

tial for coagulation of the foots. These facts were indicated in laboratory refining tests and most emphatically borne out in factory kettle refinings.

For purposes of laboratory refining, the official method for cottonseed oil designated slow-breaking fulfills both of the requirements specified above. (5) Further than this, our experience indicates that by this method good results can be obtained in the refining of kapok oil, if we are willing to use as a basis for comparison normal cottonseed oil with an equivalent free fatty acid content. We recognize that this criterion is not necessarily correct, inasmuch as even lower losses might be obtained by some other method of refining; however, until the development of more data and information point the way to a superior method, the procedure suggested heretofore does offer a uniform and equitable basis for comparison. No difficulty was encountered, when using the slow-breaking method, in obtaining good separation or settling of the foots. Some indication of the order of the results obtained may be had by referring to Table V, in which are tabulated a few results obtained by using the official refining test as designated for slow-breaking cottonseed oil.

The average loss to acid ratio for normal cottonseed oil with an equivalent free fatty acid content and without an unusual amount of extraneous matter or meal is about the same as the value found for kapok oil, that is, 3.7. This was actually calculated for cottonseed oil from averages of yearly results at a west coast refinery. It must be remembered that these values for

**TABLE V**

%	—NaOH—		Best Loss	Color (Refined Oil)		Bleach (Refined Oil)	
	%	° Be		Y	R	Y	R
3.9	13.6	14	12.8	35	4.2	20	1.7
4.1	12.0	16	14.2	35	5.0	20	2.6
5.3	12.3	18	16.9	35	5.2	20	2.4
4.5	12.7	16	12.4	35	4.8	20	2.2
4.2	12.2	16	12.3	35	5.0	20	2.3
4.2	12.2	16	12.5	35	5.0	20	2.3
2.6	10.5	12	6.7	35	4.0	20	1.5
2.6	10.5	12	8.9	35	4.6	20	1.8
3.1	12.0	14	10.6	35	4.6	20	1.9
3.8	13.4	14	16.8	35	4.8	20	2.4
3.1	7.0	14	7.6	35	2.8	20	1.7
3.3	12.4	14	14.4	35	3.5	20	1.9
2.3	7.1	16	9.3	35	4.4	20	1.6
4.9	13.5	16	18.7	35	4.2	20	1.5
2.0	6.7	16	10.0	35	5.6	20	3.2
1.8	8.9	12	6.1	35	3.2	20	1.2
3.0	11.2	12	10.8	35	4.2	20	1.5
2.3	9.9	12	9.6	35	3.5	20	1.2
2.9	11.0	12	11.2	35	4.2	20	1.4
3.9	13.6	14	15.0	35	5.0	20	2.2
2.4	10.1	12	11.1	35	4.2	20	1.9
3.7	13.2	16	11.4	35	4.0	20	1.2
3.1	12.0	14	11.1	35	3.4	20	1.5
2.9	8.0	16	10.0	35	5.6	20	2.2
3.0	8.1	16	10.6	35	5.0	20	1.6
4.9	13.5	16	15.9	35	4.8	20	1.6
Average Free Fatty Acid as Oleic .....							3.3%
Average Loss .....							11.6%
Average loss to acid ratio .....							3.5

loss to acid ratio are based on averages and as such cannot be applied to individual or specific instances or samples. However, as an average index obtained under uniform and comparative conditions of testing, it furnishes a good basis for comparison. This value, of course, varies with free fat acid content, inversely as the latter increases.

The foregoing data and information are furnished for the purpose of adding to our accumulation of knowledge pertaining to the characteristics and composition of fats and oils. The great increase in the different kinds of oils which have come into common use during the last few years has emphasized our lack of complete knowledge of the behavior of many of these. In many cases the physical constants, when available, are of such old standing as to be unreliable due to

changes in methods of analysis and changes in certain characteristics of the oils. A fact that all fat and oil chemists can well afford to remember is that one analysis of one sample of any kind of oil does not define that type of oil in general, but only the sample analyzed. Thus it is necessary to have many analyses of many samples of that kind of oil so that we may establish the practical limits of variation of the factors which define that oil and differentiate it in its action, its composition, and its characteristics from any other oil.

