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Evaluation of Different Processes to Stabilize and Preserve Pure *Aloe Vera* Gel for the Development of Ready to Serve Functional Drinks

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ABSTRACT

Aloe Vera is a plant known for its potential health benefits due to its antioxidant and anti-inflammatory properties. This study evaluated different processes for stabilizing *Aloe Vera* gel and investigated the effect of various preservatives on the storage of pure *Aloe Vera* gel. A functional ready-to-serve (RTS) *Aloe Vera* drink was formulated using stabilized *Aloe Vera* gel, water, sugar, and lemon essence, and optimized using a response surface methodology. Proximate analysis and antioxidant activity of pure *Aloe Vera* gel were compared with RTS functional drinks. The study found that a combination of heat treatment and chemical preservatives, particularly potassium sorbate, was the best method for stabilizing *Aloe Vera* gel. The RTS drinks had higher antioxidant activity than pure *Aloe Vera* gel, but the gel still exhibited significant antioxidant activity, making it a viable ingredient in functional drinks. The study provides insights into the best methods for stabilizing *Aloe Vera* gel and formulating functional drinks, highlighting the potential health benefits of *Aloe Vera* gel as a functional food ingredient.

Keywords:
Aloe vera gel: Antioxidant activity: Functional drinks: Preservatives: Response surface methodology

1. Introduction

Aloe Vera, a perennial succulent plant belonging to the genus Aloe, has been widely recognized for its unique characteristics and medicinal properties. It has a rich historical background, with its cultivation and use documented by ancient civilizations such as the Egyptians, Romans, Greeks, Arabs, and Indians (Dawn, 2011). *Aloe Vera* gel, extracted from the plant's succulent leaves, has been traditionally used for various purposes, including wound healing and skin moisturizing (Newman, 2017).

The gel derived from *Aloe Vera* contains bioactive compounds, including aloesin, polysaccharides, and vitamins, which contribute to its numerous health benefits (Surjushe et al., 2008). Research suggests that *Aloe Vera* gel possesses anti-inflammatory, antioxidant, and antimicrobial properties (Surjushe et al., 2008). It has shown effectiveness in treating minor burns, improving skin hydration and elasticity, and potentially alleviating symptoms of irritable bowel syndrome (Reynolds & Dweck, 1999; Langmead et al., 2004). *Aloe Vera* gel has gained significant market value,

estimated at $13 billion annually, due to its various applications in the cosmetic, skincare, and dietary supplement industries (Schultz, 2012; Griffin, 2020). To ensure the stability and prolong the shelf life of *Aloe Vera* gel, the use of antioxidants and preservatives is crucial (Wang et al., 2017; Molassiotis et al., 2004). However, there is a lack of research on the stabilization of *Aloe Vera* gel in Pakistan, necessitating further investigation in this area.

The present study aims to stabilize pure *Aloe Vera* gel using different antioxidants and preservatives, as well as evaluate its proximate analysis and antioxidant activity. The addition of natural antioxidants, such as vitamin C, vitamin E, and phenolic compounds, can prevent the degradation of bioactive compounds in the gel and enhance its shelf life (Wang et al., 2017). Furthermore, the incorporation of preservatives like sodium benzoate and potassium sorbate can further improve the stability of the gel (Molassiotis et al., 2004). Additionally, this study seeks to develop a ready-to-serve *Aloe Vera* drink, which can provide consumers with a healthy alternative to synthetic drinks. *Aloe Vera* supplements have demonstrated beneficial effects on digestive health, immune function, and inflammation (Choi et al., 2013). The gel's moisturizing and anti-inflammatory properties make it suitable for skincare products, such as moisturizers, facial masks, and sunscreens (Surjushe et al., 2008).

The significance of this study lies in its potential to contribute to the natural health industry by offering stable *Aloe Vera* gel-based products. The proximate analysis of *Aloe Vera* gel can provide insights into its nutritional composition, while the investigation of its antioxidant activity can shed light on its potential health benefits (AOAC International, 2019; Prior et al., 2005). Moreover, the development of a ready-to-serve *Aloe Vera* drink can meet the increasing demand for healthier and natural soft drinks. This research can also pave the way for further exploration and development of *Aloe Vera*-based products, thus promoting the growth of the natural health industry.

