Journal of
Food Chemistry & Nanotechnology

https://doi.org/10.17756/jfcn.2023-s1-015

Research Article

Open Access

Optimization of Functional Beverage Using Germinated Pseudocereals

Huraiya Habib[1], Jyoti Singh[1*], Ashwani Kumar[1], Tawheed Amin[2], Tashooq Ahmad Bhat[2], Nargis Aziz[3] and Sezai Ercisli[4, 5]

[1]*Department of Food Technology and Nutrition, Lovely Professional University, Phagwara, Punjab, India*

[2]*Division of Food Science and Technology, SKUAST-K, Srinagar, Jammu and Kashmir, India*

[3]*Division of Food Science and Technology, Islamic University of Science and Technology, Awantipura, Jammu and Kashmir, India*

[4]*Department of Horticulture, Faculty of Agriculture, Ataturk University, Erzurum, Turkey*

[5]*HGF Agro, ATA Teknokent, Erzurum, Turkey*

***Correspondence to:**
Jyoti Singh
Department of Food Technology and Nutrition,
Lovely Professional University,
Phagwara, Punjab, India.
E-mail: jyotisingh9377@gmail.com

Received: September 04, 2023
Accepted: October 23, 2023
Published: October 27, 2023

Citation: Habib H, Singh J, Kumar A, Amin T, Bhat TA, et al. 2023. Optimization of Functional Beverage Using Germinated Pseudocereals. *J Food Chem Nanotechnol* 9(S1): S108-S116.

Copyright: © 2023 Habib et al. This is an Open Access article distributed under the terms of the Creative Commons Attribution 4.0 International License (CC-BY) (http://creativecommons.org/licenses/by/4.0/) which permits commercial use, including reproduction, adaptation, and distribution of the article provided the original author and source are credited.

Published by United Scientific Group

Abstract

The functional beverages market offers many opportunities for new product development (nutraceuticals, malt drink, plant-based medicines, and products) with a desirable and effective composition of nutrients and bioactive molecules aimed at delivering health benefits, improving human well-being, and appealing to consumers due to their suitability, affordability, and appealing sensory characteristics, making functional beverages an important segment of functional food products. The use of germinated pseudocereals in the production of functional beverages has recently grown in popularity due to the special nutritional content, which is high in protein, dietary fibre, β-glucan, and bioactive compounds attributed to biological effects and health advantages. The present study aimed to develop a novel functional drink utilizing a blend of cereals and germinated pseudocereals. The buckwheat and amaranth grains were germinated at 30 °C for 96 hours and dried at 50 °C for 8 hours. Oats and germinated buckwheat, amaranth have been ground into coarse flour and mixed in different amounts ranging from 100:0:0-20:80:0 and 20:0:80.30 treatment combinations (T1–T30) were prepared and each treatment were mixed with potable water in the ratio 4:1 to prepare the malt drink. Response surface methodology was used to optimize the functional drink, using malt-drink (diluted formulation of buckwheat, amaranth and oats in the ratio 40:20:40), and double toned milk (100:0 to 20:80). The optimized functional beverage had high levels of dietary fibre (2.93 ± 0.26%), including β-glucan (0.89 ± 0.43%), antioxidants like free radical scavenging activity using DPPH assay (30.65 ± 0.24%), ABTS (19.63 ± 0.36%) and total polyphenols (248.84 ± 0.51 mg GAE/100 g). The drink was low in fat (1.23 ± 0.35%), and rich in minerals like calcium (1452.6 ± 0.37 mg/L). The innovative functional drink derived from a blend of cereals and germinated pseudocereals presents a promising avenue for expanding the range of nutritious and health-promoting beverages.

Keywords

Cereal, Pseudocereals, Oats, Malt, Functional beverage, Optimization

Introduction

In recent years, the pursuit of healthier dietary choices has led to an increased interest in functional foods and beverages that offer not only sustenance but also potential health benefits [1]. Cereals and pseudocereals, staples in the human diet for centuries, have gained attention for their nutritional richness and potential health-promoting compounds. Among the emerging strategies to enhance the nutritional content of these grains, the process of germination has shown promise

[2]. Germination involves the controlled sprouting of seeds, leading to biochemical changes that increase nutrient availability and bioactive compounds [3]. In line with this, the development of a novel functional drink by harnessing the combined nutritional power of cereals and germinated pseudocereals presents an exciting opportunity to create a convenient and palatable beverage that not only quenches thirst but also delivers a myriad of health benefits [4].

Pseudocereals are grains that lack gluten-forming proteins, making them suitable for gluten-free diets. Gluten, present in wheat, barley, and rye, triggers adverse reactions in those with celiac disease or gluten sensitivity [5]. Buckwheat is a pseudocereal known for high-quality proteins without gluten and balanced amino acids. Notably rich in lysine and arginine, buckwheat offers 11-19% protein content. Its 13S globulin, a rare vegetable protein, aids in lowering blood cholesterol. Buckwheat also provides essential fatty acids and oxidation-resistant lipids. Amaranth, another pseudocereal, boasts 4-8% dietary fiber surpassing typical cereals (2%). Its highly digestible proteins, approximately 90%, are rich in essential amino acids aligned with World Health Organization recommendations. Lysine content matches that of soy, countering cereal limitations. Notably, amaranth proteins excel in sulfur-containing amino acids (2-5%)-methionine and cysteine surpassing basic legumes averaging 1.4%. These pseudocereals offer nutrient-packed alternatives with unique benefits for those seeking gluten-free and nutritionally balanced diets [6, 7].

The (1-3), (1-4) β-D-glucans, commonly known as beta-glucans, are types of cell wall polysaccharides found in cereal grains, with oats being a particularly rich source [8]. The structure of cereal β-glucans primarily consists of cellotriosyl (a chain of three glucose units) and cellotetraosyl (a chain of four glucose units) oligomers, which are interconnected via β-(1 → 3) glycosidic linkages [9]. This arrangement of glucose units and linkages gives cereal β-glucans their unique properties and functional benefits [10]. The rheological characteristics of cereal β-glucans, such as viscosity enhancement and gelling properties, make them suitable for potential use as a food hydrocolloid. The viscosity enhancement and gelling properties of cereal β-glucans can offer several advantages in food applications like texture improvement, stability enhancement, fat and calorie reduction and emulsion stabilization [11].

