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Estimation of the Physicochemical properties in powder Infant formula available in Libyan market

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تقدير الخواص الفيزيوكيميائية لحليب الأطفال المجفف المتوفر في السوق الليبي

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Abstract:

Proper nutrition is essential for promoting an infant's growth and development throughout their first year. Since infants are limited in their food choices, the absence or insufficiency of essential nutrients can impair the body's ability to function optimally. Additionally, the presence of unnecessary components or inappropriate nutrient levels may disrupt physiological processes and metabolic functions (1). This study aimed to assess infant milk formula based on some of its physical characteristics and vitamin E level, comparing results with international standards. Ten samples (0-6 months) were collected from Libyan markets and then analyzed to know the pH and density by BOECO Ultrasonic Milk Analyzer LAC-SP, Titratable acidity (TA) was measured using titrimetric method, whereas high-performance liquid chromatography (HPLC) was used to measure the vitamin E content. The study's findings indicated that pH values in the range of 6.00 to 6.70 were within the normal range except IV, V, VI, and VIII formulas were lower than the normal range, density from 1.0025% to 1.0028% and titratable acidity from 0.12% to 0.25% were within the permissible range. Vitamin E contents from 0.44 to 6.95 mg/100 g within the permissible limits of international standards except I and VII formulas had low contents.

Key words: Infant milk formula, Physical properties, Vitamin E.

الملخص:

التغذية السليمة ضرورية لتعزيز نمو الرضيع وتطوره طوال عامه الأول. ولأن خيارات الرضع الغذائية محدودة، فإن غياب العناصر الغذائية الأساسية أو نقصها قد يُضعف قدرة الجسم على العمل على النحو الأمثل. إضافةً إلى ذلك، قد يُؤدي وجود مكونات غير ضرورية أو مستويات غير مناسبة من العناصر الغذائية إلى خلل في العمليات الفسيولوجية والوظائف الأيضية. هدفت هذه الدراسة إلى تقييم تركيبة حليب الأطفال بناءً على بعض خصائصه الفيزيائية ومستوى فيتامين E ومقارنة النتائج بالمعايير الدولية. تم جمع عشر عينات من عمر (0-6 أشهر) من الأسواق الليبية وتم تحليلها لمعرفة الرقم الهيدروجيني والكثافة بواسطة جهاز تحليل الحليب BOECO LAC-SP وتم قياس حموضة المعايرة (TA) باستخدام طريقة القياس بالمعايرة في حين تم تحليل مستوي فيتامين E من خلال كروماتوغرافيا السائل عالية الأداء (HPLC). أظهرت نتائج هذه الدراسة أن قيم الرقم الهيدروجيني (pH) في حدود (6.00 إلى 6.70) كانت ضمن المعدل الطبيعي باستثناء عينات IV وV وVI وVIII التي كانت أقل من المعدل الطبيعي، وكانت الكثافة (1.0025% إلى 1.0028%) والحموضة القابلة للمعايرة (0.12% إلى 0.25%) ضمن المعدل المسموح به. وكان محتوى فيتامين E من (0.44 إلى 6.95 ملغ/100 غرام) ضمن الحدود المسموح بها للمعايير الدولية باستثناء عينات I وVII التي كانت محتواها منخفضًا.

الكلمات المفتاحية: حليب الأطفال، الخصائص الفيزيائية، فيتامين هـ.

Introduction:

Infant formulae (IF) provide essential nutrients for a child's healthy development and act as the primary alternative or supplement to breast milk [(2)]. Infant formulas are made from modified proteins (often from cow's milk), fats (a blend of vegetable oils such as palm, coconut, and sunflower), carbohydrates (such as lactose), and essential vitamins and minerals that a baby needs for growth. Some formulas also contain additives such as prebiotics and probiotics to support the digestive and immune systems, and essential fatty acids such as omega-3 and omega-6 to support brain and eye development [(3)]. The physical characteristics of milk, including pH, density, and titratable acidity, are key factors in analyzing its physicochemical composition and nutritional properties. Composition of infant milk and processing conditions influence their physicochemical properties, which can further impact caking, Maillard reaction, and release of surface free fat. A comprehensive study of the interplay of macronutrients in infant formula powders during storage is essential to develop formulations that yield products with an extended shelf life [(4)]. As the primary source of nutrition for infants, these formulas must deliver adequate levels of vitamins essential to their diet. Vitamins, being organic compounds necessary for human survival, play a vital role in maintaining life and promoting optimal health. Vitamin E functions as an antioxidant, safeguarding the body's cells from damage inflicted by free radicals, boosting the immune system, maintaining skin and eye health, and supporting nerve and muscle function. α-tocopherol acetate is the most common type of vitamin E found in infant formulas [(5)]. Using infant formula for feeding can offer numerous advantages, but above all, it is essential that the formula meets high-quality standards to sufficiently serve as a substitute for a mother's milk. This demands the finest ingredients and the most rigorous manufacturing processes. The formula must not only provide all the essential nutritional components found in breast milk but also avoid excessive amounts of any of these nutrients. This balance is achieved through precise analysis of adequate samples to guarantee the presence of necessary nutrients and the absence of harmful substances [(6)] .This study examines significant physicochemical parameters, including pH, density, titratable acidity, and vitamin E. Comparing results to a label's values and the guidelines provided in the Codex Alimentarius of Standard Infant Formula[(7)] and the Global Standard for the Composition of Infant Formula[(8)] (the Codex Alimentarius of Standard Infant Formula agrees with the Global Standard for the Composition of Infant Formula).

