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Modification of Lipids in the Dairy Technology*

— In Relation to Manufacture of Infant Formula Milk Powder —

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1. Introduction

Bovine milk and the products are valuable food-groups for the human beings. Among the milk products, infant formula milk powder occupies a very important position as the useful nutritive source for the human babies. However, it is impossible to prepare an infant formula with ideal qualities by a simple powdering of bovine milk, because properties of bovine and human milk are considerably different from each other. Thus, it has been attempted for a long time to arrange the composition of bovine milk qualitatively and quantitatively to approach that of human milk as closely as possible, regarding the main constituents such as proteins, lipids, carbohydrates, minerals and vitamins. Speaking about the lipids, regulation of bovine milk with plant oils has widely been carried out in these years.

The present paper describes fundamental characteristics of lipid in bovine milk and discusses theoretical background of lipid modification in dairy technology, practically in manufacture of infant formula milk powder.

2. Characteristics of bovine milk lipid

Lipids of living beings usually have physico-chemical properties common to general organisms as well as specific to the biological species or tissues, and form characteristics of the individual lipid. This goes affirmatively to the bovine milk lipids. The characteristics are strongly symbolized in fat, the principal lipid class, and fatty acid, the essential lipid component, in the bovine milk.

2.1 Classification

Lipids are generally grouped into two categories, straight-chain lipid and baanched-chain lipid, from the standpoint of biochemical structures.[1] The bovine milk lipids consist almost of the straight-chain lipid with a small amount of the branched-chain lipid; acylglycerol, called historically glyceride, is overwhelmingly abundant in the lipids (Table 1). Acylglycerol involves in detail monoacylglycerol *i. e.* monoglyceride, diacylglycerol *i. e.* diglyceride, and triacylglycerol *i. e.* triglyceride, but actually and approximately only triacylglycerol, the mixture

of which makes so-called fat. The lipid classes exist in milk not in suspension, where the lipids are dispersed separately and irregularly to one another, but in emulsion, where the lipid molecules are collected together and regularly in the milk fat (correctly lipid) globules or droplets. Grossly speaking, triacylglycerols constitute a biocore of the lipid globule and the other lipid classes do a biomembrane together with protein in the form of lipoprotein.

Table 1 Classification of lipids in bovine milk

Group	Approximate %
Straight-chain lipid	
Glyceride	98-99
phospholipid	<1.0<
Glycolipid	<0.06<
Fatty acid etc*	trace
Branched-chain lipid	
Sterol	<0.3<
Non-sterol etc**	trace

* Including flavor compounds.
** Including fat-soluble vitamins.

2.2 Component fatty acids

Since almost all the lipid classes in bovine milk include fatty acids as molecular constituents, properties of the bovine milk lipid might owe principally to the component fatty acids. Characteristics of fatty acids in bovine milk are shown in Table 2, which has been figured out from the literature.(2) It is seen that number of the classes reaches more than 400, that range of the carbon number covers from 2 to 28, that there are not only even-carbon fatty acids but also some amount of odd-carbon ones, and that in addition to ordinarily-saturated and - unsaturated fatty acid there exist some quantity of acids with extraordinary structure.

These characteristics originate mainly in metabolic activities of microorganisms in the bovine rumen.

2.3 Molecular speices of fat

Fat is composed of various molecular species of triacylglycerol. Detailed analyses of the fat species are quite difficult methodologically, but general trend of the species of bovine milk fat has been clarified considerably well.

According to the studies in this field, it is said as regards three Cs in the glycerol moiety that positional distribution of the component fatty acids are asymmetry and non-random, that saturated acids are much superior to unsaturated ones at each of C-1, C-2 and C-3, and that short-chain acids are located almost exclusively at C-3, actually not at C-1 and C-2. Since the triacylglycerols containing short-chain acids at C-3 amount approximately to half of the whole triacylglycerols, the bovine milk fat can be divide into two groups(3) as shown in Fig. 1, from the angle of distribution of the molecular wight expressed by C number of the combining

