

**DEVELOPMENT AND EVALUATION OF FUNCTIONAL  
GUMMIES INCORPORATING *Alternanthera sessilis* EXTRACT**

*Dissertation submitted to Mahatma Gandhi University in  
partial fulfilment of the requirements for the award of degree  
of*

*Bachelor of Vocational Studies*

*B. Voc. Food Processing Technology*

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**ST. TERESA'S COLLEGE (AUTONOMOUS), ERNAKULAM**

**COLLEGE WITH POTENTIAL FOR EXCELLENCE**

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March 2025

## DECLARATION

We, Angie Liza Santhosh (Reg.no.VB22FPT003), Fathima Haris (Reg.no.VB22FPT011) and Namitha U M (Reg.no.VB22FPT015) hereby declare that this dissertation entitled “DEVELOPMENT AND EVALUATION OF FUNCTIONAL GUMMIES INCORPORATING *Alternanthera sessilis* EXTRACT” is a bonafide record of research work done by using the course of research and that is dissertation has not previously formed the basis for the award to us for any degree, diploma, fellowship or other title of any other university or society.

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## **CERTIFICATE**

This is to certify that the project entitled “DEVELOPMENT AND EVALUATION OF FUNCTIONAL GUMMIES INCORPORATING *Alternanthera sessilis* EXTRACT” submitted in partial fulfilment of the requirements for the Award of the degree of B.Voc Food Processing Technology to St. Teresa’s College, Ernakulam is a record of bonafide research work carried by Ms. Angie Liza Santhosh, Ms. Fathima Haris and Ms. Namitha U M under my guidance and supervision and that no part of the project has been submitted for the award of any other degree, diploma, fellowship or other similar titles or prize and that the work has been published in part or full in any scientific or popular journal or magazine.

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## **ACKNOWLEDGEMENT**

We express our heartfelt gratitude to the management of St. Teresa's College, Ernakulam, for granting us the opportunity to undertake this project. We extend our sincere thanks to Rev. Sr. Nilima CSST, our esteemed Manager, Rev. Dr. Sr. Francis Ann CSST and Rev. Sr. Tessa CSST, our esteemed Directors and Dr. Alphonsa Vijaya Joseph, our respected Principal for their constant encouragement and support throughout this endeavor.

Special thanks to our guide, Ms. Elizabeth Zarina Jacob, Assistant Professor in the Department of Food Processing Technology, for her valuable guidance, continuous support, and constructive feedback throughout the course of this research. Her expertise and encouragement have played a crucial role in shaping the direction and quality of this study.

We are also deeply thankful to the Head of the Department, Ms. Sherin Mary Simon, and to the entire faculty of the Department of Food Processing Technology for providing us with the facilities, resources, and a supportive academic environment.

Our sincere thanks to Biogenic Labs for their collaboration and assistance in this project.

## ABSTRACT

The present study aimed at developing and evaluating a functional gummy candy enriched with *Alternanthera sessilis* and sweetened with palm sugar. Four formulations incorporating 20%, 30%, 40%, and 50% leaf extract were prepared, alongside a control. The sample with 30% *Alternanthera sessilis* was selected through sensory evaluation based on a 5-point hedonic scale. Nutrient profiling revealed that the sample gummy exhibited lower moisture and protein content but higher ash, fiber, carbohydrate, energy, and total phenolic content compared to the control. The total phenolic content was three times higher in the sample, suggesting enhanced antioxidant potential. Shelf life studies over 7 days indicated stable moisture content, a slight increase in pH, a minimal rise in total plate count within acceptable limits, and absence of *E. coli*, demonstrating good physicochemical and microbial stability. Syneresis studies showed significantly lower water expulsion in the sample gummy (1.449%) compared to the control (5.50%), indicating better water retention and gel stability. The results confirm that incorporating *Alternanthera sessilis* improved the functional and nutritional properties of the gummies while maintaining desirable textural and storage qualities. This functional gummy holds promise as a nutritionally enhanced confectionery product offering both health benefits and consumer appeal.

### Keywords:

Functional gummy, *Alternanthera sessilis*, palm sugar, syneresis, total phenolic content, shelf life, nutraceuticals.

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# CHAPTER 1

## INTRODUCTION

### 1.1 INTRODUCTION TO FUNCTIONAL GUMMIES AND ITS INGREDIENTS

Foods are known to be important suppliers of vitamins and minerals, which are necessary for sustaining body processes and general health. They primarily supply energy and essential nutrients that serve as the building blocks for cells, supporting their growth, proliferation, and differentiation while also helping to safeguard against oxidative stress. Consumers' eating habits in recent years have centered on processed foods with high nutritional and functional value. Foods with natural ingredients, such as dietary fiber, natural colouring, minerals, vitamins, low calorie, low cholesterol, low fat, and no artificial additives, are now more popular among consumers (Granato et al., 2020). The phrase "functional food" was initially used in Japan in the 1980s to describe food items enhanced with specific compounds that have positive physiological benefits, like probiotics and prebiotics, as well as chemicals that lower cholesterol. Consuming these food items can help to enhance the body's overall health, lower the risk of developing certain diseases, and even cure some ailments. According to Granato et al., 2017 functional foods are defined as either processed or natural products that, when included in a balanced diet at appropriate levels, can provide health benefits that go beyond basic nutrition. Currently, nutritional and functional foods are highly sought after due to their reputation for being safe and their potential to offer nutritional benefits and therapeutic effects, including the prevention of cancer, cardiovascular diseases, obesity, and type 2 diabetes.

Due to changes in the socio-economic conditions of society, consumers are increasingly seeking products that offer health benefits along with essential nutrition. *Alternanthera sessilis*, commonly known as sessile joyweed or dwarf copperleaf, is a herbaceous plant native to tropical and subtropical regions worldwide. In South Asia, including Sri Lanka, it is widely utilized as a leafy vegetable and prized for its important therapeutic qualities. According to SravaniAbbas, and Surya (2017), *A. sessilis* is a herb that can be either annual or perennial, with dense clusters of silvery-white flowers and lanceolate to linear leaves. Along with hydrocarbons, esters, and sterols including campesterol, stigmasterol,  $\beta$ -sitosterol, and saponins, its rich phytochemical makeup also includes lupeol, a substance known for its anti-inflammatory properties, which is particularly abundant in the roots (Sravani et al., 2017).

The herb has been used traditionally to cure a number of illnesses, such as hypertension, pneumonia, diarrhea, flatulence, and wounds. Recent research has confirmed its anti-



hyperglycemic, anti-diarrheal, antioxidant, and antibacterial qualities, underscoring its potential as a natural treatment for infections and other chronic illnesses including diabetes. With these qualities and its abundance of nutrients, *Alternanthera sessilis* is positioned as a useful herbal remedy and functional food in the modern health and wellness industry.

Gummies and jellies are especially well-liked by those under the age of seventeen because of their natural and leathery qualities (Moloughney S, 2011). These goods have a gel-like structure and contain sugars (sucrose, saccharinity, and/or glucose, in the amount of approximately 55 g/100 g) and fruits (at least 45 g/100 g), along with food colorings, acids, fragrances, and gelatinizing agents (Mutlu et al., 2018).

According to a 2018 research by the Nutrition Business Journal, about 12% of all nutraceuticals sold that year were in the form of chewable gummy tablets. Chewable gummy tablets are thought to be tasty, easy to take, and to offer a pleasant experience. Chewable gummy pills feature chewable texture, color, and taste, as well as sweet qualities (because they are typically made from sugar processing), and they dissolve slowly in the mouth. Children and adults without swallowing issues may find chewable gummy tablets appealing (Arifa et al., 2018).

Due to consumer desire for healthier options, gummy formulations are being altered to suit their tastes by reducing the amount of sugar and employing plant-based gelling agents. Health and wellbeing are becoming increasingly important in the food sector, which is driving the gummy market to develop new ingredients that are both enjoyable and nutritious. Gummies can also contain probiotics, vitamins, minerals, and other bioactive substances, among other health benefits (Tarahi et al., 2023).

