LABORATORY PROCEDURES

Derivatization of Glycerol, Monoglycerides and Diglycerides for Determination by Infrared Spectrometry

Objective

Different reagents will be used to convert glycerol (Gly), monoglycerides (MG) and diglycerides (DG) into derivatives that are suitable for analytical determinations as the reaction byproducts in biodiesel production. Different functional groups such as trimethylchboro and trifluoroacetyl groups can be introduced into the Gly/MG/DG derivatives for FTIR analysis. The functional groups imparted by the derivatizing reagents allow MG and DG components to be distinguished from the main components of fatty acid methyl esters or unreacted triglycerides in the biodiesel samples.

Procedures

1. Dilution: Carefully weigh out Gly, MG, or DG (20-50mg) and dilute it with 5 mL methylene chloride. Keep the MG or DG container with a few particles left for FTIR analysis later. Make sure that all the MG/DG crystallites are fully dissolved with stirring by magnetic micro-bar and share the 5-mL standards with others. Calculate the exact MG or DG concentration based on the actual mass of MG or DG used.

2. Derivatization: At least 3 times the molar excess of reagents (i.e. TMCS, DMDCS, or TFAA) is needed to quantitatively or completely convert the Gly, MG, or DG analytes. Note that the molar ratios of the reagents to analytes for stoichiometric reaction are 3:1, 2:1, and 1:1 for Gly, DG, and MG, respectively. Mix the reagent (TMCS/TMBS/DMDCS/TFAA) and the Gly/MG/DG standards in a 3:1 mole ratio to a total volume of 1.5 mL in a 2-mL screw-cap vial followed by stirring with a magnetic micro-bar. Transfer 20 µL, 50 µL, 100 µL, 250 µL, and 1000 µL of the mixture into five separate 2-mL vials followed by the addition of 1.00 mL of methyl palmitate into each vial. Warm and stir the mixtures in order to allow methylene chloride solvent to evaporate away leaving behind the MG/DG derivative in methyl palmitate. Carry out the same derivatization reaction for a biodiesel sample assuming that it has a <3 % (w/w) each of Gly/MG/DG.

3. Analysis: Analyze the reagent (i.e. TMCS, DMDCS, or TFAA), the underivatized analyte (Gly/MG/DG), methyl palmitate, and the five mixtures containing different levels of MG/DG on the Varian 7000 FTIR after turning on the cooling water, power supply, and filling the MCT detector compartment with liquid nitrogen. A Golden Gate attenuated total reflectance accessory with a diamond crystal is used for sample analysis at a optical resolution of 4 cm⁻¹ and 50 scans.

TMCS - Trimethylchlorosilane
TMBS - Trimethylbromosilane
DMDCS - Dimethyldichlorosilane
TFAA - Trifluoroacetic anhydride