Table 2 Characteristics of fatty acids in bovine milk %

Acid group	Number of class	Range of carbon number	Even-C (E)	Odd-C (O)	E+O
Saturated: $-(CH_2)_n-$	27	2-28	60-22	2.61	62.83
Unsaturated:					
Monoenoic -CH=CH-	115	10-26	30.42	0.32	30.75
Dienoic $-(CH=CH)_2-$	42	14-26	2.97	-	2.97
Polyenoic $-(CH=CH)_n-$	18	18-22	0.85	-	0.85
Special:					
Hydroxy $-\overset{OH}{\overset{\vert}{C}}H-$	85	10-16	trace	trace	0.40
Keto $-\overset{O}{\overset{\Vert}{C}}-$	59	10-18	trace	trace	
Cyclic $-C\overset{C}{<}C-$	2	15-17	-	trace	
Monomethyl $-\overset{CH_3}{\overset{\vert}{C}}H-$	71	9-26	0.31	1.03	1.36
Polymethyl $\left(-\overset{CH_3}{\overset{\vert}{C}}H-\right)_n$	18	16-28	0.63	0.20	0.83
Total	437	2-28	95.40	4.16	99.99

acyl radicals, the one group around C38 such as C18-C16-C4 and the other around C52 such as C18-C16-C18. Existence of C38 group of triacylglycerols would be extremely characteristic for the bovine milk.

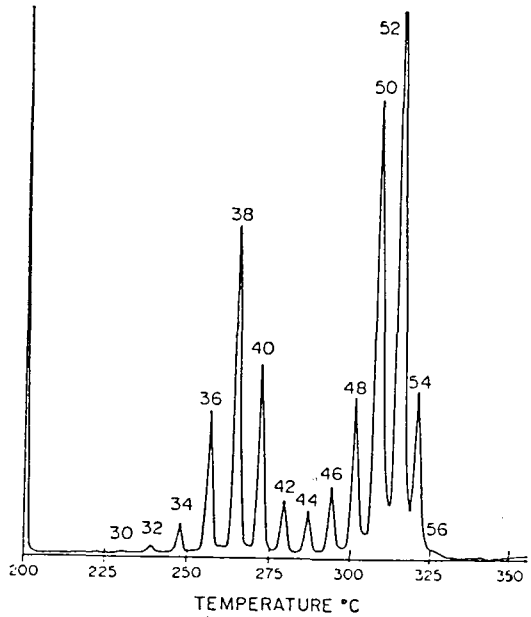

Fig.1 Gas-chromatogram of triacylglycerols in bovine milk

3. Bovine and human milk

Bovine milk is apparently similar to human milk, but componentially quite different from this. The big difference between the two milks regarding the component lipids would be that the bovine milk lipid is abundant in saturated fatty acids especially short-chain ones whereas the human milk lipid is plentiful in unsaturated fatty acids particularly the nutritively-essential ones.

3.1 Comparison of general components

Both the bovine and human milks are equally formed of 88% water and 12% solid, but chemical composition of the solid, ingredients of each component and thus nutritive properties of the milks are considerably dissimilar to each other.[4,5]

Table3 demonstrates a broad comparison of components in the two milks. As seen here, the bovine milk is rich in protein and mineral whereas the human milk in carbohydrate, and both the milks are almost equal in the amount of lipid. However, looking into the ingredients of lipid, the bovine milk is abundant in saturated acylglycerols and short-chain ones whereas the human milk in unsaturated acylglycerols and long-chain ones.

Table 3 Componential differences between bovine and human milk

Component		bovine milk	human milk
Protein	Content	2.9%	1.1%
	Casein	+++	++
	αs-Casein	+	-
	Whey protein	+	+++
	α-Lactalbumin	+	+++
	β-Lactoglobulin	+	-
Carbohydrate	Content	4.5%	7.2%
	N-Acetylglucosamine (*Bifidus factor*)	+	+++
Lipid	Content	3.3%	3.5%
	Saturated glyceride	+++	+
	Unsaturated glyceride	+	+++
	Complex lipid	+	+
	Short-chain glyceride	+++	+
Mineral	Content	0.7%	0.2%
	Calcium	+++	+
	Phosphorus	+++	+
	Iron	+	+++

3.2 Comparison of fatty acids

Properties of acylglycerols, the substantial lipid in milk, should primarily be discussed qualitatively and quantitatively on level of their molecular species, but, usually are postulated on level of their constituent fatty acids, since analyses of molecular species might be much difficult compared with those of fatty acids.

