



## Effect of Heat Drying Process on Fatty Acid Structure of Vegetable Oil in Infant Formula



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**M**ILK IS the main nutritional base which is rich in essential nutrients for the baby. Infant formula is intended as an effective substitute for infant feeding. This study aimed to determine the amount of fatty acids in cow's milk and to compare them with fatty acid profiles of breast milk, infant formula and vegetable oil used in production of baby's dry milk. In this study, 3 types of cow's milk, 3 samples of breast milk, 3 samples of infant formula and 1 and 2 samples of vegetable oil for infant formula were measured based on the presence of fatty acids in cow's milk, fatty acid profiles of breast milk and infant formula and vegetable oil. Oil samples were homogenized and prepared by Easy MIX, France. The study found that temperature on 90 °C and 180 °C had no effect on fatty acid profiles of vegetable oils and infant formula, and was very similar to breast milk, whereas the cow's fatty acid profiles differ in the amount of saturated and trans fatty acids compared to breast milk, it is clear that whole milk cannot replace breast milk just by diluting it with water and adding a little sugar.

**Keywords:** Milk, Fatty acid profiles, Pegah, Iran

### Introduction

Milk powder is a product that is made of drying fresh and healthy pasteurized milk in one of the most common industrial methods (rolling or spraying), Which can also be obtained from whole milk, low-fat milk, skim milk, or sweet butter juice [1-10].

Milk is the main nutritional base which is rich in essential nutrients for the baby. Infant formula is intended as an effective substitute for infant feeding [19]. Although production of an identical product to breast milk is not feasible, every effort has been taken to mimic the nutrition profile of breast milk for normal infant growth and development. One of the main components of milk is fat, which makes up 22 to 27 percent of all baby fat [2, 3]. Fats are important nutrients

not only because of their high energy value, but also because of their fat-soluble vitamins and the essential fatty acids  $\omega 3$  and  $\omega 6$  found in natural foods [16, 17].

Fatty acids are divided into two groups of saturated and unsaturated fatty acids. Trans fatty acids are those fatty acid isomers that have at least one double bond in the trans state. Both saturated and trans fatty acids have detrimental effects on health, but the effects of trans fatty acids are far greater than those of saturated fatty acids [15]. The fat content in a cow's milk sample consisted of 65% saturated fatty acids That mainly includes palmitic acid, stearic acid, myristic acid and 35% unsaturated fatty acids usually oleic acid [12].

It should be considered that palmitic, myristic and lauric fatty acids increase total cholesterol and

LDL cholesterol, while the effects of stearic acid and short-chain butyric, caproic and caprylic acids on blood cholesterol are neutral and even very low [11]. In addition to providing energy, milk fat has a structural role and provides essential fatty acids and fat-soluble vitamins to the human infant [10]. In the production of infant formula, since cow's milk is often used as the basis of production and cow's milk fat is different from human milk and also because it's not appropriate for human infants, Therefore, a mixture of different vegetable oils or a combination of vegetable and animal oils are used as alternative fats that's trying to make their fatty acids especially essential fatty acids such as linoleic acid and alpha linolenic acid similar to breast milk fatty acids [14]. Milk fat globule membranes are a fraction that has been previously excluded from infant formulas, but its components are active and prevent infection [15]. Milk fat globule membrane supplementation of infant formula also narrows the gap in cognitive development between breastfed and formula fed infants [16]. The monounsaturated fatty acid, oleic acid (18:1w9), is in the highest percentage (36%) in milk breast milk also contains two essential fatty acids, linoleic acid (C18:2w6) at 15% and alpha-linolenic acid (C18:3w3) at 0.35% [18], arachidonic acid (C20: 4n-6) and 0.1 to 0.4% Decanoic acid is hexane (C22: 6n-6) and much lower amounts of n-6 and n-3 fatty acids [16]. These two essential fatty acids are, respectively, converted to arachidonic acid (AA, C20:4w6) and eicosapentaenoic acid (EPA, C20:5w3), the latter of which is further converted to docosahexaenoic acid (DHA, 22:6w3). AA, EPA and DHA are important for regulating. The fatty acids found in breast milk are highly dependent on the type of breast feeding [9, 13]. Human milk fat usually contains 44% saturated fatty acids, 50% polyunsaturated fatty acids and about 5% monounsaturated fatty acids, which also follow the same pattern in breast milk production [14].

