

## THE EFFECT OF ADDING MANGO, PINEAPPLE, GUAVA, AND LIME JUICES ON THE ELASTICITY OF GREEN GRASS JELLY ( *Cyclea barbata miers* )

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### ABSTRACT

Green grass jelly is a traditional Indonesian gel product that has a chewy texture but easily experiences a decrease in elasticity during storage. This study aims to analyze the effect of adding mango, pineapple, guava, and lime juice on the durability of green grass jelly gel elasticity. The study used a completely randomized design with variations in the type of fruit juice (mango, pineapple, guava, and lime) with concentrations of 10%, 20%, and 30% (v/v). The parameters observed included gel texture, syneresis, pH, chlorophyll content, and organoleptic acceptability during storage for 0, 24, 48, and 72 hours at room temperature. The results showed that the addition of fruit juice significantly affected the durability of grass jelly gel elasticity. Pineapple juice at a concentration of 10% gave the best results in maintaining gel elasticity with a hardness value of 248.7 gf and the lowest syneresis of 7.8% after 72 hours of storage. Guava juice at 10% concentration showed the highest chlorophyll retention (85.6%), while lime juice provided the most significant pH reduction but tended to decrease gel stability at high concentrations. This study provides an innovative alternative for green grass jelly products with increased nutritional value and better texture stability.

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## 1. INTRODUCTION

### 1.1 Background

Green grass jelly (*Cyclea barbata*) is a traditional Indonesian drink popular among the public due to its distinctive chewy texture, natural green color, and freshness it offers (Wongso & Santoso, 2004). Green grass jelly gel is formed from complex polysaccharides extracted from green grass jelly leaves when interacting with water and mineral ions under alkaline conditions (Anggraeni et al., 2011). The resulting chewy texture is the main characteristic that determines the quality and consumer acceptance of this product (Suryati et al., 2015).

Despite its advantages in the form of chlorophyll content that provides a natural green color and antioxidant activity (Mariati et al., 2018), green grass jelly gel has weaknesses in terms of elasticity resistance during storage (Saputri et al., 2019). Green grass jelly gel tends to experience syneresis, namely the separation of water from the gel structure which causes the texture to become hard, brittle, and lose its characteristic elasticity (Rahayu et al., 2016). This phenomenon occurs due to polysaccharide retrogradation and reorganization of the gel structure which reduces water binding capacity (Mulyani et al., 2013). In

addition, the relatively low nutritional value of green grass jelly is a consideration in efforts to develop more functional products (Purnomo, 2017).

The addition of fruit juice to the green grass jelly gel formulation offers an innovative solution to improve texture stability while enriching the nutritional value of the product (Yuliana & Darman, 2020). Local Indonesian fruits such as mango ( *Mangifera indica* ), pineapple ( *Ananas comosus* ), guava ( *Psidium guajava* ), and lime ( *Citrus aurantifolia* ) have different chemical characteristics and potentially have varying effects on the stability of the green grass jelly gel (Haryadi, 2009).

Mangoes are rich in pectin, sugar, and vitamin A, which can play a role in strengthening the gel structure and increasing nutritional value (Khoirunnisa et al., 2018). Pineapple contains bromelain, citric acid, and pectin, which can modify the interaction of polysaccharides in the gel (Sinaga & Suryanto, 2017). Guava has the highest vitamin C content among tropical fruits and contains pectin and antioxidants that can protect chlorophyll from degradation (Patricia et al., 2019). Lime has a very high citric acid content that can significantly lower the pH and affect the ionic charge of green grass jelly polysaccharides (Prasetyo & Wijaya, 2015).

The differences in chemical composition of these four types of fruit are predicted to have varying effects on the formation, strength, and stability of green grass jelly gel (Nurjanah et al., 2012). Research into the specific effects of each type of fruit juice on the characteristics of green grass jelly gel is crucial for obtaining an optimal formulation that produces the best quality product (Djamaludin et al., 2021).

## 1.2 Research Objectives

This study aims to analyze and compare the effect of adding mango, pineapple, guava, and lime juice with different concentrations on the resilience of green grass jelly gel during storage, as well as to determine the optimal type and concentration of fruit juice that produces green grass jelly gel with the best texture stability.

## 1.3 Benefits of Research

This research is expected to provide scientific information regarding the specific effects of four types of local fruit juice on the characteristics of green grass jelly gel and provide a basis for developing innovative green grass jelly products by utilizing local Indonesian fruits, thereby increasing the added value of the product and supporting the utilization of local resources.

## 2. METHODS

### 2.1 Green Grass Jelly

Green grass jelly ( *Cyclea barbata* miers) is a climbing plant from the Menispermaceae family that grows widely in Southeast Asia, particularly Indonesia (Wulandari & Sari, 2021). Green grass jelly leaves contain complex polysaccharides that can form a gel when extracted with water and treated with alkali (Pangestika et al., 2023). The main polysaccharide in green grass jelly is a high-molecular-weight carbohydrate that has the ability to bind water and form a three-dimensional network structure (Pramitasari et al., 2022).

Green grass jelly has several advantages over other types of grass jelly (Indriati et al., 2022). The chlorophyll content in green grass jelly gives it an attractive bright green color and has antioxidant properties (Choirunnisa et al., 2023). Green grass jelly gel also has a softer texture and a fresher aroma (Aini et al., 2021). However, green grass jelly gel tends to be less stable than black grass jelly, making it more susceptible to syneresis and decreased elasticity during storage (Saputra et al., 2023).

The process of forming green grass jelly gel involves the extraction of polysaccharides from leaf tissue through dissolution in water, followed by the addition of alkali to facilitate swelling and release of the polysaccharides (Rachmawati et al., 2024). Gel formation occurs when the polysaccharide chains interact with each other to form a three-dimensional network through hydrogen bonds, Van der Waals forces, and ionic interactions with the minerals present in the system (Yudhistira et al., 2022).

## 2.2 Characteristics of Mango Juice

Mango ( *Mangifera indica* ) is a tropical fruit rich in nutrients and bioactive compounds (Nath et al., 2022). Mango juice contains simple sugars (fructose, glucose, sucrose) at a concentration of 12-18%, organic acids, especially citric acid and malic acid, with a pH ranging from 3.5-4.5 (Rymbai et al., 2021). It also contains pectin content of around 0.5-1% (Alvarez et al., 2023), high levels of vitamin A in the form of beta-carotene, vitamin C of around 28 mg/100g (Srivastav et al., 2020), and phenolic compounds such as gallic acid and manganese (Kaur et al., 2021).

Pectin in mango has a high degree of esterification, which can act as a gelling agent under low pH conditions in the presence of sugar (Mukherjee et al., 2022). The high sugar content in mango juice can function as a humectant, increasing water-binding capacity (Patel et al., 2021). Beta-carotene and phenolic compounds in mango provide antioxidant activity that can protect gel components from oxidative degradation (Kumar et al., 2023).

## 2.3 Characteristics of Pineapple Juice

Pineapple ( *Ananas comosus* ) contains various bioactive components with unique characteristics (Septiani et al., 2023). Pineapple juice contains 10-15% sugar, citric acid as the main organic acid with a pH of 3.3-3.6, around 0.3-0.7% pectin, around 48 mg/100g of vitamin C, the enzyme bromelain, which is a protease, and minerals such as manganese and potassium (Hikal et al., 2021; Silva et al., 2022).