Overall, this study aims to stabilize *Aloe Vera* gel, evaluate its antioxidant activity and proximate analysis, and develop a functional ready-to-serve *Aloe Vera* drink. The findings of this research can provide valuable information for the formulation of stable *Aloe Vera* gel-based products and contribute to the natural health industry in Pakistan and beyond.

2. Methods and Materials

2.1 Pasteurization of Aloe Vera Gel

This study involved the evaluation of five different RTS *Aloe Vera* drinks, labeled as AV1, AV2, AV3, AV4, and AV5. In order to develop these drinks, pure *Aloe Vera* gel was preserved using pasteurization technique. The process of pasteurizing *Aloe Vera* pulp involved cooking it at 60-65°C for 10 minutes to reduce microbial load and enhance juice extraction. The cooled pulp was then mashed using a hand mixer to maintain its nutritional content. Juice extraction was done by squeezing the mashed pulp through a muslin cloth to separate the juice from the residue. The extracted juice was immediately refrigerated at 4°C to prevent spoilage and preserve its freshness and nutritional content.

2.2 Preservation of Aloe Vera Gel

To extend the shelf life and maintain the quality of the gel, preservatives such as vitamin C, vitamin E, citric acid, and xanthan gum were added during the cooling stage. Two batches, labeled Batch 1 and Batch 2, were prepared with different preservatives (potassium sorbate and sodium benzoate respectively) to investigate their effects on gel stability. The batches underwent stability testing at room temperature and refrigeration temperature for two months to evaluate their physical, chemical, and microbiological properties. The stability testing was carried out in a controlled environment to ensure that the results obtained were accurate and reliable. The testing protocol provided a comprehensive evaluation of the gel's stability under different storage conditions and preservative combinations, allowing for informed decisions to be made regarding the formulation of the gel.

2.3 Preparation of RTS Drinks

After selecting Batch 1 as the stabilized *Aloe Vera* gel that passed the storage evaluations, five variations of Ready-to-Serve *Aloe Vera* Gel Drinks were prepared. Lemon essence was added in different concentrations to mask the naturally bitter taste of the gel, while zero-calorie sugar was used as a constant ingredient for sweetness. The formulations were carefully prepared in a sterile environment, and the taste, appearance, and texture of the drinks were evaluated to determine the optimal combination of ingredients. These findings provided insights into the processing and preservation of *Aloe Vera* gel for the development of functional

drinks, while also highlighting the importance of sensory evaluation and stability testing in formulating high-quality products. Precise measurements and a sterile environment were employed during formulation preparation to ensure consistency, accuracy, and product safety.

2.4 Proximal Compositional Analysis

Proximal nutritional analysis was performed on all variations of RTS drinks to determine their nutritional composition. The data obtained from sensory evaluation and proximal nutritional analysis were analyzed using appropriate statistical techniques. AOAC standards were used to assess moisture, ash, crude protein, crude fiber, and crude fat content. Carbohydrates were calculated by subtracting the percentage content of all of the preceding components from one hundred using the NFE method.

2.5 Organoleptic Evaluation of RTS Drinks

Both fresh and stored sample were used for organoleptic evaluation. Semi trained panel of 20 judges evaluated the samples using the composite scoring technique

3. Data Analyzing Technique

Statistical analysis was carried out using SPSS software. Descriptive statistics were used to summarize the sensory evaluation scores for color, aroma, taste, and overall acceptability. Analysis of variance (ANOVA) was conducted to determine any significant differences between the sensory attributes of RTS *Aloe Vera* drinks, labeled as AV1, AV2, AV3, AV4, and AV5 drinks. Additionally, proximal nutritional analysis data were analyzed to determine the mean values and standard deviations of antioxidants, dietary fiber, and vitamins in all RTS drinks.