## **1.2 RELEVANCE OF THE STUDY**

The formulation of functional gummies using *Alternanthera sessilis* and palm sugar represents an innovative effort to develop healthier and more functional confectionery products. *Alternanthera sessilis*, a plant rich in bioactive compounds like antioxidants, sterols, and saponins, offers several health benefits, including anti-inflammatory, antihyperglycemic, and antimicrobial activities. Its incorporation into gummies provides a convenient and appealing way to deliver nutritional and therapeutic advantages, particularly targeting the growing demand for functional foods.

In this study, palm sugar is used as a natural sweetener in place of refined sugars and glucose syrup. Palm sugar not only imparts a unique flavor but also offers a lower glycemic index and retains trace minerals, making the gummies a healthier option. Gelatin is utilized as the primary gelling agent to achieve the desired chewy texture and mouthfeel that consumers associate with traditional gummies. The use of gelatin also supports the structural integrity and stability of the gummies. Additionally, citric acid is incorporated to balance the flavor profile and enhance the shelf life of the product.

By combining the medicinal potential of *Alternanthera sessilis* with the natural sweetness of palm sugar and the functional properties of gelatin, this study aims to develop a nutritionally enriched, palatable, and consumer-friendly gummy product. This formulation caters to the increasing consumer interest in functional foods that are both enjoyable and beneficial to health.

### **1.3 OBJECTIVES:**

- To develop gummy candy formulations incorporating different concentrations of *Alternanthera sessilis* extract.
- To evaluate the sensory properties of the formulated gummies using a hedonic scale and to identify the best formulation based on overall acceptability.
- To analyze the nutritional composition of the developed gummies, including moisture, protein, fat, ash, fiber, carbohydrate, and energy content.
- To determine the total phenolic content of the gummies using standard analytical methods.
- To assess the microbial and physicochemical quality of the gummies during shelf life studies by evaluating total plate count, coliform count, *E. coli* detection, and pH changes.
- To monitor the physical stability of the gummies by evaluating syneresis and dispersion characteristics over a storage period.

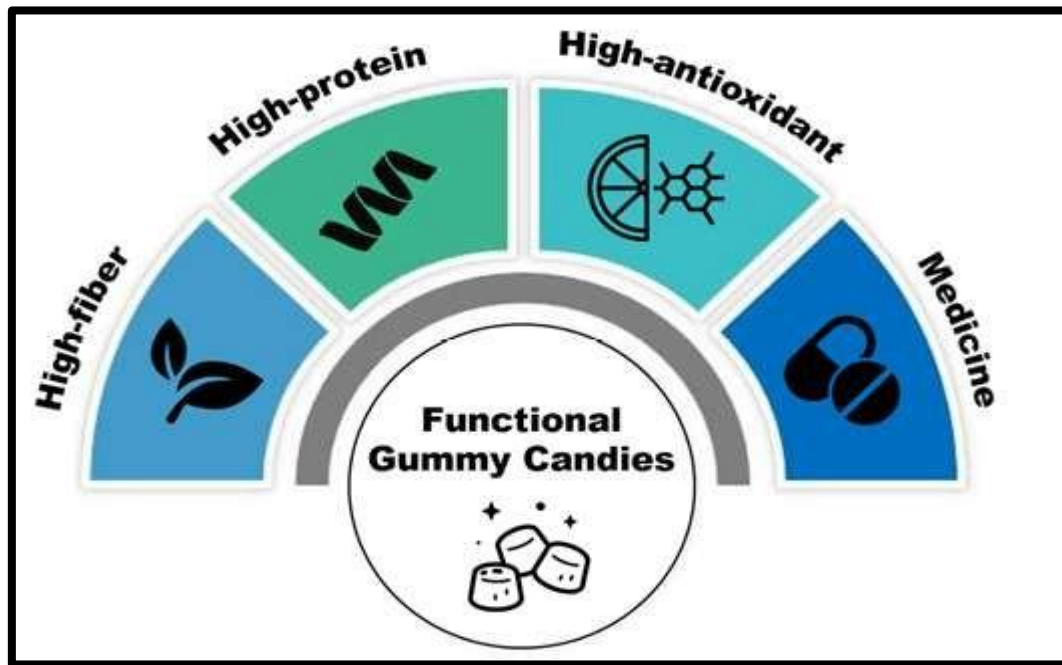
## CHAPTER 2

### REVIEW OF LITERATURE

#### 2.1 Gummy

Functional gummy candies (GCs) have emerged as a novel category combining health benefits with the traditional enjoyment of confectionery treats. The history of gummy candies dates back to 1920, when Hans Riegel in Germany, dissatisfied with his job, founded his own company and initially produced hard, colorless candies. A few years later, he innovated by creating fruit-flavored, gelatin-based candies shaped like dancing bears, leading to the birth of the Haribo brand (Gouveia et al., 2018).

Today, the innovation around GCs focuses on improving their nutritional profile by incorporating functional ingredients like natural fibers, antioxidants, proteins, and probiotics. Functional gummy candies can be classified into several categories depending on their incorporated health-promoting ingredients. The different types of Functional gummy candies are depicted in Fig 1.



**Figure 1: Types of Functional Gummy Candies (Adapted from Tarahi, M.; Tahmouzi, S.; Kianiani, M.R.; Ezzati, S.; Hedayati, S.; Niakousari, M. Current Innovations in the Development of Functional Gummy Candies. Foods 2024)**

High-fiber gummies are formulated by adding natural fibers such as inulin, konjac glucomannan, or fruit-derived fibers to improve digestive health and regulate blood glucose levels. High-protein gummies are enriched with plant-based proteins like soy protein isolate, lupine protein concentrate, or Spirulina biomass to support muscle maintenance and combat protein-energy malnutrition. Antioxidant-rich gummies incorporate natural extracts from fruits, herbs, or vegetables, such as grape skin, rosemary, or beetroot, to help combat oxidative stress and support immune health. Probiotic gummies are developed by integrating beneficial bacteria strains like *Lactobacillus* or *Bacillus* species along with prebiotic fibers to enhance gut microbiota balance. Medicinal or fortified gummies contain active pharmaceutical ingredients, vitamins, minerals, or bioactive compounds such as iron, beta-carotene, or acetaminophen, serving as effective carriers for nutrient delivery or therapeutic effects. These innovations allow gummy products to cater to a wide range of health needs while maintaining consumer appeal through enjoyable taste and texture.

Natural sweeteners such as *Stevia rebaudiana* have been used to reduce calorie content while maintaining desirable sweetness, offering benefits like improved glycemic control (Cedeño-Pinos et al., 2020). The use of alternative gelling agents, including inulin, has also enhanced the texture and stability of gummies while boosting their fiber content (Delgado & Bañón, 2018).

Furthermore, plant extracts like grape skin powder have been incorporated to increase antioxidant capacity and improve the physicochemical stability of GCs (Cappa et al., 2015). Rosemary extract has also been successfully used to enrich the antioxidant properties of gummies without negatively affecting sensory acceptance (Cedeño-Pinos et al., 2020).

Probiotic-enriched GCs are gaining attention for their ability to support gut health. Formulations containing probiotics and prebiotics like psyllium husk fiber have demonstrated enhanced antimicrobial and functional properties (Lele et al., 2018). Additionally, microencapsulation technologies, such as using amaranth carboxymethyl starch and lactoferrin for beta-carotene stabilization, offer improved nutrient protection and bioavailability in fortified gummy candies (Constantino & Garcia-Rojas, 2023).

Overall, the advancement of functional GCs offers promising opportunities for the confectionery industry by merging taste, convenience, and health benefits in one appealing product.

Overall, the advancement of functional GCs offers promising opportunities for the confectionery industry by merging taste, convenience, and health benefits in one appealing product.

