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Development of Functional Milk-based Smoothie by Incorporating Horse gram Extract

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Abstract

In the last few decades, the demand for fruits and vegetables and mixed beverages such as juices and smoothies increased rapidly, which made it one of the highest-growing segments of the food industry. The horse gram is an underutilized crop having desired nutrient and bioactive compounds essential for growth and normal body functioning. In view of the above facts, the present study aimed to develop a milk-based smoothie by incorporating horse gram extract in addition to commonly used ingredients such as sugar, dahi, stabilizer, fruits, and vegetables. In this study, three levels of horse gram extract (HGE- 5, 10, and 15%) were blended with cow milk, and other ingredients (sugar, dahi, pectin, banana, carrot juice extract) were kept constant. The 0% level of HGE was taken as a control to compare the nutritional and functional characteristics of the optimized product. The 10% HGE incorporated product was found best based on sensory and physico-chemical properties. The optimized product was found with 3.03±0.08% fat, 3.38±0.07% protein, 16.17±0.10% carbohydrate, 0.81±0.05% ash, 843.33±3.08 cp viscosity, 3.48±0.05 g/10 g whey syneresis, 12.12±0.67 g/20 g sedimentation, and 31.11 ± 4.17% 2,2-dipenylpicrylhydrazyl inhibition activity. The optimized smoothie had nine days shelf life after packing in polypropylene cups when stored under refrigerated conditions.

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Introduction

Underused/underutilized crops, also known as an orphan, neglected, or little used crops, are generally wild or semi-domesticated crops that have adapted to their local surroundings.[1] They have high nutritional content and are an excellent source of macro and micronutrients. Jack bean, winged bean, faba bean, cumina groundnut, horse gram, moth bean, adzuki beans, lima beans, etc., are the categories of underutilized crops.[2] Horse gram (*Macrotyloma uniflorum*) is one of the underutilized and unexplored legume crops grown mainly in Southeast Asia and tropical Africa. It has excellent nutritional value and is a great source of protein, carbohydrates, dietary fiber, micronutrients, and antioxidant properties. In Asian countries, particularly India, its seeds are considered the poor man's pulse crop. Water & oil absorption capacity, bulk density, and foam stability are the main functional properties of horse gram. It also contains bioactive compounds like phenolic acids, flavonoids, and isoflavonoids and anti-nutritional factors such as phytic acid and proteinase inhibitors.[3] In recent years, the isolation and utilization of potential antioxidants from legumes such as Horse gram have received much attention because it minimizes the chances of intestinal diseases, diabetes, coronary heart disease, and dental caries.[2,4] Horse gram also has other therapeutic effects in traditional knowledge systems. It has been recommended in Ayurveda medicine to treat renal stones, piles, edema, and other conditions.[3]

The health consciousness among consumers for healthy foods and the well-known fact that mixing different fruits and vegetables combines nutrients and bioactive compounds with appealing sensory attributes, and availability in the ready-to-drink form makes them very popular among consumers.[5] Smoothie is a non-alcoholic beverage generally made from a combination of fruits and vegetables processed into pulp or puree after removing the seeds and peel. They have a thicker consistency than slushies.[6] It is classified into three types: fruit itself only, fruit and dairy products, and functional smoothies. Functional smoothies are relatively new products on the market that typically include probiotics. Other ingredients, such as yogurt, milk, lentils, and cereals, may be added to smoothies to enhance their helpful functional characteristics. In view of the growing demand for food with functional properties, the current study emphasizes the importance of applying new scientific knowledge to the exploration of an underutilized crop, horse gram, is a source of functional and nutraceutical compounds.[7–9] Various challenges occur in developing functional smoothies, such as freely available anti-nutritional factors (ANFs), shelf-life, off-flavor, color degradation, and sedimentation. Therefore, several works have been done to overcome these challenges, like soaking and germination, which reduced trypsin inhibitor activity and flatulence-causing oligosaccharides, boosting protein digestibility and sensory characteristics.[10–12] Pasteurization is a process used for the shelf-life extension of liquid food products that destroy pathogens and minimizes spoilage causing microbes within limits.[13] Addition of flavoring ingredients (banana); during shelf-life storage, degradation of natural flavor occurs; therefore, to overcome this problem, natural flavoring agents are added to maintain the real flavor of food products.[14]

This natural pigment source gives desirable color to the product and improves the nutritional and functional properties of smoothies and similar products.[15] Addition of stabilizer; sedimentation and wheying off during the storage period of dairy beverages might be degraded by using pectin as a form of stabilizing agent.[16] The current study aimed to develop a functional smoothie by incorporating horse gram extract, keeping in view the maximum nutrient utilization from horse gram seeds having optimum sensorial characteristics and extended shelf life.

