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Studies on the formulation of algal products and assessments on their biochemical content

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Abstract
The present study was designed to formulate the products with using algal biomass and their pigments whose stability is compared respectively. The result shows that among varying proportions of raw, roasted and sprouted green gram, millets, white corn, green peas with 15% ashwagandha blended with either algal biomass or its pigments reported phycocyanin blended PF3 as the best powder formulation. The role of pigments, particularly phycocyanin in improving the shelf life of consumables was also found to be remarkably significant in the aspects of commercial scale of production. Efficacy of product formulations are inferred with the help of tools in biotechnology can further add to the nutritional and therapeutic value of the products to the growing demands of the population.

Keywords: *Spirulina platensis*, *Chlorella vulgaris*, biomass, algae, formulations, pigments, ashwagandha

1. Introduction
The development of functional foods offers enormous opportunities to the foods and beverage industry. In fact, functional foods have had a strong impact on global markets over the past decade and have rapidly gained market share as value added products. The use of algae as ingredients in healthy food and beverage formulations offers interesting possibilities. New healthy products can be seen as an opportunity to cater for the needs of consumers, and also to update recommendations for the action of governments, international agencies and concerned partners in the public and private sectors. The incorporation of algae in some foods and beverages that enjoy high consumer acceptance presents a good opportunity to popularize the health benefits of algae among consumers, even those unaccustomed to this food. As a result, recent years have seen efforts to incorporate algae into different foods and beverages. Algal biomass has been used for centuries as food and medicine. The health promoting effects of algae were discovered as early as 1500 BC (Shanab *et al*. 2012) [47]. However, the biomass of algae gained interest as a source of chemicals and pharmaceuticals only recently. Nowadays, the production 12 regime requires the use of extracts rather than the biomass itself, because of the formulation requirements (consistency, stability, color, flavor, etc.). Until now, algal products were available mainly as tablets, capsules, or liquid extracts, and sometimes were incorporated into food products, cosmetics, or products for plants (Spolaore *et al*. 2006) [51]. Recently, compounds derived from algae (carotenoids, β-carotene astaxanthin, long-chain polyunsaturated fatty acids (PUFAs), docosahexaenoic acid) began to be produced on industrial scale (Borowitzka, 2013) [8]. Novel compounds isolated from algae possess a great further potential to be applied for their pharmacological and biological activity. Seaweeds produce a vast spectrum of secondary metabolites because they live in non friendly environment but are not damaged photo dynamically as they synthesize protective compounds and develop protecting mechanisms. At present, the main directions in macroalgal biotechnology are biofuels, agricultural biostimulants for crop plants, probiotics for aquaculture, soil bioremediation, wastewater treatment, and biomedical applications of extracted compounds (polyphenols, polysaccharides) (Sharma *et al*. 2012) [48]. Microalgal biotechnology refers to the production of different products: phycocyanin, carotenoids (β-carotene, astaxanthin), fatty acids and lipids, polysaccharides, immuno modulators that find an application in health food, cosmetics, feed and food supplements, pharmaceuticals, and fuel production (Pulz and Gross, 2004) [42]. The products of microalgal biotechnology are coloring substances (astaxanthin, phycocyanin, phycoerythrin), antioxidants (β-carotene, tocopherol, antioxidant CO2 extract), and arachidonic acid (ARA), docosahexaenoic acid (DHA), and PUFA extracts. Algal extracts create a new market sector, because they can be used in a variety of products, for example, antioxidant capsules containing Spirulina extract, Chlorella extract in health drinks, oral capsules containing carotenoid extracts from Dunaliella. Other examples of algal extracts-based products are pet functional food, biofertilizers (which increase water-binding capacity and serve as the source of minerals and substances promoting germination, growth of leaves and stems and flowering). Polysaccharides isolated from algae are other important components of foods and cosmetics and in nutraceutical and pharmaceutical preparations and are produced mainly from seaweeds (Chaiklahan *et al*. 2013) [11]. Polysaccharides (carrageenans, alginates) are used in food industry as edible packaging materials (Gupta and Abu-Ghannam, 2011) [21]. Algal extracts are the components of functional food, because they are considered as natural, biologically active components. The latter, beside nutrition, should have the beneficial influence on functions of the body by improving health or preventing from diseases (Santoyo *et al*. 2006) [44]. Extracts from Spirulina can be added to functional foods because of antioxidant, antimicrobial, anti-inflammatory, antiviral, and antitumoral properties of the compounds (phycocyanins, carotenoids, phenolic acids, and ω-3 and six PUFAs). Algae