As both cereals and pseudocereals are recognized for their high carbohydrate content, providing a reliable source of energy [12]. Furthermore, they are rich in essential nutrients such as vitamins, minerals, and dietary fibers, which contribute to overall health and well-being [12]. However, the full nutritional potential of these grains is often hindered by certain antinutritional factors and limited nutrient bioavailability. In this context, germination emerges as a transformative process that not only mitigates antinutritional factors but also enhances the bioavailability of key nutrients, making them more accessible for human consumption [13]. The phenomenon of germination triggers enzymatic activities that break down complex compounds into simpler forms and this enzymatic activity not only reduces antinutritional components like phytic acid and tannins but also leads to an increase in certain vitamins and minerals [14]. Moreover, germination promotes the synthesis of bioactive compounds such as antioxidants and phytochemicals, which have been associated with various health benefits, including improved oxidative stress management and disease prevention. By incorporating germinated pseudocereals into a functional beverage, it is possible to deliver these bioactive compounds in an easily consumable form [15].

Pseudocereal malt contains elevated levels of protein, carbohydrates, dietary fiber, and essential minerals (like calcium, iron, magnesium, and zinc), along with vitamins such as vitamin E and B-complex vitamins. These enhanced nutrients compared to non-malted forms contribute to potential health benefits associated with pseudocereals [16]. Pseudocereals are rich in antioxidant compounds. Incorporating them in brewing significantly amplifies the final beverage's antioxidant activity [17]. Functional beverages are categorized as dairy-based, fruit and vegetable-based. These beverages address diverse needs and lifestyles, ranging from energy boosting to combating aging, fatigue, stress, and targeted disease management [12]. Recent and continuing shifts in consumer lifestyles are influenced by globalization, economic growth, advances in food science and technology, and personal lifestyle choices and religious considerations [18]. Beverages serve as an effective medium to transport nutrients and bioactive compounds into the body, enhancing their bioavailability. The global trend in functional beverage consumption is evident due to their nutrient-rich content, convenient packaging, design, portability, storage ease, and shelf-stable nature [19]. This trend reflects changing consumer preferences and evolving lifestyle patterns.

Creating innovative products from cereals and germinated pseudocereals, such as functional drinks, has the potential to significantly increase their consumption. To achieve this, a process was refined to create optimized functional beverages. While cereals and germinated pseudocereals inherently possess valuable available properties. In this study, the product's quality parameters were improved by intervening in the process using factors like malt drink, double-toned milk, and sugar. These interventions were aimed at enhancing the sensory properties of the beverage. The influence of each process parameter on the product's quality attributes was examined through response surface methodology. This approach can provide valuable insights to researchers regarding the impact of these parameters on the quality of similar beverages.

Materials and Methods

Raw material

The amaranth and buckwheat were procured from Himalayas of Gurez Bandipora, Jammu and Kashmir, India and oats was procured from local market. Rolled oats of brand Quaker double toned milk and table sugar were procured from the Law gate area, Lovely Professional University, Jalandhar, Punjab, India.

Germination

The buckwheat and amaranth grains were segregated into various groups based on germination temperatures and durations. Prior to utilization, the grains underwent meticulous cleaning and disinfection procedures. A 1% sodium hy-

pochlorite solution was employed, submerging the grains for 15 minutes. Following a thorough rinse with distilled water, the grains were soaked in 1 L of distilled water at 20 °C for a period of 12 hours. Subsequently, the soaked grains were drained and evenly distributed on germination paper placed on aluminum trays. The germination process occurred in darkness within a seed germinator (Scienocraft) under conditions of 30 °C temperature and 90% humidity. To maintain moisture, intermittent water sprinkling was applied to the grains during germination. Following the germination phase, the grains were subjected to an 18-hour drying period in a hot-air oven maintained at 50 °C. After drying, the grains were finely ground using a mechanical grinder, sifted through a sieve, and then stored in HDPE bags for further analysis.

Preparation of malt drink

According to the preliminary research, oat groats, germinated buckwheat, amaranth, and were milled individually to coarse flour. Malt was prepared as follows, in the first step, the ratio of buckwheat, amaranth and oats was standardized for the development of malt drink. To optimize the content of buckwheat, amaranth and oats, 30 treatments comprising of malted buckwheat flour, malted amaranth flour and oat flour were taken. 30 ratios of buckwheat, amaranth and oats ranging from 100:10 each, T1–T30 were prepared (Table 1). To these flour mixes, drinking water was added in the ratio of 4:1. The next step was mashing, which was done at various temperature-time combinations to facilitate the enzymatic breakdown of the starch present in the flour (Figure 1). After mashing, liquid mixture was strained to remove larger particles and obtain a clearer liquid with the help of muslin cloth. Once the liquid was strained, it was subjected to ultra-tech high-speed stirrer at a speed of 10,000 rpm for a duration of 10 minutes [20]. The sensory scores of the malt beverage samples were assessed using a sensory profiling scale with seven-point ranges, with a low score (for example, 1) denoting a negative attribute or low intensity and a high score (for example, 7) denoting a positive attribute or high intensity (Figure 2). Consistency, color, flavor, stickiness, bitterness, and odor were among the many factors taken into account.

Preparation of functional drink

The lowest and upper sensory acceptable ratios of double-toned milk and malt beverages were determined during preliminary studies. Utilizing response surface methodology, the optimization of the functional drink took place. The independent variables encompassed malt drink proportions derived from the prior experiment (40–60%), double-toned milk (30–50%) and sugar (5-10%). Dependent factors, including total soluble solids (TSS), total solids and overall acceptability (evaluated through sensory analysis), were considered. For optimizing the functional drink, a total of 20 trials were conducted. Based on the obtained responses, the software predicted the optimal combinations of malt drink, double-toned milk and sugar aiming to create a functional drink that maximizes TSS, total solids, overall acceptability, and pH.

Physio-chemical properties of optimized functional drink

TSS: TSS was determined by a portable refractometer (0 - 32 °Brix) given by Kumar et al. [20].

Table 1: Treatment combinations of buckwheat, amaranth and oats for the development of malt drink.

