

Procedure for the Extraction of Lipids from Tree Bark

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July 2002

Purpose

This method is applied to the extraction of total lipids from tree bark. The lipid content in the bark of trees is useful in determining a tree's content of lipophilic compounds.

Supplies

Aluminum Weighing Dishes: 150 ml
Beakers: 50 - 100 ml
Suction flask
Ceramic Funnel
Filter Paper - Whatman's #5 Qualitative
Glass Pipets

Reagents

1:1 (v:v) Hexane to Acetone, AR grade or higher
Dichloromethane (methylene chloride), AR grade or higher

Equipment

Drying Oven
Muffle Oven
Sonicator
Scale - weighing in grams and accurate to 4 decimal places.

Procedure

Muffle all glassware at 450°C

Rinse aluminum weighing dishes with dichloromethane. Add an extra dish for every ten samples as a laboratory blank sample.

Grind each sample in a Wiley Mill equipped with a #10 screen and return it to the sample jar. Grind a duplicate sample for every ten samples for quality control measures.

Number the aluminum weighing dishes and record in the logbook. Weigh and record the weight of each weighing dish.

Weigh out approximately 10 grams of sample into each weighing dish and record the weights, the sample identity with the corresponding weighing dish number.

Transfer the sample into a labeled beaker and cover well with 1:1 hexane : acetone (v:v). Rinse the weighing dish and add that rinse to the beaker.

Sonicate the beaker and contents for one hour.

Filter the sample using a suction flask and a funnel with #5 Qualitative Whatman filter paper. Rinse the beaker three times with a few milliliters of the solvent and then pour over the sample while it is in the funnel. Also, using a glass pipet, add a few more milliliters of solvent as a rinse over the sample. Transfer the eluent to the corresponding weighing dish and allow the solvent to evaporate in the hood.

After each sample, clean the filtration glassware by rinsing once with methylene chloride and then twice with the 1:1 hexane to acetone using a glass pipet.

Put the weighing dishes in a drying oven at 70° F for 2 hours. Cool them to room temperature and then weigh and record the weights.

Calculations

% lipid by mass:

$$\% \text{ lipid} = 100(M_1/M_b)$$

where: M_1 = net mass of lipid after drying

M_b = net mass of bark from which lipid was extracted

RPD for duplicates:

$$\text{RPD} = 100(L_1 - L_2) * A^{-1}$$

where: L_1 and L_2 = the lipid masses of duplicates 1 and 2

A = the average value of L_1 and L_2 .

Quality Control

Laboratory blanks should have concentrations that are consistent with the precision of the balance used in the laboratory.

Sample duplicates should be assessed for accuracy by having RPD values that are ≤ 20 .

Record and submit these results.

Corrective Actions

When quality control measures are out of defined QC goals, identify and define the problem, determine and implement a corrective action, and verify that the problem has been eliminated. Report the findings throughout the process.

References

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Hartman, L. and Weenink, R. O. A Note on the Fatty Acid Composition of Lipids from the Bark of *Pinus Radiata*. *New Zealand Journal of Science*, 10: 636-638, 1967.