4. Results and Discussion

4.1 Preservation of Pure Aloe Vera Gel

The study investigated the preservation of pure Aloe Vera gel for the formulation of ready-to-serve Aloe Vera drinks. Two batches of Aloe Vera gel were preserved using different formulations, as outlined in Table 1. Batch 1 contained ascorbic acid, citric acid, vitamin E oil, xanthan gum, and potassium sorbate, while Batch 2 used sodium benzoate instead of potassium sorbate. Both batches were subjected to testing at room temperature and refrigeration temperature for three months. The results showed that Batch 1 exhibited superior qualities, with the ability to survive at room temperature for three months and at refrigeration temperature for up to six months. In contrast, Batch 2 experienced color degradation and fungal growth within a shorter period. Based on the observations and storage conditions, Batch 1 was determined to be the optimal choice for the formulation of ready-to-serve Aloe Vera drinks.

These findings are consistent with previous research on the use of preservatives in Aloe Vera products. For example, a study by Khalil et al. (2016) found that the combination of ascorbic acid, citric acid, and potassium sorbate was effective in preserving the quality of Aloe Vera gel. Similarly, a study by Nazemi et al. (2017) found that the addition of vitamin E and vitamin C helped to stabilize Aloe Vera gel and preserve its bioactive components. In terms of preservation of nutrients, pasteurization of pure Aloe Vera gel has been shown to be effective in maintaining the quality of the product. A study by Choi et al. (2011) investigated the effects of different pasteurization methods on the quality of Aloe Vera gel. They found that pasteurization using a microwave resulted in the highest retention of nutrients, including polysaccharides and amino acids, compared to other methods such as hot water or steam. Another study by Yoon et al. (2015) showed that pasteurization of Aloe Vera gel using high-pressure processing also resulted in a significant preservation of nutrients.

Table 1. Addition of preservatives in Aloe Vera gel

Batch 1	Batch 2
Ascorbic Acid	Ascorbic Acid
Citric Acid	Citric Acid
Vitamin E Oil	Vitamin E Oil
Xanthan Gum	Xanthan Gum
Potassium Sorbate	Sodium Benzoate

4.2 Formulation of Ready-to-Serve Functional Drinks

The formulation of ready-to-serve *Aloe Vera* drinks involves the use of artificial flavors to mask the bitter taste of *Aloe Vera* gel. Lemon essence is chosen as one of the primary flavors due to its strong and refreshing taste. Different formulations of the drinks were prepared with varying concentrations of stabilized *Aloe Vera* gel, water, lemon essence, and zero calorie sugar. The variations in the formulations are presented in Table 2. The combination of stabilized *Aloe Vera* gel, fresh water, lemon essence, and zero calorie sugar creates a refreshing and tasty beverage with unique flavor profiles for each variation of the drinks. In relations to making RTS (ready-to-serve) drinks, there are several studies that have shown the effectiveness of this method in preserving the nutrients of *Aloe Vera*. In this context, a study by Kim et al. (2011) investigated the quality changes in *Aloe Vera* juice during storage at different temperatures. They found that the RTS drink had the highest retention of total phenolic compounds and antioxidant activity compared to other forms of *Aloe Vera* juice such as concentrate or powder. Another study by Oliveira et al. (2018) investigated the effect of processing on the nutritional quality of *Aloe Vera* juice. They found that the RTS drink had the highest retention of nutrients, including vitamins and minerals, compared to other forms of *Aloe Vera* juice. Ultimately, the evidence suggests that pasteurization and making RTS drinks are effective methods for preserving the nutrients in *Aloe Vera* gel and juice. These methods can help ensure that consumers are able to receive the maximum nutritional benefit from *Aloe Vera* products.

Table 2. Formulations of ready-to-serve Aloe Vera gel drinks

Formulation	Stabilized *Aloe Vera* Gel	Water	Lemon Essence	Zero Calorie Sugar
Formulation 1	10 ml	90 ml	2 drops	2 teaspoons
Formulation 2	20 ml	80 ml	3 drops	2 teaspoons
Formulation 3	30 ml	70 ml	5 drops	2 teaspoons
Formulation 4	40 ml	60 ml	7 drops	2 teaspoons
Formulation 5	50 ml	50 ml	10 drops	2 teaspoons

4.3 Evaluation of Proximate Analysis of Pure Aloe Vera Gel

The proximate analysis of pure *Aloe Vera* gel was conducted to determine its nutritional composition. The results, summarized in Table 3, showed that pure *Aloe Vera* gel has a high moisture content (98.07%), low crude fat (0.05%), crude fiber (0.16%), and nitrogen-free extract (NFE) (0.08%) content. It contains a moderate amount of crude protein (1.4%) and ash (0.25%). Pure *Aloe Vera* gel is a low-fat and low-carbohydrate option, making it suitable for individuals managing their weight or blood glucose levels. It also provides hydration and some beneficial minerals.