Treatments	Oats (g)	Buckwheat (g)	Amaranth (g)	Water (ml)
T1	100	0	0	400
T2	90	0	10	400
T3	90	10	0	400
T4	80	10	10	400
T5	80	0	20	400
T6	80	20	0	400
T7	70	10	20	400
T8	70	20	10	400
T9	70	30	0	400
T10	70	0	30	400
T11	60	20	20	400
T12	60	30	10	400
T13	60	10	30	400
T14	60	40	0	400
T15	60	0	40	400
T16	50	20	30	400
T17	50	30	20	400
T18	50	0	50	400
T19	50	50	0	400
T20	40	30	30	400
T21	40	40	20	400
T22	40	0	60	400
T23	40	60	0	400
T24	30	40	30	400
T25	30	30	40	400
T26	30	0	70	400
T27	30	70	0	400
T28	20	40	40	400
T29	20	0	80	400
T30	20	80	0	400

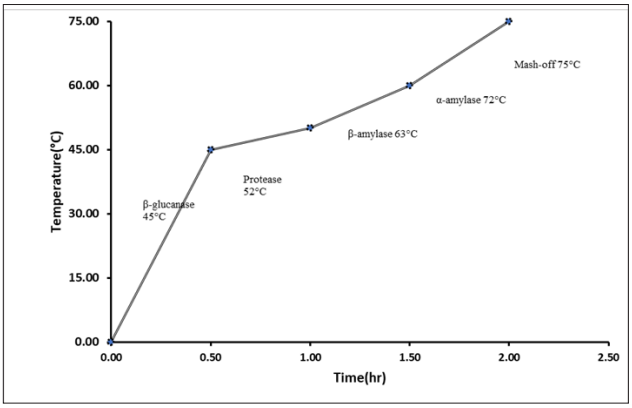


Figure 1: Enzyme action during mashing.

pH: After calibrating, the pH was determined using an Oakton digital pH meter (USA) by the method given by Kumar et al. [20].

Titrable acidity: 100 ml of boiling water and 5 ml functional drink samples were combined, and the mixture was then titrated with 0.1 M NaOH until the phenolphthalein indicator reached the pale pink end point [21].

Total solids: The weight of dried residue left was measured by evaporating the moisture from a weighed sample at a temperature of 70 °C to calculate the total solids [21].

Moisture, protein, fat, ash, carbohydrate, β- glucan content was determined as per the procedures of AACC 2000, dietary fiber (dietary fiber kit assay by Megazyme International Ireland, Ltd. 2015).

Bioactive compounds functional drink

Total phenols: The total phenols were estimated by the method given by Kumar et al. [21].

Antioxidant activity: Free radical scavenging activity using DPPH assay (2, 2-diphenyl-1-picrylhydrazyl), ABTS (2, 2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) were carried out following the procedures [22].

Viscosity

The viscosity was measured using rotational viscometer (Anton paar ViscoQC 300). Samples were put in a 500 ml beaker and viscosity readings taken on the viscometer at 200 rev/min by using Brookfield Spindle no. RH6 [20].

Measurement of color

Utilizing the Hunter color LAB system (Ultra-Scan, Hunter Lab, USA), the determination of color was accomplished, and the color value was expressed as 'L' (lightness 0–100), 'a' (+ redness, – greenness), and 'b' (+ yellowness, – blueness) [20].

Mineral analysis

The determination of mineral content was conducted using thermo electron inductively coupled plasma atomic emission spectrometry. In a conical flask, one gram of dry sample was placed, followed by the addition of 10 ml of diacid (a mixture of nitric acid and perchloric acid in a ratio of 3:1). This mixture was allowed to stand overnight, as outlined in the method by Kumar et al. [20]. Subsequently, digestion of the mixture was carried out until white fumes became visible.

Statistical analysis

The optimization of the functional beverage was done by using Design expert version 12 software through response surface methodology. Experimental analysis was carried out in triplicates and one-way ANOVA with randomized design was used to determine the data statistically followed by Duncan multiple range to measure the differences among various variables.

Results and Discussion

Screening of malt drink

The sensory profiling results are illustrated in figure 2, revealing distinctive characteristics of buckwheat, amaranth, and oats. Each ingredient showed its own advantages and disadvantages in relation to the others. Amaranth, unfortunately, exhibited an unfavorable odor that was largely unappealing. This undesirable attribute was attributed to its elevated phenols and tannins content, contributing to a pungent and bitter taste profile [23]. In contrast, oats and buckwheat brought positive

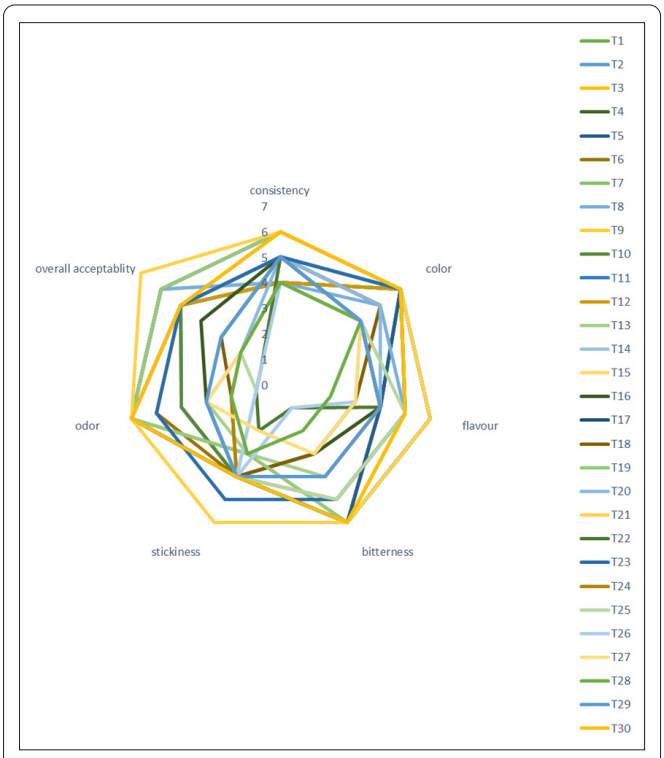


Figure 2: Sensory profiling of different treatment combinations of malt drink.

attributes such as a smooth texture and appealing color to the drink [24]. However, excessive quantities of these components led to an adverse effect, resulting in stickiness and an overly starchy flavor that was perceived as unfavorable. Moreover, oats and buckwheat managed to mitigate the unwanted odor and flavor of amaranth to a considerable degree. Considering these diverse sensory attributes, the optimal combination of buckwheat, amaranth, and oats in a ratio of 40:20:40 was optimized for formulating the composite drink.

