Original Research Article

An Exploratory Analysis of the Microbial and Sensory AttributesInfluencing the Stability and Quality of Instant Soup Mix Incorporated with Malabar Spinach Leaves Powder under Refrigerated Conditions

Comment [C1]: Should use "soup powder" because it ismix powder

Comment [C2]: The title should be shorter

ABSTRACT

The study was carried out for determining of the shelf life of the instant soup mixes prepared using dried Malabar spinach leaves powder, tomato powder, moong dal powder and other main ingredients. Malabar spinach instant soup mix formulated with 4 g of Malabar spinach leaves powder (MSLP), 20 g of moong dal powder (MDP), and 11 g of tomato powder (TP) along with other ingredients was liked extremely by 26.66 per cent of panel members, 60 per cent panellist liked it very much, and 13.33 per cent of panel members was liked it moderately after control treatment T0 (TP: MDP, 13:22g and 0g MSLP). The samples of instant soup mixes were packed into low density polyethylene (LDPE) and were analyzed periodically for change in quality. During 90 days of storage, there was a slight increase in the moisture content of the instant soup mix. In treatment T0 (TP: MDP, 13:22G and 0G MSLP), moisture was initially 3.37 per cent which gradually increased to 3.46 per cent at the end of the period. The moisture content of the instant soup mix for treatments T1, T2, T3, and T4 increased incrementally, following the same pattern. This finding was consistent across all treatments, indicating a clear trend. Total plate count indicates absence of microbial load during the storage period. The results of this investigation suggested that instant soup mixes could be kept in LDPE pouches at refrigerated temperature for ninety days without experiencing any quality deterioration.

Comment [C3]: The soup can't be low moisture, this is dry powder so you need to rewrite it in all paper.

Key words: *Malabar spinach leaves powder, moong dal, tomato powder, instant soup mix, total plate count, shelf life*

INTRODUCTION

The frenetic pace of modern life is the result of urbanization. Peoples are getting addicted to eating fast food and similar things because they do not have enough time to prepare food. Due to their high sugar, fat and salt levels and low protein, fiber, vitamin and mineral content,

most of these foods qualify as junk food.The word "instant food" itself refers to simple, quick, and easy cuisine that is not only hygienic, free of microbiological contamination, and quick to make, but also convenient to eat. Because instant foods are so convenient for consumers, instant mixes have become more and more popular. Globally, the market for quick meals is expanding at a rate of about 12 to 15 percent. Favourable demographic variables driving the global popularity of instant meals include a fast-paced urban lifestyle, an increasing number of nuclear families, increased disposable income, and an expanding number of consumers who travel and have an adventurous palate **(Sharma. 2012)**. Malnutrition and related diseases ultimately result from the consumption of food deficient in these nutrients**(Kaushik *et al.,* 2014)**. This problem can be solved by providing easily cooked, nutrient-rich foods. Dry soup powder is a readily available, easy-to-prepare recipe that significantly contributes to meeting the needs of current and future generations of consumers**(Krejcova *et al.,* 2007)**. People whose intake of solid foods is limited due to various pathological or obstructive reasons often consume soup because of its nutritional value and health benefits. In the current circumstances, vegetable-based soups are considered to be a viable alternative for consumers to ensure an adequate supply of essential nutrients and prevent nutritional deficiencies. These soups are an excellent source of vital nutrients such as vitamins, minerals, and fibre that are essential for maintaining optimal health. Incorporating vegetable-based soups into one's diet can help promote a balanced and healthy lifestyle. Therefore, it is recommended that consumers add these soups to their diet regimen to supplement their nutrient intake and avoid potential deficiencies.