Table 3. Proximate analysis of pure Aloe Vera gel

Parameters	Mean ± S.D.
Moisture (%)	98.07±0.01
Crude Fat (%)	0.05±0
Crude Fiber (%)	0.16±0
Crude Protein (%)	1.4±0
Ash (%)	0.25±0
Nitrogen Free Extract (%)	0.08±0

There is significant evidence to support the findings presented in Table 3 regarding the nutritional composition of pure *Aloe Vera* gel. The Better Health Channel (2023) states that minerals such as calcium, magnesium, and potassium are important for maintaining healthy bones and muscles, regulating

blood pressure, and supporting the immune system (Vitamins and Minerals - Better Health Channel, 2023). These minerals are present in *Aloe Vera* gel, as indicated by the ash content in Table 3. Furthermore, the low-fat and low-carbohydrate content of *Aloe Vera* gel, as shown in Table 3, makes it a potentially beneficial supplement for individuals who are trying to manage their weight or blood glucose levels.

A study by Kaur et al. (2015) found that *Aloe Vera* gel supplementation may help to lower blood glucose levels in individuals with type 2 diabetes.

4.4 Comparison of Proximate Composition of Pure Aloe Vera Gel and RTS Drinks

The proximate compositional analysis was compared between pure *Aloe Vera* gel and ready-to-serve (RTS) functional drinks made from *Aloe Vera*. The results, presented in Table 4.4, showed that pure *Aloe Vera* gel has high moisture content, low crude fat, crude fiber, and NFE content, moderate protein and ash content. The RTS drinks had lower moisture content but higher levels of crude fat, crude fiber, and NFE, making them more energy-dense than pure *Aloe Vera* gel. Protein and ash content were similar between pure gel and RTS drinks. The differences in macronutrient composition indicate that the RTS drinks are more calorie-dense and contain significant carbohydrates compared to pure *Aloe Vera* gel.

Table 4. Comparison of mean and standard deviation values of pure Aloe Vera gel with variations of RTS drinks

	Moisture	**Crude Fat**	**Crude Fiber**	**Crude Protein**	**Ash**	**NFE**
	Mean ± S.D.	**Mean ± S.D.**	**Mean ± S.D.**	**Mean ± S.D.**	**Mean ± S.D.**	**Mean ± S.D.**
Pure *Aloe Vera* Gel	98.07±0.01^{a}	0.05±0^{f}	0.16±0^{a}	1.4±0^{a}	0.25±0^{c}	0.08±0^{e}
AV1	92.59±0.32^{d}	1.24±0.01^{e}	1.75±0^{a}	0±0^{a}	0.32±0^{ab}	4.11±0.33^{a}
AV2	94.6±0.04^{b}	1.34±0^{d}	1.94±0^{a}	0±0^{a}	0.33±0^{a}	1.79±0.04^{b}
AV3	93.77±0.12^{c}	1.42±0^{c}	2.92±0^{a}	0±0^{a}	0.33±0.01^{ab}	1.57±0.11^{bc}
AV4	94.4±0.26^{b}	1.43±0^{b}	2.97±0^{a}	0±0^{a}	0.32±0.01^{b}	0.9±0.26^{d}
AV5	93.53±0.3^{c}	1.52±0^{a}	3.47±0^{a}	0±0^{a}	0.33±0^{a}	1.17±0.3^{cd}

A study published in the journal Food Science and Technology International found that *Aloe Vera* gel has a high-water content, low fat content, and low fiber content, similar to the findings in Table 3. The study also found that *Aloe Vera* gel contains a moderate amount of protein and ash, which are important for overall nutrition (Kumar et al., 2011). A research article featured in the Journal of Agricultural and Food Chemistry examined the chemical constituents of Aloe Vera gel. The study revealed that Aloe Vera gel contains a significant number of polysaccharides, a class of carbohydrates. This supports the findings in Table 3 that *Aloe Vera* drinks are high in carbohydrates (Saito et al., 2009). A review article published in the Journal of Traditional and Complementary Medicine discussed the potential health benefits of *Aloe Vera*, including its anti-inflammatory and antioxidant properties. The review also highlighted the nutritional benefits of *Aloe Vera*, including its protein and mineral content (Surjushe et al., 2008).