Infant formula is intended as an effective substitute to breast milk and is formulated to mimic the nutritional composition of breast milk. The recently updated Food and Drug Administration (FDA) rule on current Good Manufacturing Practices for infant formula, 21 CFR 106.96 [8], requires, among other things, that formulas satisfy the quality factors of normal physical growth and a sufficient biological quality of protein component (adequate amounts of protein in a form that can be used by infants). Infant formula is only for

the health of infants without unusual medical or dietary problems. The manufacturing process is highly regulated and monitored to meet national and international quality criteria [20]. Therefore, this study aimed to determine the amount of fatty acids in cow's milk and comparison them with fatty acid profiles of breast milk, infant formula and vegetable oil used in production.

## **Materials and Methods**

### *Fat extraction test and determination of fatty acid profile*

First, raw milk, infant milk, breast milk and oil samples were homogenized and prepared by Easy MIX, France. Then total fat was determined according to Gerber method according to National Iranian Standard of number of 1531 [1, 2].

### *Extraction of fatty acid profiles*

#### *Extraction of fatty acid profiles of breast milk and cow's milk*

Fulch method was used to extract fatty acids. Five grams of sample was added to 40 ml of chloroform: methanol (1: 2v/v) solution and stirred for 1 min. The resulting mixture was then passed through a filter paper to separate the solid particles. The resulting phase was divided into three equal portions in three test tubes and to each test tube was added one fifth of the volume of solution in that distilled water. After three minutes the centrifuge solution and two phases were formed, the supernatant containing water and non-lipid impurities. The supernatant was discarded and again a fifth of the remaining volume of distilled water was added to each tube and centrifuged. This was repeated three times. In the last step all three tubes were united and the supernatant was completely separated and discarded and the solution dried by a vacuum Rotary and then added to n-hexane and stored in the freezer [8].

### *Extraction of oil fatty acid profiles*

About 4 g of the sample was transferred into the balloon and added about 40 ml of methanol, 0.5 ml of methanol potassium and a few boiling stones then attach the cooling tube to the balloon until a clear solution was added (10 minutes). The balloon was then cooled under running water and the contents transferred to a separating funnel. The balloons were washed with 20 cc-hexane and 40 cc distilled water and added to the separator funnel. Shake the hopper to separate. The upper layer esters were removed and the extraction process was again performed on aqueous layer

with 20 cc-hexane and the organic layer was added to the previous contents and the solvent was extracted with a vacuum rotator [3].

#### *Methyl ester analysis of fatty acids*

Concerning the fatty methyl esters we had already prepared, 7CN-hexane grade GC was added and after homogenization the sample was added by tube shaker for methylation operation of 2cc methanol 2 molar. The tube containing the sample was then placed in a steam bath for 15 minutes at 50-55 ° C (stirred by Shaker every five minutes and each time). Finally, the upper phase was transferred to another test tube, to which was added 0.5 g of sodium sulfate without water added to remove its moisture and was isolated and analyzed by centrifugation of the supernatant. The methyl ester produced in accordance with Standard 8819 [4] was analyzed by USA GC

Thermo trace, equipped with FID detector and capillary column under the following conditions. The oven temperature started at 140 °C and its temperature program increased from 11 °C every 4 minutes to 240 °C, then at 245 °C for 2 minutes and at the end of separation (Figures 1 to 5 of acid profiles. Fatty samples, injector section temperature and FID were set at 260 °C and the helium gas flow rate constant was assumed to be 0.8 mL/min.

#### **Results**

The results of the study such as Fatty acid profile analysis and quantities of different fatty acid groups in fat extracted from breast milk and infant formula, cow's milk and two samples of infant formula were shown in table 1 and 2 and chromatogram were shown in figures 1-6.