## **2.2. *Alternanthera sessilis***

*Alternanthera sessilis* is an ancient herbaceous plant consumed for thousands of years, with its roots traced back to ancient civilizations (Jansen, 2004). The *Alternanthera* genus comprises approximately 80–200 species, predominantly originating from tropical and subtropical regions of North and South America (Singh et al., 2009). *A. sessilis* grows up to 0.2–1 meter in height, featuring creeping, slightly hairy stems that occasionally root at nodes. The plant has simple, opposite leaves, measuring 0.6–5 cm in length and 0.3–1 cm in width. Known by several local names — "Ponnanganni" in Tamil, "Ponnagantikura" in Telugu, and "Gudrisag" in Hindi — it is also commonly referred to as "Sessile Joyweed" or "Dwarf Copperleaf."

Its rising demand in food and medicinal sectors has significantly boosted its market value. *A. sessilis* is rich in minerals, vitamins, flavonoids, carotenoids, and dietary fiber, traditionally employed in treating fever, diarrhea, stomach pain, snake bites, skin diseases, and liver ailments (Debnath et al., 2014).

Nutritionally, *A. sessilis* offers a highly valuable profile. Dehydrated leaf powder was found to contain calcium (379 mg/100g), iron (5 mg/100g), protein (4 g/100g), and dietary fiber (3.56 g/100g), with low levels of fat (0.012 g/100g) and carbohydrates (2.90 g/100g) (Vijaya Vahini & Sharmila, 2023). It demonstrates a high antioxidant activity of 77.35%, indicating strong potential for reducing oxidative stress. Phytochemical analysis revealed the presence of flavonoids, phenolic compounds, alkaloids, and glycosides (Sravani et al., 2017), with total phenolic and flavonoid contents measured at 30.78 µg/mg and 16.34 µg/mg, respectively, underscoring its powerful free radical scavenging properties.

Medicinally, *A. sessilis* displays a broad spectrum of pharmacological activities. Its anti-fungal potential was demonstrated using the agar well diffusion method, where ethanol and methanol leaf extracts inhibited fungal growth against *Cyrtomium falcatum* (Ali et al., 1996; Shanthi et al., 2015). Clear inhibition zones confirmed the presence of strong anti-fungal activity.

The plant's antimicrobial capabilities have been similarly validated. Using the disc diffusion method, it was found effective against pathogens like *Salmonella typhi*, *Bacillus subtilis*, *Proteus vulgaris*, and *Streptococcus pyogenes*, exhibiting stronger growth inhibition compared to standard antibiotics like ampicillin and gentamycin (Agarwal et al., 2008). These findings highlight its potential as a natural alternative to synthetic antimicrobials.

The oil extracted from *A. sessilis* also possesses notable antioxidant activity, crucial for preventing oxidative damage and promoting wound healing. These oils are recognized for their antiviral, antimicrobial, and anticancer properties (Agarwal et al., 2008; Iqbal et al., 2011). Factors such as cultivation methods, storage, and processing techniques significantly influence the antioxidant constituents (Unni et al., 2009).

In terms of gastrointestinal health, aqueous extracts of *A. sessilis* have demonstrated significant anti-diarrheal activity. Experiments on albino Swiss mice showed strong inhibitory effects on castor oil-induced diarrhea ( $P < 0.01$ ), supporting its traditional use in treating digestive disorders (Sanjib et al., 2013). The activity is attributed to the tannins and flavonoids present in the plant.

Besides these properties, methanolic extracts of *A. sessilis* have shown antihyperglycemic activity in glucose tolerance tests, reducing blood glucose levels by up to 46.1% in experimental mice, comparable to standard diabetic medications (Sravani et al., 2017). In Taiwan, the plant is part of traditional folk medicine combined with other species to treat hepatitis, emphasizing its hepatoprotective potential (Gunasekera, 2008).

Recent technological innovations have expanded the utility of *A. sessilis*. For instance, silver nanoparticles (ASNPs) synthesized using its extracts have shown effective antimicrobial properties, confirmed through UV-visible spectrophotometry and scanning electron microscopy (Sravani et al., 2017). Additionally, tray drying techniques have been successfully employed to dehydrate the leaves, producing a microbially safe, shelf-stable powder with moisture content reduced to 6.16%, well within the limits set by FSSAI standards (Vijaya Vahini & Sharmila, 2023).

Cooking practices also influence the nutritional retention of *A. sessilis*. Studies have shown that cooking the leaves for 5–10 minutes preserves most of the protein content while only minimally

affecting carbohydrate and starch levels, indicating that light cooking is optimal for maintaining its health benefits (Sravani et al., 2017).

In conclusion, *Alternanthera sessilis* is a highly promising underutilized plant, combining a rich nutrient profile, diverse phytochemicals, and extensive medicinal properties. Its strategic inclusion in diets can address nutrient deficiencies, enhance antioxidant defenses, and support overall health. However, to ensure its safety and maximize its full potential, especially in developing countries, further studies focusing on cultivation practices, contamination management, and product development are essential.

### **2.3 Palm Sugar**

Palm sugar, traditionally derived from the sweet sap of the *Borassus flabellifer* palm, stands out as a natural and nutritionally superior alternative to refined cane sugar. Unlike refined sugar, which is heavily processed to remove all nutrients and is almost pure sucrose, palm sugar undergoes minimal processing, allowing it to retain significant phytonutrients, minerals, and antioxidants (Srikaeo, Sangkhiaw, & Likittrakulwong, 2019). This natural processing preserves health-promoting compounds such as phenolic compounds and flavonoids, leading to remarkably higher antioxidant activity compared to refined sugars.

Studies have shown that palm sugar exhibits DPPH radical scavenging activity and ferric reducing antioxidant power (FRAP) values far greater than those found in refined cane sugar (Srikaeo et al., 2019). Specifically, the antioxidant content in palm sugar syrup is nearly ten times higher than in refined sugar, contributing to its potential to protect cells from oxidative damage and even prevent DNA strand breaks caused by hydroxyl radicals. These bioactive components may play a key role in reducing the risk of chronic diseases such as cancer, cardiovascular diseases, and neurodegenerative disorders.

In addition to its superior antioxidant profile, palm sugar has a lower predicted glycemic index (pGI) compared to refined sugar. It causes a slower rate of glucose release during digestion, as shown by significantly lower pGI values when tested with corn starch mixtures (Srikaeo et al., 2019). This slow digestion rate makes palm sugar a better option for individuals managing

diabetes, obesity, or metabolic syndrome. In contrast, refined sugars cause rapid spikes in blood sugar and insulin levels, contributing to metabolic disorders (Jenkins et al., 2002).

Palm sugar's benefits also extend to gut health. It contains small amounts of natural dietary fibers like inulin, which may aid digestion and promote beneficial gut microbiota. Furthermore, palm sugars are naturally rich in glucose and fructose, and their minor content of reducing sugars supports their pleasant caramel-like taste without the need for chemical additives or bleaching agents used in refined sugars (Naknean & Meenune, 2015).

Environmentally, palm sugar production is more sustainable compared to sugarcane. Palmyra palms help restore degraded soils and require minimal water, making palm cultivation an eco-friendly practice that supports rural livelihoods (Morton, 1988). With its rich antioxidant profile, lower glycemic impact, natural fiber content, and sustainable production methods, palm sugar clearly emerges as a more wholesome and health-supportive choice over highly processed refined sugars.

## **2.4 Gelatin**

Gelatin, a natural biopolymer derived from the partial hydrolysis of collagen found in animal connective tissues, bones, and skin, has established itself as one of the most versatile gelling agents across industries. Known for its unique ability to form thermoreversible gels, gelatin undergoes solubilization upon heating and reverts to a semi-solid gel state when cooled below 35–40°C. This remarkable gel-forming capacity is a direct result of its unique triple-helix collagen structure, which, upon cooling, creates a stable three-dimensional network (Alipal et al., 2021).