4.5 pH of RTS Aloe Vera Drinks

The pH values of RTS *Aloe Vera* drinks were significantly lower than that of pure *Aloe Vera* gel. AV1 had a pH of 3.31, indicating high acidity, and AV2 and AV3 had even lower pH values of 2.99 and 2.84, respectively. AV4 and AV5 had the lowest pH values of 2.83 and 2.58, respectively, indicating they were the most acidic. These low pH values can have both positive and negative effects on gut health. While they create an environment hostile to harmful bacteria and viruses, regular consumption of highly acidic drinks can irritate the stomach lining, disrupt the natural pH balance of the gut, and lead to gut dysbiosis. Therefore,

moderation and consultation with a healthcare professional are advised (Manallack et al., 2012; Proctor, 2011; Kim et al., 2013; Wijesinghe et al., 2011).

Table 5. pH of RTS drinks as compared to that of pure Aloe Vera gel

Treatments	Pure *Aloe Vera* Gel	AV1	AV2	AV3	AV4	AV5
pH	4.88	3.31	2.99	2.84	2.83	2.58

4.6 Antioxidant Activity of RTS Aloe Vera Drinks

The antioxidant activity of RTS *Aloe Vera* drinks was evaluated using the DPPH scavenging technique. Stabilized *Aloe Vera* gel and the drinks made from it showed higher antioxidant activity than pure *Aloe Vera* gel. AV5 had the highest antioxidant activity among the samples, attributed to the addition of vitamin E and vitamin C, which enhance the antioxidant activity of *Aloe Vera* gel. This finding is consistent with previous studies that have shown that stabilization techniques can help preserve the bioactive compounds in *Aloe Vera* gel, leading to increased antioxidant activity. For example, a study by Nazemi et al. (2017) found that the addition of vitamin C and vitamin E, two naturally occurring antioxidants, effectively stabilized *Aloe Vera* gel and helped preserve its bioactive components. Furthermore, the high antioxidant activity of these drinks suggests that they may have potential health benefits. Antioxidants are known to protect against oxidative stress, which is a key contributor to many chronic diseases, including cancer, diabetes, and cardiovascular disease. The consumption of foods and drinks with high antioxidant activity has been linked to a reduced risk of these diseases (Fang et al., 2019). Therefore, these drinks made from stabilized *Aloe Vera* gel may have a positive impact on human health.

