Subject: InformConnect Open Forum Digest for Wednesday July 17, 2019
From: "American Oil Chemists' Society" <DoNotReply@ConnectedCommunity.org>
Date: Wed, 17 Jul 2019, 5:10 AM
To: joseph.diverdi@colostate.edu

Ar

Fr

InformConnect Open Forum

Post New Message

Jul 16, 2019

Discussions

started 2 days ago, Arun Sharma (1 reply)

regarding Mono , di and triglyceride analysis ex

1. Hello Arun, I don't know exactly what you are... Keith Meyer

started 6 days ago, Elizabeth Schwartz (5 replies)

Organic Compliant Bleaching Clays ex

2. Dear Manival I hear of a lot of problems with... Alan Paine

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1. Re: regarding Mono , di and triglyceride analysis

Reply to Group Reply to Sender

Keith Meyer Jul 16, 2019 3:55 PM | view attached

Keith Meyer

Hello Arun, Fr

I don't know exactly what you are trying to learn, what kind of lipid sample you want to analyze (neat oil? sample extract? origin?), or what analytical resources you have access to.

But the short answer to your question is "No", and here is why.

Let's assume that you are trying to quantify the absolute or relative amounts of monacylglyecrol, diacylglycerol, and triacylglycerol species in a lipid sample. There are several levels of detail that one can achieve in this type of analysis.

I think that a PRO and CON look at HPLC vs. GC might be most helpful to you at this stage.

Temperature programmed gas chromatographic analysis of volatile, high molecular weight lipid species at high elution temperatures using non-polar capillary GC columns coated with thermostable stationary phases. Mono- and diacylglycerol species are derivatized to render them more volatile

PROs

- Overall, much less complex than a corresponding HPLC analysis
- Quantitation using a flame ionization detector that has a very linear response and is easily calibrated
- A great approach assuming that one wishes to know how many acylglycerol species there are and not exactly which acylglycerol species are present
- Useful for animal, vegetable, and cellular lipids that contain fatty acids with about eight to twenty-four carbons
- 1,2- and 1,3-diglycerides are typically resolved as are 1- and 2-monoglycerides

CONs

- You will only learn the carbon number distribution within each acylglycerol category. For example, all the triacylglcerols with fatty acid carbon number 52, such as PSS, SPS, POO, POS, etc. will tend to elute as a single peak
- The gas chromatograph (and analysts!) need to be optimized for analysis of species with high boiling points and elution temperatures that can range from 50 to 400°C. Hints for success: Use a cold on-column injection (easy on Agilent machines from 5890 forward) and use the more stable tert-butyldimethylsilyl derivatives rather than the often mentioned trimethylsilyl ones.

Gradient normal phase HPLC analysis on polar columns such as those with silica, diol, aminopropyl, or polyvinyl alcohol functionality. Or, non-aqueous gradient reversed phase HPLC analysis on C18 phases.

PROs

- No chemical derivatization is necessary.

- There is an opportunity to resolve molecular species that have the same fatty acid carbon numbers, such as PSS, SPS, POO, and POS that have differing geometries and degrees of unsaturation
- The HPLC methods tend to be more tunable (solvents, column choices, temperature) than the GC methods so that separations can be extensively optimized according to your needs
- Non-volatile species such as phospholipids can be included in the analysis

CONs

- The solvent gradients can be very complex, especially in the case of normal phase HPLC. Also, unless one is working with very narrow bore HPLC columns, solvent usage and waste solvent disposal costs can be expensive
- There are no simple HPLC detectors with "universal" response, such as the flame ionization commonly used in GC. Refractive index detectors won't work due to the gradient elutions required; UV absorbance detectors need to be operated at short wavelengths (<250 nm) where nearly everything absorbs UV radiation. Evaporative detectors such as light scattering and charged aerosol work best in this application but generally respond non-linearly due to both their operating characteristics and response to molecules of differing sizes and volatilities; depending on the complexity of the sample; calibration can be rather nightmarish. Mass spectrometry has good universal response but, depending on the ionization mechanism, may also require careful attention to calibration if you want absolute quantitation.

I have attached a representative method that illustrates what can be achieved with the GC analysis and includes sample chromatograms. And here is a link to another viewpoint. lipidlibrary.aocs.org/lipid-analysis/...

HPLC analysis of lipid classes is well documented in this article: www.researchgate.net/profile/Christopher_Beermann/...

The long answer to your question is "Yes"; with a great deal of care, the GC and HPLC approaches can be made to yield equivalent results. Depending on your needs, my vote goes to the GC method if all you are interested in is the total amount of mono-, di-, and triacylglycerols in a vegetable oil or vegetable oil based product. Bonus: one can also measure the unesterified fatty acids, and, possibly, any methyl esters

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Keith Meyer, Ph.D.
Kraft Foods Fellow in Analytical Sciences, Retired
La Grange Park, IL USA
pinetreeln@gmail.com