Gelatin's gelling properties make it indispensable in the food industry, where it is used to stabilize, thicken, and texture a wide variety of products such as gummy candies, marshmallows, yogurts, and desserts. Its ability to trap water within a gel matrix also improves mouthfeel and shelf life (L.S. Kumosa et al., 2018). Chemically, gelatin is composed of 18 amino acids, with glycine, proline, and hydroxyproline making up around 57% of its structure. The remaining 43% is made up of amino acids like glutamic acid, alanine, arginine, and aspartic acid (Sulthana et al., 2018).

Commercial production of gelatin typically involves processing by products from cattle and pork industries. Pork skins and bones are mainly used to produce type A gelatin through acid treatment,



whereas type B gelatin is obtained from cattle hides using an alkaline treatment with agents like caustic lime or sodium carbonate to eliminate microorganisms and minerals (Mariod et al.,2013).

Compared to plant-derived hydrocolloids, gelatin gels are favored for their superior clarity, elasticity, and clean flavor release, despite being sensitive to temperature and pH variations (Alipal et al., 2021). Recent innovations also explore eco-friendly enzymatic methods to extract gelatin with enhanced gelling properties, further expanding its commercial potential.

Thus, gelatin's unmatched gelling ability continues to make it a crucial ingredient across multiple sectors, providing both functional and sensory benefits that plant-based alternatives often struggle to replicate.

## **2.5 Citric Acid**

Citric acid is one of the most important organic acids produced globally, widely used across food, beverage, and pharmaceutical industries. Its appeal in gummies lies primarily in its function as an acidulant, providing the signature tartness that balances sweetness and enhances flavor. Additionally, citric acid stabilizes pH, which is crucial for maintaining the structural integrity and shelf life of gummies, particularly those formulated with gelatin or pectin. Its chelating ability also prevents discoloration and off-flavors during storage by binding trace metals. Recognized as GRAS (Generally Recognized as Safe), citric acid is especially suited for confectionery products consumed by children. Industrial production mainly relies on microbial fermentation using *Aspergillus niger*, ensuring high purity and food safety standards (Swain et al., 2012). These properties make citric acid an indispensable ingredient in gummy formulations, contributing both functional stability and desirable sensory character.

## **2.6 Glucose Syrup**

A thick, sweet liquid called glucose syrup may be produced by hydrolyzing the starch found in carbohydrate crops including maize, rice, cassava, millet, and sorghum. One of the most profitable sugar alternatives is glucose syrup, which is primarily an aqueous blend of glucose, dextrose, and maltose (P. Hull, 2010; Graffham A. et al., 2002). In the processed food industry, it is widely used, especially as a raw ingredient for beverages, jams, and confections, as well as for chemical and medicinal uses.

Usually, either an acidic or an enzymatic process is used to hydrolyze starch in order to produce glucose syrup. Through this process, complex starch molecules are broken down into simpler sugars. A crucial indicator of the syrup's sweetness and other useful characteristics, the Dextrose Equivalent (DE), is strongly influenced by the degree of hydrolysis. Certain enzymes, including amylase, are used in enzymatic hydrolysis, whereas starch is heated with diluted acids in acid hydrolysis. Liquification and saccharification are the two primary phases of enzymatic hydrolysis. A range of items, such as baked goods, syrups, and sweets, benefit from the smoothness and texture that glucose syrup adds. By reducing water activity, it also serves as a preservative, prolonging the shelf life of the product. Additionally, because of its exceptional water solubility, it may be used in a wide variety of food and beverage compositions (O.H. Chibudike et al., 2021).

Glucose molecules compose the majority of glucose syrup; the proportion of other sugars, such as maltose, varies according to the degree of starch hydrolysis. It's somewhat acidic pH (4–6) improves stability, taste, and shelf life, while its high water solubility is a result of its carbohydrate content. Its preservation qualities are further enhanced by its low water content. High-viscosity, low-sweetness syrups have a Dextrose Equivalent (DE) of 20–40, whereas sweeter, more soluble varieties have a DE of 60–90. Glucose syrup behaves pseudoplastically rheologically, exhibiting decreased viscosity at increased shear speeds (J. Eke-Ejiofor, 2015).

## **CHAPTER 3**

### **MATERIALS AND METHODOLOGIES**

#### **3.1. INTRODUCTION**

This chapter deals with materials and method used for the development and analysis of the Gummy Candy. This study was carried out at the Department of B.Voc Food Processing Technology, St Teresa's College Ernakulam during the year 2024-2025.

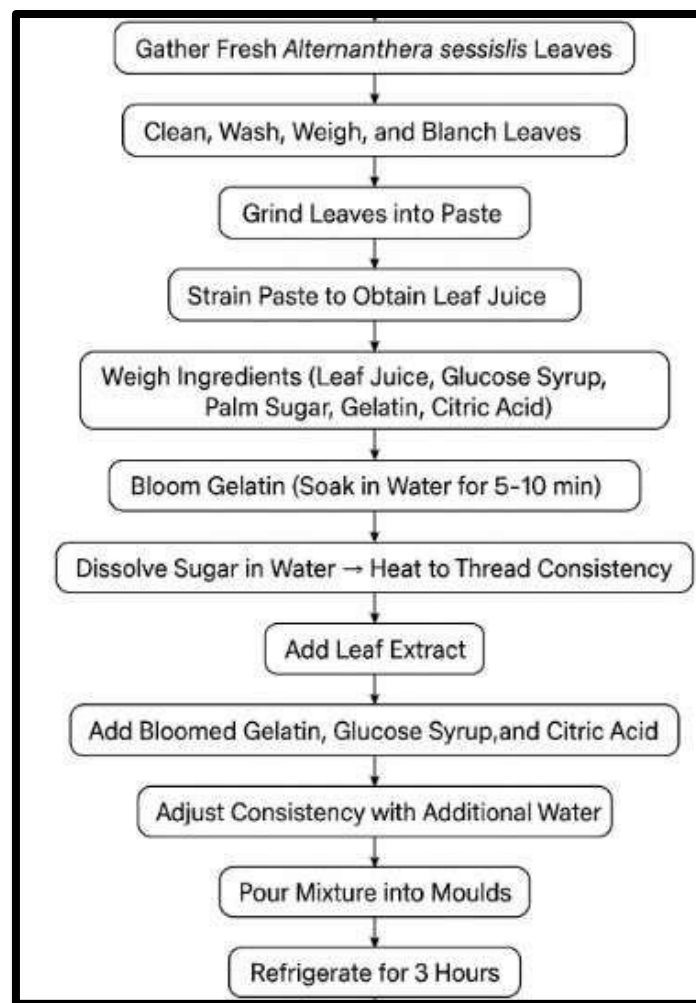
#### **3.2 DEVELOPMENT OF GUMMY CANDY USING *Alternanthera sessilis* AND PALM SUGAR:**

- 1. *Alternanthera sessilis*: Collected from garden**
- 2. GLUCOSE SYRUP: Purchased from market**
- 3. PALM SUGAR: Purchased online from Amazon**
- 4. GELATIN: Purchased from market**
- 5. CITRIC ACID: Purchased from market**

Gummy candies were prepared by mixing ingredients in five different ratios, labeled S1, S2, S3, S4 with each combination placed into separate bowls. An additional sample, labelled S0, was prepared as a control without the addition of *Alternanthera sessilis* (sessile joyweed) extract. Fresh *Alternanthera sessilis* leaves were harvested from the garden, thoroughly cleaned, washed, and sorted. They were then weighed, rewashed, and blanched to remove any impurities. Afterward, the leaves were finely ground into a paste using a mixer grinder. The paste was strained through a fine-mesh sieve to separate the juice from the pulp, resulting in a pure, fresh leaf extract. Specific amounts of the leaf extract, glucose syrup, palm sugar, gelatin, and citric acid were then measured according to the given formulations. The gelatin was soaked in a small amount of water for 5–10 minutes to allow it to bloom before cooking began.