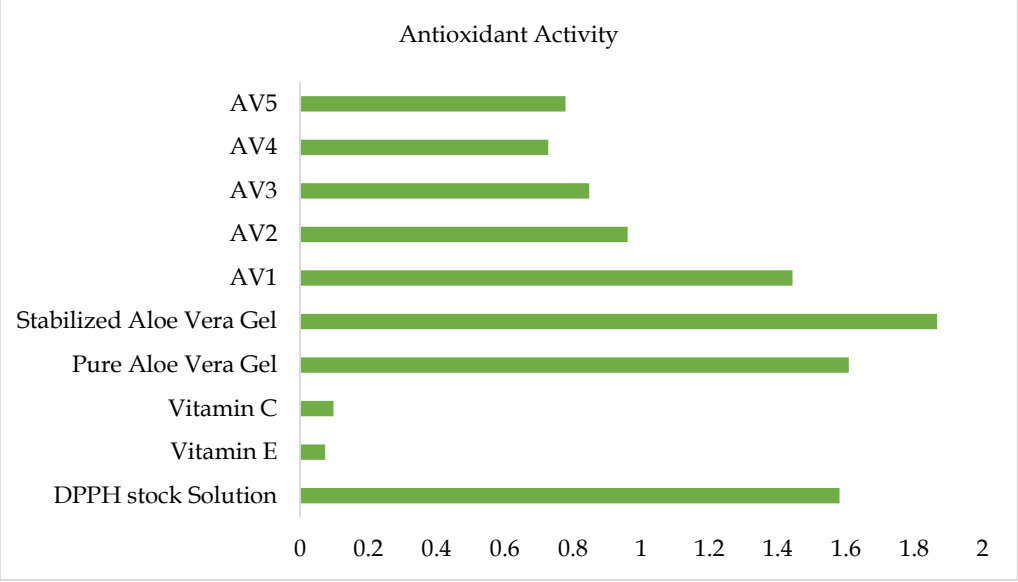

Figure 1. Antioxidant activity of pure Aloe Vera gel, stabilized Aloe Vera gel and RTS drinks made from stabilized Aloe Vera gel, determined through DPPH scavenging technique

4.7 Organoleptic Evaluation of RTS Aloe Vera Drinks

Organoleptic evaluation was performed on fresh and stored RTS *Aloe Vera* drinks using a composite scoring technique. Fresh AV1 and AV2 received the highest scores for most sensory aspects, including appearance, mouthfeel, flavor, aroma, and overall acceptability. Stored drinks showed minimal changes in sensory attributes, indicating that storage did not significantly affect their quality. The use of preservatives and carefully maintained storage conditions likely contributed to this result (Karmakar, 2017; Amanpour

et al., 2015; Nawar, 2018; Muhammad et al., 2015; Saccà et al., 2019). The organoleptic evaluation of RTS *Aloe Vera* drinks is an important aspect of product development, as it provides insight into the sensory properties of the product and allows for optimization of the formulation to ensure consumer acceptability and satisfaction (Shah et al., 2020). There wasn't much significant difference between the sensory attributes of fresh and stored (for three months) RTS drinks. This result is in line with the findings of a study by Muhammad et al. (2015), which reported that there was no significant difference in sensory characteristics of *Aloe Vera* juice stored for up to 90 days at refrigeration temperature. This may be attributed to the use of preservatives to stabilize the *Aloe Vera* gel in the formulation of the drinks, as well as the storage conditions that were carefully maintained during the three-month storage period. It is worth noting that the slight decrease in flavor scores in the stored samples may be due to the breakdown of some flavor compounds over time, as reported by Saccà et al. (2019). Nevertheless, the overall acceptability scores remain relatively high, indicating that the stored drinks are still considered to be a palatable and refreshing beverage. Therefore, it can be concluded that the sensory quality of the RTS *Aloe Vera* drinks was not significantly affected by three months of refrigerated storage.

