

Nutritional value of spirulina and its use in the preparation of some complementary baby food formulas

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Abstract

In this study use the spirulina which is one of the blue-green algae rich in protein 62.84% and contains a high proportion of essential amino acids (38.46% of the protein) and a source of naturally rich in vitamins especially vitamin B complex such as vitamin B12 (175 µg / 10 g) and folic acid (9.92 mg / 100 g), which helps the growth and nutrition of the child brain, also rich in calcium and iron it containing (922.28 and 273.2 mg / 100 g, respectively) to protect against osteoporosis and blood diseases as well as a high percentage of natural fibers. So, the spirulina is useful and necessary for the growth of infants and very suitable for children, especially in the growth phase, the elderly and the visually appetite. It also, helps a lot in cases of general weakness, anemia and chronic constipation. Spirulina contain an selenium element (0.0393 mg/100 g) and many of the phytopigments such as chlorophyll and phycocyanin (1.56% and 14.647%), and those seen as a powerful antioxidant. Finally, spirulina called the ideal food for mankind and the World Health Organization considered its "super food" and the best food for the future because of its nutritional value is very high. The American space agency is working on a project to be grown in space and regards it as the main food for astronauts. All this and more is what makes the best food spirulina exists on the ground. It ensures the whole food and alkaline balance of the body. Sixteen food formulas were prepared for as complementary food babies (1-3 years age) by using spirulina at 0, 2.5 0.5 and 7.5% for the production of two types of baby food one of them is ready to eat by using some fruits and vegetables. Papaya fruits with good nutritional values and cheap price as an essential ingredient of 30% in the four formulas and banana fruit which rich in potassium in four formulas addition to potatoes purée and carrot purée by adding 10% for each and apple purée, guava puree and mango juice by adding 15% for each been mobilized mixes in jars glass and thermal treatment was carried out at 100 °C for 40 minutes. The second type of baby foods formulas was production by using cereals, legumes and some dried green vegetables, where it was manufactured 8 dried formulas four of them by 30% wheat flour 72% and four others by 30% milled rice in addition to the 30% crushed pearl barley and dryer lentils and dried spinach dried cauliflower by adding 10% for each formulas. After produced formulass were packaged in bottles court lock. Then, evaluated all formulas microbiologically to study its safety before sensory evaluation and found to be microbiology safe. Sensory evaluation of produced formulas were acceptable sensory significantly. After that, chosen 4 formulas containing 5% spirulina based on the results of sensory evaluation was conducted analysis chemotherapy and natural for these selected formulas. The chemical composition indicated that these formulas were suitable as a food supplement for children aged 1-3 years. On the other hand, these formulas were economic cost and can produced on the scale of domestic and industrial scale, as well as can be exported to the outside.

Keywords: Spirulina; nutritional value; chemical composition; amino acids, fatty acids, vitamins, phytopigments, minerals; microbiological examination; food formulas; baby foods.

1. Introduction

Spirulina is the dried biomass of the cyanobacterium *Arthrospira platensis*, it has been widely used in several countries, it is considered GRAS (generally recognized as safe), without toxicological effects, and it is approved by the FDA (U.S.A.) and ANVISA [32]. Rich in protein (up to 65%), spirulina is considered safe for consumption by humans and animals, and it has been cultivated and used as a food source worldwide. The U.S. Food and Drug Administration [18] has not questioned the basis for the Generally Recognized as Safe designation to spirulina under the conditions of its intended use, thus restricting it as a food additive in amounts that range from 0.5 to 3.0 grams per serving. Formulators use spirulina in specialty food bars, powdered nutritional drinks, popcorn, beverages, fruit and fruit juices, frozen desserts and condiments.

Microalgae have received increasing attention due to the fact that they represent one of the most promising sources of compounds with biological activity that could be used as functional ingredients. Their balanced chemical composition (good quality proteins, balanced fatty acid profiles, vitamins, antioxidants and minerals) and their interesting attributes can be applied in the formulation of novel food products [51].

Spirulina, filamentous blue-green microalgae or cyanobacteria, is well known as a source of protein (60-70 g/100 g) of high biological value, since it is a rich source of vitamins, mainly vitamin B12 and pro-vitamin A, minerals, especially iron, and γ -linolenic acid, essential fatty acids precursor for prostaglandins [21]. Furthermore, spirulina contains such molecules as phycocyanin, β -carotene and xanthophyll pigments, α -tocopherol and phenolic compounds, which are responsible for the antioxidant activities of these microalgae, as shown by several authors for in vitro and in vivo experiments [35]. Moreover, most research has focused on the health effects of spirulina as a dietary supplement for humans and animals. Many studies have shown the effects of these microalgae that may result in significant therapeutic applications: an anti-cancer effect [29], a hypolipidemic effect [31], and a protective effect

against diabetes and obesity [3]. These advantages make spirulina a good raw material for the healthy food.

Spirulina has no cellulose in its cell walls, being composed of soft mucopolysaccharides. This makes it easily digested and assimilated. It is 85 to 95% digestible. This easy digestibility is especially important for people suffering from intestinal malabsorption. Typically, many older people have difficulty digesting complex proteins, and are on restricted diets. They find spirulina's protein easy to digest. Spirulina is effective for victims of malnutrition diseases like kwashiorkor, where the ability of intestinal absorption has been damaged. Given to malnourished children, it is more effective than milk powders because milk's lactic acid can be difficult to absorb Kelly *et al.* (2011), Parry (2014) and Robert (2010) [25, 34, 39].

Spirulina can be used at any age (from infancy to pregnancy and adulthood), but its value is particularly evident in the young growing child: during weaning and during the pre-school period (from 1 to 6 years), Dillon (2014) [12].

Spirulina offers remarkable health benefits to an undernourished children. It is rich in beta-carotene that can overcome eye problems caused by vitamin A deficiency, it provides the daily dietary requirement of beta-carotene which can help prevent blindness and eye diseases Seshadri (1993) [42]. The protein and B-vitamin complex makes a major nutritional improvement in an infant's diet. It is the only food source other than breast milk containing substantial amounts of essential fatty acid, essential amino acids and GLA that helps to regulate the entire hormone system Ramesh *et al.* (2013) [37].

Simpore *et al.* (2005 and 2006) [44,45] suggested that spirulina may be a good food supplement for undernourished children. In particular, spirulina also seems to correct anemia and weight loss in HIV-infected children, and even more quickly in HIV-negative undernourished children.

Spirulina's concentrated nutrition makes it an ideal food supplement for people of all ages and lifestyles. Spirulina is about sixty percent complete, highly digestible protein. Spirulina contains every essential amino acids. It contains more beta-carotene than any other whole food; it is the best whole food source of

gamma linolenic acid (GLA); it is rich in B vitamins, minerals, trace elements, chlorophyll, and enzymes; and it is abundant in other valuable nutrients about which scientists are learning more each year, such as carotenoids, sulfolipids, glycolipids, phycocyanin, superoxide dismutase, RNA and DNA [34].

The weaning period is the most critical phase of infant's life. During this period mother's milk is not generally adequate to cover the nutritional requirements and support body growth. With increasing the numbers of working mothers in the developing countries, the market of baby foods has been increased tremendously [2]. Breast milk alone cannot support the nutrition and other needs of the growing infant [15]. There comes a time when complementary weaning foods must be introduced into the diet to fill the gap between what is provided by milk and what the infant requires to cover his nutritional requirements [11]. In the case of baby food products, mothers often try the product first and decide whether to give it to their children or not [17,19].