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# SOYBEAN PHOSPHATIDES AND THEIR USES

## A REVIEW

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**T**HE term "lecithin," which is generally applied to the phosphatides of the soybean, is convenient, but from the scientific standpoint as misleading as are those of "stearin" and "olein" for the complex mixtures of glycerides which they represent in the fat industries. Only a minor proportion of the technical products sold as lecithin is in the form of the choline esters of diglyceride phosphoric acids. With four of the five fatty

acids detected in soybean phosphatides<sup>1</sup>, stearic, palmitic, oleic, linoleic, linolenic, including the stereo-isomers, the presence of 70 different lecithins is possible according to Suzuki and Yokoyama<sup>2</sup>. These investigators found that by treatment with warm acetone the cadmium chloride compounds with beta-lecithins (in which the phosphoric acid is linked to the center, beta, carbon atom of the glycerine radical) go into solution while those

with alpha-lecithins remain undissolved. In this way and by means of fractionation of the brominated phosphatides in various solvents they identified from among the alpha-phosphatides palmito-linoleo-, dioleo-, oleo-linoleo-, and dilinoleo- lecithin. Among the beta- compounds were palmito-oleo-, palmito-linoleo-, palmito-linolen-, dioleo-, oleo-linoleo- lecithin.

Cephalins, which differ from the lecithins in their amino-component,

which is amino-ethanol (colamine) instead of its trimethyl derivative, are also present. Two have been identified as stearo-linoleo-beta-cephalin and stearo-linoleo-beta-cephalin<sup>3</sup>. A considerable percentage of the phosphorus in soybean lecithin is perhaps present in the form of phosphatide acids; that is, base-free diglyceride-phosphoric acids, which have been detected first in plant leaves by Chibnall and Channon. For the molecular ratio of phosphorus to nitrogen in freshly prepared "lecithin" is not 1:1 but in the neighborhood of 1:8.

From the standpoint of industrial users the form of phosphatides present is in most cases of minor significance, because the properties of these compounds are alike or similar in many respects. An exception to this is their behavior towards alcohol, in which probably only lecithins are soluble. But purification by means of this solvent<sup>4</sup> is far from complete. The alcohol-insoluble phosphatides dissolved in another organic solvent and precipitated by alcohol or acetone form a powdery substance of crystalline structure<sup>5</sup>. The sols of silicic acid, oxidic aluminum or iron compounds have also been proposed to effect purification of soybean and other phosphatides without decomposition in place of cadmium chloride which is generally employed in the laboratory for that purpose<sup>6</sup>.

Another difference, of more scientific than technical interest, is the acidity of the cephalins (and phosphatide acids) in organic solvents, which is absent in lecithins with their stronger alkaline quaternary amino base choline<sup>7</sup>. However the quantitative use which has been made of this fact to determine the cephaline content of lecithin-cephalin-mixtures<sup>8</sup> is liable to be misleading. For we know of the presence of carbohydrates which apparently cannot be separated from the phosphatide complex without hydrolysis and therefore may form a part of the phosphatide molecule. These carbohydrates are a disadvantage to the lecithin, because they are the cause of fermentation and mold growth in the presence of water, if precautions against the development of micro-organisms are not taken, and also because they decompose early on heating. They also form an analytical basis, as does the nitrogen content, to differentiate vegetable phosphatides from similar synthetic compounds which have appeared on the market in recent years.

The industrial value of the phos-

phatides is based on their structure, which includes both lipophilic and hydrophilic groups. They are therefore not water-repellent like oils but swell in water to form colloidal solutions. They are able to reduce surface and interfacial tensions and thus to act as emulsifying and wetting agents. However, the claim made occasionally in the literature that soybean lecithin can take the place of egg yolk is in most cases unjustified, for lecithin dispersions are quickly subject to syneresis in acid or alkaline solution, contrary to the phosphatide-protein complex of egg yolk.

For the evaluation of lecithin products from soybeans, their phosphorus content multiplied by the factor 25.5 is frequently used<sup>9</sup>, or the proportion of matter insoluble in acetone, which removes the admixed oil, is determined.