Material and Methods

Raw Material

Fresh cow milk was collected from Dairy Farm of Banaras Hindu University and standardized at 3.0% Fat and 8.5% Solid-not fat (SNF). Dahi used in the smoothie was prepared in the laboratory of the Department of Dairy Science and Food Technology by using standardized cow milk and mixed dahi starter culture (NCDC-137). Horse gram seeds, commercial grade sugar, fruit, and vegetables are procured from the local market of Varanasi, India. Fresh black carrots (*Daucus carota ssp. sativus var. atrorubens*) and Banana (*Musa cuminate*) were procured from the local vegetable and fruit market of Varanasi. Pectin was procured from Central Drug House, Delhi, and Dehydrated Tri-sodium citrate was procured from Merck Life Science Pvt. Ltd., Mumbai.

Chemicals and Reagents

The chemicals and reagents used in the experiments were of Analytical Grade (AR).

Preparation of Horse Gram Extract

The horse gram extract was prepared by the procedure described by Verma *et al.*, (2017)[17] 250 gm of cleaned and sorted horse gram seed was taken and washed with water. Seeds were soaked in distilled water (1:3) for 16 h, and excess water was drained after soaking and then washed with water. Soaked horse gram seeds further ground in grinder with 150 ml water. The grounded paste was filtered to remove the unwanted part and heated for 8-10 minutes. Horse gram extract was cooled to room temperature and refrigerated for further use.

Black Carrot Juice Extraction

Fresh and matured locally grown black carrots (Kashi Krishna Variety) were sorted for juice extraction and washed, peeled, and extra size carrots were cut into small pieces for better juice extraction. The juice was extracted using a juicer and filtered through a muslin cloth. The filtered juice was heat treated at 63 ^{0}C for 30 min and stored in refrigerated condition (5±1 ^{0}C).

Preparation of Horse Gram Incorporated Smoothie

Cow milk (3.5% Fat and 8.5% SNF) was used as a base material for smoothie preparation. The preliminary trials have been taken to optimize the concentration of constant ingredients based on available literature and sensory evaluation, i.e., dahi (5%), black carrot juice (3%), pectin (0.2%), tri-sodium citrate (0.04%), sugar (6%), and banana (15%) (Figure 1). In this study, the Horse gram extract was taken as a variable (0%, 5%, 10%, and 15%), and the final volume of the smoothie was maintained by varying the amount of cow milk. The 0% concentration of horse gram extract was considered a control for comparing the effect. The ingredients were mixed into cow milk thoroughly using a hand blender. The prepared product was heat treated at 63˚C for 30 min and subsequently packed in polypropylene cups and stored in refrigerated condition (5±1 ^{0}C). Detailed flow diagram of horse gram incorporated smoothie given in figure 1 after mentioned highlighted content.