In most developing countries commercial weaning foods of excellent quality either imported or locally produced are presently available, but due to sophisticate processing, expensive packing, extensive promotion and solid profit margins, the price of these commercial products are generally in the order of 10-15 times the cost of the common staple foods. While these products are generally highly appreciated and their use and value are well understood, they are priced beyond the purchasing power of the majority of population in the lower income groups, Who spent already about 50-75% of their income in common foods [56].

Spirulina platensis is used in the food, medicine, and cosmetic industries, and as an additive for chips, fruit juices, sauces, spice mixtures, vegetables, soups, and other products. This investigation contributes to the determination of nutrients in *spirulina platensis* microalgae used in the food and aquaculture feed industries.

Three to ten grams a day delivers impressive amounts of beta carotene, vitamin B-12 and B complex, iron, essential trace minerals, and gammalinolenic acid. Beyond vitamins and minerals, *spirulina* is rich in phytonutrients and

functional nutrients that demonstrate a positive effect on health Robert (2010) [39].

It is legally approved as a food or food supplement in Europe, Japan and many other countries around the globe. The United States Food and Drug Administration confirmed in 1981 that *spirulina* is a source of protein and contains various vitamins and minerals and may be legally marketed as a food supplement. Many countries have set up food quality and safety standards for *spirulina*, FDA (1981) [18].

Therefore, owing to all these advantages, the present work aims to study the physicochemical and nutrition values of *spirulina*, and formulate different babies food formulas to use as a complementary for baby food formulas with lower cost. Also, to evaluate the formulas from standpoint of organolytically, physicochemical properties of the best formulas were evaluated.

2. Materials and methods

Spirulina was obtained from Aquaculture Research Center at Arab Academy for Science, Technology & Maritime Transport, Arab League.

Papaya (*Carica papaya* L.) was obtained from the farm of Horticulture department, Fac. of Agric. Moshtohor. Banana (*Musa sapientum* L.), Potato (*Solanum tuberosum* L.), carrot (*Daucus carota* L.), Anna apple (*Malus sylvestris* L.), guava (*Psidium guajava* L.) and mango (*Mangifera indica* L.), were purchased from certain farmers at Kaha city area, Qaliuobia Governorate, Egypt and immediately transported to the laboratory. Sugar was purchased from local market in Qaliuobia Governorate, Egypt.

Wheat (*Triticum* species) Wheat flour (72% ext.) was obtained from El- Mokhtar Mill, Cairo governorate, Egypt. Rice (*Oryza sativa*), Barley (*Hordeum vulgare vulgare* L.) lentil (*Lens culinaris*), chickpea (*Cicer arietinum*), peas (*Pisum sativum*), Spinach (*Spinacia oleracea*) and cauliflower (*Brassica oleracea botrytis*) were purchased from local market in Qaliuobia Governorate, Egypt.

Preparation of raw materials:

Preparation of raw materials used in formulation of spirulina with some fruits and vegetables-based baby food formulas: banana, papaya, apple, mango, guava and potato, carrot, were washed with tap water, to remove dirt, adhering latex and other foreign

matters, as well as to reduce the initial contamination with microorganisms, then papaya and banana fruits were hand peeled, papaya seeds were carefully removed and the fruits were cut into small parts. While, carrot was peeled using stainless steel peeler, the stones of mango were removed after cutting the fruits to two halves.

After that, all fruits and vegetables were blanched by using a pressure cooker where the blanching time was adjusted to be proper for each material. Potato was peeled by hand after blanching. The blanched materials were transferred to Moulinex blender (Blender Mixer, type: 741) equipped with cutters and stirrer which crushed and homogenized each of above mentioned materials into a mixture of pulp, then the mixture was passed through fine strainer to separate the pulp from any skin or seeds and then it was packed in plastic bags, sealed and frozen [20].

Preparation of raw materials used in formulation of dried spirulina with cereals-based baby food formulas: dry cereals and legumes were cleaned from impurities and then washed thoroughly with tap water, the washed cereals and legumes were separately soaked in tap water overnight, except rice was soaked for 30 minutes, according to Soliman *et al.* (1996) [50].

- The peeled chickpea, lentil, dry pea, rice and wheat were cooked separately in a pressure cooker for 5 to 10 minutes. After cooking, the remaining water was eliminated. After that, the cooked materials were dried in solar dryer at 45-60°C. Dried cereals and legumes were milled in an electrical mill, then sieved through a silk sieve (60 mesh) according to Soliman *et al.* (1996) [50].
- Spinach and cauliflower were sorted and prepared (green leaves of cauliflower were removed then edible part was cut), the prepared vegetables were washed and blanched for appropriate time (4 to 5 min) using live steam blancher then cooled down using cold water and were dried at 60 °C for 12 hrs. in an electric oven drier and ground to a particle size of 500–600 µm. to pass through 60 mesh sieve.

- All prepared materials were bottled in glass jars and stored at room temperature until using in preparation baby food formulas.

Preparation of formulated baby food formulas:

Sixteen baby food formulas were prepared as shown in Tables (A and B). The spirulina was added to the components by 0, 2.5, 5 and 7.5% to the different formulas.

To calculate those formulas, we took into account the needs of children between 1 to 3 years as defined by WHO. We also chose ingredients that are available in Egypt. We also, included results from sensory tests preliminary experiments we did with mothers, children and adults, these tests showed that formulas containing materials were accepted also indicated that those parts of materials were the best ratios.

Table (A) shows 8 prepared spirulina with some fruits and vegetables-based baby food formulas. After mixing the ingredients of the formulas, they were bottled in tight jars, and then thermally processed at 100° C for 40 min according to Soliman *et al.*, (2003) [50], Bahlol *et al.* (2007) [7] and Satter *et al.* (2013) [41]. Table (B) shows 8 prepared dried spirulina with cereals-based baby food formulas. After mixing the ingredients of the formulas were bottled in tight jars.

Methods:

Physicochemical analysis:

Moisture content, total solids, ash, fat, protein, ascorbic acid and starch were determined according to AOAC (2000) [4]. The pH value was measured with a pH meter model Consort pH meter P107. Titratable acidity was determined by titration with NaOH 0.1 N solution using phenolphthalein as indicator according to AOAC (2000) [4]. Total and reducing sugars were determined by Shaffer and Hartman method as described in the AOAC (2000) [4]. Total pectin content and fractional pectin components were determined by the method of Robertson (1979) [40]. Crude fiber was determined by Weende method in which VELP Scientifica extraction unit was used. The method is based on the solubilization of non-cellulosic compounds by sulfuric acid and hydroxide solutions as described in AOAC (2000) [4].

Total carotenoids were determined according to Harvey and Catherine (1982) [23]. Total anthocyanins was measured according to the method of Skalaki and Sistrunk (1973) [46]. Three replications of all these determinations were carried out. Carbohydrates were determined by difference from the total of dietary fibre, lipids, protein and ash contents [43].

Determination of total energy: The total energy value of the food formulation was calculated according to Sharoba *et al.* (2013) [43] using the formula as shown in the following equation:

$$\text{Total energy (kcal/100 g)} = [(\% \text{ available carbohydrates} \times 4) + (\% \text{ protein} \times 4) + (\% \text{ fat} \times 9)]$$

Amino acid analysis: The protein quantification was done with micro-Kjeldahl method. Amino acid analysis procedure involves acid/alkaline hydrolysis, separation by cation exchange column, post-column derivatization with Ninhydrin and detection using UV/Vis detector at 570 nm as described in the Korean Food Code (KFDA, 2003) [26]. These procedures in the Korean Food Code were established based on AOAC Official Method 960.52 [4] for micro-chemical determination of nitrogen (micro-Kjeldahl), AOAC Official Method 988.15 [4] for tryptophan, AOAC Official Method 985.28 [4] for sulfur amino acids, and AOAC Official Method 994.12 [4] for acid-stable amino acids. Acid (HCl) hydrolysis method was used for aspartic acid (Asp), threonine (Thr), serine (Ser), glutamic acid (Glu), proline (Pro), glycine (Gly), alanine (Ala), valine (Val), isoleucine (Ile), leucine (Leu), tyrosine (Tyr), phenylalanine (Phe), histidine (His), lysine (Lys), and arginine (Arg). And, performic acid hydrolysis method was used for sulfur-containing amino acid such as cystine (Cys) and methionine (Met), while alkaline (NaOH) hydrolysis method was used for tryptophan (Trp). After hydrolysis, amino acid analyzer (Sykam GmbH, Germany) with an integrator (Axxiom Chromatography Inc.) was used for quantification of amino acids. All results are expressed on the basis of 100 g edible portion. HPLC analysis was performed on the same day as extractions.