MATERIALS AND METHODS:

Samples:

Ten cow's milk-based, powdered infant formulas for newborns up to six months were sourced from Libyan pharmacies, selected based on market availability. These samples are as displayed in the table (1).

Table (1): Samples names for infant formulas

Sample No	Manufactured
I	Switzerland
II	Belgium
III	Europe
IV	Belgium
V	Switzerland
VI	Holland
VII	France
VIII	France
IX	Switzerland
X	France

Chemical analysis:

Determination of pH and Density:

pH and density were identified by the BOECO Ultrasonic Milk Analyzer LAC-SP. Dissolve 10 grams of infant formula sample in 50 mL deionized water. Transfer the prepared sample into the analyzer's sample holder and insert the sample holder into the analyzer's recess.

Measurment of Titratable Acidity (TA):

It entails using 0.1N sodium hydroxide NaOH to titrate 9.0 mL of dissolved infant formula sample while phenolphthalein is used as an indicator. The following formula was used to determine the total acidity that was obtained. The percentage of lactic acid was used to express it.

lactic acid= (Vg×100× 0.009)/Vm %

where Vg is the NaOH solution's additional volume (mL); Vm is the amount of milk utilized for titration (mL) and 0.009 is Lactic acid normality equivalent.

Determination of Vitamin E by HPLC:

Vitamin E was determined using the HPLC using the Thermo system (Ultimate 3000). Weigh 10 g of milk powder into a 250 mL saponification flask. Add 15 g L- ascorbic acid and 1 L ethanol into the flask, and mix the solution well. Add 25 mL of 50% potassium hydroxide solution. Thoroughly mix, and use a stopper to seal the flask. Heat the flask at 53°C, keeping it in a water bath, and continue heating for 45 minutes. Take the flask out and let it cool to room temperature. Take the saponified sample into the separating funnel. Wash the flask with 100 ml ethanol and transfer the washings into the funnel .Wash the flask with DI water and transfer the washings into the funnel, close the funnel, and shake at room temperature for 2 minutes. Add 50 ml n-hexane and shake for 10 minutes. Let the funnel stand until the hexane layer is separated, discard the lower water layer. Add another 50 ml of hexane and shake the funnel. Repeat the extraction process as before. Take about 10 g of anhydrous sodium sulfate on the filter paper, this filtration is done to remove the water. Keep the extract at 37- 40°C until the hexane is evaporated completely. Add 10 ml methanol into the beaker. Take 1 ml of solution into an amber colored injection vial [9].

Statistical Analysis:

Measurements were taken three times for each sample. The results were then processed using Microsoft Excel to calculate the average value and the variability (standard deviation) for each sample. The final results were presented as the mean values ± standard deviation.

Results and Discussion:

pH quantifies the concentration of free H+ ions resulting from the dissociation of acids in milk [10]. Table (2) and Figure (1) show that pH values of infant formulas ranged from 6.00 to 6.70. The pH values were slightly lower than observed by *Sunaric et al*, from 6.52 to 6.90[10]. pH of cow's milk is commonly in the range of 6.5 and 6.7[11]. All samples recorded pH values within the normal range except IV, V, VI and VIII formulas, were lower than the normal range, the reason may attributed to the lipolysis which leads to the release of the free fatty acids [10].

Table (2): pH values of infant formulas

Sample No	*(Mean ± STDEVA)
I	6.60±1.088E-15
II	6.60±1.088E-15
III	6.50±0
IV	6.40±1.088E-15
V	6.00±0
VI	6.31±0
VII	6.47±0.058
VIII	6.30±0
IX	6.70±0
X	6.60±1.088E-15

*Means are values derived from triplicate determinations, given as mean ± standard deviation.