are used as dietary supplements that are classified into three groups: (i) *Spirulina platensis*, (ii) Aph. flos-aquae, and (iii) *Chlorella pyrenoidosa* (Heussner *et al*. 2012) [22]. The biomass of these microalgae is obtained either from lakes or by cultivation in artificial ponds. Algae can be cultivated, in which the growth rate is high and in some cases there is a possibility of controlling the production of active compounds by adjusting cultivation conditions (Plaza *et al*. 2008) [41]. Extracts from Spirulina are active against viruses (herpes, influenza, cytomegalovirus) and inhibit carcinogenesis (Capelli and Cysewski, 2010) [10]. Spirulina is the source of vitamin A that is highly absorbable (Annapurna *et al*. 1991) [2]. Hot water extract from Spirulina supports human immune system by the improvement of immune markers in blood (higher level of gamma interferon and interleukin-12p40 and toll-like receptors) and acts directly on myeloid lineages and natural killer-cells (NK cells). Immulina is a polysaccharide found in the extract from Spirulina that activates monocytes. Water extracts also showed antiviral activity. Antioxidants present in algae and their extracts, as the use of synthetic antioxidants has been restricted because of toxicity and health risks (Farvin and Jacobsen, 2013) [18]. It is important to replace these synthetic compounds with natural antioxidants. Antioxidative compounds from marine sources include various functional compounds, for example, tocopherols (Kindleyside *et al*. 2012) [27]. Lipid-soluble algal extracts can be used as protective functional ingredients. Antioxidative properties of natural compounds from algae can prolong the shelf life of foods and cosmetics through delayed oxidation (Balboa *et al*. 2013) [6]. Natural anti-oxidants may also be useful in treating aging, UV-exposure, and diseases associated with oxidation. Extracts from Spirulina and Chlorella algae are used in cosmetics. The present study was designed to isolate the strains of Spirulina sp and Chlorella sp from Ennore estuary. This study includes product formulation using algal biomass and their pigments whose stability is compared respectively. Efficacy of product formulations are inferred with the help of tools in biotechnology can further add to the nutritional and therapeutic value of the products to the growing demands of the population.

2. Materials and Methods

2.1 Formulation of algal products

Based on the usability, low cost and high nutritional values, ingredients like millets, green grams, dry green peas, white corn and ashwagandha (*Withania somnifera*) were chosen for study and brought in one lot from the local market. Dust and waste particles were removed and cleaned from the ingredients. All ingredients were dried in shadow for 3 days in room temperature to remove the moisture content and preserved in plastic air tight containers. Various formulations of multi grains mixes were prepared by mixing the ingredients in selected proportions. Experimental formulations of powders were made with ingredients as shown in Table 1.

Table 1: various combinations of ingredients for finding best powder formulations.

	Ingredients (gm)				
	Foxtail millet	**Pearl millet**	**Green Gram**	**Green Peas**	**White Corn**
RF1	15	15	15	15	20
RF2	15	15	15	20	15
RF3	15	15	20	15	15
RF4	15	20	15	15	15
RF5	20	15	15	15	15

Raw blended formulations were made fine powdered using the grinding mill and stored in air tight containers after sieving. To analysis the effect of roasting process in nutritional values of ingredients, the best formulation of raw ingredients were roasted in frying pan at various temperatures from 40°C to 100°C. After roasting process all ingredients were made into a fine powder using the grinding mill and stored for analyzing the biochemical content. Germination is one of the best ways to improve the nutritional values in the ingredients, so the best formulation of raw ingredients were first washed in tap water to remove all dust and debris's and soaked in water for overnight. Next day morning the water was completely filtered and all raw ingredients were kept in room temperature in clean wet muslin cloth for germination. Sprouts will start developing from the ingredients and it was allowed up to 5 days from the initial day of germination. Each day the sprouted ingredients were collected and dried in room temperature without direct contact of sunlight under the shade. All collected ingredients were made powdered in grinding mill, after the moisture content was completely removed. The formulation RF2, (i.e) foxtail millet (15%), pearl millet (15%), Green gram (15%), Green peas (20%) and white corn (15%) at 5th day of sprouted was blended with algae biomass and their three pigments separately. *S. platensis* and *C. vulgaris* powder obtained from the outdoor mass cultivations were used in blended process and algae pigments like Phycocyanin (Blue pigments) was brought from Eid parry (I) Ltd, Carotenoids (Red pigments) was brought from proalgen biotech ltd and Chlorophyll (Green pigments) was brought from Camillotek India Pvt. Ltd were also blended with the best formulation. Ashwagandha (15%) was kept as a common ingredient in all formulations Table 2. Total carbohydrates, protein, fat, amino acids, moisture content and microbial count were analysed in all formulated powders.