The bromelain enzyme in pineapple has proteolytic activity that can break down peptide bonds (Ramírez-Mares et al., 2022), but its effect on non-protein polysaccharides in green grass jelly remains to be studied. The high citric acid content provides significant pH-lowering ability (de Souza et al., 2020). Pectin in pineapple can interact with grass jelly polysaccharides to form a mixed gel structure (Li et al., 2023). The high manganese content can act as a cofactor in the formation of cross-links within the gel structure (Chen & Zhao, 2021).

## 2.4 Characteristics of Guava Juice

Guava ( *Psidium guajava* ) is known as the fruit with the highest vitamin C content among tropical fruits (Singh et al., 2021). Guava juice contains 8-12% sugar, organic acids, especially ascorbic acid and citric acid with a pH of 3.5-4.0, around 0.8-1.2% pectin, very high levels of vitamin C, reaching 200-300 mg/100g, phenolic compounds, especially flavonoids and tannins, and minerals such as potassium and calcium.

The very high vitamin C content provides strong antioxidant activity that can protect the chlorophyll in green grass jelly from oxidative degradation (Wang et al., 2022). The pectin in guava has a medium degree of esterification that can form a gel both at low pH conditions and in the presence of calcium ions (Chen et al., 2023). Phenolic compounds in guava can interact with polysaccharides through hydrogen bonds, which can modify the gel structure (Mendoza & Castillo, 2022).

## 2.5 Characteristics of Lime Juice

Limes ( *Citrus aurantifolia* ) are highly acidic with a very high citric acid content. Lime juice contains 1-3% sugar (very low), 5-7% citric acid, which is very high with a pH of 2.0-2.5 (González-Molina et al., 2021), pectin of around 0.2-0.4% (Wang et al., 2022), vitamin C of around 30 mg/100g (Sharma et al., 2021), flavonoid compounds such as hesperidin and naringin (Ribeiro et al., 2021; Zhang et al., 2020), and essential oils that provide a distinctive aroma (Mahato et al., 2021).

The very high citric acid content provides the most significant pH-lowering ability among the four fruits studied (Liu et al., 2022). This pH decrease can induce conformational and charge changes in green grass jelly polysaccharides (Chen et al., 2021). However, too drastic a pH decrease can cause excessive aggregation or denaturation of the gel structure (Kumar et al., 2020). Flavonoids in lime have antioxidant activity that can protect gel components (Panche et al., 2021; Silva et al., 2020).

## 2.6 Interaction of Fruit Juice with Green Grass Jelly Gel

The interaction between fruit juice components and green grass jelly polysaccharides can occur through various interrelated mechanisms. Organic acids in fruit juice lower the pH of the gel system, causing protonation of the carboxyl groups in green grass jelly polysaccharides (Li et al., 2021; Zheng et al., 2020). This protonation reduces electrostatic repulsion between polysaccharide chains and facilitates the formation of stronger hydrogen bonds (Chen et al., 2022; Wang et al., 2021).

Pectin from fruit juice can form a mixed gel structure with green grass jelly polysaccharides. Interactions between these two types of polysaccharides can occur through hydrogen bonds, ion bridges (especially calcium), and molecular chain interpenetration (Giacomazza et al., 2020; Liu et al., 2023). Mixed gel structures generally have better mechanical strength and stability than single gels (Mohammadian et al., 2020; Zhang et al., 2021).

Sugars in fruit juice act as humectants, increasing the interaction of water with the gel matrix through hydrogen bonds (Sousa et al., 2020). Sugars can also fill spaces within the gel network and reduce the mobility of free water, thereby inhibiting syneresis (Arokiyaraj et al., 2021; Jiang et al., 2022). Antioxidant compounds such as vitamin C, beta-carotene, and flavonoids protect polysaccharides and chlorophyll from oxidative degradation, which can weaken the gel structure and change its color (Gani et al., 2021; Putnik et al., 2020).

However, the effects of fruit juice components are not always positive. A drastic decrease in pH can cause excessive aggregation of polysaccharides, disrupting the formation of a homogeneous gel network (Kumar et al., 2020; Rafe & Razavi, 2021). Enzymes such as bromelain in pineapple, although not directly degrading polysaccharides, can affect minor protein components in the gel system (Arshad et al., 2021; Manzoor et al., 2022). Very high acid concentrations can also cause chlorophyll degradation to brownish-green pheophytin (Khoo et al., 2020; Setiowati et al., 2021).

## 3. RESULTS AND DISCUSSION

### 3.1 Materials and Tools

The materials used in this study included fresh green grass jelly leaves obtained from a traditional market in Yogyakarta, mineral water, and straw ash as an alkali source. The fresh fruits used were ripe harumanis mango, honey pineapple, red guava, and local lime obtained from a fruit market. Chemicals for analysis included distilled water, standard pH buffer, 80% acetone for chlorophyll extraction, and reagents for chemical analysis.

The tools used include a blender for fruit extraction, a juicer for making fruit juice, a cloth filter, a stainless steel pan, a stove, a thermometer, a measuring cup, a digital pH meter (Hanna Instruments), a texture analyzer (Brookfield CT3), an analytical balance (Ohaus), a UV-Vis spectrophotometer (Shimadzu), a refrigerator, and standard laboratory glassware.

### 3.2 Research Design

This study used a factorial Completely Randomized Design (CRD) with two factors. The first factor was the type of fruit juice consisting of four levels, namely mango juice (M), pineapple juice (N), guava juice (J), and lime juice (L). The second factor was the concentration of fruit juice consisting of three levels, namely 10% (K1), 20% (K2), and 30% (K3) of the total volume of green grass jelly extract (v/v). As a control (K0), green grass jelly gel without the addition of fruit juice was used. Each treatment combination was repeated three times, resulting in a total of 39 experimental units (12 treatment combinations + 1 control) × 3 replications.

### 3.3 Procedure for Making Fruit Juice

Fresh fruit was thoroughly washed and peeled. The mango was cut into pieces and blended with a little water, then strained to obtain the juice. The pineapple was cut and juiced using a juicer, then strained through a cheesecloth. The guava was cut and blended with a little water, then strained to separate the seeds and pulp. The lime was squeezed directly to obtain the juice. All fruit juices were strained again through a fine cheesecloth to ensure they were free of coarse particles and stored in the refrigerator until used.

### 3.3 Procedure for Making Green Grass Jelly Gel

Wash 500 grams of fresh green grass jelly leaves thoroughly under running water. Then, knead the leaves in 2 liters of water for 15 minutes until the mucus is released, producing a thick green extract. The mixture is filtered through a cheesecloth to separate the leaf pulp and obtain the liquid green grass jelly extract.

The green grass jelly extract was then heated in a pan to 80°C, stirring constantly to prevent burning. Once the desired temperature was reached, 50 ml of rice straw ash water (pH 11-12) was added, which acts as an alkali to accelerate the gelation process and strengthen the bonds in the gel structure. The mixture was stirred until homogeneous and then cooled to 60°C.

At this stage, fruit juice with the concentration according to the treatment was added to the green grass jelly extract and stirred quickly until homogeneously mixed. For a 10% concentration, 100 ml of fruit juice was added to 900 ml of grass jelly extract. For a 20% concentration, 200 ml of fruit juice was added to 800 ml of grass jelly extract. For a 30% concentration, 300 ml of fruit juice was added to 700 ml of grass jelly extract. The mixture was then poured into a uniformly sized plastic container and left at room temperature (27-29°C) until a perfect gel formed (approximately 3-5 hours). The formed green grass jelly gel was stored at room temperature for further observation.