Total lipid and fatty acids composition analysis: The fatty acid profile was analyzed using a Gas-Chromatographic model GC-17A according to

AOAC (2000) [4]. Three samples of the oils which extracted from spirulina sample was converted to their corresponding methyl ester using boron trifluoride methanol complex (14% w/v). The mixture was maintained at 100 °C for 1 h.

The reaction was stopped with 0.5 ml of distilled water. Then, the extracted fatty acid methyl esters (FAMES) were dissolved in heptane for GC analyses. GC analyses were performed on a Hewlett–Packard 5890 Series II gas chromatograph (H.P. Co., Amsterdam, The Netherlands) equipped with a hydrogen flame ionization detector and a capillary column: HP Inovax cross-linked PEG (30 m x 0.32 mm x 0.25 lm film). The column temperature was programmed from 180 to 240 °C at 5 °C/min and the injector and detector temperature was set at 250 °C. Nitrogen was the carrier gas. FAMES were identified by comparison of their retention time with respect to pure standard FAMES purchased from Sigma and analyzed under the same conditions. Date seed FAMES were quantified according to their percentage area, obtained by integration of the peaks. The results were expressed as a percentage of individual fatty acids in the lipid fraction.

Minerals content: Minerals content were determined according to AOAC (2000) [4] using Perkin-elmer, 2380 Atomic absorption spectroscopy apparatus in central laboratory of Veterinary Faculty, Moshtohor according to AOAC (2000) [4] official method 985.01. Meanwhile phosphorus was determined by the official spectrophotometric method of the AOAC (2000) [4] using UV/visible automatic scanning spectrophotometer.

Vitamins Assay: Vitamin C was determined in all samples by dichlorophenol Indophenol dye reduction method [4].

Thiamine, Riboflavin, Niacin, Pyridoxine, Analogue, folic acid, inositol, vitamin E, vitamin K, Pantothenate and biotin were determined by the HPLC system method according to AOAC (2000) [4].

Determination of vitamin A: About 10 g of sample was homo-genized, weighed and transferred into a ground bottom flask, 30 ml of extraction solution, 0.1% antioxidant and few drops of KOH were added and reflux for 30 min at 70°C. The sample was cool down, vitamin A was extracted into hexane, and the combined hexane extract was washed with water and

then dried the hexane layer to about 2 ml on a water bath or rotary evaporator. The final volume was made up to 50 ml with the mobile phase. The mobile phase, standard and sample were filtered through 0.45 μ membrane filter and were degassed before injection.

Calibration curve was made by a standard in mobile phase with five point calibrations and analyzed independently by HPLC. A standard curve was plotted between the concentration of vitamin A and peak area obtained. For HPLC analysis, an Eelipse \times BD – C18 column (4.6 \times 250 mm 5 μ m) was used with a linear gradient of methanol: water (95:5) at constant flow rate of 1 ml /min by using a binary pump with column temperature 40°C. A multiple wavelength detector was employed for the detection of peaks using a wavelength of 325 nm and a bandwidth of 8 nm.

Phytopigments Assay: Some phytopigments content were determined according to AOAC (2000) [4] by using HPLC.

All other chemicals were obtained from Merck (Darmstadt, Germany) or Riedel-de Haen (Seelze, Germany) as HPLC-grade. All standard materials were purchased from Merck (Darmstadt, Germany) or Sigma-Aldrich Chemicals Co.(Steinheim, Germany).

Microbiological examination: Total viable bacterial count, mesophilic sporeformers bacteria, yeasts and moulds, coliform group were enumerated and the presence of (*Salmonella* spp. and *Staphylococcus aureus*) was detected according to the methods established by APHA (1992) [5] and Kang *et al.* (2003) [24]. Results were expressed as CFU g⁻¹.

Rodent hairs and Insect fragments in spirulina: Rodent hairs and insect fragments in spirulina were determined according Thind (2000) [52].

Analytical methods for heavy metals in spirulina: The determination of arsenic, cadmium and lead in spirulina sample were performed according to the methods described in the Korean Food Code (KFDA [26]) which described by Haeng-Shin *et al.* (2006) [22] by inductively coupled plasma–emission spectrometry (Model JY 38 S; Horiba, Jobin Yvon Cedex, France).

Duplicate samples were run in triplicates for the analysis of each heavy metal.

Bulk density of spirulina: Bulk density (Kg/lit) was determined by gently pouring 2 g of spirulina into an empty 10 ml graduated cylinder and holding the cylinder and tapping 10 times on a rubber mat from a height of 15 cm. The ratio of the mass of the powder and the volume occupied in the cylinder was determines the bulk density.

Sensory evaluation: Sensory evaluation was carried out by a properly well trained panel of 12 testers. They were selected if their individual scores in 10 different tests showed a reproducibility of 90%. The 12 member internal panel evaluated the different baby food formulas for color, taste, odor, texture, mouthfeel (smoothness, consistency, spreadability) and overall acceptability. Mineral water was used by the panellists to rinse the mouth between samples. Scoring was based on a 100 point scale (10-100) where (90-100) = excellent, (70-80) = very good, (50-60) = good, (30-40) = fair and (10-20) = poor.

Statistical analysis: Data of chemical composition of ingredients and formulas were expressed as mean of three replicates \pm standard error (SE). Data for the sensory evaluation of all baby food formulas were subjected to the analysis of variance followed by multiple comparison using LSD [48].

3.Results and discussion

Chemical composition and nutrition values of Spirulina: The Chemical and nutritional composition of spirulina may vary according to the growing conditions. For example, the iodine content will vary as a function when the spirulina is grown in sea water vs. fresh water. The Chemical and nutritional composition of dried powdered spirulina grown in fresh water is summarized in Tables (1, 2 and 3). It should be noted that, the cell wall of spirulina is composed of protein, carbohydrates and fat. Therefore, the bioavailability of nutrients from spirulina might be more than from other food sources, especially plant food sources.

Spirulina is the richest nutrient and complete food source found in the world. It contains over 100 nutrients, more than any other plants, grains or herbs. Today Spirulina is widely used as a food supplement to maintain health, boost energy and reduce weight.

Spirulina contains 62.84 % protein, higher than any other natural food. Spirulina contains all the essential amino acids in fairly high amounts, Spirulina is just that, a complete protein, other protein sources have very negative properties as well, such as animal fat and cholesterol. Spirulina contains essence minerals like calcium, magnesium, potassium, phosphorus, iron, and zinc as well as complete vitamin B groups and many important anti-oxidants (which protect cells). The anti-oxidant phycocyanin can only be found in spirulina. It is the richest natural source of vitamin E and beta-carotene. The results of chemical composition of spirulina are in agreement with those obtained by Branger *et al.* (2003) [10]; Habib *et al.* (2008) [21]; Vijayarani *et al.* (2012) [53] and Dolly (2014) [13]. The protein and B-vitamin complex in spirulina makes a major nutritional improvement in an infant's diet.

