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Effect of Microalgae Incorporation on the Physicochemical, Nutritional, and Sensorial Properties of an Innovative Smoothie

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Abstract: ***Smoothies are increasingly popular as convenient and nutritious meal alternatives, particularly for health-conscious consumers. The integration of spirulina, a nutrient-dense microalga, into fruit-based smoothies provides an innovative approach to enhancing their nutritional profile. This study focuses on the formulation of a spirulina-enriched smoothie containing mango, banana, and orange, fruits chosen for their complementary flavours, rich vitamin content, and natural sweetness. The objectives of this study were to optimise the formulation to achieve a balance of sensory acceptability and nutritional enhancement while maintaining the product's appeal. Various concentrations of spirulina (1-3%) were evaluated for their impact on taste, texture, colour, and overall acceptability. The smoothie was analysed for its nutritional composition, including protein content, vitamins, minerals, and antioxidant capacity. Sensory evaluation was conducted using a trained panel to assess consumer preferences. a nutrient-dense smoothie that combines the natural sweetness of mango, banana, and orange with the nutritional benefits of spirulina. This refreshing blend provides a rich source of vitamins A and C, potassium, and fibre, as well as plant-based protein and iron from spirulina. The addition of spirulina enhances the antioxidant and anti-inflammatory properties of the smoothie, supporting overall health and well-being. Smoothie is a delicious and convenient way to incorporate the benefits of spirulina into a busy lifestyle, making it an ideal choice for health-conscious individuals seeking a nutritious and revitalising beverage.***

Keywords: Smoothie, spirulina, sensory evaluation, health conscious, beverage

I. INTRODUCTION

The growing demand for functional foods and beverages has driven innovation in the formulation of health-oriented products. Smoothies, known for their convenience and dense nutrient content, have become a preferred choice for health-conscious consumers. They offer a versatile platform for incorporating fruits, vegetables, and superfoods into the daily diet. Among superfoods, Spirulina—a blue-green microalga—is recognised for its exceptional nutritional profile. It is rich in high-quality protein, essential amino acids, iron, B vitamins, antioxidants (such as phycocyanin), and anti-inflammatory compounds. Its incorporation into foods and beverages has gained attention due to potential health benefits, including immune support, detoxification, and improved energy levels. However, Spirulina's intense green colour and distinctive earthy flavour can limit its sensory appeal, making thoughtful formulation essential. Mango (Mangifera indica) and banana (Musa spp.) are popular tropical fruits widely used in smoothies due to their natural sweetness, creamy texture, and rich content of vitamins, minerals, and fiber. They can effectively mask Spirulina's strong flavor while contributing to the smoothie's palatability and consumer acceptability. Chopped dried fruits, such as raisins, dates, apricots, and cranberries, not only add natural sweetness and appealing texture but also contribute additional nutrients like iron, potassium, and dietary fiber. Their inclusion may also reduce the need for added sugars, aligning with clean-label and lowglycemic food trends.

This study aims to develop a spirulina-enriched mango-banana smoothie with the addition of chopped dried fruits and evaluate its physicochemical, nutritional, and sensory properties. The objective is to identify an optimal formulation that balances health benefits with consumer acceptability, thus contributing to the advancement of functional beverage development.

II. LITERATURE REVIEW

The development of functional beverages has seen a significant rise due to growing consumer awareness regarding health and nutrition. Smoothies, particularly fruit-based ones, have gained popularity for their convenience, palatability, and natural richness in essential nutrients. Numerous studies have focused on improving the nutritional profile of such beverages through the incorporation of functional ingredients like microalgae, plant proteins, and dried fruits.

Spirulina (Arthrosporic platensis) is a blue-green microalga recognized for its high nutritional value, particularly its protein content (60–70%), essential amino acids, iron, beta-carotene, and phycocyanin—a potent antioxidant. According to Becker (2007), Spirulina has potential as a dietary supplement due to its bioactive components and therapeutic properties, including antioxidant, anti-inflammatory, and immunomodulatory effects. However, its incorporation in food systems is limited by its characteristic earthy taste and dark green colour (Habib et al., 2008).

In fruit-based beverages, Spirulina has been successfully added in low concentrations to enhance nutritional content without severely affecting sensory quality. Yusof et al. (2019) reported that Spirulina-enriched orange and apple smoothies were acceptable to consumers when concentrations were maintained below 1.5%. Similar findings were echoed by Mishra and Kaushik (2014), who developed Spirulina-fortified fruit juices that provided improved iron and protein levels with acceptable sensory profiles.