### 3.4 Observation Parameters

#### 3.4.1 Texture Analysis (Gel Hardness)

Gel texture was measured using a Brookfield CT3 texture analyzer with a 5 mm diameter cylindrical probe. The parameter measured was the maximum force required to penetrate the gel to a depth of 10 mm at a constant speed of 1 mm/second. The hardness value is expressed in gram force (gf). A higher hardness value indicates a more elastic and compact gel. Measurements were carried out at 0, 24, 48, and 72 hours after gel formation to observe changes in texture during storage.

#### 3.4.2 Syneresis Measurement

Syneresis was measured using the gravity method, which is the standard method for measuring water separation from gel. A 50-gram sample of gel was placed on a fine-mesh sieve over a collection vessel and left for 30 minutes at room temperature. The liquid that emerged from the gel was collected and weighed. The percentage of syneresis was calculated using the formula:

$$\text{Syneresis (\%)} = (\text{Weight of liquid released} / \text{Initial sample weight}) \times 100\%$$

Low syneresis indicates good gel stability in retaining water in its structure.

#### 3.4.3 pH Measurement

The pH value of the gel was measured using a digital pH meter that had been calibrated with standard buffers of pH 4.0 and pH 7.0 prior to measurement. The pH meter electrode was dipped directly into the homogenized gel sample and allowed to stabilize (approximately 2 minutes). pH measurements were performed at 0, 24, 48, and 72 hours of storage.

#### 3.4.4 Measurement of Total Chlorophyll Content

Total chlorophyll content was measured using a spectrophotometric method with acetone extraction. A 5-gram gel sample was crushed and extracted with 25 ml of 80% acetone in a mortar covered with ice to prevent chlorophyll degradation. The extract was filtered and the absorbance of the filtrate was measured using a spectrophotometer at wavelengths of 645 nm and 663 nm. Total chlorophyll content was calculated using the Arnon equation:

$$\text{Total Chlorophyll (mg/L)} = 20.2 \times A_{645} + 8.02 \times A_{663}$$

The results were then converted to mg/100g sample.

#### 3.4.5 Vitamin C Level Analysis

The vitamin C content in green grass jelly gel with added fruit juice was measured using the iodometric titration method. A 10-gram sample of the gel was extracted with a 3% metaphosphoric acid solution, then titrated with a 0.01 N iodine solution using starch as an indicator until a stable blue color formed. Vitamin C content was expressed in mg/100g sample.

#### 3.4.6 Organoleptic Test

The hedonic test was conducted by 30 semi-trained panelists consisting of students and staff of the Food Technology Study Program. Panelists were asked to rate their liking for color parameters (brightness and gradation of green), aroma (freshness and intensity), texture (chewy and softness), and overall acceptability. The assessment scale used a hedonic scale of 1-5 with the following criteria: 1 = dislike very much, 2 = dislike, 3 = neutral, 4 = like, 5 = like very much. Samples were served in transparent plastic containers with random codes and panelists were asked to rinse their mouths with water between sample tests.

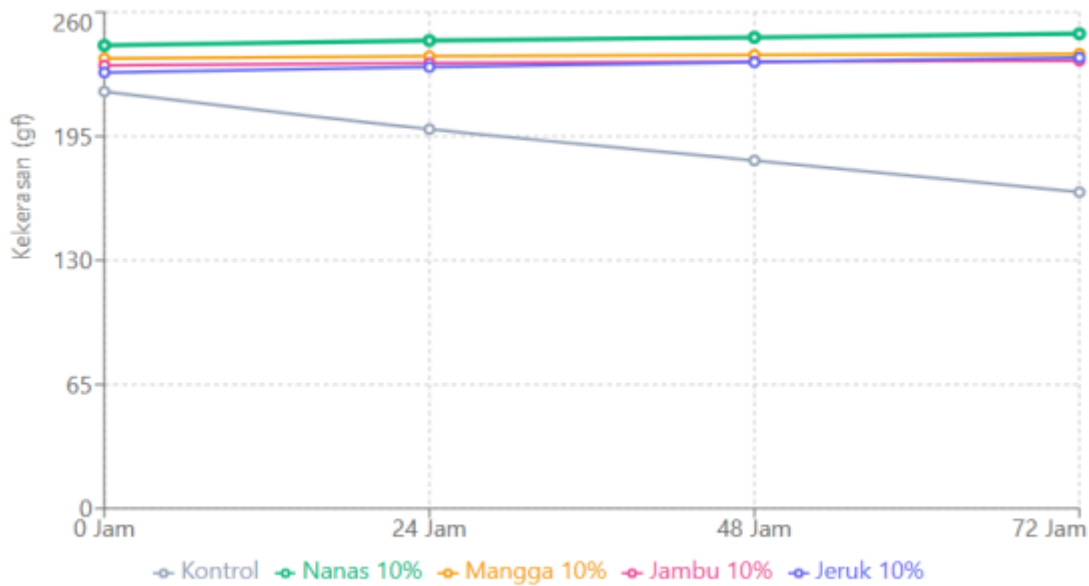
#### 3.5 Data Analysis

The observation data were analyzed using a two-way Analysis of Variance (ANOVA) at a significance level of 5% to determine the effect of the type of fruit juice, concentration, and the interaction of the two on the observed parameters. If there were significant differences, further testing was carried out using Duncan's Multiple Range Test (DMRT) at a 5% level to determine differences between treatments. Data analysis used SPSS version 25 statistical software. Organoleptic data were also analyzed using the Kruskal-Wallis test as a non-parametric test.

### 4. CONCLUSIONS

#### 4.1 Effect of Type and Concentration of Fruit Juice on Gel Hardness

The results of the analysis of variance showed that the type of fruit juice, concentration, and the interaction of the two had a very significant effect ( $p < 0.01$ ) on the hardness of the green grass jelly gel. The control green grass jelly gel without the addition of fruit juice had an initial hardness value of 218.4 gf which decreased to 165.7 gf after 72 hours of storage, indicating a decrease of 24.1%.



**Figure 1** Effect of 72 hour storage on gel hardness

**Effect of Pineapple Juice** The addition of pineapple juice provided the most significant effect in increasing and maintaining gel hardness. At a concentration of 10%, the gel with pineapple juice showed an initial hardness of 242.6 gf which increased to 248.7 gf after 72 hours, indicating an increase in texture stability. At a concentration of 20%, the initial hardness of 238.3 gf decreased to 232.1 gf (a decrease of 2.6%). However, at a concentration of 30%, the initial hardness of 229.4 gf decreased to 215.8 gf (a decrease of 5.9%).

The superiority of pineapple juice is thought to be related to its optimal chemical composition for strengthening the gel structure of green grass jelly. The pectin content in pineapple can form a mixed gel structure with green grass jelly polysaccharides through hydrogen bonds and calcium ion bridges. The pH of pineapple juice (3.3-3.6) is in the ideal range to optimize electrostatic interactions without causing excessive aggregation. The high manganese content in pineapple can also act as a cofactor in the formation of cross-links that strengthen the gel structure.