It is the only food source other than breast milk containing substantial amounts of essential fatty acids, essential amino acids and GLA that helps to regulate the entire hormone system.

Physical properties of spirulina: Spirulina offers a convenient solution to the pH problems of most diets as it is very alkaline. Because spirulina is an alkaline food (pH 6.84) that counter the acidic foods and help raise the pH level towards the alkaline side of the scale. This, in turn, promotes increased bone mass (since your body doesn't have to sacrifice calcium to balances its pH), and vastly improved metabolic functions. Consuming more alkaline foods has been strongly linked with improved immune system function, mental function, kidney function, and higher levels of energy, among other important benefits. Acidic body condition may cause many modern diseases like hypertension, cancer, diabetes, heart disease, gout and rheumatism.

Adjust the body's pH value: The ideal healthy human body's pH level should remain on low alkaline about pH 7.35~7.45. Modern day people indulge in too much acidic food like soft drinks, meat, cheese, eggs, and ham. These cause our body to become acidic (pH < 7).

Many medical research reports have proven that acidic bodies will have more chance of getting diseases or cancer.

Regular use of Spirulina can help keep your body alkaline will help you reduce this risk and is the ideal food supplement for the weight reducer.

Data in Table (1) also showed the bulk density of spirulina (0.82 Kg/lit), the bulk density of the product is affected by particle size distribution, type of agglomeration, particle porosity, and to a certain extent the moisture content. Particle size distribution is affected by the initial size of the trichomes as they are fed to the dryer and the pore diameter of the atomizer. The final quality of the product with respect to bulk density is therefore dependent on culturing, harvesting and drying conditions. To a certain extent, all these factors are harnessed in order to obtain a product that meets the requirements of formulated babies food formulas. The color of spirulina in the powder form appears a blue green to green color.

Finally Spirulina called a superfood because its nutrient profile is more potent than any other food, such as plants, grains or herbs. These nutrients and phytonutrients make spirulina a whole food alternative to isolated vitamin supplements. Protein and amino acids, vitamins and minerals, essential fatty acids and phytonutrients, comparing with other foods. Spirulina can renourish our bodies and renew our health. Spirulina has been used in preparation baby foods because of its therapeutic properties and the presence of antioxidant compounds. Babies can eat spirulina in complete safety and assimilate its nutrients without difficulty. Even malnourished babies with diminished capacity for nutrient absorption could assimilate spirulina and recover from malnutrition.

Microbiological quality and contaminant specifications of Spirulina:

Microbiological quality of Spirulina: The total viable bacterial count is widely used as an indicator microbiological quality of food. Data in Table (4) indicated that, the total viable bacterial count and mesophilic spore formers bacteria were cannot be detected. This is more acceptable for prepared food product especially baby foods. Yeast and moulds cannot be detected, this may that yeast and moulds cannot resist for drying. Count of pathogenic bacteria took the same trend of total viable bacterial count. Coliform group, salmonella and staphylococcus were not detected.

Table (A). Formulated of prepared spirulina with some fruits and vegetablesbased baby food formulas.

Formula No.	Spirulina %	Ingredients %							
		Banana	Papaya	Potato	Carrot	Apple	Guava	Mango	Sugar
Formula-1(1SFV)	0	30	-	10	10	15	15	15	5
Formula-2(2SFV)	2.5	30	-	10	10	15	15	15	5
Formula-3(3SFV)	5	30	-	10	10	15	15	15	5
Formula-4(4SFV)	7.5	30	-	10	10	15	15	15	5
Formula-5(5SFV)	0	-	30	10	10	15	15	15	5
Formula-6(6SFV)	2.5	-	30	10	10	15	15	15	5
Formula-7(7SFV)	5	-	30	10	10	15	15	15	5
Formula-8(8SFV)	7.5	-	30	10	10	15	15	15	5

Table (B): Formulated of prepared dried spirulina with cereals-based baby food formulas.

Formula No.	Spirulina %	Ingredients %						
		Wheat flour	Rice flour	Dried barley powder	Lentils powder	Dried peas powder	Dried spinach powder	Dried cauliflower powder
Formula-9(1SCP)	0	30	-	30	10	10	10	10
Formula-10(2SCP)	2.5	30	-	30	10	10	10	10
Formula-11(3SCP)	5	30	-	30	10	10	10	10
Formula-12(4SCP)	7.5	30	-	30	10	10	10	10
Formula-13(5SCP)	0	-	30	30	10	10	10	10
Formula-14(6SCP)	2.5	-	30	30	10	10	10	10
Formula-15(7SCP)	5	-	30	30	10	10	10	10
Formula-16(8SCP)	7.5	-	30	30	10	10	10	10

Table 1. Chemical composition and physical properties of Spirulina (g/100 g sample, on dry weight basis)

Chemical composition	Values %	Physical properties	Values
Moisture content	4.74 ± 0.84	pH	6.84 ± 0.14
Total solids	95.36	Bulk density	0.82 Kg/lit
Protein content	62.84 ± 1.38	Particle size	100% 60 mesh
Lipid	6.93 ± 0.57	Appearance	Fine, uniform powder
Ash content	7.47 ± 0.39	Color	Blue green to green
Crude fiber	8.12 ± 0.28	Odor and taste	Mild like sea weed
Starch	3.56 ± 0.27	Consistency	Powder

Table 2. Amino acids and fatty acids content of Spirulina (mg/100 g).

Amino acids	Values	Fatty acids	Values
Essential amino acids	%	Myristic (C14:0)	0.46
Isoleucine	6.49	Palmitic (C16:0)	40.65
Leucine	7.89	Palmitoleic (C16:1 omega-6)	6.38
Lysine	4.73	Stearic (C18:0)	1.92
Methionine	2.34	Oleic (C18:1 omega-6)	1.64
Phenylalanine	4.42	Linoleic (C18:2 omega-6)	17.95
Threonine	4.58	Gamma-linolenic (C18:3 omega-6)	24.49
Tryptophan	1.93	Alpha-linolenic (C18:3 omega-3)	traces
Valine	6.08	Erucic acid (C22:1)	5.33
Total	38.46	Lignoceric acid (C24:0)	1.18
Non-essential amino acids	%	Total saturate fatty acid	44.21
Alanine	7.52	Total unsaturated fatty acid	55.79
Arginine	7.51		
Aspartic	11.17		
Cysteine	1.11		
Glutamic	13.69		
Glycine	5.24		
Histidine	2.78		
Proline	4.35		
Serine	4.56		
Tyrosin	3.61		
Total	61.54		
Total amino acids	100 %		
% Protein	62.84 ± 1.38		

Table 3. Vitamins, phytopigments and minerals in Spirulina.