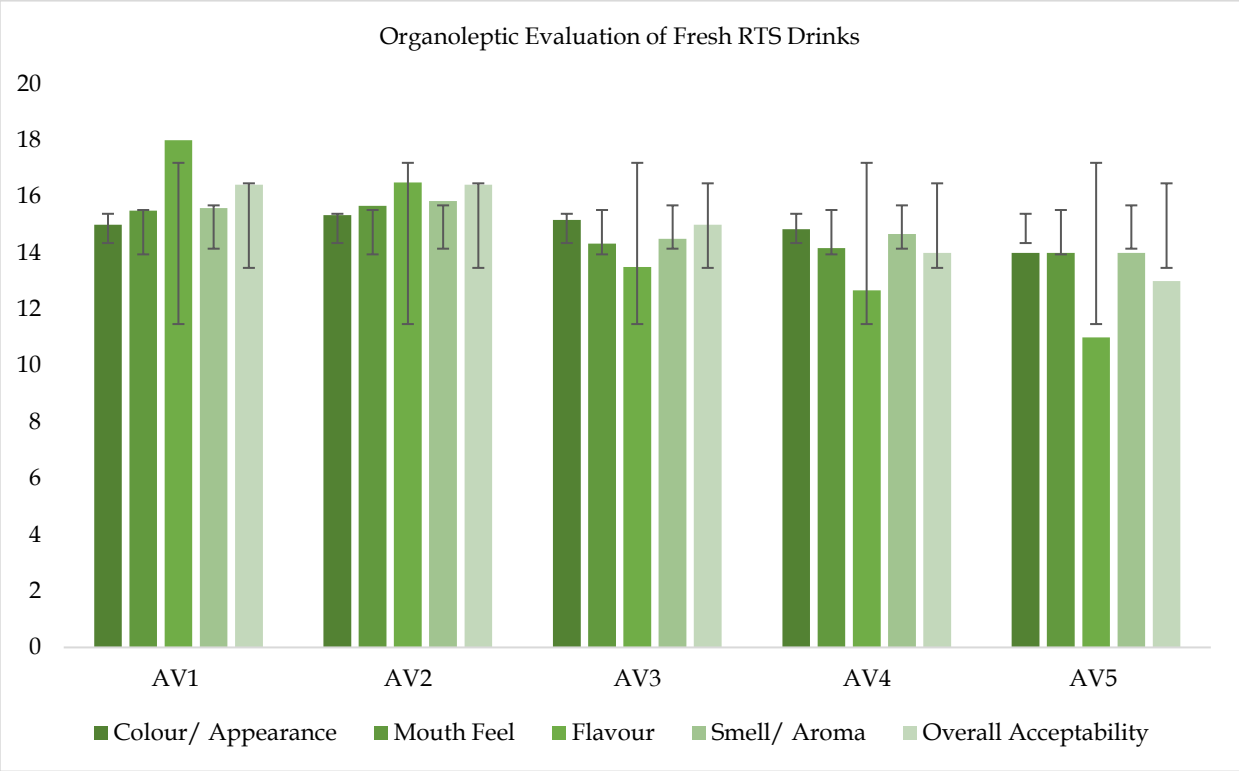

Figure 2. Results of Fresh RTS Drinks on Different Sensory Aspects

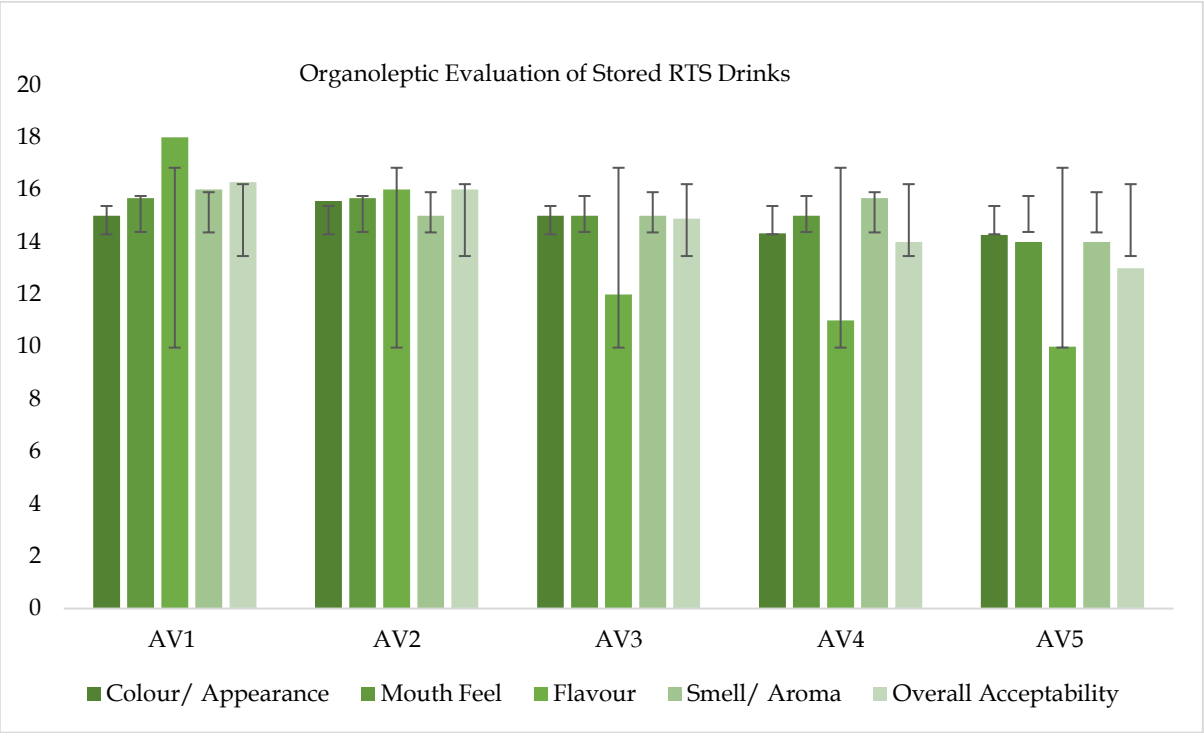

Figure 3. Results of stored RTS Drinks on Different Sensory Aspects

4.8 Sensory and Nutritional Attributes of the Most Preferred RTS Aloe Vera Drink

AV1 and AV2 RTS drinks were found to be the most liked by consumers through sensory evaluation. The sensory evaluation conducted by the panelists assessed the appearance, flavor, texture, and overall acceptability of the drinks. The high overall acceptability scores of AV1 and AV2 suggest that these drinks were well-liked by the consumers. Regarding proximal nutritional attributes, AV1 and AV2 had similar compositions. Both drinks were low in calories, with AV1 containing 24 kcal/100mL and AV2 containing 22 kcal/100mL. This low caloric value could make them suitable for consumers who are watching their calorie intake. The proximal nutritional analysis of AV1 and AV2 drinks revealed that they are a good source of antioxidants, dietary fiber, and vitamins. The drinks contain preserved *aloe vera* gel, which is known for its antioxidant and anti-inflammatory properties (Kim et al., 2009). *Aloe vera* gel is rich in vitamins A, C, and E, and minerals such as magnesium, calcium, and zinc (Salehi et al., 2020). Consuming AV1 and AV2 RTS drinks may provide several health benefits due to the presence of *aloe vera* gel. *Aloe vera* gel has been reported to have various health benefits, including wound healing, anti-inflammatory effects, and improving digestion (Al-Sadi et al., 2020). The high antioxidant content of the drinks may also protect against oxidative stress and reduce the risk of chronic diseases such as heart disease and cancer (Chávez-Santoscoy et al., 2015).

5. Conclusion

The results of this study confirm the potential of *Aloe Vera* gel as a functional food ingredient. The combination of heat treatment and chemical preservatives was found to be the best method for stabilizing *Aloe Vera* gel, effectively increasing its stability and shelf life. Potassium sorbate, Vitamin E, and citric acid were identified as effective preservatives for extending the storage life of pure *Aloe Vera* gel, while sodium benzoate showed limited effectiveness.