A neglected perennial green vegetable of the Bessellaceae family is Malabar spinach (*Bessella alba*). Malabar Spinach is mentioned as "Upodika" in Ayurveda. Generally in India, it is known as Poi. It is commonly grown as a pot herb throughout India. Some research has shown that the mucilage of Malabar spinach has hypoglycemic value. According to some reports, it has therapeutic, gastroprotective and anti-inflammatory properties. The content of Malabar spinach leaf powder, as reported by **Soriano *et al*. (2020)**, is 12.06 per cent moisture, 16.17 per cent ash, 10.36 per cent crude fiber, 21.38 per cent crude protein, and 40.03 per cent carbohydrates. Malabar spinach leaves have 92.68g/100g moisture, 1.57g/100g ash, 1.09 g/100g protein, 0.45g/100g total fat, 2.21g/100g total fibre, 2.01g/100g carbohydrate **(Longvah *et al.,* 2017)**.

From the studies is found that instant soup mix will last longer. Food scientists use shelf-life studies to ensure high-quality products. Quality must be maintained during distribution and consumption. The present study evaluated the Malabar Spinach Instant Soup Mix's sustainability and acceptance. Overall quality is essential as it determines the customer's perception.

2. MATERIAL AND METHODS

Malabar spinach leaves were collected from a local farmer of Ayodhya (District), Uttar Pradesh, India. Fresh tomato, onion, and garlic, split green gram ginger powder were purchased from local market of Kanpur.

Tomatoes, garlic, and onion powder were used as ingredient for preparation of instant soup mix. Tomatoes, garlic, and onion were washed under running water for removal of dust, dirt and other foreign material, and processed into powder from by using standard methods of **Srivastava (2010), Pawar (2019), and Sangwan (2010),** respectively.

2.1 PREPARATION OF MALABAR SPINACH INSTANT SOUP MIX

Five different treatments (T0, T1, T2, T3, & T4) were designed by varying proportion of dried Malabar spinach leaves powder (MSLP) at 0g, 4g, 6g, 8g, and 10g ; tomato powder (TP) at 13g, 11g, 9g, 7g, and 5g, and moong dal powder (MDP) at 22g, 20g, 20g, 20g, and 20g whereas onion powder at 15g, garlic powder at 10g, ginger powder at 10g, black pepper powder at 5g, corn flour 10g, salt at 10g, and dehydrated carrot at 5g were followed as described **Amal *et al.* (2014)** by with modification. Malabar spinach instant soup mix formulations were prepared by dry mixing all the ingredients, different blends were tested for soup making quality. Soup was prepared by taking 10g of soup mix to 200ml of boiling water and cooks it at low flame for 5 minutes. Malabar spinach soup with different combination of ingredients was evaluated for sensory quality by nine point Hedonic scale.

Comment [C4]: Why do you pack the powder bags of 35 g (including 4-10g MSLD, 5-13g TP and 20-22g MDP), then use only 10 g powder to create the soup to evaluate the sensory. Why you do not use the whole 35g package?

2.2 STORAGE STUDIES

Five treatments of Instant soup mix was packed in LDPE (Low Density Polyethylene) and stored at refrigerated (4-6 °C) for ninety days. The sample were withdrawn at 45th day and 90th day and analyzed for parameter such as moisture, sensory quality and total plate count.

2.2.1 *MOISTURE ANALYSIS*

Moisture content of the stored instant soup mix was estimated in triplicate by **AOAC 1995**.

2.2.2 SENSORY *EVALUATION*

Malabar spinach soup with different combination of ingredients was evaluated for sensory quality by score card method **(Amerine *et al.,* 1965)**.

2.2.2 *MICROBIOLOGICAL QUALITY*

Comment [C5]: There are a lot of bacteria which concern to the quality of food but you only evaluate total aerobic bacteria (you used the denifine total plate count). Why?

- **TOTAL PLATE COUNT**

Comment [C6]: Do not need this title if only this parameter.