Optimization of variables for the preparation of the functional drink

In table 2, the impact of three variables-namely, malt drink, double toned milk, and sugar-on several responses, including TSS, total solids, pH, and overall acceptability, is outlined. To optimize the process parameters and achieve the most favorable product composition, these factors and responses were meticulously chosen. A central composite design was employed to develop a desirability function. This function integrated the desired objectives, aiming to enhance the overall quality of the product. By utilizing this methodology, software calculated optimal proportions for blending double toned milk, sugar, and malt drink to attain the best possible overall acceptability and quality attributes. To ascertain the efficacy of the developed conditions and the experiment's overall effectiveness, a comparison was made between the predicted values and actual values of the parameters (Table 3). This analysis aimed to gauge the extent to which the experiment's outcomes aligned with predictions and met the desired goals.

Effect of variables on the TSS of the functional drink

TSS are widely utilized in the food and beverage industry to gauge the sweetness and overall quality of liquid or semi-liq-

Table 2: Effect of treatment combinations (malt drink: double toned milk: sugar) on dependent factors for optimization of functional drink.

Process variables				Responses			
Run	**Malt drink**	**Double toned milk**	**Sugar**	**TSS (°Brix)**	**Total solids**	**pH**	**Overall acceptability**
1	52.5	42.5	11.7	19	25.12	6.5	7.9
2	52.5	42.5	7.5	15	21.08	6.16	8.3
3	65	55	5	15.2	22.45	6	8.5
4	52.5	21.48	7.5	15	20.49	5.87	7.5
5	65	30	5	14.55	20.56	5.88	7.5
6	65	30	10	17.98	23.92	6	7.6
7	40	55	5	13	19.15	6.07	8.4
8	31.48	42.5	7.5	15.58	21.14	6.11	8.2
9	73.52	42.5	7.5	17.2	24.23	5.93	7.6
10	40	30	10	17.26	23.26	6.12	7.4
11	52.5	42.5	7.5	15	21.17	6.11	8.4
12	52.5	42.5	7.5	15.5	21.47	6.19	8.4
13	52.5	63.52	7.5	14.55	19.98	7	8.6
14	40	55	10	18.26	24.45	6.65	8
15	65	55	10	18	24.02	6.5	8
16	40	30	5	12.5	18.35	6	8
17	52.5	42.5	7.5	15.3	21.58	6.01	8
18	52.5	42.5	3.3	12.5	18.45	5.9	7.6
19	52.5	42.5	7.5	15.5	21.79	6.01	8.1
20	52.5	42.5	7.5	15.5	22.01	6.01	8.1

uid products, such as juices, fruits, vegetables, and more. TSS serves as a crucial indicator of flavor and sweetness in various foods and beverages [25]. Within the functional drink context, the TSS content varied between 12.5 and 19. Statistical regression analysis was conducted to establish mathematical models based on the experimental data. Notably, the blend consisting of 52.50% malt drink and 42.50% double toned milk along with sugar (11.70%) produced the highest total solid content, registering at 19 ^{0}Brix (Table 2). The significance of the fitted quadratic model's coefficients was assessed using both F-test and p value methodologies. Figure 3 visually illustrates the impact of malt drink, sugar and double-toned milk on the TSS content of the functional beverage. Figure 3 makes it clear that increasing the malt drink content in the 40% to 60% range results in a gradual rise in the TSS content of the functional drink. This increase in TSS is likely attributed to the elevated sugar content present in malt drinks [26]. This might be due to the malting process, involving germination, prompting the conversion of starch into free sugars. Consequently, this transformation results in heightened TSS within the functional drink [27]. The predicted model can be described by the following equation.

$$TSS = +15.29 + 0.54A + 0.10B + 1.99C \quad (1)$$

Effect of variables on the total solids of the functional drink

Total solid content can indicate the quality and consistency of the cereal product. Variations in total solids can affect the taste, texture, and overall quality of the cereal [28]. Carrying out regression analysis, mathematical models were fitted to the experimental data. The significant quadratic model obtained from regression analysis for TS was developed as follows:

$$\text{Total solids} = 4.59 + 0.30A - 0.11B - 0.022C \quad (2)$$

Table 3: Predicted values and actual values of the developed functional drink.

Values	TSS (^{o}Brix)	Total solids (%)	pH	Overall acceptability	Desirability
Predicted values	17.55	23.58	6.47	8.19	0.81
Actual values	15.41	6.41	6.41	7.92	
Variation (%)	0.11	0.13	0.03	0.03	

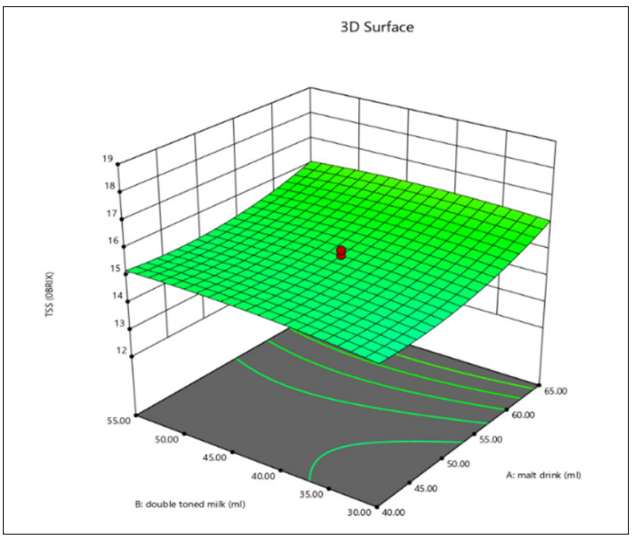


Figure 3: Effect of malt drink and double toned milk on TSS.

The total solids content of the beverage exhibited a range of 18.35% to 25.62% (Table 2). Notably, at a blend composition of 60% malt drink and 40% double-toned milk, the highest total solid content was achieved, measuring at 25.8%. Figure 4 provides a visual representation of how malt drink, sugar and double-toned milk impact the total solids content of the functional beverage. A gradual rise in the total solids content of the functional drink is observed, as indicated by the trend depicted in figure 4, by augmenting the malt drink content from 40% to 60%. It's noteworthy that double toned milk exhibited a significant negative impact on the total solid content of the functional drink. This suggests that the presence of double toned milk tends to lower the total solid content. The heightened total solid content observed can likely be attributed to the elevated solids content present in the malt drink. Therefore, with an increase in malt drink content, the total solids content of the functional drink witnessed an increment [20].