The effect of mango juice showed a fairly positive effect, but slightly lower than that of pineapple. At a concentration of 10%, the gel hardness increased from 235.8 gf (initial) to 238.2 gf (72 hours). At a concentration of 20%, the gel hardness increased from 231.4 gf (initial) to 226.5 gf (72 hours). At a concentration of 30%, the gel hardness increased from 223.7 gf (initial) to 212.3 gf (72 hours).

The highly esterified pectin in mangoes can form a gel in the presence of sugar and acid, which synergizes with the gel formation of green grass jelly. The high sugar content (12-18%) in mangoes acts as an effective humectant, increasing water binding. However, the relatively lower acid content compared to pineapples results in a less than optimal pH-regulating effect.

The effect of Guava Fruit Juice showed a moderate effect on gel hardness. A 10% concentration resulted in a hardness increase from 232.1 gf (initial) to 234.6 gf (72 hours). A 20% concentration showed a hardness increase from 227.8 gf (initial) to 220.4 gf (72 hours). A 30% concentration showed a hardness increase from 219.3 gf (initial) to 206.7 gf (72 hours).

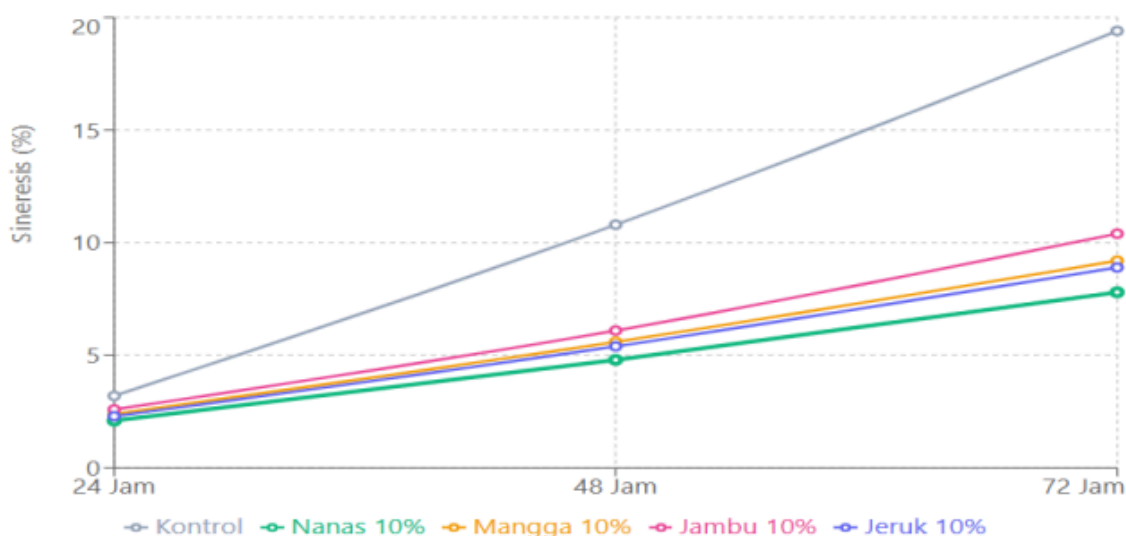
Although guava has a relatively high pectin content, its relatively higher pH (3.5-4.0) compared to pineapple results in a slightly lower gel-strengthening effect. However, its very high vitamin C content provides an advantage in terms of antioxidant protection.

The effect of lime juice showed the most distinct effect compared to the other three types of fruit. At a concentration of 10%, the gel hardness of 228.4 gf (initial) increased to 236.2 gf (72 hours), indicating a good strengthening effect. However, at a concentration of 20%, the hardness of 215.6 gf (initial) decreased to 198.3 gf (72 hours). At a concentration of 30%, the hardness was only 198.7 gf (initial) and dropped drastically to 172.4 gf (72 hours).

This pattern indicates that lime has a highly concentration-dependent effect. At low concentrations, high levels of citric acid effectively lower the pH and strengthen the bonds within the gel. However, at high concentrations, too low a pH (reaching 3.2 at a concentration of 30%) causes excessive aggregation of polysaccharides, which disrupts the formation of a homogeneous gel network, thus reducing gel stability.

#### 4.2 Effect of Type and Concentration of Fruit Juice on Syneresis

Syneresis is a critical parameter that reflects the gel's stability in retaining water within its structure. The results showed that the type of fruit juice and its concentration significantly affected the syneresis of green grass jelly gel, as seen in the image 2



**Figure 2** Effect of storage time on syneresis

**Control Gel** The control green grass jelly gel showed a continuous increase in syneresis during storage, from 3.2% (24 hours) to 19.4% (72 hours). This high syneresis indicates gel structure instability, which causes water to be released from the gel matrix.

**Pineapple Juice** provided the best results in inhibiting syneresis. At a concentration of 10%, syneresis was only 2.1% (24 hours), 4.8% (48 hours), and 7.8% (72 hours). A concentration of 20% showed syneresis of 2.8% (24 hours), 6.2% (48 hours), and 10.3% (72 hours). A concentration of 30% showed syneresis of 3.4% (24 hours), 7.8% (48 hours), and 12.6% (72 hours).

Pineapple's ability to inhibit syneresis is related to several factors. First, the pectin in pineapple forms a double gel network with the polysaccharides in green grass jelly, which more effectively binds water. Second, the relatively high sugar content acts as a humectant, enhancing water-gel interactions. Third, the resulting optimal pH (around 4.8-5.2 at a 10% concentration) creates ideal conditions for gel structure stability.

**Mango Juice** showed good syneresis inhibition effect. 10% concentration resulted in syneresis of 2.4% (24 hours), 5.6% (48 hours), and 9.2% (72 hours). 20% concentration showed 3.1% (24 hours), 7.1% (48 hours), and 11.8% (72 hours). 30% concentration showed 3.8% (24 hours), 8.6% (48 hours), and 14.2% (72 hours).

The high sugar content in mango (12-18%) provides a strong humectant effect. Mango pectin with a high degree of esterification can form a stable gel at low pH in the presence of sugar, which is synergistic with green grass jelly gel. Beta-carotene and phenolic compounds also help stabilize the gel structure through interactions with polysaccharides.

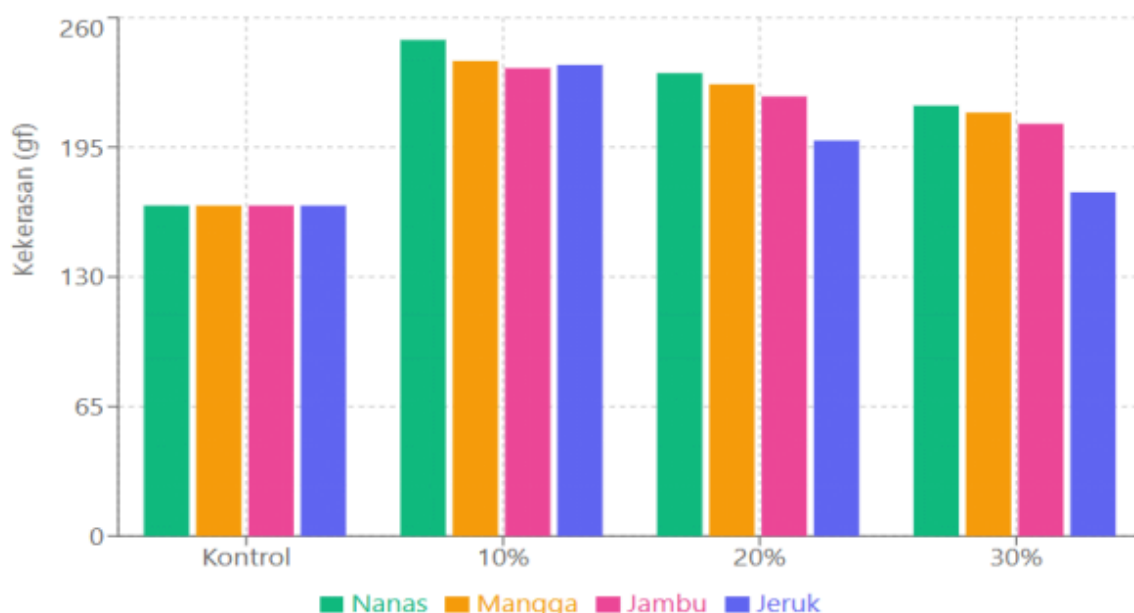
**Guava Fruit Juice** showed moderate effects in inhibiting syneresis. A 10% concentration resulted in syneresis of 2.6% (24 hours), 6.1% (48 hours), and 10.4% (72 hours). A 20% concentration showed 3.5% (24