**TABLE 1. Fatty acid profile analysis**

Iranian oil	Exterior oil	Cow's milk	Milk powder	Human breast milk	Fatty acid methyl ester percentage
0	0	0.7457	7.7998	4.0547	C4:00
0	0	0.7709	0.0000	0.0000	C6:00
0	0	0.6219	0.0000	0.0000	C8:0
0	0	1.6928	0.0000	0.5239	C10:0
0.1088	0.1377	2.2928	0.1332	3.0898	C12:0
0.5829	0.6375	9.3664	0.5933	4.2172	C14:0
0	0	1.1995	0.0000	0.1069	C14:1
0	0	1.1047	0.0000	0.2477	C15:0
0	0	0.2240	0.0000	0.0000	C15:1
26.4989	29.2876	34.7944	26.9982	21.5384	C16:0
0.1651	0	1.7197	0.1611	1.8798	C16:1
0.0784	0	0.6885	0.0000	0.3978	C17:0
0	0	0.2362	0.0000	0.2227	C17:1
1.9510	4.0743	12.2150	3.5868	5.4375	C18:0
2.3458	0.7852	26.0596	0.5755	1.7422	C18:1n9t
39.6545	41.4268	0.0861	38.3495	28.5709	C18:1n9c
0.2537	0.1547	0.1276	0.1897	0.0902	C18:2n6t
21.0111	17.97	3.5884	16.9393	22.0578	C18:2n6c
0.1254	0.5106	0.0919	0.0000	0.3903	C18:3n6
0.4895	0	0.1626	0.4745	0.0718	C20:0
2.2525	1.9485	0.7414	2.0575	1.6825	C20:1
2.4191	1.8995	0.5097	1.3705	1.4567	C18:3n3
0.0735	0	0.0000	0.0000	0.3590	C20:2
0.2372	0.2536	0.1075	0.2305	0.5667	C22:0
0	0	0.1309	0.0000	0.1068	C20:3n6
0	0	0.2133	0.0000	0.5209	C22:1n9
0.0755	0.1718	0.0000	0.0000	0.0000	C20:3n3
0	0	0.1338	0.0000	0.0836	C23:00
0.0547	0	0.0000	0.0000	0.0000	C20:4n6
0.1045	0	0.0794	0.0000	0.1014	C22:2
0.1193	0.1220	0.0000	0.1159	0.1325	C24:0
0.0621	0.2488	0.2023	0.2709	0.1713	C24:1
0.639	0.1509	0.0933	0.1538	0.1793	C22:6N3