Table 2: Formulation of algal powders.

No	**Ingredient: 1 (15%)**	**Ingredient: 2 (15%)**	**Ingredient: 3 (15%)**	**Ingredient: 4 (20%)**	**Ingredient: 5 (15%)**	**Ingredient: 6 (15%)**	**Ingredient: 7 (5%)**
PF1	Foxtail millet	Pearl millet	Green Gram	Green Peas	White Corn	Aswagandha	*Spirulina*
PF2	Foxtail millet	Pearl millet	Green Gram	Green Peas	White Corn	Aswagandha	*Chlorella*
PF3	Foxtail millet	Pearl millet	Green Gram	Green Peas	White Corn	Aswagandha	Phycocyanin
PF4	Foxtail millet	Pearl millet	Green Gram	Green Peas	White Corn	Aswagandha	Carotenoids
PF5	Foxtail millet	Pearl millet	Green Gram	Green Peas	White Corn	Aswagandha	Chlorophyll

2.2 Estimation of total proteins content (Lowry *et al*. 1951) [30]

About 10mg of formulated powder was taken in a test tube with 10 ml hot trichloroacetic acid (TCA) (55®C) and incubate in room temperature for 5 minutes. The sample mixture was centrifuged and the supernatant was discarded. The pellet was kept in room temperature for overnight with 1N of NaOH. 4ml of alkaline reagent was added with 0.1ml of pellet for dissolving TCA precipitated proteins and kept the solution in room temperature for 3 minutes. After incubation about 0.5ml of folin's reagents was added to all test tubes and mix for 5 minutes. 1N of NaOH was used as blank. Different concentrations (10-100mg) of bovine serum albumin were used as standard. All test tubes were incubated for 30 minutes in room temperature and read the absorbance. The amount of protein in 10 mg of formulated powder was measured by using standard curve.

2.3 Estimation of total carbohydrate content (Dubois, 1956) [17]

10mg of formulated powder was dissolved in 10ml of distilled water. 1ml of this extract solution was mixed with 1ml of 5% phenol and 5ml of con.c sulphuric acid. The solution was incubated at room temperature for 10 minutes. After incubation the sample was used to read the absorbance at 488nm in spectrophotometer. Glucose served as standard. Distilled water was used as blank.

2.4 Estimation of total fat content (AOAC, 1984) [4]

10mg of powder was first ground with 1.25ml of chloroform and followed by 2.5ml of methanol. The mixtures were kept in 27°C for overnight. Next day additional chloroform (1.25ml) was added to mixture and kept undisturbed for 30 minutes. The chloroform layer was removed and washed by adding 5.ml of 5% NaCl to remove methanol traces from the chloroform layer. The 63 chloroform was removed by using rotator evaporator and flask was cooled in desiccators in room temperature. Weight of total lipids from the samples was calculated and expressed as mg/g.

2.5 Moisture content (BIS, 2012) [7]

10g of powder was weighed into a petri dish which has been dried previously, cooled in desiccators and then weighed. Petri dish with powder was kept in oven at 105°C for one hour. After incubation the perti dish was cooled in desiccators in a room temperature. Loss of weight of sample taken during drying was represented as percentage of moisture content.

2.6 Microbial analysis

The microbial stability of all the formulated powders was analysed. The collected test samples were serially diluted and 1 ml of 10^{-4} dilution sample was streaked in agar plate containing in the respective media. The plates were incubated in room temperature for 2 days to determine the growth of bacteria and fungi. All growth colonies were counted and identified the organisms under microscope using standard manual.