Components	Values	Components	Values
1. Vitamins	(Values /100g)	3. Minerals	(mg/100g)
Vitamin B1(Thiamine)	5.8 mg	Ca Calcium	922.278
Vitamin B2 (Riboflavin)	4.65 mg	K Potassium	2085.28
Vitamin B3 (Niacin)	15.35 mg	Mg Magnesium	1.1902
Vitamin B6 (Pyridoxine)	0.94 mg	Na Sodium	1540.46
Vitamin B12 (Analogue)	175 µg	P Phosphprus	2191.71
Folic acid	9.92 mg	Cu Copper	1.2154
Inositol	60.45 mg	Fe Iron	273.197
Vitamin E	9.86 mg	Mn Manganese	5.6608
Vitamin K	1095 µg	Zn Zinc	3.6229
Pantothenate	108 µg	Cr Chromium	0.325
Biotin	8 µg	Se Selenium	0.0394
2. Phytopigments	(%)	B Boron	2.875
Total Carotenoids	0.573	Mo Molybdenum	0.372
beta carotenoids	0.2527		
Xanthophylls	0.2818		
Zeaxanthin	0.1331		
Chlorophyll	1.5609		
Phycocyanin	14.647		

Table 4. Microbiological quality of Spirulina (CFU/g)

Test	Values
TVBC (Total viable bacterial count)	negative
MSB (Mesophilic Spore formers bacteria)	ND
Y&M (Yeasts and Moulds)	ND
Coliform group	ND
Salmonella	ND
Staphylococcus	ND

Table 5. Contaminant specifications of spirulina

Test	Values
Arsenic	< 1.0 ppm
Cadmium	< 0.5 ppm
Lead	< 0.5 ppm
Mercury	< 0.05 ppm
Pesticides	negative
Rodent hairs	ND
Insect fragments	ND

Table 6. Chemical composition of fruits and vegetables (g/100g on wet weight basis)

Components	Banana puree	Papaya puree	Potato puree	Carrot puree	Apple puree	Guava puree	Mango puree
Moisture %	76.23±0.796	87.27±0.474	79.55±0.518	88.34±0.517	86.42±0.651	84.68±0.908	82.15±0.711
Total solids %	23.77	12.73	20.45	11.66	13.58	15.32	17.85
Ash %	0.955±0.002	0.672±0.006	1.154±0.002	0.731±0.004	0.394±0.006	0.677±0.002	0.601±0.006
Fat %	0.538±0.005	0.476±0.004	0.352±0.002	0.489±0.003	0.283±0.002	0.232±0.005	0.438±0.003
Protein %	1.409±0.065	0.739±0.012	1.550±0.003	1.432±0.008	0.236±0.005	1.091±0.021	0.953±0.023
Titratable acidity %*	0.458±0.009	0.152±0.000	0.586±0.002	0.224±0.000	0.650±0.015	0.449±0.020	0.568±0.014
pH values	4.88±0.011	5.39±0.012	5.84±0.025	6.11±0.022	3.83±0.025	4.11±0.010	4.38±0.002
Total sugars %	15.208±0.030	7.123±0.034	0.922±0.010	7.305±0.005	9.093±0.024	8.860±0.042	11.29±0.027
Reducing sugars %	9.324±0.025	2.984±0.025	0.292±0.002	2.101±0.003	5.932±0.011	3.279±0.031	2.440±0.014
Non reducing sugars %	5.884	4.139	0.630	5.205	3.161	5.581	8.850
Starch %	3.12±0.014	0.579±0.010	14.963±0.052	0.089±0.009	0.185±0.011	0.016±0.002	0.331±0.002
Fiber %	1.952±0.012	1.521±0.031	0.902±0.002	0.936±0.007	1.541±0.036	2.015±0.063	0.896±0.015
Total pectic substances %	0.774±0.004	1.914±0.004	0.742±0.002	0.714±0.006	1.504±0.002	1.314±0.004	2.331±0.002
Carotenoids (mg/l)	3.211±0.017	32.818±0.234	4.847±0.018	182.36±0.921	5.132±0.005	5.527±0.028	32.38±0.141
Anthocyanine (O.D. at 535)	-	0.0452±0.001	-	2.983±0.018	3.251±0.001	-	0.0463±0.000
Ascorbic acid (mg/100g)	17.63±0.547	91.38±1.211	12.17±0.320	9.97±0.242	15.33±0.541	79.51±1.274	39.38±1.387

Each value is the average of three replicates ± S.E.

*as anhydrous citric acid.

Table 7. Chemical composition of dried raw materials (g/100g on wet weight basis).

Raw materials	Chemical Components					
	Moisture %	Protein %	Fat %	Ash %	Total Carbohydrates	Total acidity
Wheat	9.54±0.24	12.87±0.20	3.36±0.02	1.95±0.02	72.28	0.71±0.01
Rice	9.76±0.17	9.12±0.14	0.89±0.02	0.83±0.01	79.40	0.28±0.00
Barley	9.38±0.08	13.42±0.41	3.42±0.08	2.94±0.03	70.84	0.84±0.01
Lentil	10.18±0.16	22.78±0.63	3.35±0.01	2.17±0.01	61.52	0.72±0.03
Peas	8.86±0.12	19.11±0.41	2.50±0.03	2.64±0.02	66.89	0.53±0.07
Chickpea	8.62±0.08	17.08±0.72	7.29±0.42	2.93±0.04	64.08	0.47±0.02
Spinach	6.85±0.17	15.89±0.28	1.43±0.08	9.31±0.08	66.52	0.89±0.01
Cauliflower	6.38±0.24	26.95±0.84	2.43±0.07	10.38±0.14	53.86	0.76±0.01

Table 8. Microbiological quality of formulated baby food formulas (CFU/g).

Formulas No.	TVBC [*]	MSB [†]	Yeast and mold count	Total Coliform count	Salmonella	Staphylococcus aureus
1 SFV	9.5 x10 ²	3.6 x10 ¹	4.4 x10 ¹	Nil	Nil	Nil
2 SFV	8.7 x10 ²	3.2 x10 ¹	3.9 x10 ¹	Nil	Nil	Nil
3 SFV	6.9 x10 ²	2.6 x10 ¹	3.6 x10 ¹	Nil	Nil	Nil
4 SFV	5.8 x10 ²	1.8 x10 ¹	2.8 x10 ¹	Nil	Nil	Nil
5 SFV	9.8 x10 ²	3.2 x10 ¹	5.9 x10 ¹	Nil	Nil	Nil
6 SFV	8.4 x10 ²	3.0 x10 ¹	4.7 x10 ¹	Nil	Nil	Nil
7 SFV	7.9 x10 ²	2.3 x10 ¹	3.3 x10 ¹	Nil	Nil	Nil
8 SFV	6.7 x10 ²	2.1 x10 ¹	2.9 x10 ¹	Nil	Nil	Nil
1 SCP	7.3 x10 ¹	3.5 x10 ¹	Nil	Nil	Nil	Nil
2 SCP	6.1 x10 ¹	2.9 x10 ¹	Nil	Nil	Nil	Nil
3 SCP	4.9 x10 ¹	2.8 x10 ¹	Nil	Nil	Nil	Nil
4 SCP	3.8 x10 ¹	1.9 x10 ¹	Nil	Nil	Nil	Nil
5 SCP	7.2 x10 ¹	3.8 x10 ¹	Nil	Nil	Nil	Nil
6 SCP	6.6 x10 ¹	2.3 x10 ¹	Nil	Nil	Nil	Nil
7 SCP	4.9 x10 ¹	1.7 x10 ¹	Nil	Nil	Nil	Nil
8 SCP	2.7 x10 ¹	1.3 x10 ¹	Nil	Nil	Nil	Nil

* (TVBC) Total viable bacterial count

† (MSB) Mesophilic Sporeformers bacteria

Table 9. Sensory evaluation scores of formulated baby foods formulas prepared from