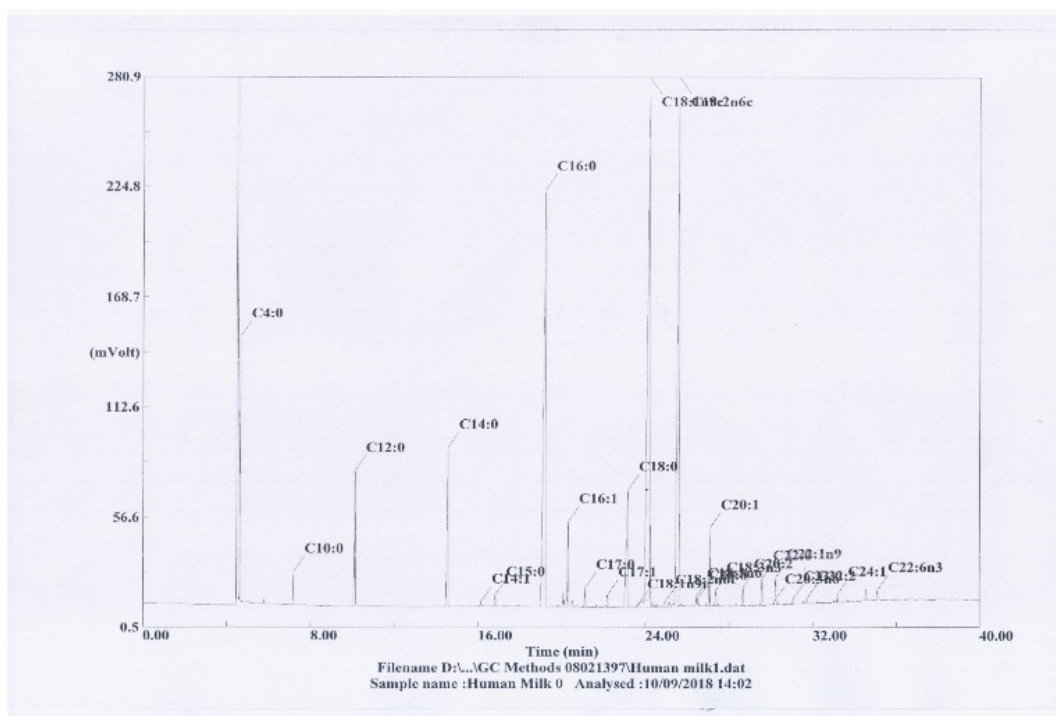


Fig. 1. Chromatogram of fatty acids derived from fat extracted from breast milk

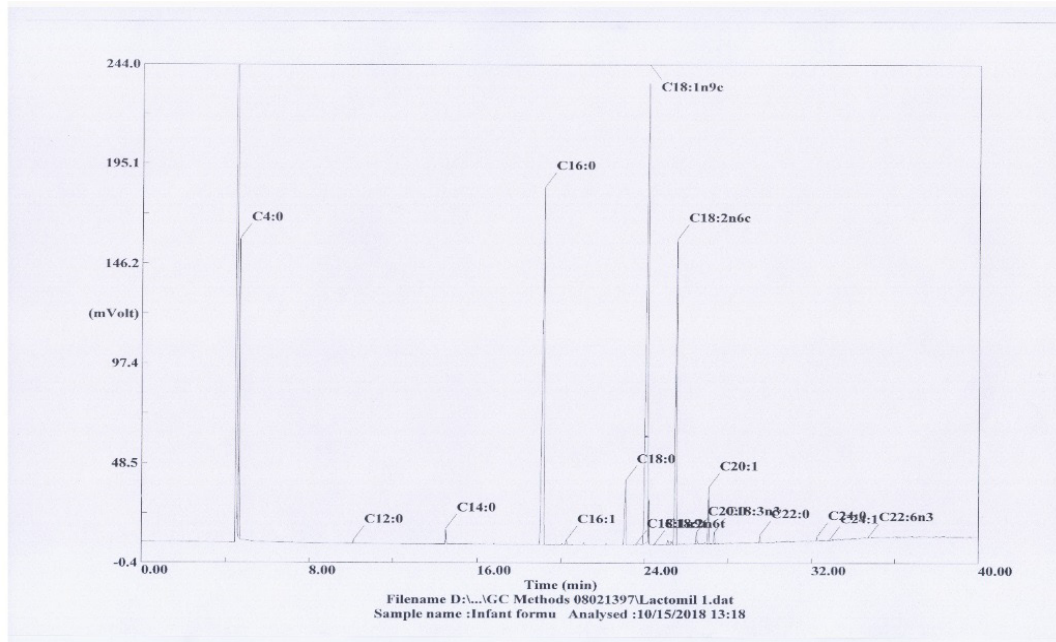


Fig. 2. Chromatogram of fatty acids derived from fat extracted of neonatal dry milk (Lactomil1)