2.7 Organoleptic evaluation of the formulated powders

Formulated powders were anyalsed for its colour, smell and texture by the five registered dietitian members in the panel. The formulated powders were subjected for the organoleptic evaluation was carried out Hedonic scale score card was used for the overall acceptability of the formulated powders. (Key score: 0-poor, 5-fair, 10-good, 15-very good, 20-excellent).

2.8 Statistical analysis

The experiments conducted in triplicates in this study were subjected to various statistical analysis using ANOVA software that the standard error mean and P value significance were estimated and the results were inferred for logical interpretations.

3. Results

3.1 Powder formulations

The data on the biochemical analysis of raw powder formulations RAF1-RAF4 are shown in the table 3 and graphically presented in fig 1. Among the tested raw powder formulations RAF1-RAF4, the maximum carbohydrate content (681.4mg/g) was obtained in RAF3 and minimum carbohydrate content (385.8mg/g) was seen in RAF1. Data from the table 3 clearly shows that the maximum protein content (427.0mg/g) was obtained in RAF1 and minimum protein content (284.1mg/g) was obtained in RAF3. Fig 1 shows that the highest fat content (180.1mg/g) was obtained in RAF2 and the lowest fat content (100.3mg/g) was obtained in RAF1. Since the RAF2 showed the optimal content of carbohydrate, protein and fat, when compare to other formulation it was taken for further studies. From the Table 3, it is seen that mean values of carbohydrate, protein and fat content of raw formulations RAF1- RAF4 showed significant, since the P value is less than 0.05.

The mean value of biochemical analysis of roasted powder formulations RSF1-RSF4 are given in the table 4 and data are also graphically presented in fig 2. Among the roasted powder formulations RSF1-RSF4, the maximum carbohydrate content (430.2mg/g) was obtained in RSF1 and minimum carbohydrate content (372.6mg/g) was obtained in RSF4. Data from the table 4 clearly show that the maximum protein content (342.8mg/g) was observed in RSF1 and minimum protein content (280.9mg/g) was observed in RSF4. Fig 2 shows that the highest fat content (172.3mg/g) was obtained in RSF1 and the lowest fat content (123.2mg/g) was obtained in RSF4. From the Table 4, it is seen that mean value of carbohydrate, protein and fat content of roasted formulations RSF1-RSF4 showed significance among the values, since the P value is less than 0.05.

The data on biochemical analysis of sprouted powder formulations SSF1-SSF5 are presented in the table 5 and data were also graphically shown in fig 3. Among the sprouted powder formulations SSF1-SSF5, the maximum carbohydrate content (430.8mg/g) was seen in SSF1 and minimum carbohydrate content (289.4mg/g) was seen in SSF5. Table 5 shows that the maximum protein content (485.0mg/g) was obtained in SSF5 and minimum protein content (378.1mg/g) was obtained in SSF1. Fig 3 shows that the highest fat content (287.9mg/g) was obtained in SSF5 and the lowest fat content (190.4mg/g) was obtained in SSF1. From the Table 5, it is seen that mean values of carbohydrate, protein and fat content of sprouted formulations SSF1- SSF5 showed significance in their concentration, since the P value is less than 0.05.

The mean values of the biochemical analysis of algae and its pigments blended powder formulations PF1-PF5 are given in the table 6 and graphically presented in fig 4. Among the

algae and its pigments blended powder formulations PF1-PF5 the maximum carbohydrate content (309.8mg/g) was obtained in PF1 and minimum carbohydrate content (262.1mg/g) was obtained in PF2. Data from the table 6 clearly show that the maximum protein content (512.4mg/g) was seen in PF3, while minimum protein content (476.6mg/g) was observed in PF5. Fig 4 shows that highest fat content (291.4mg/g) was obtained in PF2 and lowest fat content (264.1mg/g) was obtained in PF1. The mean values of carbohydrate, protein and fat content of final formulations PF1-PF5 shows significance in their concentration, since the P value is less than 0.05. Microbial analysis of formulated powders was examined and the results show that there is no detectable bacterial and yeast count of formulated powders PF1-PF5 after formulation at 10^{-4} dilution. The moisture content of the formulated powders PF1-PF5 was determine about 3.2%, 3.0%, 4.0%, 4.1% and 3.6% respectively.