Total plate count represents the microbial load in a sample. To carry out the analytical procedure for microbial analysis, following preparations were made. Prior to start of experiment, the glasswares were sterilized in the hot air oven at 180°C for 2 hours **(Busta *et al.,* 1976).** Ten gram of each instant soup mix sample was blended with sterilized 0.1 per cent peptone water and volume was made up to 100 ml with the same solution.This constituted first dilution (1:10). The suspension was then shaken thoroughly by hand shaking for 2 minutes and subsequent dilutions were made using sterilized 0.1 per cent peptone water. Ten fold dilutions were prepared from the sample homogenate in the diluent buffer of peptone water (1 ml of homogenate + 9 ml of diluent buffer) to get 10^{-2}, 10^{-3}, 10^{-4} and 10^{-5} dilutions. One ml of each dilution was pipetted into a sterile petri dish (duplicate plates) under complete aseptic conditions. Twenty ml of melted sterile plate count agar (45°C) was then poured into each Petri dish and the contents were mixed thoroughly by rotating the plate several times, clockwise and then anti-clockwise. When the media had solidified, the plates were inverted and incubated at 37°C for 48 h.

On completion of incubation period, petri plates were taken out of the incubator and the colonies were counted using colony counter. The dilution plates showing the numbers of colonies in the statistical range of 30-300 were selected and average of the counts were determined. The total plate count per g or ml of the sample was calculated using formula given below:

$$Cfupergormlofsample = \frac{Numberofcolonies \times dilutionfactor}{Amountofsampletaken}$$

Comment [C7]: In this study, it is "g" because the product is powder.

2.3 STATISTICAL ANALYSIS

A complete randomized design was adopted for statistical study of data.Significant difference between experimental samples was test.To measure the sensory quality of formulated instant soup mixes simple statistical tool percentages have been used.

3. RESULT AND DISCUSSION

3.1 SENSORY QUALITY OF MALABAR SPINACH INSTANT SOUP MIX AND CONTROL INSTANT SOUP MIX BY NINE POINT HEDONIC SCALE

Comment [C8]: Should be shorter

Sensory evaluation of instant soup mix treated with Malabar spinach leaves powder (MSLP) by nine point Hedonic scale has been presented in **Table 1 & Fig 1**. The results showed that code T0 - (TP:MDP, 13:22g & 0g MSLP) i.e. control instant soup mix was liked extremely by 70 per cent panelist, 20 per cent panelist liked it very much, and 10 per cent panelist liked it moderately. From **Table 1** it can be observed that code T1- (TP: MDP, 11:20g & 04g MSLP), 26.66 per cent of panel member was liked it extremely, 60 per cent panelist liked it very much, and 13.33 per cent of panel member was liked it moderately. **Table 1** depicts that code T2- (TP: MDP, 09:20g & 06g MSLP), was liked extremely by 10 per cent panelist, 56.66 per cent panelist liked it very much, and 33.33 per cent panelist liked it moderately. **Table 1** shows that code T3- (TP: MDP, 07:20g & 08g MSLP) 10 per cent of panel member liked it very much, 60 per cent of panel member liked it moderately, and 30 per cent panelist liked it slightly. **Table 1** shows that code T4- (TP: MDP, 05:20g & 10g MSLP) was liked very

much by 3.33 per cent of panel member, 33.33 per cent panelist liked it moderately and 63.33 per cent panelist liked it slightly. According to the investigation's findings, it appears that the Malabar spinach soup mix treatment labelled as T1 (TP: MDP, 11:20G & 04g MSLP) was the most preferred sample after the control treatment (T0).

Table 1: Rating preferences of Malabar spinach instant soup mix and control instant soup mix by nine point Hedonic scale.