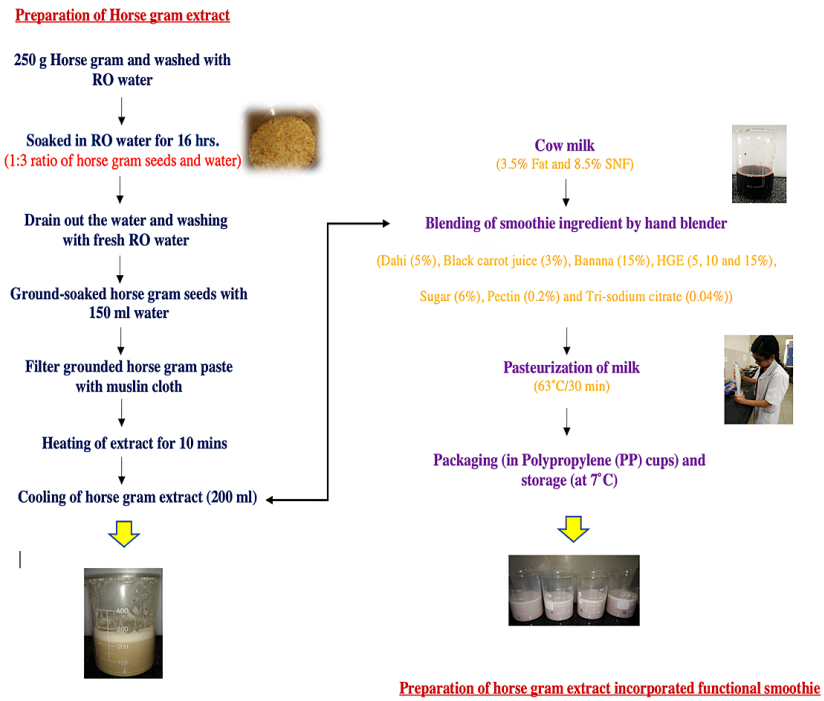

Fig. 1: Process flow diagram for preparation of horse gram incorporated Functional Milk-based Smoothie

Table 1: Raw ingredient formulation of different levels of horse gram extract incorporated smoothie (100g)

Sample Code	Treatment	Cow Milk (g)	Dahi Juice (g)	Carrot (g)	Pectin (g)	Sodium tri-citrate(g)	HGE (g)	Banana (g)	Sugar (g)
S1	5% HGE	65.76	5.00	3.00	0.20	0.04	5.00	15.00	6.0
S2	10% HGE	60.76	5.00	3.00	0.20	0.04	10.00	15.00	6.0
S3	15% HGE	55.76	5.00	3.00	0.20	0.04	15.00	15.00	6.0

Proximate Aanalysis

Proximate analysis of horse gram incorporated smoothie and control smoothie samples were carried out by standard method used for similar products. Fat (%), Protein (%), Total solids (%), Crude fibre (%), Carbohydrates (%), Ash (%), and pH value (pH meter make: Thermo Scientific, Singapore; Model: Sn B21899) of horse gram extract and horse gram incorporated smoothie was determined by AOAC, (2000)[18] methods.

Antioxidant activity Analysis

Free radical scavenging activity (RSA) of HGE and smoothie was measured by using the method of [19](List the method). 2 gm of sample was taken in a conical flask, and 25 ml of methanol added, then placed the conical flask into the shaker machine for 2 h, then centrifuged the sample at 6000 rpm for 10 min at 27 ^{0}C. 2.5 ml of centrifuged supernatant was taken and mixed with 5 ml of 2 mM DPPH in methanol solution vortexed. The mixture was incubated at room temperature in dark conditions for 30 min. Absorbance was measured at 517 nm, and 80% methanol was used as a blank. Antioxidant activity was expressed as percentage inhibition of the DPPH radical and was determined by the following equation.

DPPH inhibition (%) =(Absorbance of blank-Absorbance of sample)/(Absorbance of blank)×100

Viscosity

The viscosity of the optimized and control sample of the smoothie was determined at 26 °C by using a 1-1 system and TL-7 spindle of Viscostar plus Viscometer.

Sedimentation

The sediments of the horse gram incorporated smoothie were determined by using [20] method (list the method), with slight modifications. About 20 g of sample was taken in a centrifuge tube and centrifuged at 6000 rpm for 20 min at 27 ºC. The sediment content was calculated as a percentage on a weight basis.

Sedimentation (g/g of sample) = ((Weight of Sample-Weight of Whey))/(Weight of Sample)×100

Whey Syneresis

Whey syneresis of the product samples was determined by the centrifugation method Bahrami *et al*., (2013) [21] with slight modification. About 10 g of the sample was centrifuged at 6000 rpm for 15 min at 10°C. The supernatant was removed from the centrifuge tube, and the weight of the suspended particle was measured.

Whey syneresis (g/g of sample) = (Initial weight-Final weight)/(Initial weight ×100)

Sensory Evaluation

Horse gram incorporated smoothies were subjected to sensory evaluation by an expert and semi-trained panel of judges (n=30) for various sensory attributes, viz., flavour, color and appearance, body and texture, sweetness and overall acceptability criteria using a 9-point hedonic scale described by Stone and Sidel, (2004).[22] Labeled samples of freshly prepared products were given to the panel of judges. The judges were asked to rank the products from 1 to 9 according to their liking preference using a 9-point hedonic scale rating.