3.2 Organoleptic evaluation

Organoleptic evaluation of formulated powders were analysed and the average values of organoleptic scores are presented in table 7. Data from table 7 shows that the colour of the formulated powders PF1-PF5 was found 14.9, 15.5, 16.1, 14.8 and 14.0 respectively. The average values of organoleptic score of odour of the formulated powders PF1-PF5 were observed 8.4, 7.5, 10.2, 9.4 and 9.7 respectively. The mean values of organoleptic score for texture of formulated powders PF1-PF5 was found 8.1, 9.0, 11.7, 9.5, and 9.8 respectively. From the Table 7, it is inferred that mean values of organoleptic evaluation score of the formulated powders showed significance, since the P value is less than 0.05; organoleptic score of physical stability for formulated powder shows significance in their colour, odour, and texture.

Table 3: Biochemical parameters of raw powder formulated with different raw ingredients.

Formula No	Carbohydrates (Mg/g) (Mean ± SEM)	Proteins (Mg/g) (Mean ± SEM)	Fat (Mg/g) (Mean ± SEM)
RAF1	385.8 ± 12.54	427.0 ±14.24	100.3 ±3.47
RAF2	459.8 ±9.60	364.0 ±5.79	180.1 ±6.20
RAF3	681.4 ±19.02	284.1 ±6.00	145.0 ±4.55
RAF4	500.0 ±8.75	300.8 ±9.27	138.7 ±6.49

From the Table: 3, it is concluding that standard error mean of carbohydrate, protein and fat content of raw formulations shows significance in their concentration. Since the P value is less than 0.05, carbohydrates, protein and fat content shows significance in their concentration in various combinations of raw ingredients.

Table 4: Biochemical parameters of roasted RAF2 powder formulated with different roasting temperature.

Formula No	Carbohydrates (Mg/g) (Mean ± SEM)	Proteins (Mg/g) (Mean ± SEM)	Fat (Mg/g) (Mean ± SEM)
RSF1 (40°C)	430.2±19.0	342.8±10.7	172.3±2.41
RSF2 (60°C)	422.7±14.7	328.0±17.5	159.0±5.82
RSF3 (80°C)	400.1±12.8	302.4±22.0	137.7±3.00
RSF4 (100°C)	372.6±28.6	280.9±12.4	123.2±1.99

From the Table: 4, it is concluding that standard error mean of carbohydrate, protein and fat content of roasted formulation shows significance in their concentration. Since the P value is less than 0.05, carbohydrates, protein and fat content shows significance in their concentration during roasting of raw ingredients at different temperatures.

Table 5: Biochemical parameters of sprouted RAF2 powder formulated with different sprouting days.

Formula No	Carbohydrates (Mg/g) (Mean ± SEM)	Proteins (Mg/g) (Mean ± SEM)	Fat (Mg/g) (Mean ± SEM)
SSF1 (Day:1)	430.8±10.41	378.1±5.47	190.4±6.75
SSF2 (Day:2)	417.1±12.17	390.4±9.88	201.7±3.22
SSF3 (Day:3)	351.2±9.65	414.0±14.21	234.0±9.01
SSF4 (Day:4)	321.6±18.11	440.8±16.91	276.3±11.0
SSF5 (Day:5)	289.4±8.57	485.0±12.30	287.9±8.77

From the Table: 5, it is concluding that standard error mean of carbohydrate, protein and fat content sprouted formulation shows significance in their concentration. Since the P value is less than 0.05, carbohydrates, protein and fat content shows significance in their concentration during sprouting of raw ingredients at different days.

Table 6: Biochemical parameters of SSF5 powder formulated with algae and its pigments.

Formula No	Carbohydrates (Mg/g) (Mean ± SEM)	Proteins (Mg/g) (Mean ± SEM)	Fat (Mg/g) (Mean ± SEM)
PF1 (Spirulina)	309.8±10.14	500.8± 7.29	264.1±6.49
PF2 (Chlorella)	262.1±18.70	501.0±12.57	291.4±12.55
PF3(Phycocyanin)	294.1±12.51	512.4±19.17	272.0±16.17
PF4 (Carotenoids)	297.3±9.89	489.0±10.00	280.6±9.99
PF5 (Chlorophyll)	280.6±17.01	476.6±15.90	276.5±20.27

From the Table: 6, it is concluding that standard error mean of carbohydrate, protein and fat content of final formulation shows significance in their concentration. Since the P value is less than 0.05, carbohydrates, protein and fat content shows significance in their concentration after blending with algae and its pigments.