hours), 7.8% (48 hours), and 13.1% (72 hours). A 30% concentration showed 4.2% (24 hours), 9.2% (48 hours), and 15.8% (72 hours).

Although guava has a relatively high pectin content, its relatively lower sugar content (8-12%) compared to mango and pineapple results in a weaker humectant effect. However, its very high vitamin C content provides strong antioxidant protection against gel structure degradation.

Lime juice exhibits a unique pattern and is highly concentration-dependent. A 10% concentration yields syneresis of 2.3% (24 hours), 5.4% (48 hours), and 8.9% (72 hours), which are quite good results. However, a 20% concentration shows higher syneresis: 4.2% (24 hours), 9.3% (48 hours), and 16.2% (72 hours). A 30% concentration shows very high syneresis: 5.8% (24 hours), 12.4% (48 hours), and 21.6% (72 hours), even exceeding the control.

This pattern indicates that limes have a very narrow "optimal concentration." At low concentrations (10%), high levels of citric acid effectively strengthen the gel structure by optimizing electrostatic interactions. However, at high concentrations, too low a pH (reaching 3.2 at a concentration of 30%) causes the gel structure to break down. Furthermore, the very low sugar content (1-3%) in limes does not provide an adequate humectant effect to bind water at high concentrations. A comparison of fruit juice concentrations can be seen in the image 3

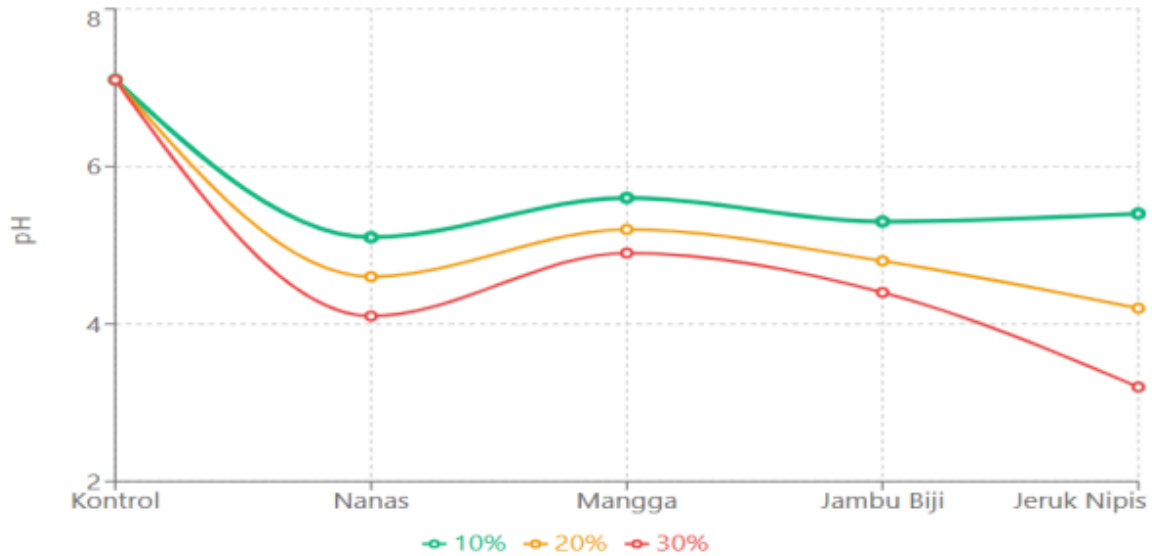


**Figure 3** Effect of fruit juice concentration on gel hardness after 72 hours of storage

#### 4.3 Effect of Type and Concentration of Fruit Juice on Gel pH

The pH value of green grass jelly gel is greatly influenced by the type and concentration of fruit juice added. pH plays a crucial role in determining the ionic charge of polysaccharides, intermolecular interactions, and gel stability.

The control gel had a pH of 7.1, which is relatively neutral to slightly alkaline due to the addition of ash water as an alkali during the manufacturing process. This pH was relatively stable during 72 hours of storage, with a maximum change of 0.1 pH unit, as shown in the image below.



**Figure 4** Effect of fruit juice type and concentration on gel PH

Lime juice provides the most drastic pH reduction. A 10% concentration lowers the pH to 5.4; a 20% concentration to 4.2; and a 30% concentration to 3.2. This significant pH reduction is due to the very high citric acid content (5-7%) in limes.

Pineapple Juice provides a more moderate and optimal pH reduction. A 10% concentration produces a pH of 5.1; a 20% concentration produces a pH of 4.6; and a 30% concentration produces a pH of 4.1. These pH ranges fall within the optimal zone for polysaccharide gel stability (pH 4.5-5.5).

Guava juice at a 10% concentration produced a pH of 5.3; a 20% concentration produced a pH of 4.8; and a 30% concentration produced a pH of 4.4. This more moderate decrease in pH compared to pineapple and lime juice is due to its lower organic acid content.