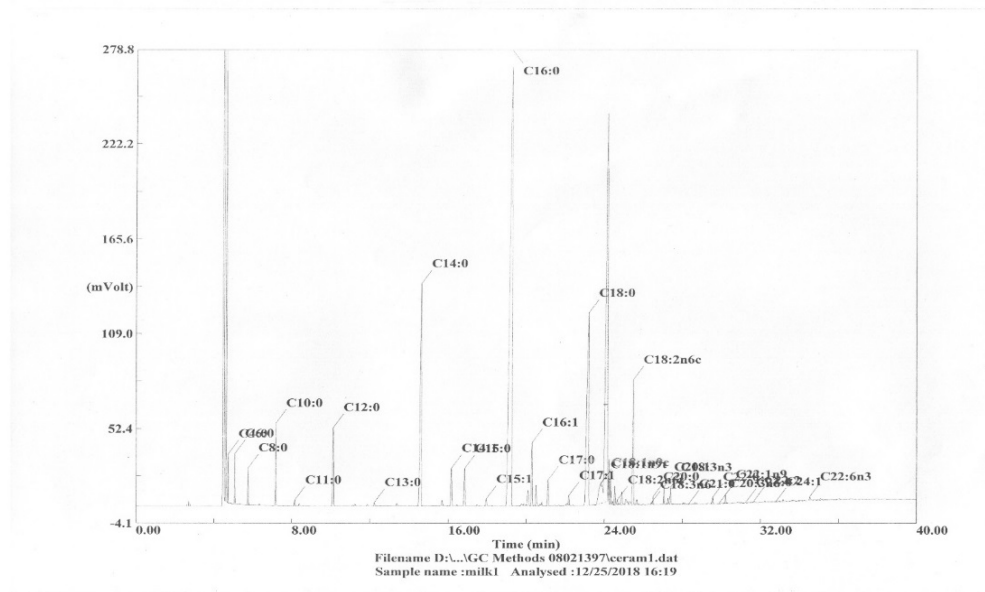


Fig. 3. Chromatogram of fatty acids derived from fat extracted of cow's milk

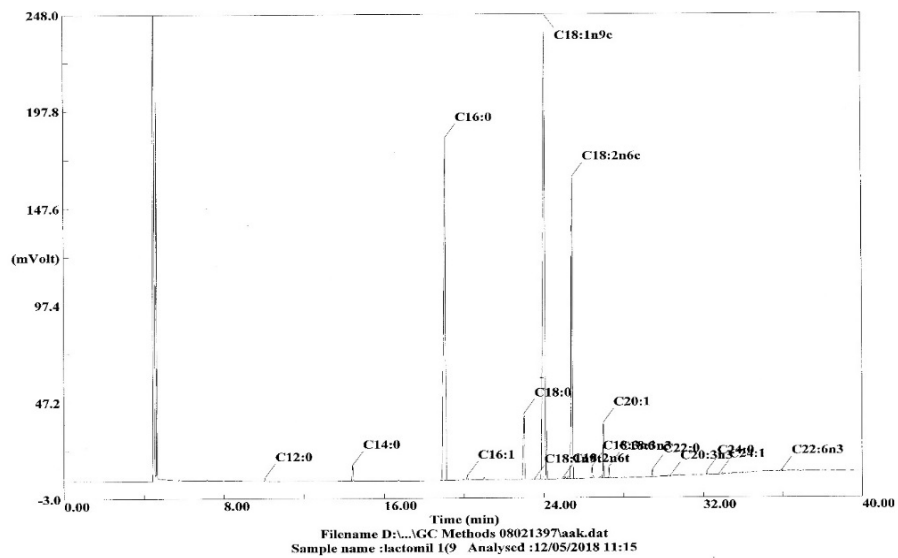


Fig. 4. Chromatogram of fatty acids derived from fat extracted of foreign oil



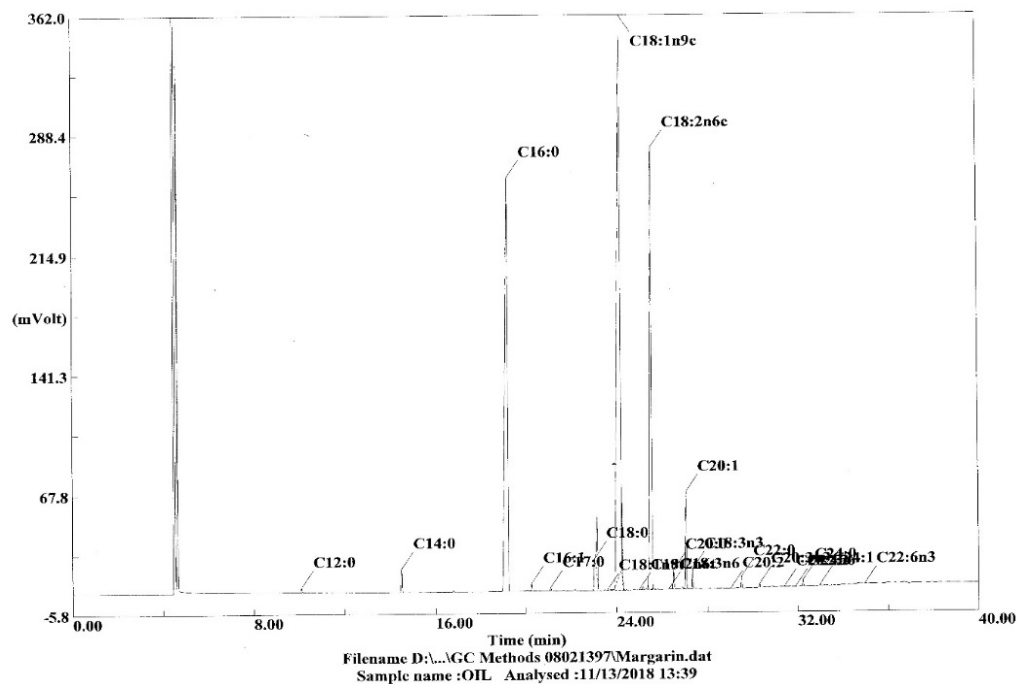


Fig. 5. Chromatogram of fatty acids derived from fat extracted of Iranian Oil Company