Table4 indicates composition of the principal fatty acids constituting lipid in bovine and human milks. Main fatty acids are oleic and palmitic ones in both cases, but followed by saturated acids such as stearic and short-chain acids in bovine milk whereas by unsaturated acids such as linoleic and linolenic acids in human milk.

According to the Table, it may be characteristic that the ratio of saturated and unsaturated fatty acids is 60:40 in bovine milk and 50:50 in human milk and that the nutritively-essential fatty acids such as linoleic, linolenic and arachidonic ones are several times more in human milk than in bovine milk.

Table 4 Composition of fatty acids in milk

Fatty acid	C:△	Bovine	Human
		%	%
Saturated:			
Butyric	4:0	2.8	0.1
Caproic	6:0	2.3	0.1
Caprylic	8:0	1.1	0.2
Capric	10:0	3.0	2.3
Lauric	12:0	2.9	9.5
Myristic	14:0	8.9	10.4
Pentadecanoic	15:0	0.8	-
Palmitic	16:0	23.8	22.2
Heptadecanoic	17:0	0.7	-
Stearic	18:0	13.2	5.5
Nonadecanoic	19:0	0.3	-
Arachidic	20:0	0.3	1.0
Behenic	22:0	0.1	-
Unsaturated:			
Decenoic	$10:1^{9}$	0.3	0.1
Dodecenoic	$12:1^{9}$	0.1	0.1
Tetradecenoic	$14:1^{9}$	0.7	0.6
Hexadecenoic	$16:1^{9}$	1.5	4.5
Oleic	$18:1^{9}$	25.5	27.9
Linoleic	$18:2^{9,12}$	2.1	13.0
Linolenic	$18:3^{9,12,15}$	0.4	2.5
Arachidonic	$20:4^{5,8,11,14}$	0.1	-

3.3 Compensation of fatty acids

It is perhaps impossible to make artificially the bovine milk lipid equal to the human milk lipid, since both lipids are quite different from each other qualitatively and quantitatively as well as in major lipid classes and minor ones. Only thing

to be able to do is to approximate the bovine milk lipid to the human as closely as possible. In this meaning, it has been practically carried out that the bovine milk lipid is substituted and mixed industrially by plant lipid, which is generally less in saturated acids whereas much more in unsaturated ones, particularly in the essential acids such as linoleic and linolenic acids, than animal lipid, as shown in Table 5. This compenstion would be one step for the bovine milk lipid to approach to the human milk lipid.

Table 5 Fatty acid composition of edible fat (%)

Fatty acid / Fat	Butyric 4:0	Caproic 6:0	Caprilic 8:0	Capric 10:0	Lauric 12:0	Myristic 14:0	Palmitic 16:0	Palmitoleic 16:1	Stearic 18:0	Oleic 18:1	Linoleic 18:2	Linolenic 18:3	Arachidic 20:0	Eicosenic 20:1
Animal fat:														
Butter	2-5	1-3	1-3	1-4	2-5	7-11	24-29	1-2	7-13	30-40	2-4	1-2	-	-
Pork	-	-	-	-	-	1-2	24-33	2-3	8-15	40-60	7-12	-	-	-
Beef	-	-	-	-	-	2-8	24-35	1-3	14-30	30-50	1-5	-	-	-
Plant oil:														
Soybean	-	-	-	-	-	-	5-12	-	2-7	20-35	50-57	3-8	0-1	0-1
Sunflower	-	-	-	-	-	-	3-8	-	2-5	15-35	50-75	0-1	-	-
Cottonseed	-	-	-	-	-	0-3	20-30	0-2	1-5	15-30	40-52	-	0-1	-
Safflower	-	-	-	-	-	-	4-8	0-1	1-4	8-25	60-80	0-1	-	-

4. Biochemical function of fatty acids

The nutritively-essential fatty acids are actually polyunsaturated acid such as linoleic and linolenic ones in plant and animal lipid or arachidonic and icosapentanoic ones in animal lipid. These acids are biologically of importance in generating energy as well as in constituting biomembrane in human tissues. On the other hand, polyunsaturated acids have a weak aspect to subject to peroxidation to produce the compounds, some of which are physiologically toxic for human tissues.