Effect of variables on the pH of the functional drink

pH is a measure of the acidity or alkalinity of a solution. It's an important parameter to consider in various food and beverage products, including cereal-based drinks [29]. The pH of a cereal-based drink can affect its taste, stability, shelf life, and overall consumer acceptability [30]. In some cases, the pH of a cereal-based drink can affect its texture or mouthfeel. For example, a slightly acidic pH might contribute to a smoother mouthfeel.

The pH of drink was found in the range of 5.87 to 7 (Table 2). The quadratic model obtained from regression analysis for pH was developed as follows:

$$pH = +6.08 - 0.0558A + 0.2285B + 0.1705C \quad (3)$$

It could be seen from the figure 5 that there was gradual decrease in pH with an increase in malt drink because malt drinks are slightly acidic due to the presence of organic acids, and other acidic compounds that are naturally present or may be introduced during the brewing or manufacturing process [31]. The pH of a malt drink can influence its flavor, stability, and overall sensory perception.

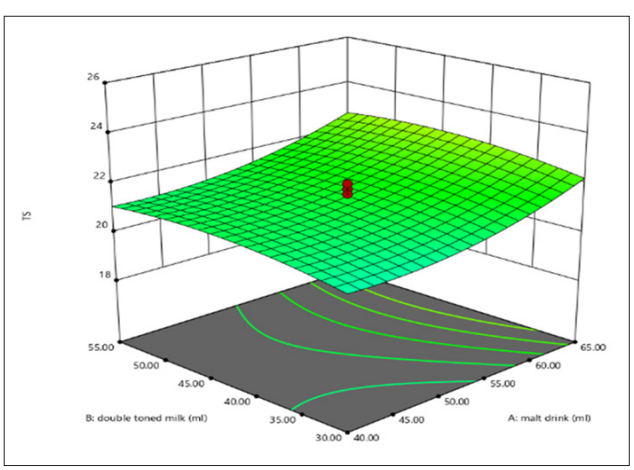


Figure 4: Effect of malt drink and double toned milk on total solids.

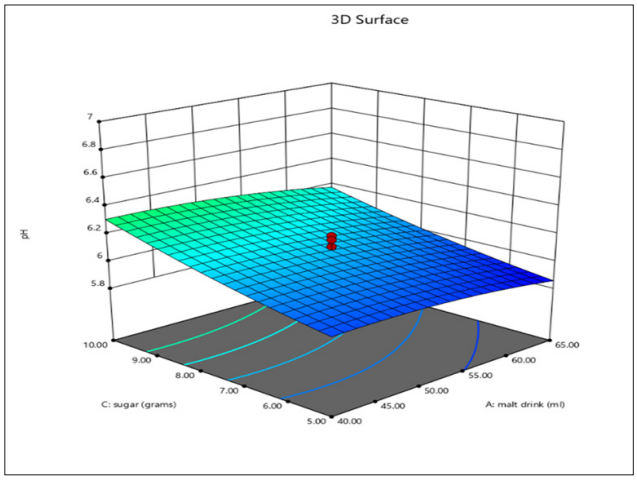


Figure 5: Effect of malt drink and double toned milk on pH.

Effect of variables on the overall acceptability of the functional drink

The overall acceptability of a drink, including cereal-based drinks, refers to how well the product is received and liked by consumers in terms of taste, appearance, aroma, texture, and other sensory attributes [32]. The significant quadratic model obtained from regression analysis for overall acceptability was developed as follows:

$$\text{Overall acceptability} = +8.22 - 0.0885A + 0.3112B - 0.0656C \quad (4)$$

Figure 6 shows with an increase in double toned milk up to 55% and sugar up to 10% there was an increase in overall acceptability of the functional drink [20]. The pleasant malt aroma at this concentration might be responsible for this effect. Increasing the concentration of amaranth drink resulted in amaranth flavor and odor, but a higher milk concentration suppressed the amaranth odor [33].

Optimization of process for the preparation of synbiotic drinks

Preparation of functional drink/base-product

The optimum levels of malt (65 ml), double toned milk

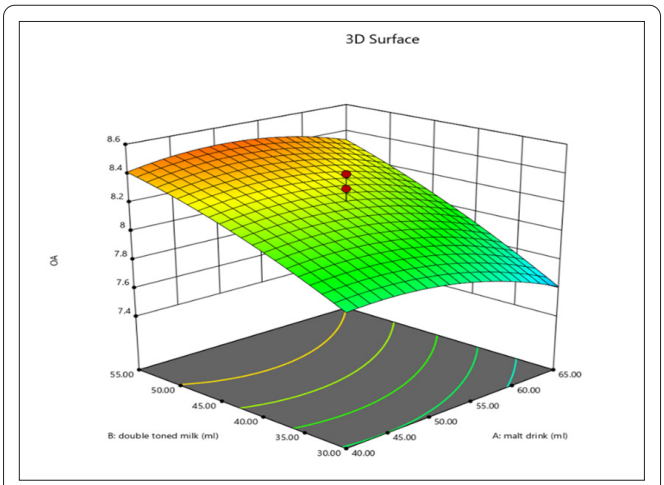


Figure 6: Effect of malt drink and double toned milk on overall acceptability.

(55 ml) and sugar (5 g) predicted through response surface methodology were used for the preparation of functional drink (Figure 7). A standard method as described with some modifications, was followed for its preparation [20].

Physicochemical composition of the functional drink

Table 4 depicts the nutritional composition of the functional drink and sweetened double-toned milk. Pseudocereal based functional drink had a total solid content of 23.44 ± 0.28% as compared to 17.33 ± 0.60% in sweetened double toned milk used as control. The TSS, sugar content and total carbohydrate content of the functional drink was 15.22 ± 0.63 °Brix, 6.23 ± 0.16% and 17.76 ± 0.23%, respectively, while that of control was 13.74 ± 0.11 °Brix, 5.06 ± 0.13% and 12.14 ± 0.83%, respectively. Functional drink showed significantly higher values of total solids, TSS, sugars and total carbohydrates, which might be due to the presence of pseudocereal malt because malting breaks down the starch to free sugar molecules, which results in higher total solids and thus enhanced flour solubility, higher TSS and total carbohydrate and eventually in the functional drink [34]. The protein content of the functional drink was found (4.16 ± 0.36%) This might be because buckwheat and amaranth grains have a high amount of protein content which was further enhanced by the process of malting/germination [35].