Many products are being offered to manufacturers under the name of lecithin which differ widely in appearance and quality. The main product of commerce is manufactured on the basis of U. S. Pat. 1,464,557 (H. Bollmann) by causing phosphatides in the warm extracted oil to absorb water and swell, then centrifuging them out and drying under vacuum.<sup>9a</sup> They can be bleached by means of peroxides<sup>10</sup>. The resulting material is of waxy to fluid consistency. It contains 25-35 per cent of soybean oil, which can be washed out by means of acetone or ethyl acetate, a process which naturally adds considerably to the price, and is therefore resorted to only where the introduction of the highly unsaturated oil is not desired. To facilitate the handling and improve the keeping quality of the acetone-washed phosphatides, other fats are usually added to replace the soybean oil<sup>11</sup>. Another product has been prepared with water and alcohol, thus making the phosphatides more easily miscible with aqueous solutions. Some patents use such agents as glycerine or sugar syrup to remove most of the water from the hydrated phosphatides, another stabilizes the emulsion without drying by the addition of alkali peroxides<sup>12</sup>. A number of patents have been granted for lecithin mixtures with other substances; for example alkalis, which increase the solubility, sodium silicate, flour, carbohydrates, milk, cocoa, grain germs, lime, sulphur, rubber, mineral oil, benzyl and other alcohols. Halogenation and hydrogenation have also been proposed, but probably have never attained commercial significance. A greater

effect in the reduction of the surface tension of cacao butter and increased emulsifying power as measured by the drop number have been reported after hydrogenation<sup>13</sup>, whereas the effect of lecithin in margarine was found destroyed by this process<sup>14</sup>.

In the oil industry phosphatides are generally considered a nuisance, because they are liable to cause cloudiness in oils on storage, interfere with hydrogenation, and lower the water-resistance of lacquer coatings. But they also have beneficial effects in these fields, which form the basis of several patents. According to one of them<sup>15</sup> .2—1.5% of phosphatides are added before the pressing of oils or fats, especially cacao butter, in order to save power and increase the yield of fat. Pupa oil is deodorized by solvent extraction after the addition of lecithin according to Jap. Pat. 101,846<sup>16</sup>. In refined winterized cottonseed oil a small proportion of vegetable lecithin retards the crystallization of solid glycerides and thus improves the cold test<sup>17</sup>. .2-1% lecithin is claimed to increase the creaming quality of shortenings, their shortening power, and to reduce the danger of over-creaming<sup>18</sup>.

One of the oldest and most important fields of application is that of margarine, where the phosphatides reduce the interfacial tension between fat and aqueous phase, so that a few tenths of one per cent lecithin are able to reduce or eliminate the spattering at rapid heating of the margarine in the frying pan<sup>19</sup>. It also prevents the adhesion of milk solids to the pan and their burning. Halogenated lecithin with 10-20% bromine or iodine also acts as preservative in margarine<sup>20</sup>.

Similar to this use is the addition of .1-2% to butter in order to improve color and other properties<sup>21</sup>, and if added in larger amounts to the cream before ripening, to increase the butter flavor.

The work done on the anti-oxidative properties of soybean phosphatides, also the basis of several patents, was discussed in this journal not long ago<sup>22</sup>. The authors came to the conclusions that the cephalins are the active part of the lecithin. However, egg lecithin does not act as an anti-oxidant, although egg yolk does so very actively and although cephalins are present here too. It is not unlikely that another, as yet unknown, constituent is the active one. That a heavily peroxide-bleached lecithin would give disappointing results as a rancidity retarder is but natural. In this connection it is of

interest that .01-.15% lecithin has also been patented as an anti-oxidant for gasoline to prevent gum formation<sup>28</sup>.

The application of soybean lecithin in the field of soaps and cosmetics is perhaps the one most frequently discussed in the technical literature. In soap, where lecithin (being itself saponifiable it must be added after saponification) has been found to increase the surface tension and the drop number against kerosene in low soap concentrations. Foam number and stability are increased, but the foam volume is lower. Turbidity and transparency temperatures of soap solutions are lowered, the gold number is increased<sup>24</sup>. An increased cleaning efficiency is ascribed to the lecithin addition because of its emulsifying power, the formation of a soft clinging foam, and an alkalinity-depressing effect which make the phosphatides suitable as superfatting agent<sup>25</sup>. From 1-10% are recommended<sup>26</sup>. It is also used as an emulsifier for solvents in soaps<sup>27</sup>. Its activating and nourishing effect on the skin is made use of in beauty creams, with or without the addition of cholesterol, salves, bath salts<sup>28</sup>, and pine needle extracts<sup>29</sup>.