4.1 Generation of energy

Fatty acid-containing compounds taken by human body and/or synthesized by human tissues metabolically liberate in time the component saturated and unsaturated fatty acids, which are usually degraded principally by β-oxidation finally to CO_2 and H_2O to generate 9 kcal of energy in average per one gram of fat (Fig. 2). In case of the unsaturated fatty aceds including the essential ones, not only β-oxidation but the additional reactions such as isomerization and hydroxylation (Fig. 3) take part in the whole degradative pathway, because the acids have the double bond with *cis* configuration in their molecule.

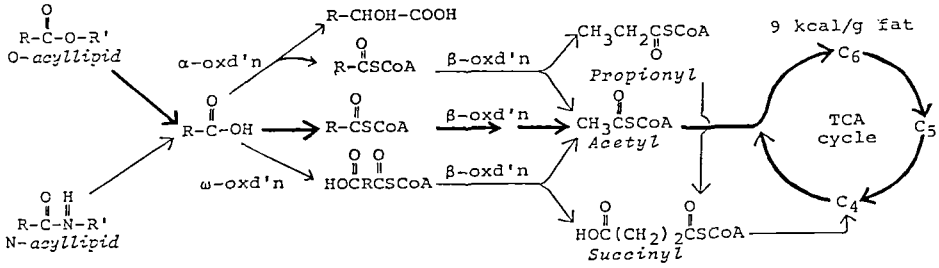

Fig. 2 Generation of energy from fatty acids in animal tissue. β-Oxidation with bold arrows is the major pathway, whereas α- and ω-Oxidations with fine arrows are only the minor.

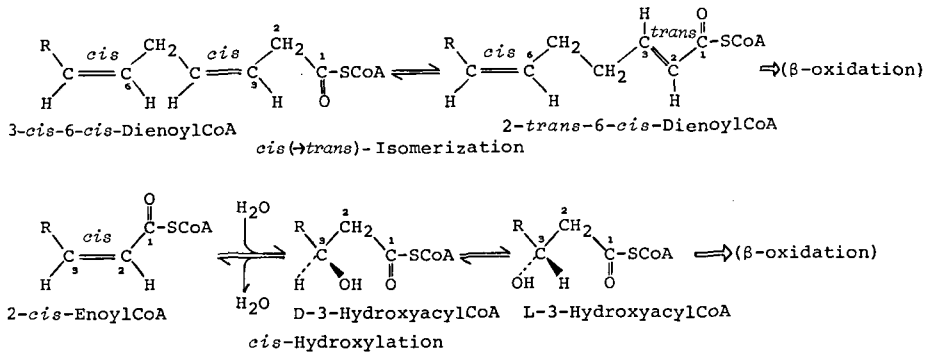

Fig. 3 Introductory reactions in β-Oxidation of unsaturated acid

4.2 Constitution of biomembrane

The biomembrane has basically a structure,[6] where bilayer of the polar lipids, which consist mainly of phospholipids, additionally of glycolipids, sterols and so forth, locate the internal and peripheral protein particles at places to constitute lipoprotein as a whole (Fig. 4). A greater part of the hydrophobic ridicals facing each other in the bilayer is formed of the fatty acid moieties, namely the long-chain acyl radicals, the composion of which largely relates to the existing state and biological activity of the biomembrane. Looking over the molecular forms of fatty acids (Fig. 5), saturated and *trans*-unsaturated fatty acids are rather straight like the cudgel whereas the *cis*-unsaturated acids, naturally-occurring unsaturated ones, are bending like the crescent in accordance with number of the double bonds in the molecule. Therefore, the biomembrane rich in saturated acids, which are located tightly in parallel, is apt to become solid and crystalline, while the biomembrane rich in unsaturated acids, which are placed loosely in disorder, is prone to get fluidic and liquid-crystalline. Accordingly, the biomembrane abundant in essential fatty acids could be softer, more flexible and more transportable of materials than that dificient in the essential acids.