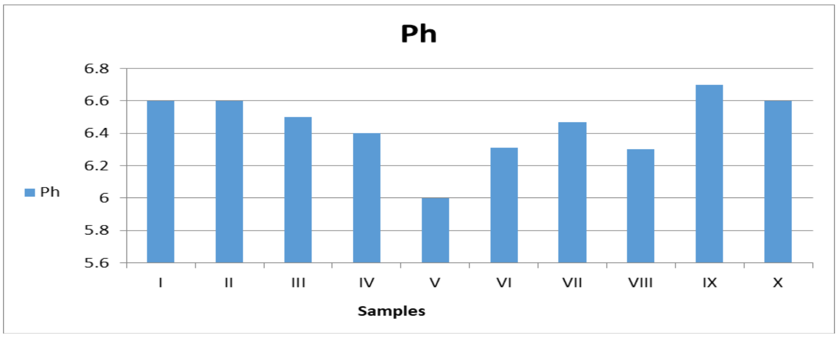

Figure (1): pH values of infant formulas

Table (2) and Figure (3) indicate that the infant formulas density values were in the range of 1.0025% to 1.0028 g/cm³. One of milk powder's most complicated characteristics is its bulk density, which is important for both pragmatic and economic reasons. A high bulk density is preferred in order to save money transportation and packing costs. However, low bulk density, which is present in agglomerated materials, affects other powder qualities like flowability and instant characteristics (12). All samples recorded values of density within the normal range 1.0025 to 1.0035 g/cm^3.

Table (3): Density values of infant formulas

Sample No	(Mean ± STDEVA) g/cm^3
I	1.0028±0
II	1.0027±5.77E-5
III	1.0027±0
IV	1.0026±5.77E-5
V	1.0028±5.77E-5
VI	1.0026±0
VII	1.0026±0
VIII	1.0026±5.77E-5
IX	1.0028±0
X	1.0025±0

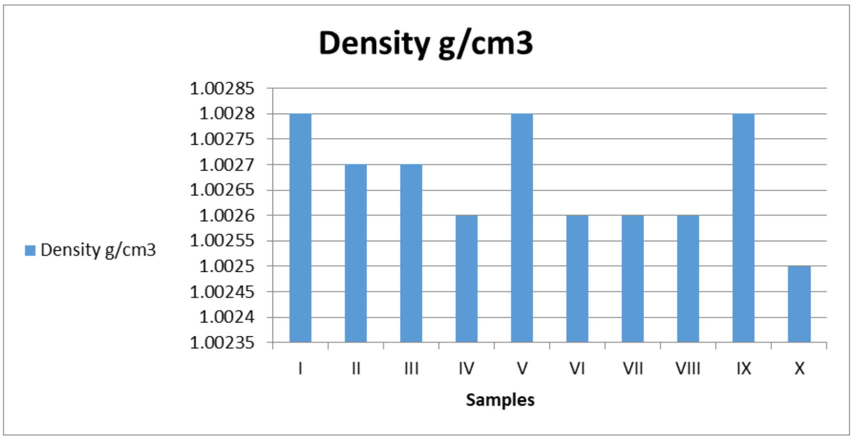

Figure (2): Density values of infant formulas

Titratable acidity (TA), since it measures the total acid content, is a parameter that can be used to express natural acidity and buffering capability (10). Table (4) and Figure (3) show the percentage of acidity based on lactic acid of infant formula ranged from 0.12% to 0.25%. Formula II had the highest acidity 0.25%, whereas VI and VIII formulas had the lowest acidity 0.12%. A different study found that the average lactic acid acidity percentage was 0.0608%, which is less than the current study's (10). According to the current study, the infant formula's acidity is less than 1.5%, which satisfies USA criteria (13).

Table (4): The percentage of titratable acidity of infant formulas

Sample No	(Mean ± STDEVA)%
I	0.21±0.006
II	0.25±0.006
III	0.19±0.011
IV	0.16±0.011
V	0.16±0.015
VI	0.12±0
VII	0.17±0.006
VIII	0.12±0.006
IX	0.15±0.015
X	0.15±0.011