Table 7: Organoleptic evaluation of powders formulated with algae and its pigments.

	Colour (Mean ± SEM)	Odur (Mean ± SEM)	Texture (Mean ± SEM)
	1st Month	1st Month	1st Month
PF1 (Spirulina)	14.9±0.0 17	8.4±0.029	8.1±0.030
PF2 (Chlorella)	15.5±0. 052	7.5±0.014	9.0±0.019
PF3 (Phycocyanin)	16.1± 0. 84	10.2±0.091	11.7±0.055
PF4 (Carotenoids)	14.8±0.022	9.4±0.033	9.5±0.027
PF5 (Chlorophyll)	14.0±0.037	9.7±0.060	9.8±0.070

From the Table: 7, it is inferred that standard error mean of organoleptic evaluation of the formulated powders show significance. Since the P value is less than 0.05, organoleptic score of physical stability for formulated powder shows significance in their colour, odour, and texture before and after storage.

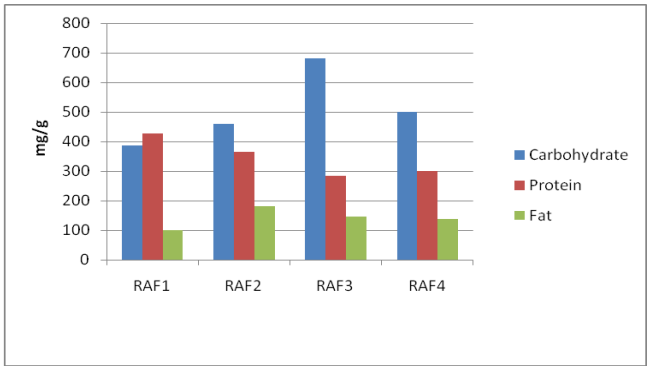

Fig 1: Biochemical parameters of raw powder formulated with different raw ingredients.

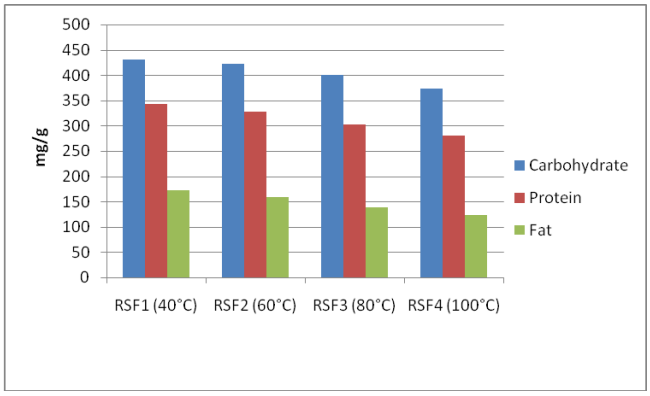

Fig 2: Biochemical parameters of roasted RAF2 powder formulated with different roasting temperature.

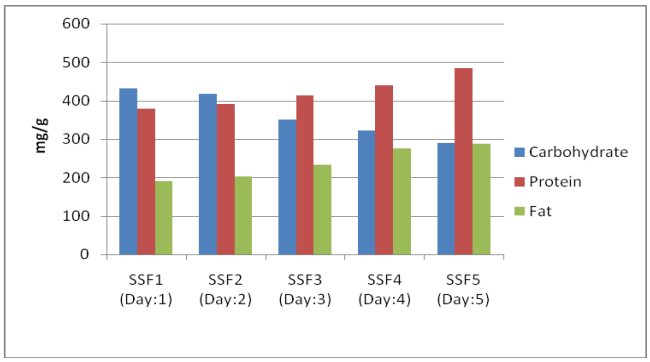

Fig 3: Biochemical parameters of sprouted RAF2 powder formulated with different sprouting days.