Further, in the protein particle buried in the lipid bilayer, the peptide chain

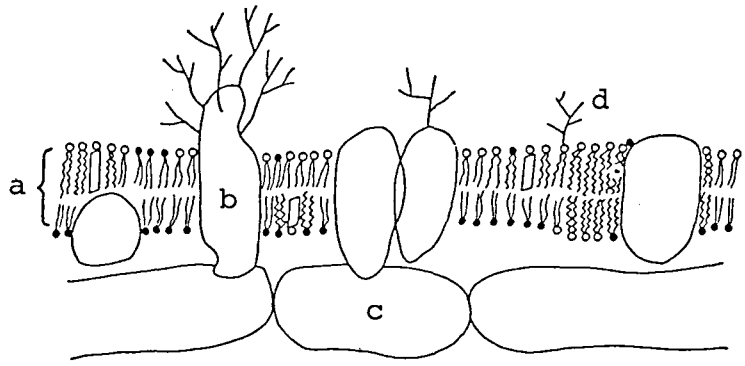

Fig. 4 Structural model of biomembrane
a. Lipid bilayer b. Integral protein
C. Peripheral protein d. Sugar-chain residue

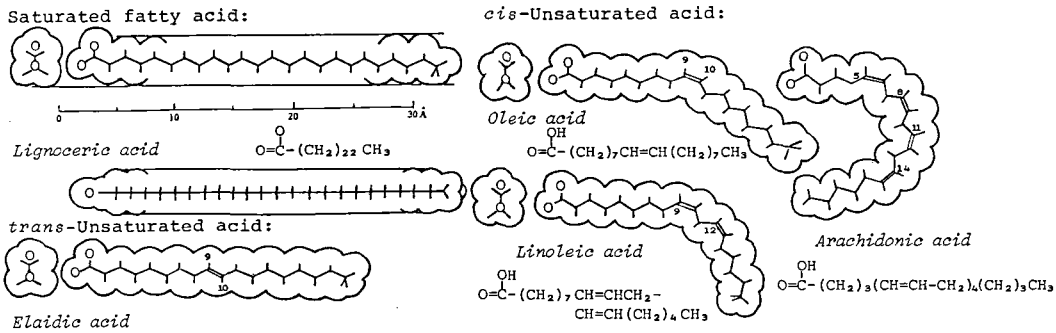

Fig. 5 Molecular form of fatty acids

and the acyl chain are molecularly located in adjacency, where the bending structure of the peptide and that of the *cis*-unsaturated acid are almost parallel with each other[7] (Fig. 6). This means that the essential fatty acids might protect the protein from undesirable oxidation which lead protein to age and retrogradation.

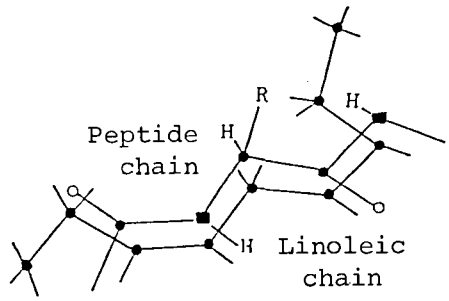

Fig. 6 Parallel co-existence of peptide chain and linoleic chain

4.3 Formation of lipid peroxide

Unsaturated fatty acids, especially polyunsaturated ones such as the essential fatty acids are liable to subject to peroxidation by various oxidative mechanisms, particularly by the autoxidation to from lipid peroxides, which are further degraded or polymerized to produce many kinds of compounds with lower or higher molecules. Fig. 7 shows that a poly-unsaturated acid forms lipid peroxides by automatic oxidation and that the lipid peroxide produces further the degradative compounds such as malonic aldehyde. It is probable that the lipid peroxides and the degradatives would react with proteins,nucleic acids, biomembranes and so on to become cause of the liver disease or arterial sclerosis and further of age of cancer.(8)