Evaluation of Shelf Life of Optimized Product

The optimized products were stored in cleaned and sterilized 200 ml polypropylene (PP) bottles at refrigerated temperature (7±0.5 °C) and room temperature (30±0.5 °C). The samples were analyzed for changes in sensory parameters (color

and appearance, flavor, consistency, sweetness, and overall acceptability), acidity, pH, and microbial counts at three-day intervals to determine the product's shelf life. The standard plate count, coliform count, and fungal count were performed as per the method described by APHA, (1992).[23]

Statistical Analysis

The data of different tests were analyzed using one-way ANOVA by SPSS 16.0 software (SPSS INC, Chicago, IL, USA), and all the tests were performed in triplicate.

Result and Discussion

Proximate and Functional Analysis of Horse Gram Extract (HGE)

The horse gram extract was evaluated for fat, protein, carbohydrates, ash, total solids, crude fibre, and antioxidant activity, and the results are shown in Table 1. Sivakumar, (2020)[24] reported horse gram extract having fat 0.25%, fibre 0.80%, protein 3.64% and carbohydrate 3.19%. Ojha *et al*., (2020)[25] reported 52.68% DPPH antioxidant inhibition for horse gram.

Table 2: Proximate and functional analysis of horse gram extract

Parameters	Ash	TS	Protein	Fat	Carbohydrate	Crude Fiber	Antioxidant activity
Results	0.22±0.03	9.02±0.91	3.53±0.13	0.26±0.14	5.00 ± 0.95	0.72±0.07	54.38 ± 1.59

Values are Mean±SD (n=3)

Optimization Level of Horse Gram Extract in Smoothie

In this study, the level of horse gram extract was optimized on the basis of sensory, physical, and functional properties of the smoothie. Horse gram incorporated smoothies were subjected to sensory evaluation by an expert and semi-trained panel of judges (n=10) for various sensory attributes, viz., flavor, color and appearance, body and texture, sweetness, and overall acceptability criteria using a 9-point hedonic scale. Results of sensory analysis of horse gram incorporated smoothie sample and control sample are presented in figure 2.

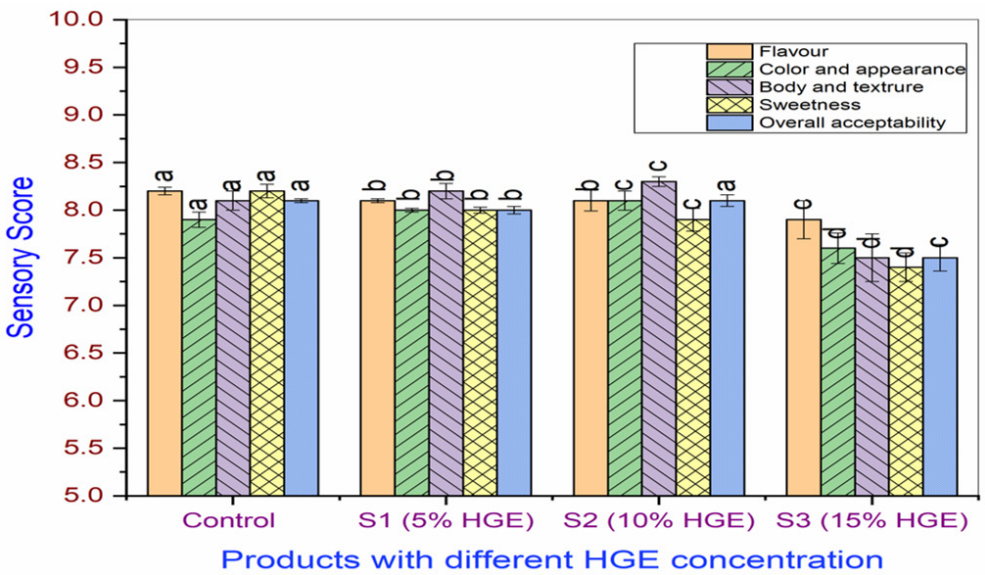

Fig. 2: Sensory evaluation of smoothies with different levels of Horse gram extract (HGE), Mean ± SD values used whereas a, b, c, d are different superscript differ significantly ($p < 0.05$)

The overall acceptability score of control and horse gram incorporated smoothie combination; S1 (5% HGE), S2 (10% HGE), and S3 (15% HGE) was 8.1±0.02, 8.0± 0.04, 8.1± 0.06 and 7.5 ± 0.14, respectively. Overall acceptability score of control and 10% HGE added smoothie was statically non-significantly different ($p < 0.05$). The panelists mostly liked control, and 10% HGE added smoothie due to better sensory parameters. However, all the sensory scores decreased with the increased rate of horse gram extract. A similar reason of acceptability was reported by Adebayo-Oyetoro *et al*., (2016)[26] for pawpaw juice milk blends and Hassan *et al*., (2015)[27] for fruit-flavored milk-based beverages.

