

Lipid reaction experiments



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Lipids are necessary to diverse forms of organisms and they possess specific functions for every specific kind of lipids. One common contribution of lipids is that it is the secondary source of energy. In this experiment, different reactions of lipids were observed to identify some of the properties of lipids through the use of different tests such as Iodine, Acrolein, Saponification and Liebermann-Burchard test. These tests are completed to perceive what kind of lipid is applicable to be a substitute to some important components of certain material that are widely used throughout the world.

INTRODUCTION

Detection of unsaturation in lipids made possible by adding Iodine solution which yields the iodization of the double bond. On the other hand, glycerol component of acylglycerides is determined by addition of potassium bisulphate which induces the formation of acrolein. Aroma of acrolein is irritating. Saponification which refers to the reaction of acylglycerides with bases which economically essential in making soap because of its lather-bubble properties. Cholesterol forms deeply colored condensation product with acetic anhydride under acidic conditions in Liebermann-Burchard test.

MATERIALS AND METHODS

There are four tests which required reagents:

Iodine test

A pinch of butter was dissolved in 1.00 ml of CHCl_3 in a test tube. Five drops of egg yolk, coconut oil, and cod liver oil were dissolved in 1.00 ml CHCl_3 in separated tubes. 1.00 ml each of lecithin and cholesterol were placed in separate test tubes. To each sample, Iodine solution was added drop by drop

while shaking in between addition until no further discoloration was observed. Number of drops was recorded.

Acrolein Test

Lipid samples were prepared like in procedure number 1. A pinch of KHSO_4 was added to each sample 250-ml beaker used as water bath to heat the samples gently at first then vigorously. The tubes were removed from water and vapour wafted. Smell is recorded produced in each sample.

Saponification

The lipid samples were prepared same as in first procedure, 2. 00ml of ethanolic KOH. Water heated for 5 minutes. Tubes cooled and soap precipitate was observed.

Liebermann-Burchard Test

For this test, almost the same procedure as the saponification and 5 drop of acetic anhydride to each samples the 2 drops of concentrated H_2SO_4 . The color changed as observed.

RESULTS AND DISCUSSION

Four tests in this experiment were applied in order to observe whether there is a reaction or not in lipids. These are the Iodine Test, Acrolein Test, Saponification, and the Liebermann-Burchard Test. The following reagents/materials that were used to identify the appearance of the lipids are the butter, egg yolk, coconut oil, olive oil, cod liver oil Lecithin and Cholesterol.

The lipids are a large and a diverse group of naturally occurring organic compounds that are related by their solubility in non-polar organic solvents

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and general insolubility in water. On the table below, the other chemicals have these different reactions of lipids.

TEST FOR UNSATURATION:

The Iodine Test or the Unsaturation of Lipids is used to experiment the presence of starch. It is also dissolved in an aqueous solution of potassium iodide that reacts with starch resulting into a purple-black color. The colour can be detected visually with concentrations of iodine as low as 0.00002M at 20°C. Despite the intensity of the colour decreases with increasing temperature and with the presence of water-miscible, organic solvents such as ethanol. Also, the test cannot be done at a very low pH due to the hydrolysis of the starch under these conditions

This test recognizes the level of saturation and the number of bonds of an oil, fat, or lipid it has. The more unsaturated, multi-bonded the lipid is, the more it absorbs the iodine. The less iodine it absorbs, the lipid is taken for to be saturated, single bonded.

Acrolein Test is the simplest unsaturated aldehyde. It is a relatively electrophilic compound and a reactive one, hence its high toxicity. It is a good Michael acceptor and it is a useful reaction with thiols. It forms acetals readily, a prominent one being the spirocycle derived from pentaerythritol, diallylidene pentaerythritol. Acrolein participates in many Diels-Alder reactions, even with itself. Via Diels-Alder reactions, it is a precursor to some commercial fragrances, including lylal, norbornene-2-carboxaldehyde, and myrac aldehyde. Below is the table for the Acrolein Test.

ACROLEIN TEST:

- SAMPLES
- CHARACTERISTIC SMELL

Butter

Bad (tolerable); Burnt butter

Egg yolk

Disinfectant (alcohol-like); Burnt grease

Coconut Oil

Bad (tolerable); Burnt grease

Olive Oil

Bad (tolerable); Burnt grease

Cod Liver Oil

Obnoxious (awful); Fishy smell

Lecithin

Foul (semi-tolerable); Burnt grease

Cholesterol

Foul (semi-tolerable); Minty-like smell

The principle behind this test is a specific chemical reaction. This reaction is utilized to determine the presence of glycerin in a fat. By heating the fat

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sample in the presence of potassium bisulfate (KHSO_4), which acts as a dehydrating agent, acrolein ($\text{C}_3\text{H}_4\text{O}$, or $\text{CH}_2=\text{CH}-\text{CHO}$) is formed and can easily be detected by its odor. Whenever fat is heated in the presence of a dehydrating agent, the fat molecule will shed its glycerol in the form of the unsaturated aldehyde-acrolein.

Saponification of oils means the hydrolysis of triglycerides in the oil in the presence of an alkaline medium into glycerol and fatty acids with the production of sodium salt of fatty acids or 'soap'. Saponification is also the alkaline hydrolysis of the fatty acid esters.

Liebermann-Burchard Test is a test for unsaturated steroids and triterpenes was based on the formation of series of colors with acetic anhydride in the presence of concentrated sulfuric acid.

SAPONIFICATION & LIEBERMANN-BURCHARD TEST:

The number of milligrams of alkali required to neutralize the fatty acids containing one gram of fat is used as an indicator of fatty acid chain length in trycylglycerols and triacylglycerol containing long fatty acids having lower saponification number than those with shorter fatty acids is called as Saponification Number.

The Liebermann-Burchard or Acetic anhydride Test is used for the detection of cholesterol. The formation of a green or green-blue colour after a few minutes is positive. This colour begins as a purplish, pink colour and progresses through to a light green then very dark green colour. The colour is due to the hydroxyl group ($-\text{OH}$) of cholesterol reacting with the reagents

and increasing the conjugation of the un-saturation in the adjacent fused ring.

The following pictures that are shown below are the appearance of the lipids in each sample. The first picture shows the appearance before heating it into the water bath and the second picture shows the final result of the reacted sample after heating it into the water bath.