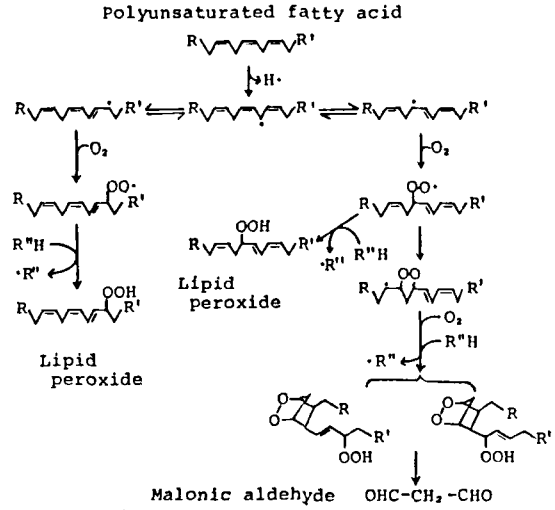

Fig. 7 Peroxidation of polyunsaturated lipid

5. Situation of commercial fat

Unsaturated lipid is originally defended from peroxidation in biological tissues by coexistence of the antioxidative compounds. However, commercial edible fat is ordinarily disposed to partial peroxidation, since attention is not paid specially to the antioxidative materials, which might be tolerably removed in the process of manufacturing.

5.1 Antioxidative material

Living being has usually mechanisms to defend the generation of lipid peroxide by enzymes or antioxidative compounds. General antioxidants in nature are known to be uric acid, ascorbic acid (vitamin C), glutathione, carotenoid, tocopherol(vitamin E) and so on (Fig. 8*), among which cartenoid and tocopherol would especially be effective to antioxidative reaction in the hydrophobic system of cells.In addition, special antioxidants such as gallic acid(tannin component),sesamol(in sesame),

quercetin(kind of flavonoid), gossipol(in cottonseed), nordihydroguajaretic acid(in creosote bush) and so forth have been found in plant kingdom. Further, in imitation of the natural antioxidative compounds, several artificial antioxidants such as butylhydroxyanisol *i. e.* BHA, dibutylhydroxytoluol *i. e.* BHT, ethylprotocatechuate *i. e.* EP, propylgallate *i. e.* PG and so on have as well been synthesized for the practical use.

*Carotenoid and tocopherol involved respectively some analogues, among which the representative one is shown for each in Fig. 8.

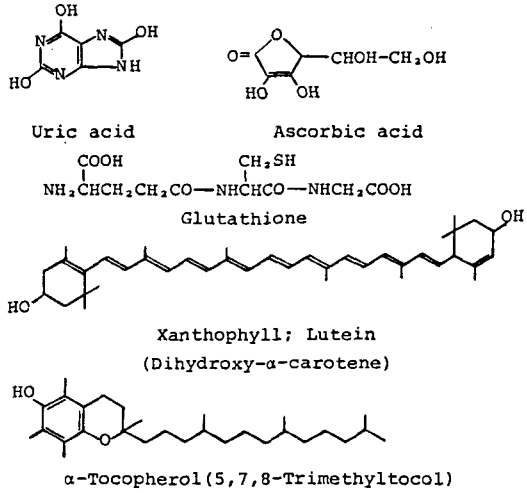

Fig. 8 Antioxidative compounds in nature

5.2 Mechanism of antioxidation

The antioxidative compound usually possesses functional groups such as -OH, -SH and so on in the molecule. A representative group will be the phenolic OH, which reacts with the active lipid-radical R. to produce the stable lipid RH and stabilize itself by resonance to stop the propagative autoxidation of lipid(Fig. 9).

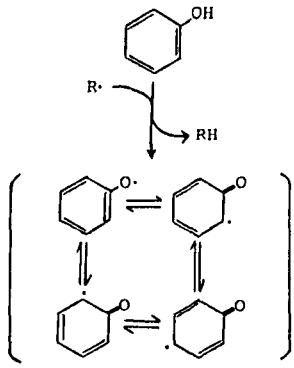

Fig. 9 Mechanism of antioxidation

5.3 Stability of market fat

Edible fats are usually manufactured by collection of fat from the animal or plant materials and then by purification of the crude fat. The methods of collection involve rendering (for beef, pork, fish etc.), compression(for coconut cottonseed etc.), extraction(for rice-bran etc.), press-extraction(for soybean, rapeseed etc.) and centrifugation(for bovine-milk etc.).