In paint manufacture lecithin increases the wettability of pigments and facilitates their grinding and re-mixing<sup>30</sup>. The hygroscopicity introduced by a small amount of lecithin makes the paint films more elastic and prevents cracks. Casein for paints is solubilized with alkali and finally emulsified with lecithin<sup>31</sup>; improved durability and resistance to rubbing are claimed. In printing inks for textiles lecithin is used with starch as a thickening agent, replacing some or all of the more expensive gums<sup>32</sup>. In nitrocellulose lacquers lecithin acts as a protective colloid and stabilizer<sup>33</sup>.

It was natural that the replacement of the costly egg yolk used in tanning processes by soybean lecithin was tried to facilitate the absorption of oils from liquors by the leather, but because of the lack of resistance to syneresis of lecithin emulsions against alum and other substances additional protective colloids have to be used<sup>34</sup>. A greater affinity for leather than that of sulphonated oils is claimed, and so lecithin as an aqueous emulsion or in mixture with oils<sup>35</sup> or water-miscible solvents<sup>36</sup> is also used as a leather dressing. After penetration of the skin lecithin becomes fixed and is not washed out.

In the textile industries lecithin is

of value as a wetting and especially as a softening agent<sup>37</sup> for all kinds of materials, foremost among them rayon. It is not suitable for acid baths because of the instability of its solution but may be added to dye baths as wetting agent or to prevent the premature oxidation of the leuco form of vat dyes<sup>38</sup>. Lecithin effects more even and thorough dyeing, greater brilliancy of coloration, flexibility, and softer feel<sup>39</sup>.

In rubber compositions soybean lecithin facilitates mixing, accelerates vulcanization, and acts as a softener<sup>40</sup>. Its addition is recommended to hard rather than to soft rubber. The dielectric properties are reduced, and excessive proportions cause low tensile strength, but powdered rubber made with lecithin has been found suitable for filters, diaphragms, paints, and chlorinated rubber<sup>41</sup>. Other plastics for which lecithin has been recommended as a homogenizing agent are resinous mixtures for phonograph records<sup>42</sup> and linoleum cement where a small amount of lecithin reduces the need for softening agents<sup>43</sup>.

While in other industries the use of soybean phosphatides often meets the competition of synthetic, frequently more effective, compositions, this is less the case with food-stuffs. From the work done in recent years it appears that phosphatides are not an essential food and that they have no tonic effect, except in special cases, such as malnutrition, but they are unquestionably a valuable foodstuff. In the feed of animals lecithin is said to produce an abundant growth of hair and a skin of good quality<sup>44</sup>.

The replacement of egg yolk by lecithin as an emulsifying agent in mayonnaise is impossible<sup>45</sup> because of the high acidity, whereas neutral emulsions can be prepared with lecithin, for example of mineral oils. In cake, too, soybean phosphatides cannot take the place of eggs, but the advantages of their use in small quantities are described in several patents. One recommends the addition of .1-.3% lecithin to the ingredients to increase the sugar and moisture tolerance of cake when the percentage of egg whites to yolks is increased above the proportion found in whole eggs (3:2), preferably with a small amount of free fatty acids<sup>46</sup>. Another uses lecithin to increase the fat tolerance<sup>47</sup>.

The use of lecithin in bakeries is not confined to cake, but extends also to bread where  $\frac{1}{4}$ -1% is added to the dry flour to promote the nourishment of yeast and to in-

crease the dilatibility of the gluten<sup>48</sup>. A similar improvement in the gluten is obtained in the manufacture of alimentary pastes<sup>49</sup>. Noodles made from middlings of wheat grown in Germany are liable to disintegrate during cooking but the addition of 1% soybean lecithin is able to prevent this and preserve the shape of the pastes. Zieglmayer explains this action by the formation of a protein-lecithin-fat adsorption complex which retards the access of water to the swelling colloids and thus offsets the disadvantage of the inferior quality due to the irregular dehydration of the grain during ripening in the relatively cool and wet European climate. The retarding effect which (synthetic) lecithin and cephalin have on the activity of trypsinases and peptidases, explained by the formation of adsorption complexes<sup>50</sup>, is possibly another factor in this improvement of inferior wheat products.