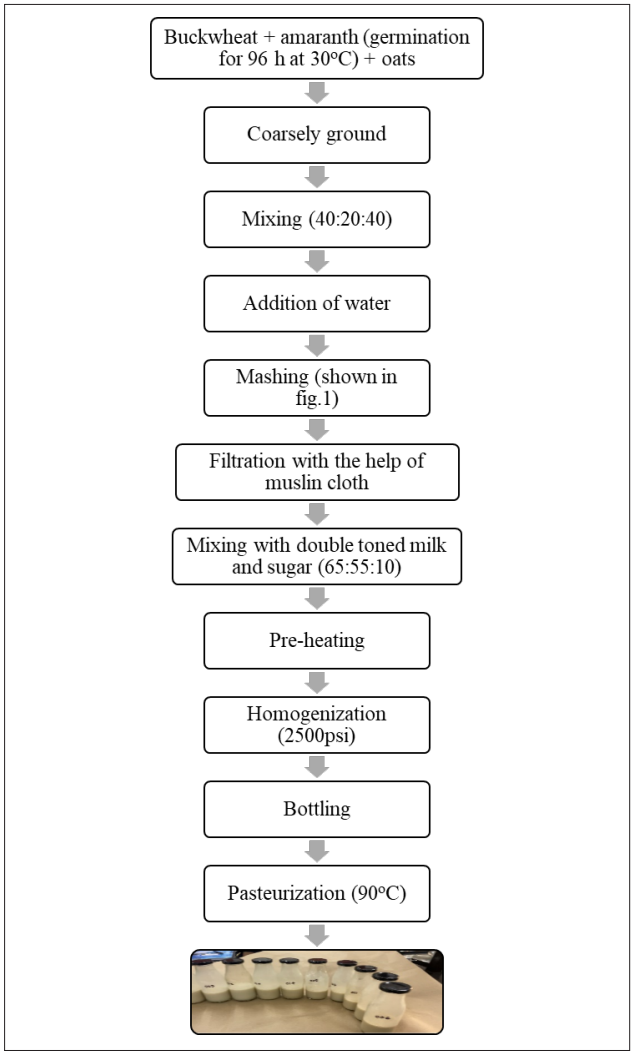


Figure 7: Flow chart of preparation of functional drink.

Table 4: Physiochemical and nutritional composition of laboratory prepared functional drink.

Parameters	Values
TSS (°Brix)	15.22 ± 0.63
Total solids (%)	23.44 ± 0.28
Sugars (%)	6.23 ± 0.16
Total carbohydrates (%)	17.76 ± 0.23
Fat (%)	1.23 ± 0.35
Protein (%)	4.16 ± 0.36
Ash (%)	0.83 ± 0.13
pH	6.01 ± 0.11
Energy (kcal/100 g)	98.81 ± 0.22
Total phenol content (mgGAE/100 g)	248.84 ± 0.51
Free radical scavenging activity (DPPH %)	30.65 ± 0.24
Antioxidant activity (ABTS %)	19.63 ± 0.36
β-Glucan (%)	0.89 ± 0.43
Dietary fibre (%)	2.93 ± 0.26
Viscosity	7.89 ± 0.74
L*	76.49 ± 0.96
a*	−1.83 ± 0.4
b*	11.52 ± 0.39

The ash content of the functional drink was found to be 0.83 ± 0.13% which might be due to malt addition possibly and higher mineral content of buckwheat and amaranth [36]. The total fat content of the functional drink was found to be 1.23 ± 0.35%. The fat content of germinated buckwheat and amaranth decreased by germination and, moreover, the mixture was diluted to 4 times with potable water for the preparation of the malt [37]. This could possibly result in lower fat content of the malt, and hence, of the functional drink. The addition of malt to double toned milk enhanced the total phenolic content (248.84 ± 0.51 mg GAE/100 g) and free radical scavenging activity using (DPPH 30.65 ± 0.24% and ABTS 19.63 ± 0.36%) of the functional drink.

Buckwheat, amaranth and oats are rich source of phenolic compounds and antioxidants [7, 20] and germination further enhanced the antioxidant and total phenolic content of the grains [38]. The malt addition also increased the β-glucan (0.89 ± 0.43%) and dietary fiber (2.93 ± 0.26%) content in the functional drink [20]. This is because the malt had higher dietary fiber and β-glucan content due to buckwheat, amaranth and oats [39]. Possibly due to the transfer of cereal solids like sugars and starch into the malt drink, the viscosity of the functional drink increased [40]. Substituting double-toned milk with malt drink resulted in notable alterations in color values. The addition of malt drink led to a decrease in lightness and an increase in both redness and yellowness. These changes in lightness and the 'a' and 'b' values of the functional drink were associated with the dark brown color of the malt drink [20].

Minerals

Minerals play a significant role in human nutrition and are required for the proper maintenance of various physiochemical functions that are important to life [41].The functional drink was analyzed for minerals like iron, calcium, potassium, phosphorus, magnesium and zinc, and the results were presented in table 5. The iron content of the functional drink was 163.54 mg/L calcium (1452.6 mg/L), potassium (104 ± 0.26 mg/lit) phosphorus (17.28 ± 0.21 mg/L) magnesium (7.65 ± 0.33 mg/L) and zinc (4.125 ± 0.41 mg/L). All these minerals present in the functional drink are important for various metabolic functions and hence, functional drink may offer a good mineral source. A significant increase in major mineral content was observed upon the addition of malt drink [20].

Table 5: Mineral content of functional drink.

Mineral (mg/L)	Values
Iron	163.54 ± 0.35
Potassium	104 ± 0.26
Phosphorus	17.28 ± 0.21
Calcium	1452.6 ± 0.37
Magnesium	7.65 ± 0.33
Zinc	4.125 ± 0.41

Conclusion

In conclusion, this study successfully developed a novel functional drink by harnessing the nutritional advantages of cereals and germinated pseudocereals. Through a meticulous process of germination, drying, and blending, a range of treatment combinations were formulated. Employing response surface methodology, the optimal composition was determined, leading to the creation of a malt drink containing a balanced blend of buckwheat, amaranth, and oats. The optimized drink exhibited remarkable qualities, boasting substantial dietary fiber content (2.93%), including notable β-glucan (0.89%) content. Furthermore, its antioxidant capacity, as measured by free radical scavenging activity using DPPH (30.65%) and ABTS (19.63%) assays, showcased its potential health benefits. The drink's richness in total polyphenols (248.84 mg GAE/100 g) further underlined its potential contribution to overall well-being. Impressively, the drink retained its low-fat profile (1.23%), while also offering a noteworthy supply of essential minerals, prominently calcium (1452.6 mg/L). By introducing this innovative beverage derived from a thoughtful fusion of cereals and germinated pseudocereals, this study opens up an exciting avenue for the diversification of nourishing and health-enhancing drink options. The concoction's diverse nutritional composition, highlighted by its fiber, antioxidant, and mineral content, makes it a promising candidate for those seeking both flavor and wellness in their beverages.