The crude fat collected by the respective method should be refined through procedures of degumming, deacidification, decoloration and deodorization.

Fig. 10 illustrates a gross picture of manufacture of the soybean oil, which is a representative plant oil utilised most not only in Korea or Japan but also in the world. In the course of manufacture, the fat is elaborated to the edible article of commerce, in which the original antioxidative compounds may have been removed through the refining procedures. It is likely that carotenoids would be excluded principally in the stage of decoloration and tocopherols of deodorization.

Thus, the plant oils finely prepared are rather unstable for the peroxidation, so that even addition of antioxidant, usually α-tocopherol, may be carried out for the commercial use.

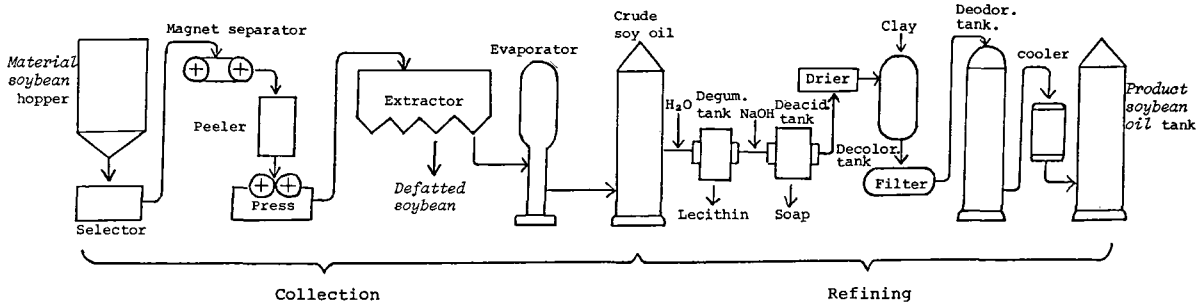

Fig. 10 Flow sheet for manufacture of soybean oil

6. Control of milk-powder lipid

Considering the facts above mentioned, at least not only plant oil but also antioxidant should be added to the bovine milk in manufacture of the infant formula milk powder to approach the composition to that of human milk. This is only a treatment adopted from the angle of component fatty acids of both the milks, but many other problems remain from the standpoint of whole lipids of the two.

6.1 Manufacture of infant formula

The infant formula milk powder is generally manufactured industrially by adding or mixing the defatted, sometimes decaseinated or desalted bovine milk to various compensatory nutrients such as protein, carbohydrate, fat, vitamin, mineral and so forth(Fig. 11). The compensatives may be added usually in the stages of standardization or blending;particularly fat also of spray-drying. The most useful fats to be substituted are currently plant oils such as soybean, cottonseed and coconut oils.

In every case, the substituted plant oil should be dispersed and emulsified as the fine lipid droplets where the oil is covered and protected by the membrane of bovine-milk protein. Tocopherol *i. e.* vitamin E, quite poor in bovine milk compared with human one, are originally added as one of the fat-soluble vitamins and contributes simultaneously to antioxidation of unsaturated lipids in the infant formula.

Therefore, it should be desirable and reasonable that plant oil and tocopherol, practically the α-form, are added at the same time in manufacture of the infant formula.

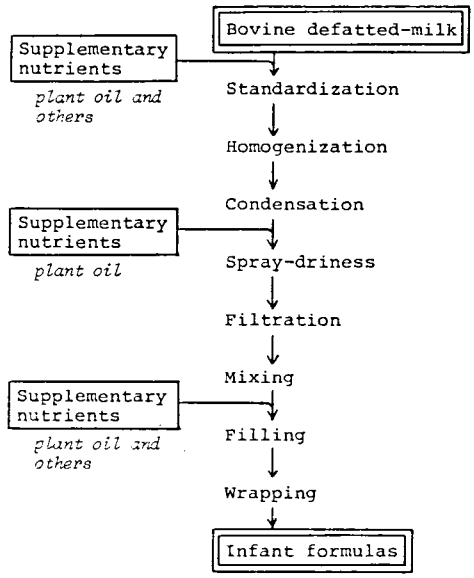

Fig. 11 Manufacture of infant formula milk powder

6.2 Composition of infant formula

Table 6 shows the nutritional composition of preparations of infant formula manufactured by several dairy companies in Japan. As seen here in the preparations, 80-100% of fat (lipid) of the bovine milk have been substituted by plant oil and some amounts of tocopherol have been added, so that the contents of linoleic acid and tocopherol are approximately 3 g and 4-6mg, respectively, in 100 g of the milk powder.

