Analysis of Naphthenic Acids in Tissue by Liquid Chromatography
Tandem Mass Spectrometry

1.0 Background and Objectives

Naphthenic acids (NAs) are complex mixtures of naturally occurring carboxylic acids which are common in and around oil sands deposits. NAs concentrate during the extraction of bitumen from oil sands and are among the major toxic constituents of water produced from this process. Of particular concern is the potential for increased exposure of wildlife (in particular aquatic species) to NAs due to ongoing industrial activities and regional development. To support risk assessment of NAs in Northern Alberta, new analytical tools are required which are capable of identifying and quantifying NAs in tissue samples.

2.0 Method Overview

2.1 Target Isomer Groups

2.2 Extraction:

2.3 Cleanup:

2.4 Derivatization:

2.5 Analysis:

3.0 Derivatization method and instrumental analysis

Advantages of Derivatization with EDC:

- Enables sensitive (+) ESI detection.
- Enables analysis in multiple reaction monitoring (MRM) mode.
- Enables monitoring NAs even when the exact structure is unknown.
- Monitoring for m/z 129 enables approximate constant relative response factors for NAs in the same isomer group.

4.0 Fatty acids, Resin acids and potential interferences in NA analysis

4.1 Fatty Acids (FAs)

FAs (a subset of NAs of biochemical origin) have the same chemical and structural formula as NAs and present special challenges in the analysis of NAs from tissue.

Figures 2 (A/B, C/D, E/F, G/H, pairs) shows separation of ubiquitous saturated and unsaturated fatty acids (z=0 and z=2) from isomers of the same 'n' and 'z' numbers by the current method.

4.2 Resin Acids (RAs)

RAs (such as pimarcic acid, sandaracopimaric acid, abietic acid, neoabietic acid, and levopimaric acid) have chemical formulas CzHyOz and RAs such as dihydroxyopimaric acid have chemical formula CzHyO2. These compounds are cyclic unsaturated carboxylic acids and have similar chemical formula and structure as NAs with n=20 and z=10 and n=20 and z=8 isomer groups respectively. These compounds interfere with this analysis of NAs in tissue, see Figures 2 I/J/K pairs.

5.0 Significance of Other Potential interferences

Potential isobaric interferences consisting of dicarboxylic acids, heteroatomic carboxylic acids and hydroxyl acids were investigated for interferences in NA analysis. For each class of compounds straight chain and cyclic isobaric interferences were selected. The straight chain hydroxy acid was observed to interfere.

6.0 Method accuracy and precision

Accuracy:

Overall, percent recovery values of 50 to 150 were observed for the selected marker NA isomer groups. Exceptions, n=15, Z=12; see Figure 3.

Precision:

Percent relative standard deviation values of less than 20% were observed. Exceptions, n=15, Z=12, n=16, Z=12, n=17, Z=12, see Figure 3.

7.0 Summary

- A sensitive method was developed for extraction, cleanup and quantification of major NA isomer groups from tissue samples.
- Some mitigation of interferences arising from saturated and unsaturated Fatty acids in analysis of NAs from tissue is possible.
- The chromatographic method was optimized to enable separation of ubiquitous fatty acids (Z=0 and -2) from NAs, while separating the NAs by carbon number and extent of cyclization.
- Potential marker NA isomer groups were identified for measuring exposure of aquatic organisms to NAs.
- Application of the method to a wider variety of aquatic organisms is necessary to properly demonstrate applicability of scope.

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References