A functional ready-to-serve *Aloe Vera* drink was successfully formulated using *Aloe Vera* gel, water, sugar, and other ingredients. The optimized formulation exhibited acceptable taste, color, and aroma. The RTS functional drinks had higher antioxidant activity compared to pure *Aloe Vera* gel, suggesting that the inclusion of other ingredients enhanced the antioxidant properties. Nevertheless, pure *Aloe Vera* gel still exhibited significant antioxidant activity, supporting its use as an ingredient in functional drinks. The findings have significant implications for the food industry and consumers, highlighting the best methods for stabilizing *Aloe Vera* gel and formulating functional drinks. *Aloe Vera* gel

offers potential health benefits as a functional food ingredient, contributing to overall health and wellness. Further research is recommended to explore the effects of different preservatives on gel quality, sensory attributes of RTS drinks, additional health benefits of preserved *Aloe Vera* gel, utilization in other food products, and the use of natural preservatives. Implementing these recommendations can lead to improved quality, acceptability, and nutritional value of *Aloe Vera* gel products, satisfying consumer demand for natural and healthy food options and promoting better health outcomes.

Conflict of Interest

The authors declare no conflict of interest.

Acknowledgment

The work presented in this paper, "Evaluation of Different Processes to Stabilize and Preserve Pure *Aloe Vera* Gel for the development of Ready to Serve Functional Drinks" is my own effort. This work has not been submitted previously, in whole or in part, to qualify for any other academic awards. The content of the thesis is the result of the work which has been carried out since the official commencement date of the approved research program. Any editorial work, paid or unpaid, carried out by a third party must be acknowledged. Ethical procedures and guidelines have been followed.

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