The cooking process started by dissolving sugar in an equal amount of water and heating it until it reached a thread-like consistency. The leaf extract was then incorporated, followed by the gelatin, glucose syrup, and citric acid. Additional water was added to adjust the final consistency of the mixture. Once properly combined, the mixture was poured into molds while still warm to allow it

to set. The molds were placed in a refrigerator and chilled under controlled conditions for 3 hours. Following this, the gummies underwent a deep-freezing process for around 12 hours to achieve the desired gummy texture and firmness. After the setting process was complete, the gummies were gently removed from the molds to maintain their shape and structure. At this stage, the gummies had reached the intended texture and appearance, making them ready for packaging and consumption.



**Figure 2: Flowchart for the Preparation of Functional Gummy Candy using *Alternanthera sessilis* and Palm Sugar**

### 3.3 SELECTION OF THE BEST FORMULATION OF GUMMY:

Sensory evaluation is a key area in assessing the acceptability of food products, and its significance is anticipated to increase considerably in the future. It involves the scientific analysis of food characteristics—including taste, smell, texture, and appearance—using human senses, typically through techniques such as tasting panels and consumer trials (Murray et al., 2003). Sensory evaluation of the five different samples (S0, S1, S2, S3, and S4) was conducted by a panel of 10 judges using a 5-point hedonic scale. Based on the mean overall acceptability scores, the best formulation of the gummy was identified. A model of the score card used is given in Appendix

### 3.4 NUTRIENT PROFILE OF THE DEVELOPED SAMPLE

The nutrient profiling of the functional gummy formulated with *Alternanthera sessilis* was carried out to assess its nutritional value and functional potential. *Alternanthera sessilis*, known for its rich content of bioactive compounds, was incorporated into the gummy formulation. Standard analytical methods were employed to determine the macronutrient and micronutrient composition, including moisture, ash, protein, fat, carbohydrate and fiber. The results provide a comprehensive understanding of the nutritional enhancements achieved through the incorporation of *Alternanthera sessilis* into the gummy matrix.

#### 3.4.1 DETERMINATION OF MOISTURE CONTENT:

The total moisture content of the Gummy sample was determined using, Weigh to the nearest 0.001 g. -5.0 g of the laboratory sample in the dish previously dried and weighed, together with its lid, to the nearest 0,001 g. Place the dish with its lid underneath in the oven for 2h. The time should be reckoned from the moment the oven attains 130°C after the dishes have been placed. After 2h, cover dish while still in oven, transfer to desiccator, Cool in the desiccator. When the dish has cooled to room temperature (25 ± 3 °C) (generally 30-45 min after has been placed the desiccator), weigh it to the nearest ( 0.001g). The dish should be placed back in the oven till a constant weight is achieved.

#### Calculation

The moisture content, expressed as a percentage by mass of the of expression product, is given by the following equations:

$$\text{Moisture (\%)} = \frac{W_1 - W_2}{W_1 - W} \times 100$$

Where:

W = Mass in g of the empty dish

W<sub>1</sub> = Mass in g of the dish with the test portion before drying

W<sub>2</sub> = Mass in g of the dish with the material after drying

### 3.4.2 DETERMINATION OF TOTAL ASH:

The total Ash content of the gummy sample was determined by, Take fresh sample for the determination of Total ash, rather than left over after determination of moisture. Weigh a previously clean and dried dish (W<sub>1</sub>). Weigh accurately about 5 g of powdered sample into the dish. Place with its lid underneath in the oven maintained at 130-133 °C for 2h. The time should be reckoned from the moment the oven attains 130°C after the dishes have been placed. Remove the dish after 2 h, cool in the desiccator and weigh (W<sub>2</sub>). Ignite the dried material in the dish left after the determination of moisture with the flame of a burner till charred.

Note: This step must be carried out in a fume hood. Ignition till grey ash is obtained.

Transfer to a muffle furnace maintained at 550 ± 25 °C and continue, Cool in a desiccator and weigh, Repeat the process of heating cooling and weighing at 30 min intervals till the difference in weight in two consecutive weighing is less than 1 mg. Note the lowest weight (W<sub>2</sub>). ash still contains black particles add 2-3 drops of pre-heated water at 60 °C. Break the ash and evaporate to dryness at 100-110 °C. Re-Ash at 550 °C. Until ash is white or slightly grey.

Calculation

$$\text{Total ash on dry basis (\% by weight)} = \frac{W_1 - W}{W_1 - W} \times 100$$

Where.

W=Mass in g of empty dish

W<sub>1</sub>=Mass in g of the dish with the dried material (moisture free) taken for test

W<sub>2</sub>= Mass in g of the dish with the ash

Calculate the mean of two determinations and express the result to one decimal place.

### **3.4.3 DETERMINATION OF TOTAL FAT:**

The Total Ash Content of the gummy sample was determined by, Dissolve 10 gm sample in 10 ml warm water, and introduce into Mojonnier fat extraction tube or similar apparatus. Add 25 ml peroxide free ethyl ether. Cork the tube and shake vigorously for 1 minute. Add 25 ml of Petroleum ether and shake again for 30 seconds. Let stand for 30 minutes or until separation is complete. Draw off the ether layer containing fat in a previously dried and weighed flask. Repeat the extraction twice, Pool the ether extract, recover excess solvent and dry the fat for 1 hour at 100 °C. Cool and weigh. Fat must be dried by keeping the flasks for 30 minutes and weighed, till constant mass is achieved.

Calculation

Fat, % on dry basis=  $M_1 \times 100 \times 100$

Where,

$M_1$  = Weight in g of the fat

$M_2$  = Weight in g of sample take

$M$  = Moisture % in the sample

### **3.4.4 DETERMINATION OF PROTEIN CONTENT:**

The protein content of the gummy sample was determined by Transfer carefully about one to two grams of the sample accurately weighed, to the Kjeldhal flask, taking precaution to see that particles of the material do not stick to the neck of the flask. Add about 10 g of anhydrous sodium sulphate, 0.2 to 0.3 g of copper sulphate and 20 ml of concentrated sulphuric acid. Place the flask in an inclined position. Heat below the boiling point of the acid until frothing ceases. Increase heat until the acid boils vigorously and digests for 30 minutes after the mixture becomes clear and pale green in colour. Cool the flask. Transfer quantitatively to the round-bottomed flask with water the total quantity of water used being about 200 ml. Add a few pieces of pumice stones to avoid bumping. Add about 50 ml of Sodium hydroxide solution (which is sufficient to make the solution alkaline) carefully through the side of the flask so that it does not mix with the acid solution but forms a separate layer below the acid layer. Assemble the apparatus as shown above taking care that the dip tube extends below the surface of the standard sulphuric acid solution contained in the

beaker. Mix the contents of the flask by shaking and distill until all the ammonia has passed over into the standard sulphuric acid. Missing from the document. Shut off the burner and immediately detach the flask from the condenser. Rinse the condenser thoroughly with water into the beaker. Wash the dip tube carefully so that all traces of the condensate are transferred to the beaker. 8. When all the washings have been drained into the beaker, add two or three drops of methyl red indicator solution and titrate with the standard sodium hydroxide solution. Carry out a blank determination using all reagents in the same quantities but without the sample to be tested.

Calculation

Total Protein ( $N \times 6.25$ ), % by mass =  $8.75 \times (B-A) \times N$

Where,

B = volume in ml of the standard sodium hydroxide solution used to neutralize the acid in the blank determination

A = volume in ml of the standard sodium hydroxide solution used to neutralize the excess of the acid in the test with the material

N= Normality of the standard sodium hydroxide solution

M = mass in g of the material taken for the test

### **3.4.5 DETERMINATION OF CRUDE FIBRE**

The crude fiber content of the functional gummy was estimated using the method prescribed by IS 11062:1984. A known weight of the sample was subjected to sequential digestion with 1.25% sulfuric acid ( $H_2SO_4$ ) followed by 1.25% sodium hydroxide (NaOH) under controlled conditions. After digestion, the residue was filtered, thoroughly washed, dried at  $105^\circ C$ , weighed, and subsequently ashed in a muffle furnace at  $550^\circ C$  to remove inorganic matter. The crude fiber content was calculated based on the loss in weight after ashing.