Flavour score of control, S1, S2 and S3 are 8.2 ± 0.04, 8.1 ± 0.02, 8.1 ± 0.11 and 7.9 ± 0.20, respectively. The Flavour score of 5% and 10% horse gram incorporated smoothie was non-significantly different ($p < 0.05$), but the 15% HGE flavor score reduced significantly. The flavor score decreased with increased fortification which may be due to the typical beany flavor of bean crops.[28,29.] Narwal and Yadav, (2022)[30] reported a decrease in the flavor score of horse gram flour supplemented noodles. The color and appearance score of the control and different level of horse gram extract added smoothie was significantly different ($p < 0.05$). Sensory scores of color and appearance for 5 and 10% HGE incorporated smoothies were higher compared to the control sample. Joshi and Dubey, (2018)[31] study finding revealed that horse gram dal added laddo had better color and appearance compared to control (without horse gram) laddo·. Body and texture score of control, S1, S2 and S3 was 8.1 ± 0.10, 8.2 ± 0.08, 8.3 ± 0.05 and 7.5 ± 0.25, respectively. Body and texture sensory scores increased significantly on horse gram addition, but the scores decreased significantly after adding more than 10% horse gram extract. The sedimentation increased with an increasing amount of HGE due to the insoluble matter of horse gram extract. The sweetness score of smoothie decreased on horse gram incorporation; it was observed that with horse gram incorporation, total solids of smoothie increased, which led to a decrease in sweetness score of horse gram incorporated smoothie.

Physicochemical characteristics of Smoothie

The viscosity of control and 5%, 10%, and 15% incorporated HGE was 718.64±4.89, 740.05±5.24, 843.33±3.08, and 880.00±6.42, respectively. The viscosity of horse gram incorporated smoothie increased significantly ($p > 0.05$) with an increasing rate of HGE, and this increase was attributed to increasing the total solids and protein content of the smoothie. A similar trend of viscosity report by Rani *et al*., (2016)[20] for ready-to-serve breakfast smoothies prepared from the composite milk-sorghum base. Kumar, (2012)[32] reported that a higher amount (5.68%) of green gram flour added breakfast smoothie had higher viscosity compared to a low level of green gram addition (2.31%).

Table 3: Physico-chemical and antioxidant activity of smoothie samples

Sample Code	Viscosity (Cp at 20 rpm)	Whey Syneresis (ml/10g)	Sedimentation (ml/20g)	DPPH Inhibition activity	pH (%)	Acidity (%)
Control	718.64±4.89^{a}	4.21±0.04^{a}	9.68±0.48^{a}	24.59 ± 3.49^{a}	5.86 ± 0.03^{a}	0.39 ± 0.05^{a}
S1	740.05±5.24^{b}	3.89±0.06^{b}	10.56±0.32^{a}	28.18 ± 2.86^{a}	5.87 ± 0.05^{a}	0.40 ± 0.03^{a}
S2	843.33±3.08^{c}	3.48±0.05^{c}	12.12±0.67^{b}	31.11 ± 4.17^{a}	5.93 ± 0.03^{a}	0.41 ± 0.01^{a}
S3	880.00±6.42^{d}	2.92 ±0.11^{d}	14.14±0.51^{c}	38.20 ± 5.04b	5.98 ± 0.04^{a}	0.41 ± 0.02^{a}

Mean ± SD (n=3); a, b, c, d different superscript differ significantly ($p < 0.05$)

Whey syneresis of horse gram smoothie decreased significantly ($p > 0.05$) with an increase in horse gram addition, lowest whey syneresis (2.92 ±0.11 ml/10g) was observed in 15% HGE added smoothie. Similar findings were reported by Rani *et al*., (2016) [20] for sorghum fortified breakfast smoothies and Kumar, (2012) [32] for germinated green gram flour incorporated breakfast smoothies. Sedimentation of control and 5%, 10%, and 15% HGE incorporated smoothie was 9.68±0.48, 10.56±0.32, 12.12±0.67, and 14.14±0.51, respectively. Sedimentation of the smoothie was increased significantly ($p > 0.05$) with

the addition of HGE. An increased sedimentation rate leads to an increase in the insoluble matter in horse gram extract at a higher rate of HGE incorporation. Sedimentation of the smoothie can be reduced by increasing the rate of pectin addition during smoothie preparation, as Rani *et al.*, (2016)[20] suggested that smoothie-prepared pectin helps improve sedimentation of smoothie.