Mango (Mangifera indica) and banana (Musa spp.) are ideal bases for smoothies due to their natural sweetness, viscosity, and high vitamin and mineral content. Mango is a rich source of vitamin C, beta carotene, and polyphenols, while banana contributes potassium, dietary fiber, and natural sugars. Their combination helps in masking the off-flavor of functional additives like Spirulina (Giri & Prasad, 2007).

III. MATERIALS AND METHODS

Raw materials:

1) Mango powder
2) Orange juice
3) Banana puree
4) Spirulina powder
5) Honey
6) Dry fruits

IV. METHODOLOGY

To formulate a spirulina smoothie, the process begins with the selection of ingredients. The main components include mango, banana, orange, spirulina powder, and optional ingredients such as water, natural sweeteners like honey, and ice cubes. The next step is ingredient preparation, which involves washing and peeling the fruits (mango, banana, and orange), then cutting them into smaller pieces to facilitate easy blending. The required amount of spirulina powder is also measured at this stage.

In the formulation development phase, the prepared fruits are blended together in a blender along with a pre-determined amount of spirulina powder (ranging from 1–3% concentration). Water, ice cubes, or sweeteners may be optionally added to adjust the consistency and taste of the smoothie.

The optimization process involves preparing multiple formulations with varying spirulina concentrations (e.g., 1%, 2%, and 3%) and blending each mixture until smooth and consistent. Sensory evaluation is then conducted using a trained panel to assess factors such as color, flavour, aroma, texture, and overall acceptability.

Once the optimal formulation is selected, the smoothie is poured into appropriate packaging, such as bottles, and stored under refrigerated conditions. Finally, shelf-life testing is conducted to assess the product's stability and sensory quality over a designated storage period.

V. METHODS

1) Determination of Energy Value:

The energy value of the formulated product was calculated as per the method given in IS 13285:1992 (RA 2022) – Energy Foods – Specification. The energy (calorific) value was derived using the Atwater general factors, which estimate the metabolizable energy provided by macronutrients. Protein, fat, and carbohydrate contents are expressed in grams per 100 grams of the product. The constants 4, 9, and 4 represent the energy values (in kcal/g) of protein, fat, and carbohydrates, respectively. This method provides an estimate of the energy content in kilocalories per 100 grams of the product.

CALUCULATION:
The following formula was used:
Energy (kcal/100g) = (Protein × 4) + (Fat × 9) + (Carbohydrate × 4)
Determination of Carbohydrate (IS 1656:2022):
Determine moisture, protein, fat, and ash content of the sample using standard methods Like Calculate carbohydrate content by subtracting the sum of these components from 100.
CALUCULATION:
Carbohydrate (%) = 100 – (Moisture + Protein + Fat + Ash)
Determination of Protein (IS 7219:1973, RA 2020):
Procedure (Kjeldahl Method):
Weigh the sample accurately (usually 1g) and digest it with strong sulfuric acid and catalyst to convert nitrogen to ammonium sulphate. Neutralize the digest using sodium hydroxide and distil the ammonia released over a boric acid solution, Titrate the distillate against a standard acid (usually HCl or H2SO4) to determine the nitrogen content. To calculate crude protein content, multiply the result for nitrogen by a conversion factor (usually 6.38 for dairy products).
CALUCULATION:
Protein (%) = Nitrogen (%) × 6.38
Where 6.38 is the nitrogen-to-protein conversion factor for milk and milk-based products.

2) Determination of Total Sugars (Based on IS Method):
To find the total sugar content, the known weight of the sample was initially diluted with distilled water. The solution was then hydrolysed using dilute hydrochloric acid to transform all the non- reducing sugars into reducing sugars. The solution was then neutralized cautiously and filtered to eliminate any solid impurities after the hydrolysis. The resulting filtrate was titrated against Fehling's solution to approximate the overall amount of reducing sugars, which now indicates the total sugar content of the sample.
CALUCULATION:
Total Sugar (%) = Volume of sample used × Factor / Weight of sample × 100

3) Determination of Fat by Gerber Method (FSSAI Manual):
In order to estimate the total fat content, the sample was first acidified and subjected to sulfuric acid to hydrolyse proteins and liberate fat. Amyl alcohol was then added to aid in the separation of fat. The contents were transferred to a butyrometer and tightly capped. The butyrometer was centrifuged to cause the fat to separate, rising to the calibrated neck of the butyrometer. Finally, the fat layer was taken directly from the graduated scale. This method, the Gerber method, is officially recognized by FSSAI for estimating fat in milk and milk products.