The crude fiber percentage was determined using the formula:



$$\text{Crude Fiber (\%)} = \left( \frac{\text{Weight of dried residue} - \text{Weight of ash}}{\text{Weight of sample}} \right) \times 100$$

### 3.3.6 DETERMINATION OF CARBOHYDRATE

The total carbohydrate content of the functional gummy was determined by difference, following the procedure specified in IS 1656:2007, Annex C. In this method, carbohydrate percentage was calculated indirectly by subtracting the sum of percentages of moisture, protein, fat, ash, and crude fiber from 100. The formula used was:

$$\text{Carbohydrate (\%)} = 100 - (\% \text{Moisture} + \% \text{Protein} + \% \text{Fat} + \% \text{Ash} + \% \text{Crude Fiber})$$

### 3.4.7 DETERMINATION OF ENERGY

The energy (calorie) content of the functional gummy was calculated using the standard FAO method, based on the Atwater general factors. The energy value was determined by multiplying the amounts of protein, fat, and carbohydrate by their respective energy conversion factors and summing the results. The following formula was used:

$$\text{Energy (kcal/100g)} = (\text{Protein (g)} \times 4) + (\text{Fat (g)} \times 9) + (\text{Carbohydrate (g)} \times 4)$$

where:

- Protein provides 4 kcal per gram
- Fat provides 9 kcal per gram
- Carbohydrate provides 4 kcal per gram

The values of protein, fat, and carbohydrate were obtained from the proximate analysis, and the calculation was performed per 100 g of sample.

### **3.5 DETERMINATION OF PHENOLIC COMPOUNDS**

The total phenolic content of the functional gummy was determined according to the procedure outlined in IS 14502:1998, which specifies the method for evaluating phenolic compounds in plant-based products. The estimation was carried out using the Folin-Ciocalteu reagent method. An aliquot of the gummy extract was mixed with Folin-Ciocalteu reagent and allowed to react, followed by the addition of sodium carbonate solution. After incubation at room temperature for a specified time, the absorbance was measured spectrophotometrically at 760 nm. A standard curve was prepared using gallic acid, and the results were expressed as milligrams of gallic acid equivalents (mg GAE) per 100 g of sample.

### **3.6 DETERMINATION OF pH**

The pH of both the control and sample gummy samples was determined on day 0 and after 7 days of storage using a calibrated digital pH meter. Approximately 10 g of each gummy sample (control and sample) was homogenized separately in 90 mL of distilled water to prepare a 10% (w/v) suspension. The suspension was mixed thoroughly, and the pH was measured by immersing the electrode directly into the solution. Prior to measurement, the pH meter was calibrated using standard buffer solutions of pH 4.0 and pH 7.0 to ensure accuracy.

### **3.7 SHELF LIFE STUDY**

To assess the microbiological and physicochemical stability of the functional gummy during storage, shelf life studies were conducted over a period of 7 days under ambient conditions ( $25 \pm 5^\circ\text{C}$ ). Both the control and sample gummies were evaluated. The parameters analyzed included pH, moisture content, Total Plate Count (TPC), coliform presence, and *Escherichia coli* detection. The methods for pH measurement and moisture content determination were described previously.

#### **3.7.1 DETERMINATION OF TOTAL PLATE COUNT**

Microbial load in the functional gummy was evaluated by performing a Total Plate Count at day 0 and after 7 days of storage. One gram of the sample was aseptically transferred into 9 mL of sterile peptone water and serially diluted up to  $10^{-6}$ . From appropriate dilutions, 0.1 mL was spread onto Plate Count Agar (PCA) plates in duplicate. The plates were incubated at  $37^\circ\text{C}$  for 24–48 hours.

After incubation, colonies were counted and results were expressed as colony-forming units per gram (CFU/g) of the sample.

### **3.7.2 DETERMINATION OF E.Coli**

The presence of *Escherichia coli* in the functional gummy samples was evaluated following the Bacteriological Analytical Manual (BAM), 8th Edition, procedure. A 25 g portion of the gummy sample was aseptically homogenized with 225 mL of sterile Buffered Peptone Water (BPW) and incubated at 35°C for 24 hours for pre-enrichment. After incubation, 1 mL of the enriched sample was transferred into Lauryl Sulfate Tryptose (LST) broth and incubated at 35°C for 24–48 hours. Tubes showing gas production were considered presumptive positive for coliforms. Positive samples were further inoculated into EC broth and incubated at 44.5°C for 24 hours. Gas production in EC broth indicated presumptive *E. coli* presence. A loopful from positive EC broth was streaked onto Eosin Methylene Blue (EMB) agar and incubated at 35°C for 24 hours. Colonies exhibiting a metallic green sheen were taken as presumptive *E. coli* and further confirmed by Gram staining and IMViC biochemical tests (Indole positive, Methyl Red positive, Voges-Proskauer negative, Citrate negative). Results were reported qualitatively as either detected or not detected.

### **3.8 SYNERESIS STUDY**

Syneresis refers to the expulsion or drainage of water from a contracting or shrinking structure, which can negatively affect the quality of the gummy bears. To evaluate syneresis, the test was conducted at room temperature ( $25 \pm 5^\circ\text{C}$ ) by weighing the samples. An absorbent paper was placed on the surface of each gummy, and the samples were reweighed after a set period. A noticeable difference between the initial and final weights indicated the occurrence of syneresis.

### **3.9 DISPERSION TIME TEST**

The dissolution behavior of both control and sample gummies was evaluated to assess their dispersion characteristics. The test was performed using a flask containing 100 mL of purified water maintained at  $37^\circ\text{C} \pm 0.5^\circ\text{C}$ . Each gummy sample (control and functional formulation) was placed separately into the water and subjected to continuous stirring using a magnetic stirrer. The time taken for each gummy to completely disperse was recorded visually with the aid of a stopwatch.

## CHAPTER 4

### RESULTS AND DISCUSSION

The present investigation was undertaken to develop a functional gummy using *Alternanthera sessilis* and evaluate their sensory, nutritional and functional properties.

#### 4.1 DEVELOPMENT OF GUMMY CANDY USING *Alternanthera sessilis* :

A total of four formulations of gummy candies were developed by incorporating *Alternanthera sessilis* leaf extract at varying concentrations of 20%, 30%, 40%, and 50% (w/w). In addition to these, a control gummy formulation without any leaf extract was also prepared for comparison. This approach was undertaken to evaluate the impact of different levels of *Alternanthera sessilis* on the sensory properties of the gummies. The detailed formulation compositions for both control and treated samples are presented in Table 1.

SAMPLE	LEAF EXTRACT	PALM SUGAR	GLUCOSE SYRUP	GELATIN	CITRIC ACID	WATER
S0	0	11%	16%	5%	0.3%	39%
S1	20%	11%	16%	5%	0.3%	24%
S2	30%	11%	16%	5%	0.3%	12%
S3	40%	11%	16%	5%	0.3%	0
S4	50%	11%	16%	5%	0.3%	0

**TABLE 1: FORMULATIONS OF FOUR DIFFERENT GUMMY CANDY**

#### 4.2 SELECTION OF THE BEST FORMULATION OF GUMMY:

The acceptability of the five samples were found by a panel of 10 semi trained members using a five-point hedonic scale. It was found that sample S2 with 30% *Alternanthera sessilis* leaf extract was accepted when compared to other samples. Image of the final product is attached in appendix number 2.