Horse gram-added smoothie was evaluated for antioxidant potential, and the study revealed that up to 15% horse gram extract incorporation DPPH inhibition activity was non-significantly different ($p > 0.05$). It may be attributed to the low amount of phenolic compound available in horse gram extract. Germination of horse gram increased the antioxidant activity compared to ungerminated horse gram; germinated horse gram flour had 60.76 ±0.64% antioxidant activity (in terms of DPPH inhibition) while ungerminated horse gram flour had 52.56±0.75 antioxidant activity (in terms of DPPH inhibition) as reported by Moktan, (2016).[33] The pH and acidity of control and different level (5%, 10%, and 15%) of horse gram incorporated smoothie was non-significantly different ($p > 0.05$).

Compositional Analysis of Control (Without Hge) and 10% Hge Incoporated Smoothie

10% HGE incorporated smoothie and control sample different components such as fat, protein, carbohydrates, ash, and total solids presented in Table 4.

Table 4: Chemical composition of control (without HGE) and 10% HGE incorporated smoothie

Parameter	Control (without HGE)	Optimized (10% HGE)
Fat	2.66±0.11^{a}	3.03±0.08^{b}
Protein	2.78±0.05^{a}	3.38±0.07^{b}
Carbohydrate	18.20±0.06^{a}	22.17±0.10^{b}
Ash	0.72±0.02^{a}	0.81±0.05^{a}
TS	24.38±0.09^{a}	29.40±0.04^{b}

Values are reported as Mean ± SD (n=3) a,b,c,d different superscript differ significantly ($p < 0.05$)

The composition of control and 10% HGE incorporated smoothie had 2.66±0.11, 3.03±0.08 fat, 2.78±0.05, 3.38±0.07 protein, 18.20±0.06, 22.17±0.10 carbohydrates, 0.72±0.02, 0.81±0.05 ash and 24.38±0.09, 29.40±0.04 total solids, respectively. All the composition (fat, protein, carbohydrates, and total solids, except ash content) of the control and 10% HGE incorporated smoothie significantly differ ($p < 0.05$). The increase in different constituents, i.e., fat, protein, carbohydrates, and total solids, may be attributed to a good amount of fat, protein, and carbohydrates in horse gram extract, banana, and other ingredients that are used in the preparation of smoothies. Finding of compositional analysis of horse gram incorporated smoothie in line with the smoothie prepared by several authors.[20,32] Ash content of 10% HGE incorporated smoothie and control sample was non-significantly differ ($p > 0.05$), it may be due to lesser ash content in HGE. Ash content of HGE incorporated smoothie was higher compared to multi-fruit smoothie.[34]

Storage Study

Pasteurized 10% HGE incorporated smoothie samples were packed in polypropylene (PP) cups in hygienic conditions and stored in refrigerated conditions (7˚C). During the storage study, different physico-chemical (pH and acidity) and microbial (Total plate count (TPC), Yeast and mold (YM), and Coliform count) parameter was analyzed at an interval of 3 days up to 15 days, results are showed in Table 5. In addition, sensory analysis of the product was done to check the acceptability of the control and optimized smoothie sample.

During the storage, the pH value of control and 10% incorporated smoothie decreased significantly for the 0th day and 15th days of storage. The pH of the

control sample non-significantly ($p > 0.05$) differs up to 12 days of storage, but on 15 days of storage pH value changed and differed significantly. At the same time, 10% HGE incorporated smoothie pH value differed significantly after three days of storage and reduced significantly ($p < 0.05$) up to a pH of 5.32 on 15 days of storage. The acidity of control, as well as 10% HGE, the incorporated smoothie was non-significantly ($p > 0.05$) different throughout the storage.