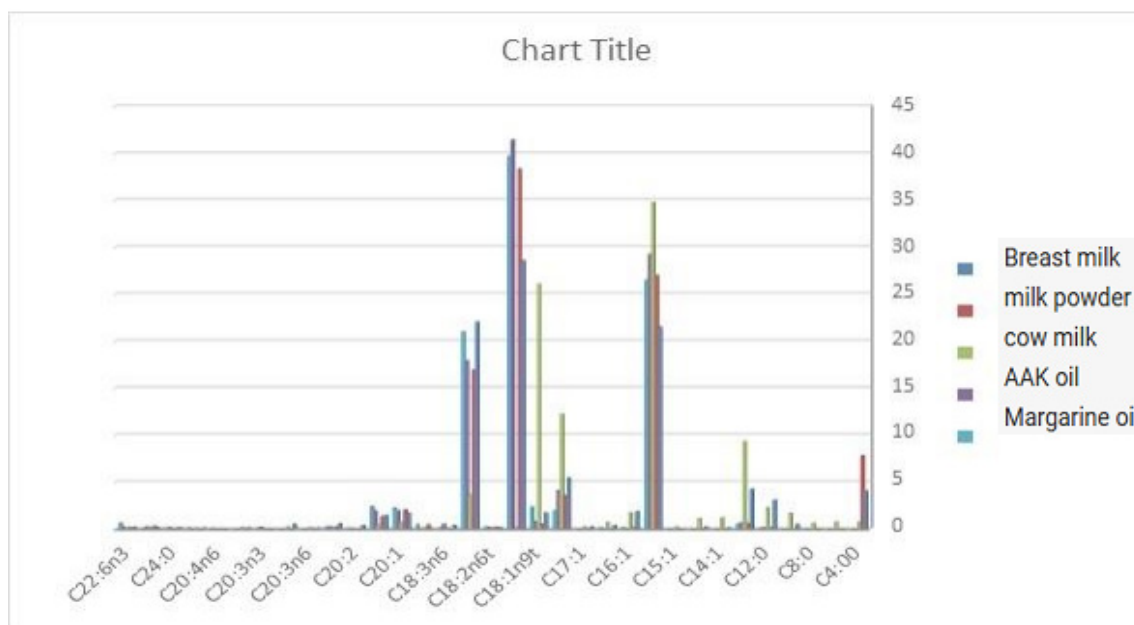


Fig. 6. Comparison of the amount of fatty acids extracted from the samples

**TABLE 2. Quantities of different fatty acid groups in fat extracted from breast milk and infant formula, cow's milk and two samples of infant formula (by percentage)**

Iranian oil	Exterior oil	Cow milk	Milk powder	Human breast milk	Fatty acid methyl ester percentage
0.69	0.77	16.69	8.52	11.99	≤14 C
99.31	99.23	83.31	91.48	88.01	≥16 C
30.07	34.51	64.69	39.93	40.36	Saturated fatty acid
69.93	65.49	35.31	60.07	59.64	Unsaturated fatty acid
2.66	2.2	4.40	2.49	4.17	Monounsaturated fatty acid
67.27	63.29	87.98	57.58	55.47	Polyunsaturated fatty acid
2.6	0.94	26.19	0.76	1.83	Trans fatty acids
8.74	9.72	7.22	12.35	15.41	(Linoleic acid/ linolenic acid)
0.69	0.77	11.66	0.73	7.3	Lauric fatty acid & myristic fatty acid
0.00	0.00	0.21	0.00	0.52	The amount of erolic acid

### Discussion

Considering that the spray dryer temperature is 180 °C and based on the results, it can be concluded that the temperature does not have a significant effect on the vitamins and fatty acid elements and structure and all results are in accordance with the standard. The amount of fat should not be less than 4.4 and more than 6 g/kg of infant formula fed from birth until 6 months of age. In infant formula, total fat content is 5.27 g/100 kcal for infants fed up to 6 months of age, which is 5.77 g/100 kcal.