SL NO	QUALITY PARAMETERS	S0	S1	S2	S3	S4
1	APPEARANCE	4.8	4.7	4.8	4.6	4.3
2	AROMA	3.8	3.7	3.8	3.6	3.4
3	TEXTURE	4.6	4	4.5	3.8	3.5
4	TASTE	4.1	4	4	3.6	3.2
5	MOUTH FEEL	4.4	3.8	4.1	3.8	3.5
6	OVERALL ACCEPTANCE	4.34	4.04	4.24	3.88	3.58

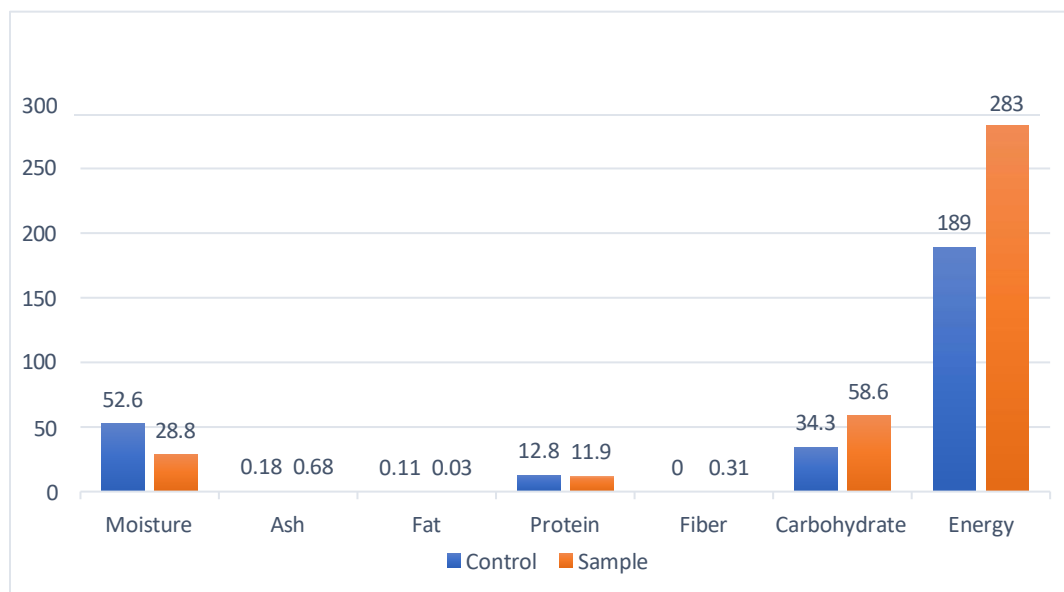
**TABLE 2: MEAN SENSORY SCORES OF FOUR DIFFERENT FORMULATIONS OF GUMMY ALONG WITH CONTROL**

### 4.3 NUTRIENT PROFILE OF THE SAMPLE AND CONTROL

The nutrient composition of both the control gummy and the *Alternanthera sessilis*-enriched gummy S2 with 30% leaf extract was evaluated and is presented in Table 3. The addition of *Alternanthera sessilis* notably influenced the nutritional profile compared to the control. Each parameter was analyzed and discussed as follows:

SL.No	PARAMETERS	UNIT	CONTROL	SAMPLE
1	MOISTURE	g/100g	52.6	28.8
2	ASH	g/100g	0.18	0.68
3	FAT	g/100g	0.11	0.03
4	PROTEIN	g/100g	12.8	11.9
5	FIBRE	g/100g	0	0.31
6	CARBOHYDRATE	g/100g	34.3	58.6
7	ENERGY	Kcal/100	189	283

**TABLE 3: NUTRIENT PROFILE OF FUNCTIONAL GUMMIES USING *Alternanthera sessilis* AND CONTROL**



**Figure 3: Graphical representation of Nutritional Profile of functional gummies using *alternanthera sessilis* and control**

#### 4.3.1 MOISTURE CONTENT

Moisture content plays a crucial role in determining the texture, stability, and shelf life of gummy candies. An optimal moisture level helps maintain the chewiness of the gummy while preventing microbial spoilage. The sample gummy showed a lower moisture content compared to the control gummy. This decrease could be attributed to the addition of *Alternanthera sessilis* leaf extract, which may have contributed solids that reduced the relative water content in the matrix.

Additionally, the phytochemical compounds in *Alternanthera sessilis* may have led to increased binding of free water molecules, thereby lowering the measurable moisture content (Kumar et al., 2015). A reduced moisture level is beneficial for improving shelf life and microbial safety of the functional gummy.

#### **4.3.2 ASH CONTENT**

Ash content represents the total mineral content of a food product, indicating its nutritional value in terms of essential minerals. The ash content of the sample gummy was higher than that of the control. This increase is due to the natural mineral richness of *Alternanthera sessilis*, which is known to contain calcium, potassium, and magnesium (Karthikeyan et al., 2009). The higher mineral content enhances the nutritional value of the functional gummy.

#### **4.3.3 FAT CONTENT**

Fat content affects the mouthfeel, texture, and energy density of the product. Lower fat levels are generally desirable in health-oriented confectionery products. There was no significant difference in fat content between the control and the sample gummy, and both showed low values. This is expected as neither *Alternanthera sessilis* nor the base ingredients (palm sugar, glucose syrup) are significant fat sources. Maintaining a low-fat content aligns with the development of a healthy, functional gummy.

#### **4.3.4 PROTEIN CONTENT**

Protein is an essential macronutrient that contributes to growth, repair, and overall body maintenance. In functional foods, maintaining or enhancing protein content is often desired to improve nutritional value. The protein content of the sample gummy was slightly lower than that of the control gummy. This reduction could be attributed to the dilution effect caused by the incorporation of *Alternanthera sessilis* leaf extract, which is higher in fiber and other non-protein components. The increase in crude fiber content may have displaced the protein fraction within the gummy matrix (Jain et al., 2011). Additionally, the original palm sugar base used in the control might have contributed minor protein fractions that became relatively reduced when substituted with the leaf extract.

#### **4.3.5 FIBRE CONTENT**

Fiber is crucial for digestive health and functional food claims. In gummies, higher fiber enhances nutritional benefits without affecting taste if formulated properly. The fiber content was significantly higher in the sample gummy compared to the control. *Alternanthera sessilis* leaves are a known source of dietary fiber (Jain et al., 2011), which explains the observed increase. This enhancement adds value by promoting digestive health benefits through the functional gummy.

#### **4.3.6 CARBOHYDRATE CONTENT**

The carbohydrate content of the sample gummy was significantly higher compared to the control gummy, with almost a 25% increase observed. This increase can be attributed to the additional natural carbohydrates present in *Alternanthera sessilis* leaves along with the existing palm sugar and glucose syrup used in the formulation (Karthikeyan et al., 2009). In contrast, the control gummy, formulated solely with palm sugar as the sweetening agent, exhibited a relatively lower carbohydrate content. Although there was an increase, it remains within acceptable limits for functional confectionery products, especially when considering the added health benefits conferred by the bioactive and fiber components introduced by the leaf extract.



#### 4.3.7 ENERGY

Energy value is a critical parameter that represents the total caloric contribution of a food product, calculated based on the content of protein, fat, and carbohydrates. It is important for nutritional labeling and dietary planning, especially for functional foods. The energy value of the sample gummy was higher than that of the control gummy. This increase is primarily attributed to the higher carbohydrate content in the sample, despite the slight decrease in protein levels. Since carbohydrates provide 4 kcal per gram, their significant rise (~25% higher compared to control) contributed more to the total energy. Fat content remained low and almost similar in both formulations, indicating that the energy difference was mainly driven by the carbohydrate variation (FAO, 2012). Overall, the energy value of the sample gummy remained within acceptable limits for functional confectionery products while offering enhanced nutritional and health benefits.

#### 4.4 TOTAL PHENOLIC CONTENT

Phenolic compounds are important bioactive constituents in foods known for their antioxidant, anti-inflammatory, and antimicrobial properties. Measuring total phenolic content is critical to assess the functional potential of nutritionally enhanced products like gummies. The total phenolic content of the sample gummy formulated with *Alternanthera sessilis* extract was found to be 0.30%, whereas the control gummy without extract showed a much lower value of 0.10%. This threefold increase in phenolic content is attributed to the incorporation of *Alternanthera sessilis*, which is naturally rich in polyphenolic compounds (Karthikeyan et al., 2009).

