	1
SD-9600-DHS-NA	Biotage® SPE Dry Sample Concentrator System 100/120 V	1
C103198	TurboVap® LV, Evaporator 100/120V	1
C103199	TurboVap® LV, Evaporator 220/240V	1

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