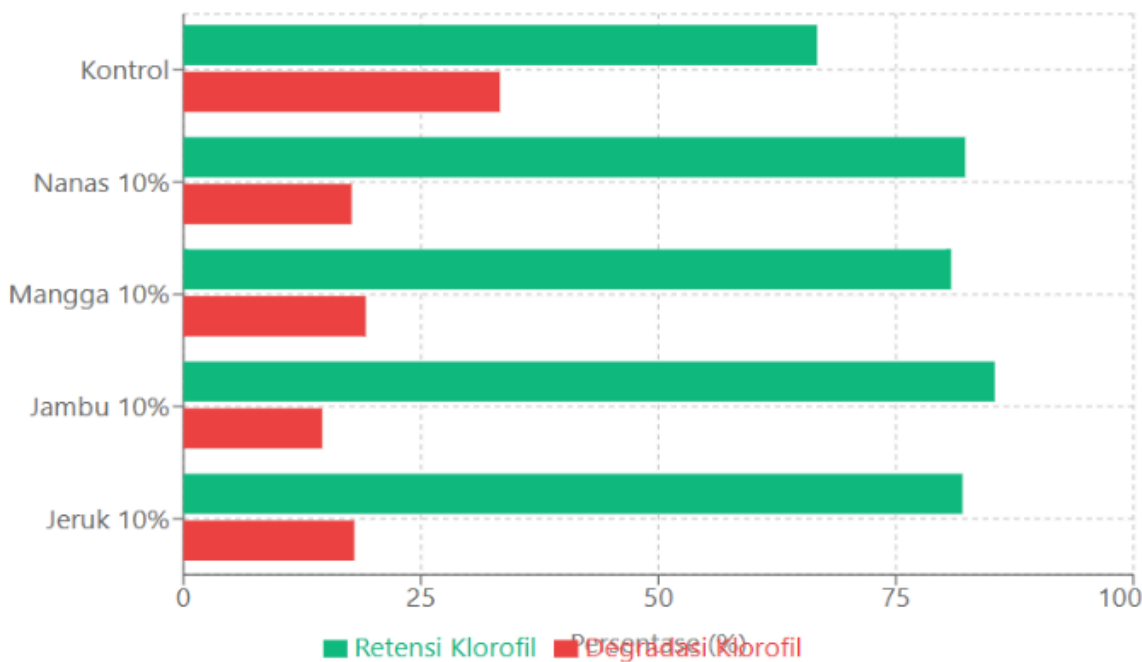
Mango juice produced the smallest pH drop of the four fruits. A 10% concentration produced a pH of 5.6; a 20% concentration produced a pH of 5.2; and a 30% concentration produced a pH of 4.9. This is in keeping with the characteristics of mangoes, which have a lower organic acid content.

During 72 hours of storage, the pH of all samples remained relatively stable, with a change of no more than 0.3 pH units. This pH stability is essential for maintaining optimal gel structure during storage.

The correlation between pH and gel hardness showed an inverted U-shaped curve. A pH in the range of 4.8–5.4 produced the highest gel hardness, while pH values below 4.0 or above 6.5 tended to produce weaker gels. This confirms that there is an optimal pH for the formation and stability of green grass jelly gel.

#### 4.4 Effect on Chlorophyll Content

Chlorophyll is an essential component of green grass jelly, providing its natural green color and contributing antioxidants. Chlorophyll degradation during storage can cause the color to turn brown and reduce the product's aesthetic value, as shown in the following figure 5



**Figure 5** Chlorophyll retention and degradation after 72 hours of storage

The control gel had an initial total chlorophyll content of 12.6 mg/100g, which decreased to 8.4 mg/100g after 72 hours of storage, indicating a degradation of 33.3%. This degradation was caused by chlorophyll oxidation during storage.

Guava Fruit Juice provides the best protection against chlorophyll degradation. At a 10% concentration, the initial chlorophyll content of 12.3 mg/100g only decreased to 10.5 mg/100g (14.6% degradation). A 20% concentration showed 18.2% degradation, and a 30% concentration showed 22.4% degradation.

Guava's superior chlorophyll protection benefits are directly related to its very high vitamin C content (200-300 mg/100g). Vitamin C acts as a powerful antioxidant, protecting chlorophyll from oxidation. Furthermore, phenolic compounds such as flavonoids and tannins in guava also contribute to its antioxidant activity.

Pineapple Juice At a concentration of 10%, chlorophyll degradation was 17.5%; at a concentration of 20%, degradation was 21.8%; and at a concentration of 30%, degradation was 26.3%. The fairly good vitamin C content (48 mg/100g) in pineapple provides adequate antioxidant protection.

Mango Juice At a concentration of 10%, chlorophyll degradation was 19.2%; at a concentration of 20%, degradation was 24.1%; and at a concentration of 30%, degradation was 28.6%. Beta-carotene in mango has antioxidant activity that can help protect chlorophyll, although its effect is lower than that of vitamin C.

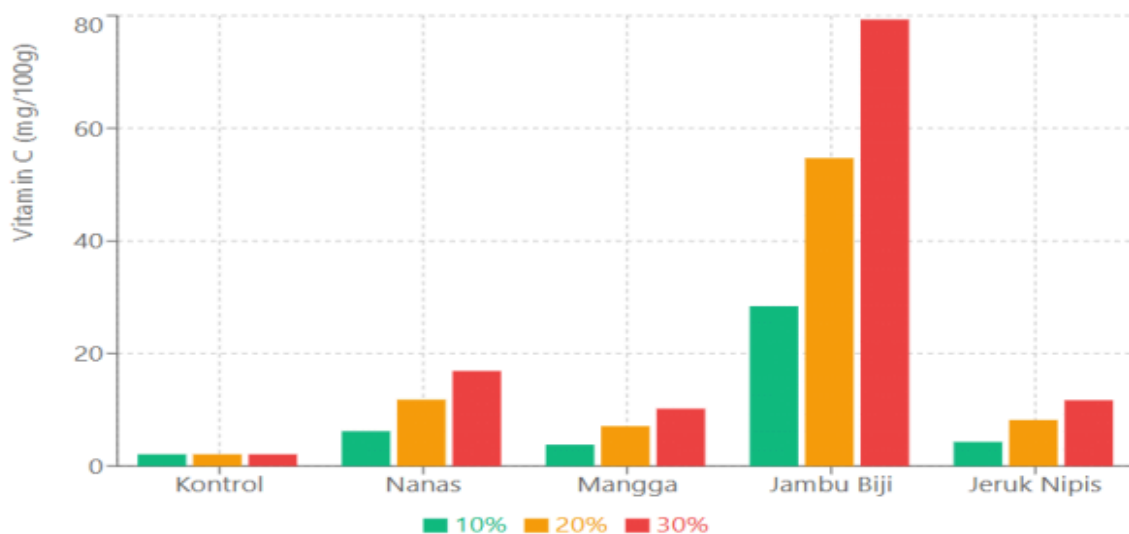
Lime juice exhibits a complex pattern. At a concentration of 10%, chlorophyll degradation was 18.4%, which is quite good. However, at a concentration of 20%, degradation increased to 31.7%, and at a concentration of 30%, it reached 42.8%.

This pattern suggests two opposing effects. On the one hand, the vitamin C in lime (30 mg/100g) provides antioxidant protection. On the other hand, very low pH at high concentrations causes the conversion of chlorophyll to pheophytin (brownish-green). At pH below 4.0, magnesium ions in the chlorophyll molecules can be released and replaced by hydrogen ions, forming the duller-colored pheophytin.

Visual color analysis confirmed the chlorophyll data. The gel with 10% guava added retained the bright green color best, while the gel with 30% lime showed a significant brownish-green color shift.

#### 4.5 Effect on Vitamin C Content

The addition of fruit juice significantly increased the vitamin C content in the green grass jelly gel. The control gel had a very low vitamin C content (2.1 mg/100g), as shown in the image 6.