Formulas No.	Sensory attributes					
	Color (20)	Taste (20)	Odor (20)	Texture (20)	Mouth feel (20)	Overall acceptability (100)
1 SFV	17.92 ^c ± 0.35	17.90 ^b ± 0.32	17.31 ^b ± 0.47	17.14 ^d ± 0.27	18.76 ^{ab} ± 0.26	86.34 ^c ± 1.24
2 SFV	18.95 ^b ± 0.39	18.05 ^b ± 0.30	17.64 ^{ab} ± 0.30	18.86 ^b ± 0.35	18.94 ^{ab} ± 0.40	90.35 ^b ± 1.02
3 SFV	19.55 ^{ab} ± 0.23	18.66 ^a ± 0.31	17.48 ^{ab} ± 0.32	19.15 ^{ab} ± 0.27	18.92 ^{ab} ± 0.30	95.47 ^a ± 0.92
4 SFV	18.47 ^{bc} ± 0.30	18.14 ^{ab} ± 0.30	17.10 ^b ± 0.31	19.26 ^{ab} ± 0.31	18.42 ^b ± 0.34	89.13 ^{bc} ± 1.30
5 SFV	18.22 ^c ± 0.21	17.12 ^c ± 0.35	17.69 ^{ab} ± 0.30	17.89 ^c ± 0.33	19.11 ^a ± 0.31	87.12 ^c ± 1.21
6 SFV	18.45 ^{bc} ± 0.23	17.85 ^b ± 0.34	17.95 ^{ab} ± 0.31	19.05 ^{ab} ± 0.30	18.75 ^{ab} ± 0.31	89.84 ^b ± 1.21
7 SFV	19.85 ^a ± 0.38	18.45 ^{ab} ± 0.38	18.21 ^a ± 0.23	19.15 ^{ab} ± 0.27	18.50 ^b ± 0.27	96.37 ^a ± 1.25
8 SFV	18.85 ^{bc} ± 0.33	18.27 ^{ab} ± 0.30	17.84 ^{ab} ± 0.30	19.53 ^a ± 0.26	18.43 ^b ± 0.24	88.17 ^{bc} ± 1.07
L.S.D at p ≤ 0.05	0.69	0.53	0.86	0.62	0.57	2.51
1 SCP	17.47 ^c ± 0.27	17.45 ^{bc} ± 0.09	16.75 ^{bc} ± 0.20	16.70 ^d ± 0.41	18.29 ^b ± 0.30	82.72 ^d ± 1.12
2 SCP	18.76 ^{ab} ± 0.34	17.58 ^b ± 0.21	17.26 ^b ± 0.24	18.45 ^b ± 0.37	18.47 ^{ab} ± 0.33	85.36 ^c ± 1.82
3 SCP	19.26 ^a ± 0.23	18.55 ^{ab} ± 0.37	16.95 ^{bc} ± 0.27	18.67 ^{ab} ± 0.20	18.78 ^a ± 0.41	92.85 ^a ± 1.32
4 SCP	18.05 ^{bc} ± 0.28	17.85 ^b ± 0.23	16.55 ^c ± 0.21	18.75 ^{ab} ± 0.28	17.95 ^b ± 0.18	88.47 ^b ± 1.58
5 SCP	17.85 ^{bc} ± 0.26	16.79 ^c ± 0.14	17.62 ^{ab} ± 0.35	17.17 ^c ± 0.14	18.78 ^a ± 0.42	83.17 ^{cd} ± 1.05
6 SCP	18.18 ^{bc} ± 0.28	17.93 ^b ± 0.19	17.65 ^{ab} ± 0.29	18.29 ^b ± 0.29	18.38 ^{ab} ± 0.37	87.11 ^{bc} ± 1.47
7 SCP	19.45 ^a ± 0.12	18.76 ^a ± 0.31	17.95 ^a ± 0.38	18.84 ^{ab} ± 0.23	18.13 ^b ± 0.28	93.48 ^a ± 1.41
8 SCP	18.47 ^b ± 0.14	17.96 ^b ± 0.26	17.48 ^{ab} ± 0.18	19.28 ^a ± 0.15	18.06 ^b ± 0.26	91.85 ^a ± 1.28
L.S.D at p ≤ 0.05	0.74	0.68	0.55	0.64	0.47	2.38

*Values represent of 12 panellists (Mean ±S.E.);

* a, b,....: There is no significant difference (p ≥ 0.05) between any two means have the same superscripts, within the same acceptability attribute.

Table 10. Some physicochemical properties and nutritional value of prepared spirulina, fruits and vegetables-based baby food formulas.

Components	Formula No. 3 SFV	Formula No. 7 SFV
Moisture %	72.41±0.43	72.56±0.28
Total solids %	27.59	27.44
Ash %	1.89± 0.02	1.82±0.02
Fat %	0.543± 0.007	0.516±0.005
Protein %	4.64±0.31	4.51±0.25
pH values	5.23±0.03	5.45±0.09
Titrateable acidity % ⁺	0.39±0.003	0.35±0.006
Starch%	2.24±0.07	2.19±0.06
Total sugars %	16.07±0.64	15.82±0.43
Reducing sugars %	9.64±0.416	9.72±0.023
Non reducing sugars %	6.43	6.10
Total pectic substances %	1.86±0.01	1.93±0.02
Fiber %	1.637±0.02	1.796±0.02
Carotenoids (mg/l)	21.79±0.069	25.87±0.130
Anthocyanine (O.D. at 535)	0.192±0.002	0.489±0.005
Ascorbic acid (mg/100g) *	26.28±0.89	31.16±0.53
Minerals content (mg/100g)		
K Potassium	720.86	625.47
Ca Calcium	240.85	239.62
Na Sodium	369.20	497.74
Mg Magnesium	12.054	19.958
Fe Iron	60.121	48.981
Mn Manganese	1.444	1.977
Cu Copper	0.527	0.635
Zn Zinc	1.360	1.610
P Phosphorus	482.18	386.74
Cr Chromium	0.0715	0.0972
Se Selenium	0.0087	0.0069
B Boron	0.6325	0.5060
Mo Molybdenum	0.0818	0.0655
Energy values k.cal/100 g	104.127	102.444

Each value is the average of three replicates ± S.E.
Chemical composition on wet weight basis.

*as anhydrous citric acid

Table 11. Some chemical composition and nutritional value of prepared dried spirulina with cereals-based baby food formulas.

Components	Formula No. 3 SCP	Formula No. 7 SCP
Moisture %	6.12±0.08	5.89±0.05
Total solids %	93.88	94.11
Ash %	3.69±0.04	3.48±0.07
Fat %	3.02±0.05	2.84±0.04
Protein %	20.04±0.34	21.57±0.47
Titratable acidity %	0.24±0.00	0.21±0.01
Total carbohydrates %	66.89	66.01
Minerals content (mg/100g)		
K Potassium	2271.494	1982.855
Ca Calcium	471.702	479.242
Na Sodium	240.517	298.643
Mg Magnesium	56.162	59.872
Fe Iron	61.072	63.388
Mn Manganese	3.8885	3.9538
Cu Copper	1.5535	1.2704
Zn Zinc	2.7205	2.5193
P Phosphorus	579.176	586.741
Cr Chromium	0.1430	0.1944
Se Selenium	0.0173	0.0139
B Boron	1.265	1.212
Mo Molybdenum	0.1437	0.1409
Energy values k.cal/100 g	374.9	375.88

Each value is the average of three replicates ± S.E.; Chemical composition on wet weight basis.

Table 12. Amino acids content of formulated baby food formulas (mg/100g formulas).