In sausages, also in cheese<sup>52</sup>, lecithin is recommended to increase cohesion and prevent crumbliness<sup>51</sup>.

Considerable quantities of lecithin are used in the candy industry, especially chocolate manufacture. The addition of .3% soybean phosphatides lowers the viscosity of the chocolate mass considerably and thus makes possible notable savings of the valuable cacao butter, which, for example, may be reduced from 35 to 30%<sup>53</sup>. Also the detrimental effect of small amounts of moisture on the viscosity is reduced, the temperature range for enrobing processes extended and the shelf life of the finished products increased. The same principle of surface reduction of fats in mixture with sugar is made use of in cake fillings.

In the manufacture of various types of candy such as taffies, caramels, nougats, and others, lecithin facilitates the distribution of the fatty materials, prevents graining and streaks, and acts as an anti-oxygen<sup>54</sup>. .1-25% lecithin aids in the distribution and fixation of volatile flavoring agents<sup>55</sup>. As a protective colloid lecithin has been patented in cocoa powder<sup>56</sup> and in the dry ingredients of soups and sauces<sup>57</sup>. In coffee infusions lecithin in combination with bromide and pH adjustment is claimed to reduce the injurious effect of caffeine<sup>58</sup>.

Other uses for soybean lecithin have been found in the mineral oil industry: as emulsifying agent for asphalt and tar emulsion<sup>59</sup>, to facilitate the penetration of creosote dur-

ing the impregnation of wood<sup>60</sup>, and for insecticides, where the copper compounds of lecithin also may be used<sup>61</sup>. Another patent applies lecithin emulsion with solid poisons, such as sodium arsenate or Paris Green to increase the dispersion of the active substances and improve their adhesion to plants<sup>62</sup>. As the essential ingredient of a contact insecticide harmless to man and mammals lecithin serves in connection with a dispersing agent such as butanol or sulphonated oils<sup>63</sup>.

In electroplating .01% lecithin as a protective colloid is claimed to give finer, denser, more uniform coatings<sup>64</sup>.

In pointing out the more or less successful, and wide application of soybean phosphatides, this survey also shows the need for further research, primarily towards gaining a clearer conception of the actual composition of the acetone-insoluble material of the "lecithin" and of its effects, investigations of which have so far been undertaken in but a few fields.

(The large majority of patents has been secured by Hanseatische Muehlenwerke A.-G., abbreviated in the following survey, where the

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## THE RELATION OF LIPIDS TO PHYSIOLOGICAL ACTIVITY

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ABOUT a century ago studies on the physiological rôle of the lipids of the brain were conducted by investigators interested in the relationship between chemical composition and mental activity. The complexity of the brain lipids and the difficulty of their separation retarded further physiological study at that time, but did stimulate chemical investigation which resulted eventually in an elucidation of the various lipids and their classification. This early conception of the lipids as essential constituents of protoplasm was revived recently by the French school of workers, Mayer, Schaeffer and Terroine. As a result of their work certain lipids have been found to play an essential rôle in the vital economy of living cells. The early studies have been interestingly summarized by Bloor et al. (1)

Two methods of attack have been

followed in the development of this concept. The French workers have shown that phospholipids and cholesterol are present in constant amounts in living tissue irrespective of the state of nutrition. They designate the phospholipid and cholesterol as the *élément constant*, considering these lipids to be constant and integral components of protoplasm essential to the vital functions of the living cell. Bloor (2), in this country, has conducted extensive studies which further substantiate their thesis. He has shown that the constituent which was most constant for each kind of organ was the total phospholipid. Analyses on the organs of the beef are in agreement with those of the French authors on the phospholipid and cholesterol content of the organs of different small animals. "The brain, which has the greatest variety and extent of function—that of con-

trolling all the activities of the organism—has the highest content of phospholipid and cholesterol. The liver, which is next in importance in the successful functioning of the organism, comes next to the brain in its content of phospholipids, then the pancreas with two important secretory functions, then the kidney and lung, each with one important function. In the case of the various muscles of beef it was found that the phospholipid content ran parallel with the probable activity of the muscle—those muscles which were more continuously and strongly active having a higher phospholipid (and unsaponifiable) content than those which were less active." In general, changes in cholesterol do accompany alterations in phospholipids, but are definitely less extensive.

A second method of attack developed by Bloor and extended by