When vegetable fat replaces milk fat, the fat formula has lower amounts of short fatty acids and higher amounts of C12: 0 and C18: 1, which is more similar to breast milk fat. Human milk fat has a shorter chain fatty acid than cow's milk. According to Table 1 and accordance with the Food Standards for Medicines [5] and National Standard 1-2202 [6], the amount of linoleic acid (C18: 2n6c) (component of omega-6 fatty acids commonly found in vegetable oils such as sunflower, corn and soybean) Should be at least 300 and a maximum of 1,400 mg/kg of breast

milk for infant feeding from birth to 6 months of age. As the fat content of infant formula is 27%, infant formula contains 16.94% fat, which is 893.3 mg/100 calories for infant formula. If this level is 3.59% in cow's milk, since the total fat content of the cow's milk was 3.4% by weight, then 23.93 mg/100 kg of cow's milk is linoleic acid, which is due to the low percentage. It is also unsaturated fatty acid in cow's milk, as well as 27 grams of baby oil added per 100 grams of infant formula, well enough to meet the baby's need. The mean total fat [1] in the three breast milk samples was 3.8%. Given the amount of linoleic acid in the fatty acid profile structure of 22.0578%, this amount equals 163.71 with respect to the amount of fat in the dry matter, equivalent to 1364.25 mg/100 kcal of infant formula fed from birth to 6 It's a month. Alpha-linoleic acid (omega-3 fatty acids), which is essential in addition to fatty acids, plays a vital role in maintaining human health and the human body is unable to produce it, mainly in seeds, nuts, rapeseed oil, flaxseed, Cannabis, and Fatty Fish Found in Human breast milk It should be 0.3 g/100 g of Human breast milk for infants up to 6 months of age [5, 6]. This value in Human

breast milk tested is 1.4567% of total fat in milk the dry weight of the newborn is 1.3705 and its value in cow's milk is 0.5097%.

The ratio of linoleic acid to alpha linolenic acid should be between 5 and 15 [5, 6]. According to Table 1, this ratio is present in breast milk, cow's milk and cow's milk, but in cow's milk, this ratio is within acceptance due to low levels of linoleic acid (3.5874) and alpha linolenic acid (0.5097), but it is very high. Down.

According to Table 2, saturated fatty acid content in cow's milk is 59.78% more than that of mother's milk, while infant formula is not significantly different in milk, and is 39.93%. Trans fatty acids content should not be more than 3% of total fatty acids [5]. In fatty acid profiles of breast milk and dry milk, trans fatty acids content is less than 2%, whereas in cow's milk this amount is more than 26%. Percentage reaches, The amount of erasic acid should not be more than 1% of total fatty acids [5, 6]. This value is 0.52% in mother's milk 0.52% in cow's milk and 0.21% in dry milk.

The amount of lauric and myristic fatty acids alone or in total should be up to 20% of the total fatty acids [5, 6]. This amount was 7.3% in Human breast milk, 0.7% in infant formula and 0.66% in cow's milk.

The process of producing powder products using spray drives as one of the best production methods with minimal changes in the chemical and physical structure of the product. The process of producing powdery products in this way is such that it can be well dissolved in water and with minimal pre-process changes.

All the effective parameters in these products are controllable, including inlet air temperature, outlet air temperature, inlet valve inlet, evaporator concentrated milk inlet, All parameters in the product are analyzed and evaluated, taking into account the quadrants obtained from the average of three replicates. The analysis software used is US-based Design Expert. The results showed that by increasing the inlet drying material, increasing the water

separation capacity of the evaporator system, as well as decreasing the inlet fan diameter to reduce the powder escape from the dryer system, the conversion coefficient decreased and the efficiency increased.

The results of this study showed that temperature had no effect on fatty acid profiles of vegetable oils and infant formula and some minerals and vitamins, and was very similar to breast milk, whereas the cow's fatty acid profiles differ in the amount of saturated and trans fatty acids compared to breast milk, it is clear that whole milk cannot replace breast milk just by diluting it with water and adding a little sugar.

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#### *Conflict of interest*

The authors declare no conflict of interest.

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