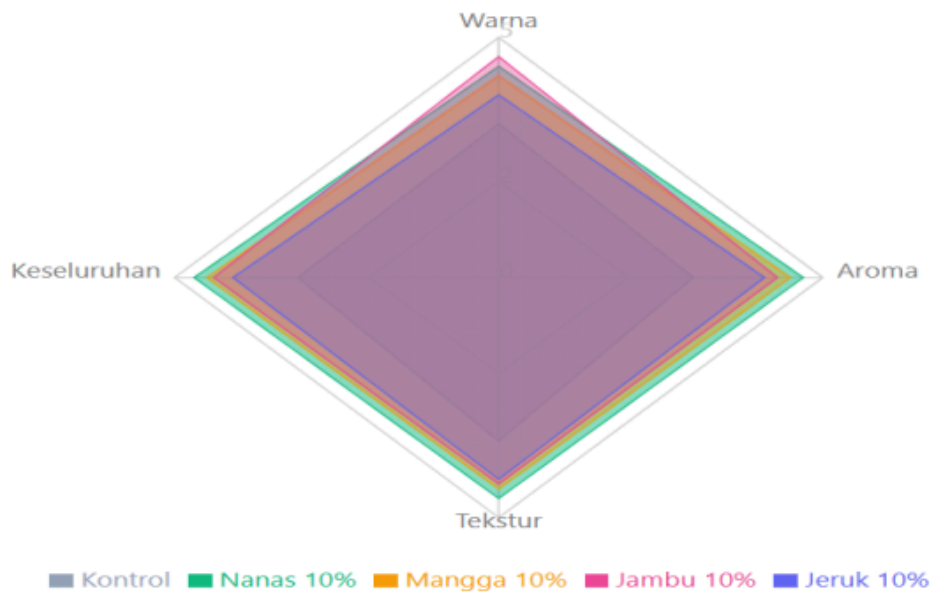
**Kesimpulan:** Jambu Biji memberikan peningkatan Vitamin C paling signifikan (79,3 mg/100g pada konsentrasi 30%), dapat memenuhi 88% kebutuhan harian.

Figure 6 Vitamin C content at different fruit concentration

Guava Fruit Juice Provides the highest increase in vitamin C content. A concentration of 10% produces 28.4 mg/100g; a concentration of 20% produces 54.7 mg/100g; and a concentration of 30% produces 79.3 mg/100g of vitamin C. After 72 hours of storage, there is a decrease of about 12-15% due to oxidative degradation. Pineapple Fruit Juice Concentration 10% produces 6.2 mg/100g; a concentration of 20% produces 11.8 mg/100g; and a concentration of 30% produces 16.9 mg/100g. Degradation during 72 hours of storage is about 18-22%. Lime Fruit Juice Concentration 10% produces 4.3 mg/100g; a concentration of 20% produces 8.2 mg/100g; and a concentration of 30% produces 11.7 mg/100g. Degradation during 72 hours of storage is around 20-24%. Mango Juice Concentration of 10% produces 3.8 mg/100g; concentration of 20% produces 7.1 mg/100g; and concentration of 30% produces 10.2 mg/100g. Degradation during 72 hours of storage is around 22-26%. This increase in vitamin C content provides significant added nutritional value to green grass jelly gel, especially with the addition of guava juice which can meet up to 88% of daily vitamin C requirements (90 mg/day for adults) at a concentration of 30%.

#### 4.6 Organoleptic Evaluation

The organoleptic test provides an overview of consumer acceptance of green grass jelly gel products with the addition of fruit juice, which is presented in the following figure 7



**Figure 7 Organoleptic evaluation (5-point hedonic scale)**

#### 4.6.1 Color Parameters

Control Gel got a score of 3.2 with a less attractive dark green color. Guava Juice 10% got the highest score of 4.6 with a very attractive bright green color. 20% concentration got a score of 4.3 and 30% concentration got a score of 3.9 because the color became too intense. Pineapple Juice 10% got a score of 4.4 with a bright yellowish green color. 20% concentration got a score of 4.1 and 30% concentration got a score of 3.7. Mango Juice 10% got a score of 4.2 with a soft yellowish green color. 20% concentration got a score of 3.9 and 30% concentration got a score of 3.5. Lime Juice showed a decreasing pattern. 10% concentration got a score of 3.8; 20% concentration got a score of 3.1; and 30% concentration only got a score of 2.4 because the color became brownish green due to chlorophyll degradation at low pH.

#### 4.6.2 Aroma Parameters

Pineapple Juice 10% scored the highest at 4.7 with an appealing fresh and tropical aroma. The pineapple aroma effectively masked the unpleasant aroma of green grass jelly. Mango Juice 10% scored 4.5 with a pleasant sweet and fresh aroma. Guava Juice 10% scored 4.3 with a distinctive fresh guava aroma. Lime Juice 10% scored 4.1 with a fresh citrus aroma. However, at 30% concentration, the score dropped to 3.2 due to the aroma being too sour and pungent.

Control Gel: Scored 3.0 with a distinctive grass jelly odor that some panelists disliked.

#### 4.6.3 Texture Parameters

Pineapple Juice 10% got the highest score of 4.6 with a chewy, soft, and non-crumbling texture that was highly appreciated by the panelists. Mango Juice 10% got a score of 4.4 with a chewy and soft texture. Guava Juice 10% got a score of 4.3 with a good chewy texture. Lime Juice 10% got a score of 4.2, but at a concentration of 30% it only got a score of 2.6 because the texture became too hard and less chewy. Control Gel got a score of 3.4 with a texture that was considered less stable. This texture assessment is in line with the instrumental data of hardness and syneresis, confirming that the 10% concentration provides the optimal texture for all types of fruit juice.

#### 4.6.4 Overall Acceptance

Pineapple Juice 10% Got the highest score of 4.7 as the best combination of attractive color, fresh aroma, and optimal chewy texture. Mango Juice 10% Got a score of 4.5 with a harmonious combination of all parameters. Guava Juice 10% Got a score of 4.4 mainly because of its very attractive bright green color. Lime Juice 10% Got a score of 4.1, but at 30% concentration only got a score of 2.5 because of the combination of dull color, too sour aroma, and hard texture. Control Gel: Got a score of 3.1 as a standard product without innovation.

#### 4.7 Gel Stability Enhancement Mechanism

Based on the research results, the mechanism of increasing the stability of green grass jelly gel by adding fruit juice can be explained through several interrelated aspects:

##### 1. pH Modification and Electrostatic Interactions

Organic acids in fruit juice (citric acid, malic acid, ascorbic acid) lower the pH of the gel system from neutral-alkaline conditions (pH 7.1) to acidic conditions (pH 4.0-5.6 depending on the type and concentration). The decrease in pH causes protonation of the carboxyl group ( $-\text{COO}^-$  to  $-\text{COOH}$ ) in the green grass jelly polysaccharide chain.

This protonation has two main effects. First, it reduces the electrostatic repulsion between the negatively charged polysaccharide chains, allowing them to move closer together. Second, it increases the ability to form hydrogen bonds because the  $-\text{COOH}$  group can act as both a hydrogen bond donor and acceptor.

However, too low a pH (below 4.0) can cause excessive protonation that induces irregular aggregation and precipitation of polysaccharides, as observed in high concentration lime treatments.

##### 2. Formation of Dual-Network Gel Structure

Pectin contained in fruit juice (the highest in guava 0.8-1.2%, then mango 0.5-1%, and pineapple 0.3-0.7%) can form a mixed gel structure with green grass jelly polysaccharides. Interaction occurs through several mechanisms: Hydrogen bonds between the hydroxyl groups of pectin and grass jelly polysaccharides. Calcium ion bridges that connect the carboxyl groups of both types of polysaccharides. Chain interpenetration where the pectin and grass jelly polysaccharide chains penetrate each other and become entangled.

This double gel structure has higher mechanical strength and better stability than a single gel. The junctional zones between the two polysaccharide networks create a denser and more organized structure.

##### 3. Humectant Effect of Sugar

Sugar in fruit juice (highest in mango 12-18%, then pineapple 10-15%, guava 8-12%, and lowest in lime 1-3%) acts as a humectant that increases water binding capacity through hydrogen bonds between the hydroxyl groups of sugar and water molecules.