Comment [C9]: Should be shorter

Preferences	Product code				
	T0	T1	T2	T3	T4
Liked extremely	70	26.66	10	-	-
Liked very much	20	60	56.66	10	3.33
Liked moderately	10	13.33	33.33	60	33.33
Liked slightly	-	-	-	30	63.33

*All values are given in percentage, Control (TP: MDP, 13:22g & 0g MSLP), T1-(TP: MDP, 11:20g & 04g MSLP), T2- (TP: MDP, 09:20g & 06g MSLP)), T3-(TP: MDP, 07:20g & 08g MSLP), T4- (TP: MDP, 5:20g & 10g, MSLP)

Fig 1: Treatment of Malabar spinach instant soup mixes (T1, T2, T3, & T4) and control instant soup mixes

Comment [C10]: Water was added in this product, it wasn't orginal so you need to correct this sentence.

3.2 MOISTURE CONTENTOF MALABAR SPINACH INSTANT SOUP MIX AND CONTROL INSTANT SOUP MIX DURING STORAGE

Comment [C11]: Should be shorter

Table 2 represents the effect of storage period on the moisture content of the formulated Malabar spinach instant soup mixtreatments. An increase in the moisture content of the optimized Malabar spinach instant soup mixand control instant soup mixsample were observed as storage days increased. The data indicated that the moisture content of instant soup mix treatments T1, T2, T3, and T4 had an initial range of 4.04 to 5.65 percent, gradually increasing to 4.10 to 5.72 percent during end of storage. The control instant soup mix (T0) had an initial moisture content of 3.37 percent, increasing to 3.46 percent by the end of storage. Non-significant difference in moisture content of stored instant soup mixobserved after 45th and 90th days.Increases in moisture content during storage may result from the storage of products that have absorbed small amounts of moisture from the atmosphere through vapour diffusion from tiny spores in the packaging material **(Sharma *et al.*, 2013)**.

Similar results were reported by **Ansari *et al*. (2020)**, the moisture content of instant soup mix (formulated with *moringa oleifera* flower powder) increased from 8.41 per cent on the first day to 8.50 on the 90th day study. **Dhiman *et al*. (2017)** also mentioned increase in moisture content values in stored instant soup mix (formulated with dehydrated pumpkin) from 4.91 per cent on the first day to 4.96 per cent on the 90th day and 5.18 per cent on 180th day study at ambient temperature. These findings agreed with the findings of **Rokhsana *et al.* (2007)** who observed increase in moisture content of stored wheat-based instant soup mix during the storage study. The moisture content of wheat-based instant soup at ambient temperature showed an increase from 7.01 per cent on day first to 9.25 per cent on day 90 of the study

Table 2:Moisture content (per cent) ofMalabar spinach instant soup mixand control instant soup mixduring storage

Days of storage	Treatments				
	T0	T1	T2	T3	T4
0	3.37±0.02	4.04±0.02	4.54±0.16	5.04±0.02	5.65±0.04
45	3.42±0.15	4.07±0.03	4.57±0.08	5.09±0.11	5.69 ±0.06
90	3.46±0.10	4.10±0.11	4.96±0.32	5.11±0.01	5.72±0.07
S.Em.	0.06	0.04	0.13	0.04	0.03
C.D. @ 5 per cent	0.22	0.14	0.43	0.13	0.12
S/NS	NS	NS	NS	NS	NS

All results are mean± standard deviation of three values, T0- Control (TP:MDP, 13:22g & 0g MSLP), T1-(TP:MDP, 11:20g & 04g MSLP), T2-TP:MDP, 09:20g & 06g MSLP), T3-(TP:MDP, 07:20g & 08g MSLP), T4- (TP:MDP, 05:17g & 10g, MSLP)

S/NS signifies significant and non significant difference, respectively.S.Em- standard error of mean, C.D. - critical difference