4) Estimation of cholesterol (FSSAI Manual of Methods of Analysis of Foods – Oils and Fats (2021):
For analysis of the cholesterol content, a definite amount of the fat or oil sample was saponified with alcoholic potassium hydroxide to release the unsaponifiable material, i.e., cholesterol. The unsaponifiable fraction was then dissolved out with a suitable organic solvent such as ether or hexane. The solvent on evaporation left behind the residue with cholesterol, which was then purified if necessary. Cholesterol was then measured by a colorimetric assay based on reaction with Liebermann-Burchard reagent, which gave a colour that was read spectrophotometrically at a particular wavelength (most often 620 nm) and had the cholesterol concentration calculated from a standard curve calibration.
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CALUCULATION:

Cholesterol (mg/100g) = As/Astd × Cstd× v/w ×100

Where:

- As = Absorbance of sample
- Astd = Absorbance of standard
- Cstd = Concentration of standard cholesterol solution (mg/ml)
- V = Final volume of extract (ml)
- W = Weight of sample

6) Estimation of calcium content:

In order to obtain the calcium content, a specified volume of the sample solution prepared was taken and taken to around pH 12 with a suitable buffer solution such that titration conditions were optimal. Suitable indicator like Patton and Reeder's was added to the solution so that visual detection of endpoint could be made. The sample was titrated against a standard EDTA solution. As it is titrated, EDTA binds to the calcium ions of the sample. Endpoint was signalled by a distinctive change in colour that guaranteed complete titration of all the calcium ions with EDTA. By then, the amount of EDTA taken during the endpoint had been noted down. Calcium content of the sample was then calculated from the amount of EDTA solution taken, its molarity, and the weight or volume of the sample, whichever is applicable.

CALUCULATION:

Calcium (%) = (V×M×40.08) × 100 100

Where: V = Volume of EDTA solution used (in liters)

M = Molarity of the EDTA solution (mol/L)

40.08 = Molar mass of calcium (g/mol)

W = Weight of the sample (in grams)

7) Estimation of Moisture content by IS 12711:1989 (RA 2020):

For determining the moisture content, an accurately weighed known quantity of the sample was taken in a clean, dry, and pre-weighed dish for moisture. The sample was dried in a hot air oven at 105 ± 1°C for 4 to 6 hours or until constant weight is obtained. After drying, the dish was cooled in a desiccator and reweighed. The weight loss was measured as the moisture content of the sample.

CALUCULATION:

Moisture (%) = W1-W2 / W1-Wo × 100 Where:

W_0 = Weight of empty dish (g)

W_1 = Weight of dish + sample before drying (g)

W_2 =Weight of dish + sample after drying (g)

8) Determination of ash content procedure based on IS 12711:1989 (RA 2020):

For an estimation of the ash content, a definite quantity of the sample was accurately weighed and put in a pre-ignited and tared silica crucible. The sample was then burned gently over a mild flame so that it would not be lost by spattering. After initial charring, the crucible was placed in a muffle furnace and subjected to firing at 550 ± 25°C for 5 to 6 hours, or until the ash coloured white or light grey, which indicates total ashing. The crucible was cooled in a desiccator and weighed. The difference in weight before and after ashing was recorded as the total ash content of the sample.

CALUCULATION:

Ash(%) = W2-Wo × 100 W1-Wo

Where:

W_0 = Weight of empty crucible (g)

W_1 = Weight of crucible + sample before ashing (g)

W_2 =Weight of crucible + ash after ignition (g)

VI. SAMPLE FORMULATIONS

The three formulations are made using the plant-based alternatives.

INGREDIENTS	VARIATION 1	VARIATION 2	VARIATION 3
MANGO POWDER	23 gm	15 gm	30 gm
BANANA PUREE	10 ml	20 ml	15 ml
ORANGE JUICE	18 ml	20 ml	15 ml
HONEY	2 tbsp	2 tbsp	2 tbsp
DRY FRUITS	50 gm	50 gm	50 gm
WATER	50 ml	40 ml	60 ml
SPIRULINA	1 gm	1 gm	1 gm

TABLE 1 SAMPLE FORMULATIONS

TRAIL 1 TRAIL 2 TRAIL 3

FIG. 1 SAMPLE FORMULATIONS

VII. RESULTS AND DISCUSSIONS

1) Nutrient Analysis:

Test Results					
Sr. No.	Test Parameter	Unit	Specification	Result	Test Method
Table 1: Nutritional Value					
1	Energy	Kcal/100 gm	100 to 200gm	102.77	IS 13285 : 1992 (RA 2022)
2	Carbohydrates	gm/100 gm	15 to 25 gm	19.38	IS 1656 : 2022
3	Protein	gm/100 gm	2 to 10 gm	2.15	IS 7219 : 1973 (RA 2020)
4	Total Sugar	gm/100 gm	6 to 10gm	6.14	IS 6287 : 1985 (RA 2020)
5	Total Fat	gm/100 gm	0.8 to 4.5gm	1.85	FSSAI Manual
6	Cholesterol	mg/100 gm	Nil	Nil	FSSAI Manual(Oils and Fats): 2021
7	Calcium (as Ca)	mg/100 gm	50 to 100 mg	65.10	IS 5949 : 1990 (RA 2024)
8	Sodium (as Na)	mg/100 gm	10 to 15 mg	12.10	IS 9497 : 1980 (RA 2024)
9	Moisture	gm/100 gm	65 to 90 gm	76.10	IS 12711 : 1989(RA 2020)
10	Total Ash	gm/100 gm	<1.0 gm	0.52	IS 12711 : 1989(RA 2020)

TABLE 2 NUTRIENT ANALYSIS

The comprehensive analysis of the food sample has yielded promising results, affirming its nutritional value and safety for consumption. The sample exhibits a favorable nutritional profile, characterized by a moderate energy content, substantial carbohydrate and protein levels, and a notable presence of essential minerals such as calcium. Furthermore, the absence of cholesterol enhances its appeal as a heart-healthy option.

The microbiological assessment has provided assurance regarding the sample's safety, as it revealed undetectable levels of coliforms, yeast, and mold. This suggests a low risk of foodborne illnesses, thereby endorsing its suitability for human consumption.

In conclusion, the findings of this analysis substantiate the food sample's nutritional adequacy and microbiological safety, rendering it a viable choice for those seeking a healthy and safe dietary option.

2) Microbial Analysis:

Test parameter	Incubation period	Media used	Specification limit	Results (cfu/2 ml)
Coliform	35±2°C at24 hrs	Violet red bile agar	1cfu/2ml	<1 (at 24,48,72,96,and120hrs)
Total plate count (TPC)	35±2°C at48 hrs	Plate count agar	1cfu/2ml	<1 (at 24,48,72,96,and120hrs)
Yeast and mould	25±2°C at120 hrs	Potato dextrose agar	1cfu/2ml	<1 (at 24,48,72,96, and120hrs)

TABLE 3 MICROBIAL ANALYSIS

In an effort to create a healthier beverage option, we have developed a nutrient-rich smoothie by combining spirulina, a nutrient-dense blue-green microalga, with mango, banana, and orange. After testing various formulations, they identified an optimal blend (sample T2) that stood out for its exceptional nutritional profile and taste. Notably, this smoothie boasts a higher protein content (14.3%) and lower acidity (0.43% citric acid), making it a more appealing choice. To overcome the strong flavour of spirulina, the researchers added toned milk and pineapple essence, which significantly improved the smoothie's sensory appeal. Crucially, microbiological analysis confirmed the product's safety for consumption. Overall, this study showcases the potential of spirulina as a valuable ingredient in creating nutritious and enjoyable beverages, paving the way for innovative, health-promoting drinks.

VIII. SUMMARY AND CONCLUSION

A. Summary:

The present study was undertaken to formulate and evaluate a nutrient-rich, spirulina- enriched smoothie using mango, banana, and orange as the base fruits. Spirulina, a blue-green microalga, is known for its exceptional nutritional profile, particularly its high protein, iron, antioxidant, and vitamin content. The incorporation of spirulina into a fruit-based smoothie was aimed at enhancing the functional and nutritional value of the beverage without compromising sensory acceptability.

Multiple formulations were prepared by varying the concentration of spirulina (e.g., 0.5%, 1%, and 1.5%) and evaluated for their physicochemical characteristics (pH, total soluble solids, acidity, viscosity), nutritional composition (moisture, protein, carbohydrate, energy, vitamin C), and sensory attributes (color, taste, aroma, texture, and overall acceptability).

Among the different variations, the formulation with 1% spirulina was found to be the most acceptable in terms of taste, aroma, appearance, and overall sensory score, while also showing a significant improvement in protein content and antioxidant potential compared to the control. The fruity flavor of mango, banana, and orange successfully masked the mild earthy note of spirulina, making it palatable and appealing.