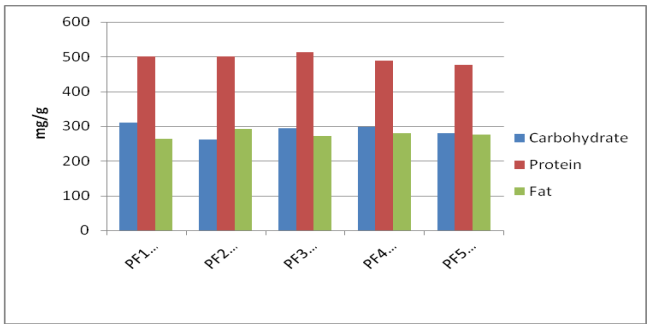

Fig 4: Biochemical parameters of SSF5 powder formulated with algae and its pigments.

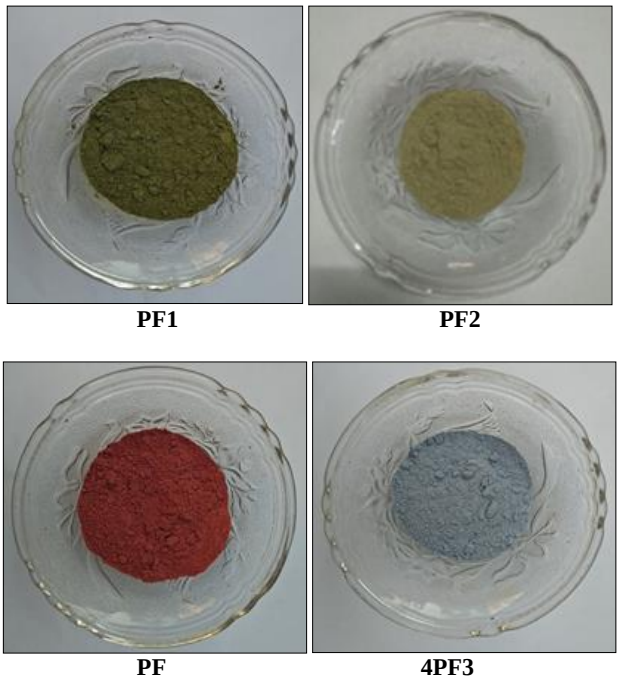

Fig 5: SSF5 Powder formulated with algae and their pigments.

4. Discussion

The importance of grains and legumes in the world is high due to their significance in human and animal nutrition. Hence different combination of grains and legume seeds were analysed for their carbohydrate, protein and fat content under varying conditions of roasting and sprouting. Among the raw formulations, RAF3 showed the balanced carbohydrate, protein and fat content. Grains and legumes, being less expensive, but rich source of good quality 40% protein and 20% fat, have been recognized as a vital ingredient for protein enrichment for ready to eat food product (Kulkarni 1994) [28]. Several workers have explored the possibilities of using soy flour with cereal flour to prepare a variety of food products of Indian taste such as snacks, baby foods, chapatti, beverages and bakery products (Rathod and Williams 1976 [43]; Tsen *et al*. 1973 [53]; Sushma *et al*. 1979 [52]; Okeiyi and Futrell 1983 [40]; Chauhan and Bains, 1985 [12]; Singh and Singh 1989 [50]; Chauhan and Santosh Kumari 1990 [13]; Vimala *et al*. 1990 [55]; Deshpande *et al*. 2004 [16]; Mridula and Wanjari, 2006 [33]). Jayasinghe *et al*. (2016) [25] formulated of nutritionally superior and low cost seaweed based soup mix powder, with balanced nutrients. The results of present study confirm the observations of Amal *et al*. (2014) [1] where they observed similar biochemical contents in the diets from dried health mixture. Satusap *et al*. (2014) [46] also formulated the cereal and legume based food products for the elderly people with the balanced nutritive value.

Roasting has a significant impact on the overall quality of grain and the final product. The roasting odour, a most important quality of powder is basically depends on the roasting temperature and time combination. Hence, roasting temperature and time combination are of great importance in grain processing for making product. While roasting at

varying time intervals decreased the content of carbohydrates, protein and fat content. Similar results were obtained in groundnuts by Uche Samuel Ndidi *et al*. (2014) [54]. Mridula *et al*. (2007) [34] states that the roasting process may affect the colour, texture of formulated powder. But an experiment conducted by Ocheme *et al*. (2015) [38], shows that during roasting, the protein content showed an increasing trend. Kavitha *et al*. (2014) [26] states that the protein, carbohydrate and fat content was gradually increased during roasting. Pearl millet, teff, cowpea, and peanut used in the formulation of experimental weaning foods were evaluated for changes in physicochemical properties resulting from roasting and malting (Griffith *et al*. 1998) [20].