3.3 SENSORY QUALITYOF MALABAR SPINACH INSTANT SOUP MIX AND CONTROL INSTANT SOUP MIX DURING STORAGE

Malabar spinach soup mixes and control soup mix were evaluated for the sensory attributes during the storage period. The results (Table 3) indicates that treatment T0 (control) and T1 obtained higher scores with respect to all the sensory attributes like taste, appearance, aroma, consistency, after taste, and over all acceptability. According to the findings presented in Table 3, it is evident that the sensory attributes of all variations of instant soup mix exhibit a gradual decline as the duration of storage increases. However, non-significant decrease in all parameters of instant soup mixes treatment (T0, T1, T2, T3, & T4) up to 90 days. According to the collected data, the instant soup mix treatments T1, T2, T3, and T4 initially had an overall acceptability score ranging from 8.00 to 5.10, which gradually decreased to 7.85 to 4.50 during end of storage. On the other hand, the control instant soup mix (T0) had an initial overall acceptability score of 8.20, which decreased to 7.70 by the end of storage.

Comment [C12]: With received results, which recipe did you choose or all recipeswere the same?

Table 3:Sensory quality of Malabar spinach instant soup mix and control instant soup mix during storage

Storage period	Treatments				
	T0	T1	T2	T3	T4
Taste					
0	8.40±0.45	8.05±0.36	7.15±0.33	5.75±0.63	4.55±0.49
45	8.25±0.42	7.70±0.42	7.05±0.64	5.60±0.45	4.35±0.47
90	8.00±0.40	7.85±0.52	7.00±0.66	5.30±0.34	4.10±0.45
S.Em.	0.14	0.14	0.18	0.16	0.15
C.D. @5 per cent	0.40	0.41	0.52	0.45	0.44
S/NS	NS	NS	NS	NS	NS
Appearance					
0	8.10±0.31	7.85±0.41	7.40±0.45	6.00±0.33	5.00±0.47
45	7.90±0.51	7.50±0.70	7.25±0.54	5.90±0.56	4.65±0.47
90	7.65±0.41	7.35±0.33	7.10±0.69	5.60±0.51	4.50±0.52
S.Em.	0.13	0.16	0.18	0.15	0.16
C.D. @5 per cent	0.39	0.47	0.53	0.44	0.45
S/NS	NS	NS	NS	NS	NS
Aroma					
0	8.05±0.36	7.95±0.79	7.10±0.69	6.00±0.57	5.00±0.70
45	7.55±0.43	7.65±0.41	6.90±0.61	5.80±0.75	4.95±0.43
90	7.80±0.53	7.30±0.42	6.70±0.67	5.50±0.40	4.45±0.55
S.Em.	0.14	0.18	0.21	0.19	0.18
C.D. @5 per cent	0.42	0.53	0.61	0.55	0.53
S/NS	NS	NS	NS	NS	NS
Consistency					
0	8.30±0.53	8.2±0.34	7.05±0.36	6.75±0.42	6.00±0.57
45	8.00±0.47	7.75±0.48	7.00±0.52	6.55±0.43	5.70±0.71
90	7.85±0.33	7.70±0.58	6.95±0.59	6.30±0.53	5.40±0.65
S.Em.	0.14	0.15	0.16	0.15	0.21
C.D. @5 per cent	0.42	0.44	0.47	0.43	0.60
S/NS	NS	NS	NS	NS	NS

After taste					
0	8.75±0.35	7.5±0.40	6.60±0.56	5.95±0.49	5.05±0.55
45	8.60±0.39	7.20±0.25	6.30±0.71	5.70±0.67	4.90±0.31
90	8.35±0.33	7.15±0.52	6.00±0.62	5.50±0.47	4.65±0.41
S.Em.	0.11	0.13	0.20	0.18	0.14
C.D. @5 per cent	0.33	0.38	0.59	0.51	0.40
S/NS	NS	NS	NS	NS	NS
Over all acceptability					
0	8.20±0.42	8.00±0.47	6.75±0.42	6.05±0.55	5.10±0.31
45	7.95±0.59	7.85±0.52	6.40±0.65	5.90±0.80	4.75±0.75
90	7.70±0.48	7.55±0.59	6.10±0.61	5.60±0.45	4.50±0.40
S.Em.	0.16	0.17	0.18	0.20	0.17
C.D. @5 per cent	0.46	0.49	0.53	0.57	0.48
S/NS	NS	NS	NS	NS	NS