B. Conclusion:

The study demonstrated that spirulina can be effectively incorporated into a fruit-based smoothie to create a functional beverage with enhanced nutritional benefits. The optimal formulation balanced nutritional enhancement with consumer acceptability, offering a promising option for health-conscious individuals seeking plant-based, protein-rich drinks.

This spirulina-enriched smoothie could serve as a value-added functional product suitable for a wide range of consumers, including vegetarians, athletes, and individuals with iron or protein deficiencies. Further studies can explore shelf-life stability, fortification with probiotics or fibers, and commercial scale-up potential.

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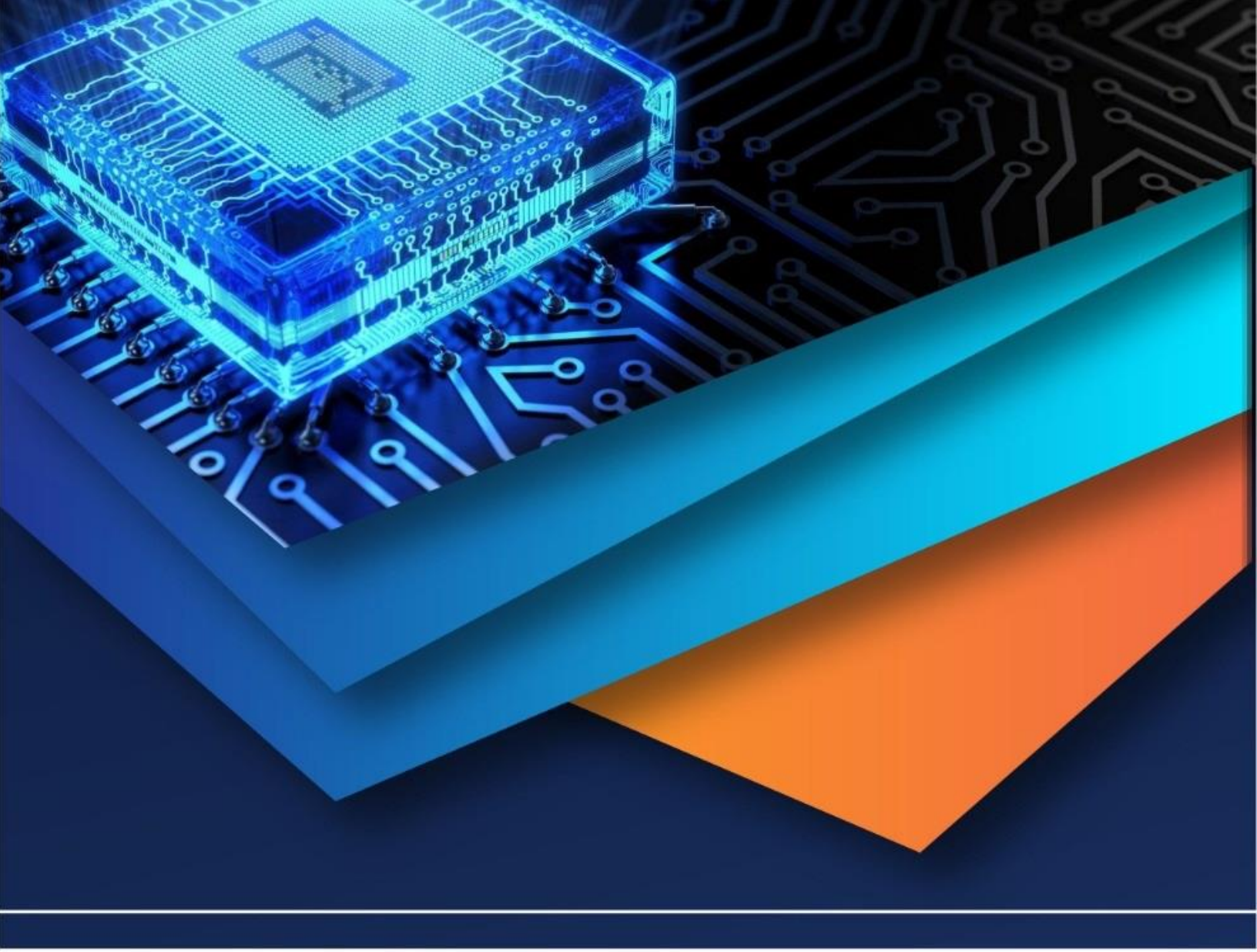

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