Germination is an inexpensive and simple method of improving nutritive value and several studies have reported higher levels of nutrients and lower values of anti-nutrients in germinated food grains compared to the un-germinated originals. While searching for new sources of functional foods, special attention has been paid to sprouts from the legumes seeds which are more and more often used in human diets throughout the world in general and in the subcontinent in particular. In the present study among the sprouted formulations, SSF5 shows the balance nutrients. During the sprouting the protein and fat content was gradually increased but carbohydrate showed a decreasing trend. Similar results were reported by Ihsanullah Daur (2008) [24] and Dagnia *et al*. (1992) [15], who reported that germination for 6 days of *L. angustifolius* increased the protein content by about 10%. The increase in protein content after germination was also found in other legumes such as soybean (Mostafa *et al*. 1987) [32], mungbean (Mubarak 2005) [36], fenugreek (Martinez-Villaluenga 2008) [31] and dry bean, lentils, faba beans (Hsu 1980) [23]. Increase in protein content was also noted by Camacho *et al.* (1992) [9] during germination of beans, lentils, chick pea and pea's seeds. Ohtsubo *et al*. (2005) [39] found an increment in crude protein of germinated brown rice. Data regarding the effect of sprouting on the proximate composition of SSF5, agree with Obizoba (1991) [37] who reported increase in 4.0% moisture, 52.1% crude protein and 0.08 % ash. Our results were in opposite line with Badshah *et al.* (1991) [5] and Chung *et al.* (1998) [14] who noted significant losses in lipid content during canola sprouting. The decrease in fat content of seed could be due to total solid loss during soaking prior to germination (Wang *et al*. 2005) [56]. Amongst various cereals and legumes, barley and bengal gram are the most liked grain for making health powder (Mridula *et al*. 2007) [35]. Bengal gram is commercially produced in Bihar and Eastern Uttar Pradesh, which is the most liked form of health powder but in other parts of Uttar Pradesh, barley and bengal gram based health powder is preferred by the population. Earlier health mix was considered as food of the poor but now a day, it is gaining popularity amongst all because of its protein value and suitability of this product for diabetics due to lower glycemic index of bengal gram and barley (Shukla *et al*. 1991) [49]. Anonymous (2000) [3] developed an acceptable quality product from roasted flour mixture using wheat, bengal gram, defatted soyflour, groundnut and jaggery called amirtham.

Among various powder formulations of algal biomass and the chosen pigments: Phycocyanin, Chlorophyll and Carotenoids PF3 showed the balanced nutritive value when compare to others. Saraswathi *et al*. (2010) [45] formulated the multifaceted *Spirulina* health drink with the aim of targeting diabetic population. Lisiane Fernandes *et al*. (2016) [29] formulated the novel foods for physical activity practitioners using *Spirulina*. Supportive studies reports that incorporation of *Chlorella* and *Spirulina* in pasta products can enhance the nutritional and sensorial quality of pasta, without affecting its cooking and textural properties (Fradique *et al*. 2010) [19]. The stability factors elucidates PF3 to be the best that had retained its color and odor with least reduction rates of 20.8% and 21.5% although PF2 showed a better texture (31.1% reduction). Likewise the carbohydrate, protein and the fat content were retained higher in PF3 during formulation. Recent use of algae protein powder from *Chlorella* commercially available as Algavia has been used in milkshakes, cheese crackers, salad dressings, soups etc. that the whole algal protein is easily dispersed, has no precipitation, has a minimal impact on viscosity and does not require the addition of other ingredients such as stabilizers.

5. Conclusion

The formulation products via algae and algae metabolics blended with powder is a novel aspect appended to the nutritional and therapeutic value to the humans as well as targets the industrial markets aiming for production of value added consumables. Advanced studies proving the biochemical efficacy of the consumable product would be highly appreciable. The role of pigments, particularly phycocyaninin in improving the shelf life of consumables was also found to be remarkably significant in the aspects of commercial scale of production.

6. References

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