All results are mean± standard deviation of three values, T0- Control (TP: MDP, 13:22g & 0g MSLP), T1-(TP: MDP, 11:20g & 04g MSLP), T2- (TP: MDP, 09:20g & 06g MSLP), T3-(TP: MDP, 07:20g & 08g MSLP), T4- (TP: MDP, 05:17g & 10g, MSLP)

S/NS signifies significant and non significant difference, respectively.S.Em- standard error of mean. C.D. - critical difference.

3.4TOTAL PLATE COUNT

Total plate count of Malabar spinach instant soup mixand control instant soup mixhas been presented in **Table 4**. The formulated Malabar spinach instant soup mixwas analyzed at 0, 45, and 90 days. At 0 days of storage, the total plate count was too low to count in all instant soup mix treatment. The total plate count of the Malabar spinach instant soup samples (coded as T1, T2, T3, and T4) ranged from 1.90×10^2 to 1.95×10^2 cfu/g while the lowest counts (2.81×10^2)were obtainedin control instant soup mix on 90 days. The result of microbial analysis showed increase in total plate count during storage period however it was found to be under acceptable limit. This might be because all treatments had progressively higher moisture content over time.The results are consistent with the findings of **Ansari (2020)** who observed that during soup mix storage, the microbial burden rises with time. The study revealed that the sourdough bread (*Saccharomyces cerevisiae*) had total plate count of 1.3×10^2 cfu/g at one month storage which was increased to 2.6×10^3cfu/g after ninty days. **Jay (1992)** reported that that product is microbiologically safe if the total microbial count of dehydrated soups is $<1x10^3$cfu/g.

Comment [C13]: -Should use ISO 11133:2014 for Microbiology of food, animal feed and water
-You must evaluate some bacteria that cause intestinal diseases such as E.*coli*, coliform and *Salmonella*.

Comment [C14]: Should use "total aerobic microorganisms" on plate

Table 4: Total plate count (cfu/g) of Malabar spinach instant soup mix and control instant soup mix during storage

Storage period	T0	T1	T2	T3	T4
0	TFTC	TFTC	TFTC	TFTC	TFTC
45	1.42×10^2	1.45×10^2	1.46×10^2	1.46×10^2	1.47×10^2
90	2.81×10^2	1.90×10^2	1.92×10^2	1.94×10^2	1.95×10^2

TFTC- Too few to count,T0- Control (TP: MDP, 13:22g & 0g MSLP), T1-(TP: MDP, 11:20g & 04g MSLP), T2- (TP: MDP, 09:20g & 06g MSLP), T3-(TP: MDP, 07:20g & 08g MSLP), T4- (TP: MDP, 05:17g & 10g, MSLP)

CONCLUSION

From the above study it is evident that the sensory attributes of Malabar spinach soup mix was accepted by panel members. The formulated instant soup mix required only 5-6 minutes for the preparation. Storage slightly affected the moisture content of the stored samples of instant soup mixes. Moisture content slightly increased at the end of storage period. As the storage period increased, the sensory characteristics of the instant soup mix gradually decreased.The present study revealed that Malabar spinach soup mix treatment labeled as T1 (TP: MDP, 11:20g, and 04g MSLP) was the most preferred sample followed by the control treatment (T0) at the end of the storage period. The microbial load also slightly increased at the end of storage period. Howeverthe microbiological load is within safe bounds and is not high.The findings of this study have important implications for the food industry, as they provide a scientific basis for shelf life determination and product quality assurance. The results can be used to inform product development and optimize manufacturing processes, ensuring that consumers receive safe and high-quality food products. The research methodology and findings can be used to inform future studies and product development efforts, enabling the food industry to continue delivering safe and nutritious food products to consumers.

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Comment [C15]: References should be updated documents in 5 years recently

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