Sugar also fills the spaces within the gel network and reduces the mobility of free water. Water tightly bound to sugars and polysaccharides becomes more difficult to release from the gel structure, thus inhibiting syneresis. Furthermore, sugar increases the viscosity of the liquid phase in the gel, slowing the diffusion of water out of the gel matrix.

##### 4. Antioxidant Protection

Antioxidant compounds in fruit juice (vitamin C, beta-carotene, flavonoids, tannins) protect polysaccharides and chlorophyll from oxidative degradation through several mechanisms: scavenging free

radicals that can break bonds in the polysaccharide chain. Chelation of metal ions that can catalyze oxidation reactions. Protection of chlorophyll from oxidation that can change the color of the gel.

Guava with the highest vitamin C content (200-300 mg/100g) showed the best chlorophyll protection, while mango with high beta-carotene also provided good protective effects.

## 5. Optimizing Ionic Balance

Minerals in fruit juice, such as calcium, potassium, and magnesium, play a role in cross-linking and gel stabilization. Calcium, in particular, can form an "egg-box structure" with the carboxyl groups in pectin and grass jelly polysaccharides, strengthening the gel structure. Manganese in pineapple can act as a cofactor, facilitating bond formation within the gel structure.

## 6. Specific Interactions of Each Type of Fruit

- a) **Pineapple** memberikan kombinasi optimal dari pH (3,3-3,6 → gel pH 4,8-5,2), pektin, gula, dan mineral yang menghasilkan stabilitas gel terbaik. Enzim bromelain yang ada dalam nanas tidak signifikan mempengaruhi polisakarida non-protein cincau hijau.
- b) **Mangoes** provide the strongest humectant effect due to their highest sugar content, as well as the contribution of pectin with a high degree of esterification which forms a stable gel under acid-sugar conditions.
- c) **Guava** provides the strongest antioxidant protection due to its highest vitamin C content, resulting in the best chlorophyll and color retention although the gel strengthening effect is slightly below that of pineapple.
- d) **Lime** provides the most drastic pH reduction which is effective at low concentrations (10%) but damaging at high concentrations (>20%) because the pH is too low and the sugar content is very low.

## 4.8 Correlation Analysis Between Parameters

Pearson correlation analysis showed a significant relationship between the observed parameters:

- a) Hardness vs Syneresis : Strong negative correlation ( $r = -0.87$ ,  $p < 0.01$ ), indicating harder gels tend to have lower syneresis.
- b) pH vs Hardness : Curvilinear correlation with optimal pH 4.8-5.4 producing the highest hardness.
- c) Chlorophyll vs Vitamin C Content : Strong positive correlation ( $r = 0.82$ ,  $p < 0.01$ ), indicating that vitamin C is effective in protecting chlorophyll.
- d) Organoleptic Acceptance vs Hardness : Moderate positive correlation ( $r = 0.64$ ,  $p < 0.05$ ), indicating that consumers prefer a stable, chewy texture.

## 5. CONCLUSION AND SUGGESTIONS

### 5.1 Conclusion

Based on the research results, it can be concluded that: The addition of mango, pineapple, guava, and lime juice significantly affected the elasticity of the green grass jelly gel. The type of juice, concentration, and their interaction significantly affected all tested parameters.

- a) **Pineapple juice at a concentration of 10%** gave the best overall results in maintaining gel elasticity with a hardness value of 248.7 gf (an increase of 13.9% from the control), the lowest syneresis of 7.8% (a decrease of 59.8% from the control), an optimal pH of 5.1, chlorophyll retention of 82.5%, and the highest organoleptic acceptance (score of 4.7) after 72 hours of storage.

- b) **Guava juice 10%** provides the advantage in terms of the best chlorophyll retention (85.4%) and the highest increase in vitamin C content (28.4 mg/100g), making it the best choice for both nutritional and color aspects.
- c) **10% mango juice** gave good results with advantages in humectant effect due to high sugar content, resulting in a hardness of 238.2 gf and syneresis of 9.2%.
- d) Lime juice exhibits a highly concentration-dependent effect. A 10% concentration yielded good results (hardness of 236.2 gf), but 20% and 30% concentrations significantly reduced gel stability due to the low pH (3.2-4.2) (Mahato et al., 2021; González-Molina et al., 2021) and very low sugar content (Sharma et al., 2021).

The optimal concentration for all types of fruit juice is 10% (v/v) (Mohammadian et al., 2020; Zhang et al., 2021). Increasing the concentration to 20% and 30% tends to decrease gel stability, except for some specific parameters such as vitamin C content (Li et al., 2021; Chen et al., 2022).

The mechanisms of increasing gel stability involve pH modification that optimizes electrostatic interactions (Wang et al., 2021; Zheng et al., 2020), formation of a double gel structure with pectin (Liu et al., 2023; Giacomazza et al., 2020), the humectant effect of sugar (Sousa et al., 2020; Jiang et al., 2022), antioxidant protection against degradation (Gani et al., 2021; Putnik et al., 2020), and optimization of ionic balance in the gel system (Rafe & Razavi, 2021; Kumar et al., 2020).

The addition of fruit juice not only increases texture stability but also enriches nutritional value (vitamin C, antioxidants) (Ribeiro et al., 2021; Panche et al., 2021), maintains the natural green color (Khoo et al., 2020; Setiowati et al., 2021), and improves the sensory acceptability of green grass jelly gel products (Huang et al., 2021; Duan et al., 2022).

## 5.2 Suggestions

Based on the research results, some suggestions for further research and practical applications are:

### **Further Research:**

Microstructural analysis of the gel using Scanning Electron Microscopy (SEM) to visualize changes in the gel matrix structure with the addition of fruit juice. Study of chlorophyll degradation kinetics and product shelf life at various storage temperature conditions (room temperature, refrigeration, freezing). Complete texture profile analysis (TPA - Texture Profile Analysis) for parameters of elasticity, cohesiveness, springiness, and chewiness. Formulation optimization using Response Surface Methodology (RSM) to obtain the optimal combination of various types of fruit juice. Analysis of total antioxidant activity (DPPH, ABTS, FRAP methods) to evaluate potential health benefits. Study of specific interactions between fruit juice components and grass jelly polysaccharides using FTIR and rheological analysis. Evaluation of the content of specific bioactive compounds (beta-carotene, flavonoids, tannins) in the final product.

### **Practical Applications:**

Formulation of a commercial product of green grass jelly gel with 10% pineapple juice as a superior product with the best texture stability. Development of a product variant with 10% guava juice as a functional product high in vitamin C and antioxidants. Fruit juice combinations (e.g., pineapple 7% + guava 3%) to obtain an optimal balance between texture, nutrition, and color. Development of appropriate packaging to maintain product quality during distribution and storage. Economic feasibility study for industrial-scale production.

### **Explore Other Local Fruits:**

Testing the potential of other local Indonesian fruits such as soursop, starfruit, passion fruit, or dragon fruit as fortifiers for green grass jelly gel. Evaluation of combinations with local herbal extracts to enhance functional value.

### **Education and Dissemination:**

Dissemination of research findings to grass jelly micro, small, and medium enterprises (SMEs) to improve product quality and competitiveness. Training on green grass jelly gel production techniques using fruit juice to improve the skills of local producers. Publication of recipes and production guides for household consumers.

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