Table 4.8.1 Changes in pH, acidity, and microbial parameter of control and 10% HGE incorporated smoothie during the shelf-life study

Parameters	Samples	Number of days of storage study					
		0	3	6	9	12	15
pH	Control	5.63 ± 0.05^{a}	5.58 ± 0.07^{a}	5.53 ± 0.03^{a}	5.44 ± 0.06^{a}	5.38 ± 0.05^{a}	5.24 ± 0.03^{b}
	Optimized	5.97 ± 0.07^{a}	5.86 ± 0.04^{a}	5.66 ± 0.05^{b}	5.55 ± 0.03^{c}	5.43 ± 0.07^{d}	5.32 ± 0.05^{d}
Acidity	Control	0.39 ± 0.06^{a}	0.41 ± 0.04^{a}	0.43 ± 0.01^{a}	0.45 ±0.02^{a}	0.48 ± 0.03^{a}	0.52 ± 0.01^{a}
	Optimized	0.50 ± 0.05^{a}	0.51 ± 0.03^{a}	0.54 ± 0.04^{a}	0.56 ± 0.09^{a}	0.61 ± 0.06^{a}	0.64 ± 0.05^{a}
YM (log_{10}	Control	nd	nd	nd	1.63 ± 0.10^{a}	1.74 ± 0.08^{a}	1.89 ± 0.13^{a}
cfu/ml)	Optimized	nd	nd	nd	1.56 ±0.14^{a}	1.66 ± 0.08^{a}	1.75 ± 0.15^{a}
TPC(log_{10}	Control	0.10 ± 0.04^{a}	1.18 ± 0.08^{b}	1.23 ± 0.11^{b}	1.34 ± 0.06^{b}	1.67 ± 0.05c	1.84 ± 0.10^{c}
cfu/ml)	Optimized	0.02 ± 0.06^{a}	1.11 ± 0.03^{b}	1.19 ± 0.08^{b}	1.27 ± 0.10^{b}	1.45 ± 0.17^{b}	1.77 ± 0.07^{c}
Coliform	Control	nd	nd	nd	nd	0.42 ± 0.01^{a}	0.57 ± 0.02^{b}
(log_{10}cfu/ml)	Optimized	nd	nd	nd	nd	0.38 ± 0.06^{a}	0.45 ± 0.02^{a}

Values are reported as Mean ± SD (n=3) a,b,c,d different superscript differ significantly ($p < 0.05$), nd-not detected

Total plate count increased significantly ($p > 0.05$) throughout the storage period from initial count of 0.10 ± 0.04 and 0.02 ± 0.06 log10cfu/ml to 1.84 ± 0.10 and 1.77 ± 0.07 log10cfu/ml for control and 10% HGE incorporated smoothie, respectively. Bhardwaj, (2019) [34] reported a similar finding for a multi-fruit smoothie. The coliform and fungal count was found to be nil at the initial stage of the packed sample. The coliform count was observed after nine days of storage in control, and 10% HGE incorporated smoothie. Coliform count of control and 10% HGE incorporated smoothie was 0.57 ± 0.02 log_{10} cfu/ml and 0.45 ± 0.02 log_{10} cfu/ml on 15 days of storage. Yeast and mold count was absent for up to 6 days of storage for both samples, after which yeast and mold count were observed in both samples. Yeast and mold count for control and 10% HGE incorporated smoothie was 1.63 ± 0.10 log_{10} cfu/ml and 1.56±0.14 log_{10} cfu/ml on 9 days of storage; further changes in yeast and mold count of both the sample were statically non-significant ($p > 0.05$).

Horse gram contains good amount of nutrient and posses various health benefits on consumption, but several researcher reported possible side effects of horse gram crops. Excessive consumption of horse gram cause gas and bloating, it may be due to presence of specific oligosaccharide i.e. raffinose Prasad and Singh, (2015) [3]; Mao *et al*., (2018)[35] Few study, Pramod *et al*., (2006)[36] reported that horse gram had allergic reactions. Horse gram had several anti-nutrient factors such as phytic acid that restrict absorption of minerals present in horse gram Prasad and Singh, (2015)[3], but in present study horse gram seed soaked overnight to minimize anti-nutrient factors Gupta *et al*., (2015).[37]

Conclusion

Horse gram is an underutilized crop, and its application in food formulation is very limited. In recent years several researchers attempted to fortify or utilize horse gram in food formulation and preparation. The current study revealed that the horse gram has immense potential due to its nutritional and functional properties, such as being rich in protein, carbohydrates, and a good amount of antioxidant activity. Incorporating horse gram improves the physical and functional properties of the smoothie. Horse gram can be utilized in

smoothie-like products to improve its nutritional and sensorial properties.

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Conflict of Interest

The author declares that the research was conducted in the absence of any commercial or financial relationship that could be constructed as potential conflict of interest.

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