Acknowledgement

None.

Conflict of Interest

None.

References

1. García-Oliveira P, Fraga-Corral M, Pereira AG, Prieto MA, Simal-Gandara J. 2022. Solutions for the sustainability of the food production and consumption system. *Crit Rev Food Sci Nutr* 62(7): 1765-1781. https://doi.org/10.1080/10408398.2020.1847028
2. Ram A, Thattantavide A, Kumar A. 2023. Re-emergence of Psedocereals as Superfoods for Food Security and Human health: Current Progress and Future Prospects. In Kumar A, Singh P, Singh S, Singh B (eds) Wild Food Plants for Zero Hunger and Resilient Agriculture. Plant Life and Environment Dynamics. Springer, Singapore, pp 207-236.
3. Rollán GC, Gerez CL, LeBlanc JG. 2019. Lactic fermentation as a strategy to improve the nutritional and functional values of pseudocereals. *Front Nutr* 6: 98. https://doi.org/10.3389/fnut.2019.00098
4. Graziano S, Agrimonti C, Marmiroli N, Gullì M. 2022. Utilisation and limitations of pseudocereals (quinoa, amaranth, and buckwheat) in food production: a review. *Trends Food Sci Technol* 125: 154-165. https://doi.org/10.1016/j.tifs.2022.04.007
5. Ranftler C, Röhrich A, Sparer A, Tschegg C, Nagl D. 2022. Purified clinoptilolite-tuff as an efficient sorbent for gluten derived from food. *Int J Mol Sci* 23(9): 5143. https://doi.org/10.3390/ijms23095143
6. Malik M, Sindhu R, Dhull SB, Bou-Mitri C, Singh Y, et al. 2023. Nutritional composition, functionality, and processing technologies for amaranth. *J Food Process Preserv* 2023: 1753029. https://doi.org/10.1155/2023/1753029
7. Dabija A, Ciocan ME, Chetrariu A, Codină GG. 2022. Buckwheat and amaranth as raw materials for brewing, a review. *Plants* 11(6): 756. https://doi.org/10.3390/plants11060756
8. Wouk J, Dekker RFH, Queiroz EAIF, Barbosa-Dekker AM. 2021. β-glucans as a panacea for a healthy heart? Their roles in preventing and treating cardiovascular diseases. *Int J Biol Macromol* 177: 176-203. https://doi.org/10.1016/j.ijbiomac.2021.02.087
9. Peesapati S, Sajeevan KA, Patel SK, Roy D. 2021. Relation between glycosidic linkage, structure and dynamics of α-and β-glucans in water. *Biopolymers* 112(5): e23423. https://doi.org/10.1002/bip.23423
10. Shoukat M, Sorrentino A. 2021. Cereal β-glucan: a promising prebiotic polysaccharide and its impact on the gut health. *Int J Food Sci Technol* 56(5): 2088-2097. https://doi.org/10.1111/ijfs.14971
11. Bangar SP, Sandhu KS, Trif M, Manjunatha V, Lorenzo JM. 2022. Germinated barley cultivars: effect on physicochemical and bioactive properties. *Food Anal Methods* 15: 2505-2512. https://doi.org/10.1007/s12161-022-02311-5
12. Rawat M, Varshney A, Rai M, Chikara A, Pohty AL, et al. 2023. A comprehensive review on nutraceutical potential of underutilized cereals and cereal-based products. *J Agric Food Res* 12: 100619. https://doi.org/10.1016/j.jafr.2023.100619
13. Raungrusmee S. 2023. Underutilized Cereals and Pseudocereals' nutritional Potential and Health Implications. In Anal AK (ed) Pandemics and Innovative Food Systems. CRC Press, Boca Raton, pp 163-193.
14. Lan Y, Zhang W, Liu F, Wang L, Yang X, et al. 2023. Recent advances in physiochemical changes, nutritional value, bioactivities, and food applications of germinated quinoa: a comprehensive review. *Food Chem* 426: 136390. https://doi.org/10.1016/j.foodchem.2023.136390
15. Aloo SO, Ofosu FK, Oh DH. 2023. Effect of germination on alfalfa and buckwheat: phytochemical profiling by UHPLC-ESI-QTOF-MS/MS, bioactive compounds, and *in-vitro* studies of their diabetes and obesity-related functions. *Antioxidants* 10(10): 1613. https://doi.org/10.3390/antiox10101613
16. Martínez-Villaluenga C, Peñas E, Hernández-Ledesma B. 2020. Pseudocereal grains: nutritional value, health benefits and current applications for the development of gluten-free foods. *Food Chem Toxicol* 137: 111178. https://doi.org/10.1016/j.fct.2020.111178
17. Terpinc P, Cigić B, Polak T, Hribar J, Požrl T. 2016. LC-MS analysis of phenolic compounds and antioxidant activity of buckwheat at different stages of malting. *Food Chem* 210: 9-17. https://doi.org/10.1016/j.foodchem.2016.04.030