Since it is advised in the International Standards by the FAO-WHO that linoleic acid should be more than 0.3 g per 100 kcal of energy and the ratio of polyunsaturated fatty acids(g) to tocopherols(mg) should be more than 0.6, the Japanese infant formulas seem to satisfy the conditions well.

6.3 Problems in milk powder lipid

The infant formula milk powder is widely manufactured in the world, covering Korea, Japan and Western countries. The common theatment for lipids in the manufa-

Table 6 Composition of infant foumula milk powder in Japan
(in 100g of product, 1979)

Component \ Product		M_1	M_2	W_1	W_2	Y
Energy	*Kcal*	508	515	521	520	520
Water	*g*	2.0	2.7	2.0	2.0	2.0
Protein	*g*	13.0	13.7	13.9	12.0	13.5
Fat	*g*	25.0	27.0	27.7	28.0	27.3
Bovine	*g*	5.0	8.1	7.1	—	5.0
Substituted	*g*	20.0	18.9	20.6	28	22.3
Linoleic acid	*g*	2.5	3.3	3.5	—	3.3
Carbohydrate	*g*	57.7	54.2	54.1	56.0	55.0
Lactose	*g*	51.0	49.2	54.1	56.0	55.0
Others	*g*	6.7	5.0	—	—	—
Ash	*g*	2.3	2.4	2.3	2.0	2.3
Mineral						
Calcium	*mg*	410	340	380	330	370
Phosphorus	*mg*	270	280	300	260	216
Iron	*mg*	6	6	7	6.2	6
Vitamin						
A	*IU*	1,700	2,000	1,700	2,071	1,500
B_1	*mg*	0.3	0.4	0.4	0.55	0.3
B_2	*mg*	0.4	0.7	0.6	0.82	0.6
B_6	*mg*	0.3	0.3	0.3	0.32	0.4
B_{12}	*μg*	2	2	2	0.82	1
C	*mg*	45	50	46	40.0	48
D	*IU*	400	400	420	331	400
E (Tocopherol)	*mg*	4	6	6	4.85	3.1
Niacin	*mg*	6	5	6	4.06	5
Folic acid	*mg*	0.2	0.05	0.1	—	0.1
Pantothenic acid	*mg*	—	—	2	—	2

cture is humanization of bovine milk by substitution with plant oil.

This could lead to the partial approach of bovine milk to human milk in the composition of fatty acids and the amount of linoleic acid, but, of course, not to the perfect approach, because the molecular species and their compositions of triacylglycerol, phospholipid, glycolipid and the other minor lipids (Table 1) are considerably different between bovine and human milk. An attempt to approximate the composition of molecular species of triacylglycerols in bovine milk to that in human milk through the ester exchange reactions and to improve the degree of digestion and absorption of fats in bovine milk to that of human, is now being carried out somewhere, but appears to be still difficult to achieve success.

The level of blood cholesterol is usually lower in sucklings with infant formula than in those with mother milk, the right or wrong of which can not be determined yet.

Babies nursed with the modified milk containing plenty of vitamin D are to suffer more often from rickets than thouse with the mother milk, the reason of which can not be understood yet. Thus, many problems remain to be studied as regards the artificial control of lipids in manufacture of infant formula.

7. Conclusive note

Compositional differences between bovine and human milk have been emphasized and artificial modification of lipid in bovine milk to approach to that in human milk have been discussed in this paper. It is basically desirable that foods should be taken as in natural conditions as possible.

Thus, human babies should originally be nursed with mother milk. However,babies who are very poor in growth, or abnormal in metabolism, or lacking in mother milk must depend inevitably upon the artificial nursing. For these cases, better humanization of bovine milk will be required in future as well. Therefore, concerning the milk lipid, it might be an obligation for lipid scientists to modify the bovine milk lipid to approach the human milk lipid as closely as possible.

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