Amino acids	Formulas			
	Formula No. 7 SCP	Formula No. 3 SCP	Formula No. 7 SFV	Formula No. 3 SFV
Essential amino acids	%	%	%	%
Isoleucine	2.819	2.655	6.0908	6.6505
Leucine	5.064	4.175	7.7253	8.3147
Lysine	3.359	3.917	6.7575	7.3045
Methionine	2.618	2.347	2.5762	2.3133
Phenylalanine	3.185	2.701	6.5597	6.5385
Threonine	2.803	3.339	4.8293	5.2788
Tryptophan	2.006	2.567	0.936	0.9954
Valine	4.219	4.284	5.7693	6.083
Total	26.073	25.985	41.244	43.479
Non-essential amino acids				
Alanine	5.944	5.366	7.0242	6.13
Arginine	4.141	4.916	7.753	5.357
Aspartic	21.194	19.969	6.1128	6.533
Cysteine	2.586	2.647	2.0059	2.506
Glutamic	13.802	14.724	12.005	14.15
Glycine	4.382	3.702	4.6927	5.426
Histidine	3.077	2.869	4.4637	4.697
Proline	13.107	13.897	6.3654	3.512
Serine	3.584	2.775	4.5655	4.68
Tyrosin	2.12	3.159	3.7675	3.534
Total	73.937	74.024	58.756	56.52
Total amino acids	100 %	100 %	100 %	100 %
% Protein	4.51	4.64	21.57	20.04

Contaminant specifications of Spirulina: Data in Table (5) indicated that, the spirulina free from pesticides, rodent hairs and insect fragments. On the other hand, the level of heavy metals in line with the specifications of the global food this results are in agreement with those obtained by Haeng-Shin *et al.* (2006) [22] and Llobet *et al.* (2003) [27].

Use spirulina in production of some food formulas as complementary food for babies: In Egypt, most of the time, the formulas given to babies are of poor nutritional quality: they are mainly cereal flours with sugar, sometimes some fruits, and rarely, when the mothers can afford it, powdered milk. Those formulas do not cover the babies' needs in proteins, lipids and micro-nutrients.

Babies need to have enough calories and fat in order to grow normally. As babies move from a liquid diet to a more solid diet, using some higher calorie foods can help to meet their needs. Nutritious foods for older vegetarian babies include mashed tofu, bean spreads, avocado, and cooked dried fruits. Fat intake should not be limited. Fat sources for older infants include avocado, vegetable oils and soft margarine.

Nutrition plays an important role for fundamental vital functions. Many nutritionists have focused on naturally occurring components (e.g. vitamins, fatty acids, proteins, amino acids, phenolic compounds and dietary fibre) in foods that have a positive effect on target functions beyond nutritive value and provide health benefits, as well as possibly reducing the risk of diseases. The term functional food originates from Japan and generally represents the category of foods that contain biologically active compounds with potential to enhance health or reduce risk of serious diseases and finally, may improve the quality of life. Furthermore, foods identified as "Food for specified health use (FOSHU)" should be in the form of naturally occurring food or drink products, but not pills or capsules.

In recent years, different healthy ingredients have been used in the production of baby foods to enhance its nutritional profile or to confer functional properties.

However, the amount of raw material that can be used as a substitute or can be added to baby foods represents a compromise between nutritional improvement and satisfactory sensorial properties of baby foods, from the previous results in Tables (1 to 5) we can be certain that spirulina of the best raw materials that can be used in the manufacture of baby food.

Chemical properties of ingredients used in baby food formulas:

Chemical composition of ingredients used to prepared spirulina, fruits and vegetables-based baby food formulas: Chemical properties of ingredients used for the preparation of the baby food formulas are presented in Table (6). The results demonstrated that the moisture content of ingredients varied from 76.23% to 88.34% in banana puree and carrot puree, respectively. Potato puree had the highest level of ash being about 1.154% while, the lowest level of ash was found in apple puree being about 0.394%. Also, potato puree had the highest level of protein (1.55%). So, spirulina and potato puree were the main source of protein in formulated baby food formulas. Potato puree was the main source for starch. The pH value for papaya puree was 5.39. The pH value of ingredients ranged from 3.83 to 6.11 for apple puree and carrot puree, respectively. Titratable acidity for all ingredients was less than 1% . With regard to total sugars the data showed that the banana puree had the highest amount of total sugars, (15.208%). Pectin ranged from 0.714 to 2.331% in carrot puree and mango puree, respectively. The pectin can hold the water in baby stomach. So, pectin is very important for children especially when they have diarrhea. On the other hand, guava puree had the highest level of fiber (2.015%) followed by banana puree (1.952%), while potato puree had the lowest level of fiber being (0.902%). Papaya puree contained amount of carotenoids less than carrot puree. So, adding of carrot puree will increase the percentage of carotenoids in all formulas. As known that, the carotenoids help the baby as color to attract any foods. Carrot puree had high percentage of anthocyanin more than other fruits or vegetables ingredients. Results appeared that ascorbic acid content was ranged from 9.97 to 91.38 mg/100g in carrot and papaya puree, respectively.

These results of chemical composition for ingredients used for the preparation of baby food formulas were in agreement with those obtained by MaCance and Widdowson's (1992) [28]; Ramulu and Rao (2003) [38]; El-Mansy *et al.* (2005) [14]; Wall (2006) [55] and Bahlol *et al.*, (2007) [7].

Chemical composition of ingredients used to prepared dried spirulina with cereals-based baby food formulas:

The proximate chemical analysis were carried out on the original raw materials used in this research i.e. cereals (wheat, rice and Barley), legumes (dried peas, hulled chickpea and lentil), vegetable (Spinach and Cauliflower). The results are illustrated in Table (7). It noticed that, lentil and rice had the higher moisture content being 10.18 and 9.76%, respectively. Meanwhile, dried cauliflower had the lower moisture content. From the results in the same table, it could be noticed that dried cauliflower had higher protein content (26.95%). Dried cauliflower and dried spinach had higher protein and ash contents. Total carbohydrate were calculated for raw materials, total carbohydrate content ranged from 53.86% in dried cauliflower to 79.40 in rice. These results are in agreement with Soliman *et al.* (1996) [49]; Atwa (2003) [6]; Abd El-Salam (2005) [1] and Baik and Ullrich (2008) [8].

Microbiological quality of the formulated baby foods formulas:

The overall bacteriological status of the formulated prepared spirulina with some fruits and vegetables-based baby food formulas was observed to be satisfactory. The microbiological quality attributes of different prepared formulas calculated as CFU g⁻¹ are shown in Table (8). The obtained results revealed that the total viable bacterial count was ranged from 5.8 x10² to 9.8 x10² CFU/g for formulas No. (4SFV) and (5SFV), respectively as indicated in Table (8).

The low counts of the examined formulas for total viable bacterial, yeasts & moulds indicated adequate thermal process, good quality of raw materials and as a result of the good different processing conditions under which the production of formulas was carried out. Mesophilic sporeformers bacterial count was 1.8 x10¹ and 3.6

x10¹ CFU/g for formulas No. (4SFV) and (1SFV), respectively. However, coliform group, *Salmonella* and *Staphylococcus aureus* were found to be absent in all formulas. The microbiological results are in agreement with many authors such as Wadud *et al.* (2004) [54]; Soliman (2003) [50] and Bahlol *et al.* (2007) [7]. The formulated of prepared dried spirulina with cereals-based baby food formulas were tested for the same microbiological tests. The obtained results in Table (8) reveal that the total bacterial count ranged from 2.7 x 10¹ to 7.3 x 10¹ cfu/g. The low total bacterial counts of the examined baby food formulated might be due to their low moisture content. The current results were within the advisable standards reported by Skovgaard (1989) [47], who recommended that a total bacterial count up to 104 per gram for dried baby foods might be save enough to be used by babies. The current results were less than those allowable in many international standards in other foods. The obtained results are also agree with those obtained by Radi *et al.* (2003) [36] who produced new production from siwi date for young children. These results are in agreement with those reported by Soliman *et al.* (1996) [49]. The yeast & molds, coliform group, *Salmonella* and *Staphylococcus aureus* did not appear in any dried baby food formulas, this may be related to low moisture content of all mixtures. This may be due to the effect of good processing and good ingredients to decrease the total bacterial count. Otherwise, the drying steps in prepared dried formulas this may be reduce its number under the detection limit. Also, spirulina was also reported to present antimicrobial activity as well as to inhibit the replication of total bacterial count. The microbiological results suggested that, the formulas are suitable to be submitted for sensory evaluation by babies, these results are in agreement with those obtained by Ozdemir *et al.* (2004) [33]. Who studied the antimicrobial activity of spirulina against various gram-positive, gram-negative bacteria and fungal species. The methanol extract showed maximum antimicrobial potency, and Bhowmik *et al.* (2009) [9] who found that Spirulina was had probiotic efficacy and inhibitory effect against several pathogens.