18. de Palma A, Vosough S, Liao F. 2022. An overview of effects of COVID-19 on mobility and lifestyle: 18 months since the outbreak. *Transp Res Part A Policy Pract* 159: 372-397. https://doi.org/10.1016/j.tra.2022.03.024
19. Pérez-Marroquín XA, Estrada-Fernández AG, García-Ceja A, Aguirre-Álvarez G, León-López A. 2023. Agro-food waste as an ingredient in functional beverage processing: sources, functionality, market and regulation. *Foods* 12(8): 1583. https://doi.org/10.3390/foods12081583
20. Kumar A, Kaur A, Tomer V, Rasane P, Gupta K. 2020. Development of nutricereals and milk-based beverage: process optimization and validation of improved nutritional properties. *J Food Process Eng* 43(1): e13025. https://doi.org/10.1111/jfpe.13025
21. Kumar A, Kaur A, Tomer V, Gupta K, Kaur K. 2021. Effect of rose syrup and marigold powder on the physicochemical, phytochemical, sensorial and storage properties of nutricereals and milk-based functional beverage. *J Am Coll Nutr* 40(2): 133-140. https://doi.org/10.1080/07315724.2020.1744487
22. Singh J, Inbaraj BS, Kaur S, Rasane P, Nanda V. 2022. Phytochemical analysis and characterization of corn silk (*Zea mays*, G5417). *Agronomy* 12(4): 777. https://doi.org/10.3390/agronomy12040777
23. Akkad R, Buchko A, Johnston SP, Han J, House JD, et al. 2021. Sprouting improves the flavour quality of faba bean flours. *Food Chem* 364: 130355. https://doi.org/10.1016/j.foodchem.2021.130355
24. Kumari R, Gupta M. 2022. Elucidating the techno-functional, morphological and phenolic properties of hull less barley and buckwheat incorporated pasta. *Food Chem Adv* 1: 100055. https://doi.org/10.1016/j.focha.2022.100055
25. Barbhuiya RI, Singha P, Singh SK. 2021. A comprehensive review on impact of non-thermal processing on the structural changes of food components. *Food Res Int* 149:110647. https://doi.org/10.1016/j.foodres.2021.110647
26. Navyashree N, Buvaneswaran M, Sunil CK, Rawson A, Natarajan V. 2022. Development of white finger millet probiotic beverage using *Lactocaseibacillus rhamnosus* (LGG): process optimization and study of physicochemical and nutritional properties. *J Food Process Eng* 46(10): e14200. https://doi.org/10.1111/jfpe.14200
27. Kewuyemi YO, Kesa H, Adebo OA. 2022. Biochemical properties, nutritional quality, colour profile and techno-functional properties of whole grain sourdough and malted cowpea and quinoa flours. *Int J Food Sci Technol* 57(3): 1527-1543. https://doi.org/10.1111/ijfs.15512
28. Ganguly S, Sabikhi L, Singh AK. 2022. Effect of probiotic fermentation on physico-chemical and nutritional parameters of milk-cereal based composite substrate. *J Food Sci Technol* 59(8): 3073-3085. https://doi.org/10.1007/s13197-021-05350-8
29. Dymchenko A, Geršl M, Gregor T. 2023. Trends in bread waste utilisation. *Trends Food Sci Technol* 132: 93-102. https://doi.org/10.1016/j.tifs.2023.01.004
30. Kokwar MA, Arya SS, Bhat MS. 2022. A cereal-based nondairy probiotic functional beverage: an insight into the improvement in quality characteristics, sensory profile, and shelf-life. *J Food Process Preserv* 46(1): e16147. https://doi.org/10.1111/jfpp.16147
31. Dysvik A, Liland KH, Myhrer KS, Westereng B, Rukke EO, et al. 2019. Pre-fermentation with lactic acid bacteria in sour beer production. *J Inst Brew* 125(3): 342-356. https://doi.org/10.1002/jib.569
32. Yao Y, He W, Cai X, Bekhit AEDA, Xu B. 2022. Sensory, physicochemical and rheological properties of plant-based milk alternatives made from soybean, peanut, adlay, adzuki bean, oat and buckwheat. *Int J Food Sci Technol* 57(8): 4868-4878. https://doi.org/10.1111/ijfs.15814
33. Hernández-García Y, Melgar-Lalanne G, Téllez-Medina DI, Ruiz-May E, Salgado-Cruz MP, et al. 2022. Scavenging peptides, antioxidant activity, and hypoglycemic activity of a germinated amaranth (*Amaranthus hypochondriacus* L.) beverage fermented by *Lactiplantibacillus plantarum*. *J Food Biochem* 46(7): e14139. https://doi.org/10.1111/jfbc.14139
34. Chawanda ET, Manhokwe S, Jombo TZ, Mugadza DT, Njini M, et al. 2022. Optimisation of malting parameters for quinoa and barley: application of response surface methodology. *J Food Qual* 2022: 5279177. https://doi.org/10.1155/2022/5279177
35. Dabija A, Ciocan ME, Chetrariu A, Codină GG. 2022. Buckwheat and amaranth as raw materials for brewing, a review. *Plants* 11(6): 756. https://doi.org/10.3390/plants11060756
36. Duliński R, Zdaniewicz M, Pater A, Poniewska D, Żyła K. 2020. The impact of phytases on the release of bioactive inositols, the profile of inositol phosphates, and the release of selected minerals in the technology of buckwheat beer production. *Biomolecules* 10(2): 166. https://doi.org/10.3390/biom10020166
37. Thakur P, Kumar K, Ahmed N, Chauhan D, Eain Hyder Rizvi QU, et al. 2021. Effect of soaking and germination treatments on nutritional, anti-nutritional, and bioactive properties of amaranth (*Amaranthus hypochondriacus* L.), quinoa (*Chenopodium quinoa* L.), and buckwheat (*Fagopyrum esculentum* L.). *Curr Res Food Sci* 4: 917-925. https://doi.org/10.1016/j.crfs.2021.11.019
38. Zhang G, Xu Z, Gao Y, Huang X, Zou Y, et al. 2015. Effects of germination on the nutritional properties, phenolic profiles, and antioxidant activities of buckwheat. *J Food Sci* 80(5): H1111-H1119. https://doi.org/10.1111/1750-3841.12830
39. Dziadek K, Kopeć A, Pastucha E, Piątkowska E, Leszczyńska T, et al. 2016. Basic chemical composition and bioactive compounds content in selected cultivars of buckwheat whole seeds, dehulled seeds and hulls. *J Cereal Sci* 69: 1-8. https://doi.org/10.1016/j.jcs.2016.02.004
40. Fernandesa CG, Sonawaneb SK, Arya SS. 2021. Cereal based functional beverages: a review. *J Microbiol Biotechnol Food Sci* 8(3): 914-919. https://doi.org/10.15414/jmbfs.2018-19.8.3.914-919
41. Hussain A, Kausar T, Sehar S, Sarwar A, Ashraf AH, et al. 2022. Utilization of pumpkin, pumpkin powders, extracts, isolates, purified bioactives and pumpkin based functional food products: a key strategy to improve health in current post COVID 19 period: an updated review. *Appl Food Res* 2(2): 100241. https://doi.org/10.1016/j.afres.2022.100241