164. On the Lecithins of Egg Yolk.

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The author has communicated already to this "Proceedings" about a method of isolating each simple α- or β-lecithins. The process is applicable to both the lecithins of animal as well as of vegetable origin without making any difference, the fact being verified in the cases of soy bean1) and human brain lecithins.2) In the previous studies, however, analyses were made with amorphous specimens, though the author found later3) that lecithins crystallize some times in the free state and at other times as complexes of the salts of heavy metals. The present study has been undertaken to identify each simple lecithin in a crystalline state, leaving no room for suspecting its purity.

The outlines of the preparation of lecithin mixture are shown in the following diagram:

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Egg yolk (6.5 kg)

Acetone

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Light yellow powder (2.5 kg)

↓

Hot alcohol

Afterward ether

↓

Solution

(Neutral fats, cholesterol,
pigments etc.)

↓

Residue

(Proteins, carbo-
hydrates etc.)

↓

Solution

The solvents were evaporated
A yellowish red semisolid matter remained (600 g)
Treated with cold alcohol

↓

Residue

(Kephalins etc.)

↓

Solution

A saturated alcoholic solution of CdCl₂
was added

↓

Precipitates (786 g)

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Solution

(Cholesterol)

The precipitates were dried
Extracted with ether repeated by

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Residue (747 g)

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Solution

(Kephalin-CdCl₂)
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1) This Proc. 6 (1930), 341; 7 (1931), 12, 226.
2) " 8 (1932), 183, 358.
3) " 8 (1932), 490.
The analyses of the lecithin mixture: Total N: 1.87% (1.75 calc. as dioleo-lecithin), P: 3.99% (3.86 calc. as above), N: P = 1:1.04, Amino N: 0.01% (that is practically zero). Ba-glycerophosphates (P: 10.10; Ba: 44.73% (calc. 10.10, 44.69)) have been recovered from the hydrolysates with a yield of 92%, ca. 73% of which consisted of β-form, the remaining portion having been α-form. Only choline (chloroplatinate, Pt: 31.50% (calc. 31.68), decomposed at 232°C) and no other bases have been found.

The separation of each of the simple lecithins was carried out by the following scheme:

1. The mixture of α-lecithins. From the hydrolysate of the mixture, only α-glycerophosphoric acid has been recovered, the barium salt solution giving no turbidity with Ba(NO₃)₂. As fatty acid components, oleic acid (its dibromocompound contained Br: 36.04% (calc. 36.15)), clupanodonic acid (identified as decabrombehenic acid; Br: 70.79% (calc. 70.76)), and iso-palmitic acid (m.p: 57-57.5°C; C: 74.99, H: 12.44% (74.96, 12.59% calc. as C₁₆H₃₂O₂)) were identified. The proportions of the 3 acids were roughly 72:2:26.

2. The mixture of β-lecithins. By hydrolysing the mixture with...
baryta, $\beta$-glycerophosphoric acid which formed insoluble complex Ba-salt with Ba(NO$_3$)$_2$ to the extent of 99% of calc. amount and two fatty acids, namely, oleic (Br: 36.05% as dibromstearic acid) and isopalmitic acid (C: 74.97, H: 12.61%) were identified.

Isopalmitic acid is an acid found and named by Kawase$^1$ in the oil of silk worm pupa. It has the same composition with palmitic acid but with a lower melting point. Later Kato$^2$ found the same substance in the oil of giant silk worm pupa (Antheraea Pernyi Guirin), K. Suzuki$^3$ and Sueyoshi and Furukubo$^4$ in the egg yolk oil. The present author, also, has separated an acid which is considered to be identical with the so-called isopalmitic acid from mixed lecithins as well as from simple lecithins. Suspecting the purity of the substance, he strived to fractionate, but each fraction had the same melting point. Our laboratory studies are, at present, being made to ascertain whether or not there exists any independent substance as isopalmitic acid. For the time being, however, the author adopts the name following the senior authors referred to above.

(3) Dioleo-$\alpha$-lecithin tetrabromide. This fraction was dissolved in alcohol and HgCl$_2$-complex was precipitated by adding an alcoholic solution of HgCl$_2$. The complex was recrystallized from hot alcohol (Plate 1). Hg: 31.21% (calc. 31.01), N: 0.70% (calc. 0.72) and P: 1.58% (calc. 1.60). Therefore the composition: bromlecithin-3HgCl$_2$. The crystals as well as free lecithin bromide were hydrolysed with HCl and only dibromstearic acid (Br: 36.03 and 35.94% respectively), and no other acid, has been recovered.

(4) Diisopalmito-$\alpha$-lecithin. Recrystallized from acetic acid (Plate 2). By hydrolysing the lecithin, isopalmitic acid and only this acid (m.p: 57–57.5°C, C: 75.05, H: 12.77%) has been found. The contents on P and N were 1.94% (calc. 1.86) and 4.03% (calc. 4.12) respectively.

(5) Oleo-clupanodono-$\alpha$-lecithin dodecabromide. A colourless, twig like crystal (Plate 3). Soluble in hot toluene and xylene but insoluble in alcohol, ether, pet. ether, acetone and benzene. m.p: 180°C (decomposing). Br: 52.80% (calc. 52.96), P: 1.69% (calc. 1.71), N: 0.84% (calc. 0.77). On hydrolysing the lecithin, dibromstearic acid (Br: 35.80%) and decabrombehenic acid (Br: 70.65% (calc. 70.76)) were obtained. An equimolecular mixture of di-(dibromstearo)- and di-(decabrombeheno)-$\alpha$-lecithin should show the same analytical results. However, di-(dibromstearo)-$\alpha$-lecithin was identified already in (3) as a semisolid matter soluble in CHCl$_3$ but the lecithin under discussion had shown no sign of such solubility.

3) 3 (1927), 530.
(6) Oleo-isopalmito-β-lecithin dibromide. A semisolid substance at room temperature, soluble in most of the ordinary organic solvents excluding acetone. Hg-complex was formed and recrystallized from alcohol (Plate 4). Hg : 34.00% (calc. 34.31), N : 0.87% (calc. 0.80), P : 1.79% (calc. 1.77). These figures well conform with the formula, the bromlecithin-3HgCl₂. The complex was hydrolysed with HCl and dibromstearic acid (Br : 36.12%) and isopalmitic acid (m.p : 57–57.5°C, C : 74.68, H : 12.78%) have been identified in the molar proportion of 1 : 1.03. The free lecithin, also, gave the same acids. This lecithin could not be an equimolar mixture of di-(isopalmito)- and di-(dibromstearo)-β-lecithin as the latter substance is hardly soluble while the lecithin in question is easily soluble in pet. ether.

(7) Dioleo-β-lecithin tetrabromide. Soluble in alcohol, ether, chloroform and acetone but hardly soluble in pet. ether. At about 2°C, it solidifies in feather-like crystals (Plate 5). Br : 28.29% (calc. 28.46), N : 1.32% (calc. 1.25), P : 2.79% (calc. 2.77). On hydrolysing the lecithin, only dibromstearic acid (Br : 35.81%) was identified as the fatty acid component.

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