Sensory evaluation:

Cereals in the form of paps prepared with milk are usually one of the first foods added in the diversification of the infant diet from the 5th/6th

months. Milk and cereal based ready-to-eat infant foods are presently available on the market. These products have a long shelf-life and can be consumed for up to one year after manufacture. Due to their composition. When cereals are well accepted, we add fruit and vegetables. One new food can be started every 3-4 days. This way we can see if baby has a reaction to a new food. Mash or puree fruits and vegetables. As our baby gets better at chewing.

For this reason, experiments were in this research concentrated on the production of two types of baby food and one dependent on grains and other certified on fruits and vegetables and using a variety of materials in order to give some kind of change in the diet of infants and children. Sensory tests were conducted for them and the results were good where they were prepared to accept all the formulas.

Data in Table (9) show the analysis of variance for data of sensory evaluation between the 16 prepared baby food formulas. The averages of the overall acceptability obtained scores were in the range from 82.72 to 96.37. These means that all the prepared baby food formulas were accepted with significant differences. On the other hand, analysis of variance for obtained scores for overall acceptability indicated significant differences ($P > 0.05$) between the different formulas (Table 9). So, LSD test was applied to carry out the multiple comparisons which indicated that, the different formulas could be divided into some significant groups ($P > 0.05$) (LSD = 2.51, 2.38 for fruits and vegetables based baby food formulas and cereals-based baby food formulas, respectively), where there are no significant differences ($P > 0.05$) between the different formulas inside every group. The high scores in both types of baby foods groups included formulas No. (3 SFV, 7 SFV, 3 SCP and 7 SCP) which had the 5% of spirulina.

This four baby food formulas were selected which obtained high scores in both types of baby foods groups. The study was continued on the selected formulas that their physicochemical properties were determined.

Physicochemical properties and nutritional value of formulated baby food formulas:

Moisture, crude protein, fat, crude fiber, ash, carbohydrate, some vitamins and minerals which were thought to be great importance in infant feeding from 6 – 36 months where determined. The food formulas were prepared to produce as complementary baby food using some fruits, vegetables, cereals and legumes with spirulina. But the visibility of chemical composition is too important. Therefore some chemical analyses were carried out. Data in Tables (10 and 11) indicated that moisture and total solids content in food formulas nearly varied in type 1 and type 2 prepared formulas. This is due to the adding kind of fruits, vegetables, cereals and legumes. It is clear that the total solids in cereals and legumes formulas was the highest among other fruits and vegetables formulas. cereals and legumes formulas had the highest level of ash content, while fruits and vegetables formulas had the lowest level of ash. The same results were obtained with fat, protein and carbohydrates, on the contrary, it was the titratable acidity and phytopigments which are important as it affected on the taste and flavor. The obtained data indicated that the total sugars and total carbohydrates were the major components in total solids in all formulas and the main source of energy value.

The percentages of total pectic substances and fiber were acceptable and suitable for babies related to the important of those for exertion. Energy values for formulate baby food formulas were estimated from the percentage of total carbohydrate, protein and fat contents and were higher in cereals and legumes formulas.

The mineral composition of fruits and others plants ingredients can reflect the trace mineral of soil in a geographic region and varies with climate, maturity, cultivars, and agricultural practices. Some minerals content of the babies food formulas are shown in Tables (10 and 11). The obtained data revealed that the highest potassium and calcium are particularly essential for infant and young children. The variation of the minerals content in all formulas, may be due to the different content of these elements in raw ingredients.

From the results of minerals it could be concluded that the formulas are considered as a source for some minerals. These formulas are not totally balanced in micronutrients but they are: balanced in macro-nutrients, rich in micro-nutrients, produced with foods available locally and rather cheap, and they are a considerable improvement compared to most local baby foods. The results of physicochemical properties and nutritional value of formulated baby food formulas were in agreement with those obtained by Bahlol *et al.* (2007) [7] and Mehder (2009) [30].

Amino acids content of formulated baby food formulas:

Protein is the major functional and structural component of all the cells of the body; for example, all enzymes, membrane carriers, blood transport molecules, the intracellular matrices, hair, fingernails, serum albumin, keratin, and collagen are proteins, as are many hormones and a large part of membranes. Moreover, the constituent amino acids of protein act as precursors of many co-enzymes, hormones, nucleic acids, and other molecules essential for life. Thus an adequate supply of dietary protein is essential to maintain cellular integrity and function, and for health and reproduction. Data in Table (12) shows the amino acids content of formulated baby food formulas. From the obtained results; it could be observed that the different baby food blends contained good proportions of essential amino acids. Comparing the essential amino acids pattern of the formulated formulas with hen's egg protein as a standard, it was found that the essential amino acids content of the blends have a good percent from their corresponding quantities in egg's protein. It may be noted that the total essential amino acids of the different baby food blends also suitable for babies in the age 1 to 3 years. This could be explained that although egg have much higher percentage of protein, but the percentage of protein content in the formulated baby food formulas was at in the range of recommended dietary allowance (RDA) for protein and amino acids for babies at age 1-3 years according to FAO/WHO, 1991, [16].

Much has been written about the health benefits of spirulina, of all the humans that can benefit from taking Spirulina, children can benefit the most.

Children love spirulina and it is safe and highly nutritious for them. Children of all ages can eat spirulina in complete safety and assimilate its nutrients without difficulty. Even malnourished children with diminished capacity for nutrient absorption could assimilate spirulina and recover from malnutrition. Spirulina can build up tissue growth, improve vision, strengthens body's immune system there by improves resistance to chronic infections, ability to heal and ability to concentrate in children. For baby, who are not able to swallow the capsules, the baby can be used this spirulina formulas. The powder spirulina formulas can also be mixed with fruit juice, milk, salads and convenient soups.

The amount of spirulina needed depends on metabolism degree to physical exertion, lifestyle, and an individual baby's unique body needs. By starting with a small amount and gradually increasing until the optimal daily amount is found, babies can enjoy the benefits of this super nutritious food from babyhood throughout their lives. (Children of all ages, including infants can be given 2 to 5 gms of spirulina/day). Spirulina is not a drug, but a natural food supplement, and is not habit forming. Its effects can be sustained by taking it regularly at approx. 2 to 5 g/day. To see any benefits of spirulina, it should be taken at least for 6-8 weeks.

4. Conclusions

A spirulina farm is an environmentally sound green food machine. Cultivated in shallow ponds, this algae can double its biomass every 2 to 5 days. This productivity breakthrough yields over 20 times more protein than soybeans on the same area, 40 times corn and 400 times beef. Spirulina can flourish in ponds of brackish or alkaline water built on already unfertile land. In this way, it can augment the food supply not by increase the agricultural area or increasing agricultural intensification in Egypt, or clearing the disappearing rainforests in the world, but by cultivating the expanding deserts. Finally from this research we can use as spirulina recommended by the United Nations World Health Organization (WHO) which confirmed that spirulina represents an interesting food for multiple reasons, rich in iron and protein, it can be safely administered to babies without any risk.

Compliance with Ethics Requirements. Authors declare that they respect the journal's ethics requirements. Authors declare that they have no conflict of interest and all procedures involving human / or animal subjects (if exist) respect the specific regulation and standards.

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