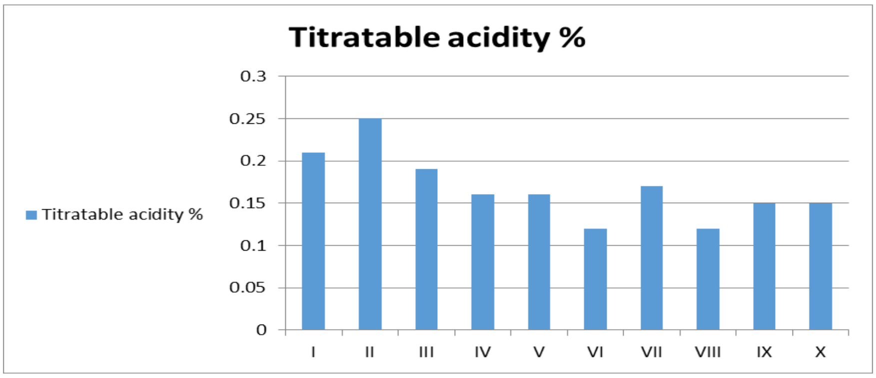

Figure (3): The percentage of titratable acidity of infant formulas

Vitamin E acts as a powerful antioxidant for infants, protecting their cells from damage. Its main function is to support the immune system and strengthen immunity in infants [5]. Table (5) and Figure (4) show that the vitamin E contents of infant formulas ranged from 0.44 to 6.95 mg/100 g. Formulae VI and II had the highest amounts of vitamin E (6.95 mg/100 g and 5.81 mg/100 g, respectively), whereas formulae VII and I had the lowest amounts (0.44 mg/100 g and 2.69 mg/100 g, respectively). All formulas had vitamin E levels lower than those declared on the label. Vitamin E contents of infant formulas set in Codex Alimentarius range between 0.5-5 mg/100 kcal, about 3.33-33.3 mg/100 g. All samples in this study had vitamin E contents meet Codex Alimentarius requirements except I and VII formulas, which had low contents.

A study by *Feras et al*, reported that vitamin E contents from 0.33 to 4.12 mg/100g no significant differences observed with our study. Since human milk has a sufficient amount of vitamin E (5.4 mg/ml) for infants, while cow milk has a low amount (1.1 mg/ml), it is added to infant formulas to boost vitamin content and reduce lipid peroxidation, hence extending the product's shelf life [14]. The notable discrepancies in vitamin E content relative to the specified values may result from an error in the fortification process or inaccurately labeled values. Companies typically report the added amount of each vitamin rather than the total content. Also, differences in fortification procedures may have a large impact upon vitamin content, and the amount of vitamin E is reduced when fat is removed from cow's milk[15]. Iron and polyunsaturated fatty acid (PUFA) concentrations may also be linked to decreased vitamin E levels in newborns. Therefore, a given formula fortified with PUFAs and iron may have reduced vitamin E levels [16].

Table (5): Vitamin E contents of Infant formulas as compared to label value

Sample No	Vitamin E mg/100g	Label value mg/100g
I	2.69	8.0
II	5.81	8.2
III	4.18	8.2
IV	3.58	5.0
V	3.85	8.0
VI	6.95	10.0
VII	0.44	10.0
VIII	3.07	8.5
IX	4.85	8.0
X	4.63	8.0

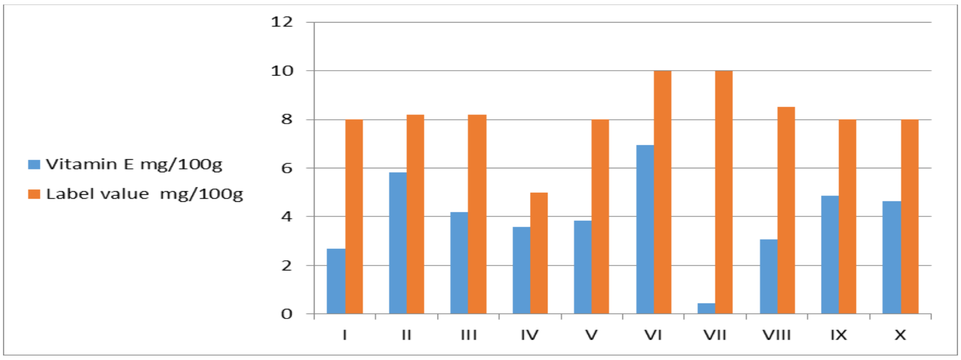

Figure (4): Comparison between vitamin E contents of infant formula and label value

Conclusions:

Infant health plays a fundamental role in every healthcare organization, serving as a critical focus area. Evaluating nutritional status is essential to safeguard the well-being of our infants. Fortunately, all brands in this study were safe, Inconsistencies existed between the concentration of infant formula's chemical composition and the values claimed on the manufacturers' labels, which did not affect the total result. pH values were within the normal range except IV, V, VI and VIII formulas were lower than the normal range. The acidity and density were within the permissible range. Vitamin E contents were within Codex Alimentarius requirements except I and VII formulas showed low contents. Nutrition analysis is a critical step for infant formula, and quality control. Additional research is necessary to assess the infant formula's chemical composition over a larger variety of brands and more samples to verify the correctness of the contents stated on their labels.

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