The significant enhancement in total phenolics suggests that the sample gummy may offer improved antioxidant properties compared to the control. This aligns with the objective of developing a functional gummy with potential health benefits beyond basic nutrition. The presence of higher phenolic compounds also contributes to better oxidative stability, which may indirectly improve the shelf life of the product.

#### 4.5 pH

The pH of the control and sample gummies was measured to evaluate the influence of formulation on the physical characteristics and setting behavior of the gummy matrix. The control gummy (without *Alternanthera sessilis* extract) exhibited a pH of 4.52, whereas the sample gummy (with leaf extract) showed a higher pH of 4.72.

pH plays a crucial role in gummy formation, particularly in the setting, texture, and overall gel strength. Gelatin, the primary gelling agent used in gummy formulations, requires an optimal pH range between 4.0 and 5.5 for effective gelation. A pH that is too low can weaken gelatin networks, resulting in softer, less cohesive products, while a slightly higher pH within the optimal range promotes better gelation and firmer texture (Karim & Bhat, 2009).

In this study, the higher pH observed in the sample gummy may be attributed to the buffering effect of natural compounds present in *Alternanthera sessilis* leaves. The favorable pH contributed to improved gel strength and structural stability, enhancing the physical characteristics of the functional gummy.

#### 4.6 SHELF LIFE STUDY

Shelf life evaluation of the functional gummy developed with *Alternanthera sessilis* was conducted over a period of 7 days at ambient conditions ( $25 \pm 5^\circ\text{C}$ ) to assess its microbiological and physicochemical stability. The results for pH, moisture content, microbial load and *E. coli* detection are presented below.

SL.No	PARAMETERS	SAMPLE (AT DAY 0)	SAMPLE (AT DAY 7)
1	MOISTURE	28.8	28.8
2	Ph	4.72	5.33
3	TPC	<100	130
4	E.Coli	ABSENT	ABSENT

**TABLE 4: PHYSICOCHEMICAL AND MICROBIOLOGICAL CHANGES IN SAMPLE GUMMY DURING 7 DAYS OF STORAGE**

The sample gummy exhibited a slight increase in pH from 4.72 at day 0 to 5.33 at day 7, possibly due to the degradation of organic acids or minimal microbial activity during storage. Moisture content remained stable at 28.8%, indicating good water retention and structural integrity of the gummy matrix. Total Plate Count (TPC) showed a slight rise from <100 CFU/g to 130 CFU/g over the 7 days but remained within acceptable microbiological limits. *E. coli* was absent throughout the storage period, confirming the microbiological safety of the functional gummy under ambient conditions.

#### 4.7 SYNERESIS

Syneresis refers to the expulsion of water from a gel matrix, which affects the texture, stability, and overall acceptability of gummy products. Evaluating syneresis is essential to assess the water-holding capacity and structural integrity of the formulation during storage.

BATCH	INITIAL WEIGHT	FINAL WEIGHT	SYNERESIS %
CONTROL	1.09g	1.03g	5.50
SAMPLE	1.38g	1.36g	1.449

**TABLE 5: SYNERESIS PERCENTAGE OF CONTROL AND SAMPLE GUMMIES**

The syneresis percentage was measured for both the control and sample gummies. The control gummy exhibited a syneresis of **5.50%**, while the sample gummy formulated with *Alternanthera sessilis* showed a significantly lower syneresis value of **1.449%**.

The lower syneresis observed in the sample gummy suggests that the incorporation of *Alternanthera sessilis* improved the water retention ability of the gummy matrix. This can be attributed to the presence of dietary fibers and bioactive compounds from the leaf extract, which can bind water effectively and enhance the gel network's stability (Jain et al., 2011).

The reduced syneresis in the sample gummy indicates better structural integrity and improved shelf stability, making it more suitable for functional food applications where moisture retention is critical for texture and sensory quality.

#### 4.8 DISPERSION TIME

To evaluate the disintegration behavior of the gummy formulations, a dissolution test was performed under controlled conditions. This test aimed to compare the time required for the complete dispersion of the standardized gummy against that of the original gummy sample.

BATCH	DISPERSION TIME
CONTROL	4 min 20 sec
SAMPLE	3 min 7 sec

**TABLE 6 : DISPERSION TIME OF CONTROL AND SAMPLE**

From the data, it is evident that the sample gummy dispersed within 3 minutes 7 seconds and the control gummy dispersed in 4 minutes 20 seconds. This difference in dissolution time may be attributed to variations in the composition, texture, or binding agents used in the formulation process.

## CHAPTER 5

### SUMMARY AND CONCLUSION

The present study was undertaken to develop a functional gummy candy enriched with *Alternanthera sessilis* and palm sugar as a healthier base. Four formulations with varying concentrations of *Alternanthera sessilis* extract (20%, 30%, 40%, and 50%) were prepared along with a control. Sensory evaluation identified the best-accepted formulation i.e S2 with 30% of *Alternanthera sessilis* leaf extract, which was selected for further nutrient profiling and physicochemical and microbiological analysis.

The nutrient analysis revealed that the sample gummy had a lower moisture and protein content but a higher ash, fiber, carbohydrate, energy, and total phenolic content compared to the control. In particular, the total phenolic content was three times greater in the sample, indicating enhanced antioxidant properties. These improvements align with the objective of developing a nutritionally superior gummy product with functional benefits. Moisture retention was satisfactory, and the syneresis study showed that the sample gummy had significantly lower water expulsion (1.449%) compared to the control (5.50%), suggesting better structural integrity and stability. Shelf life evaluation over 7 days at ambient temperature demonstrated that the sample gummy maintained its physicochemical quality, with only a slight increase in pH and microbial load, both of which remained within safe and acceptable limits. No *E. coli* was detected at any point, confirming the microbiological safety of the product.

However, during the course of evaluation, stickiness of the gummy candy was observed as a notable issue, particularly after storage. This stickiness could affect handling, packaging, and consumer acceptance, and it highlights an important area for improvement in future studies. Potential strategies could include optimizing the drying process, modifying the formulation with natural anti-sticking agents, or exploring better packaging materials to maintain product quality.

In conclusion, the incorporation of *Alternanthera sessilis* into gummy formulations proved effective in enhancing the nutritional and functional properties without significantly compromising sensory quality. The developed gummies are promising as value-added functional confectionery products catering to health-conscious consumers. Future research could focus on extending the

shelf life further with natural preservation methods, improving the textural characteristics to address stickiness, and conducting clinical evaluations to validate the health benefits of *Alternanthera sessilis*-based gummies. Scaling up production and broader consumer acceptance studies would also be valuable steps toward commercial application.

## CHAPTER 6

### REFERENCES

1. Alipal, J., Mohd Pu'ad, N. A. S., Lee, T. C., Nayan, N. H. M., Sahari, N., Basri, H., Idris, M. I., & Abdullah, H. Z. (2021). A review of gelatin: Properties, sources, process, applications, and commercialisation. *Materials Today: Proceedings*. <https://doi.org/10.1016/j.matpr.2020.12.922>
2. Ali, A. M., Mackeen, M. M., El-Sharkawy, S. H., Abdul Hamid, J., Ismail, N. H., Ahmad, F., & Lajis, M. N. (1996). Anti-viral and cytotoxic activities of some plants used in Malaysian indigenous medicine. *Pertanika Journal of Tropical Agricultural Science*, 19(2/3), 129–136.
3. Arifa-Begum, S. K., Padma, S. V., Anusha, V., Keerthi, V. Z., Vinitha, S. P., Prameela, K., Nazeema, M. D., & Padmalatha, K. (2018). Formulation and evaluation of pediatric oral soft jellies of salbutamol sulphate. *Research Journal of Pharmacy and Technology*, 11(11), 4939–4945. <https://doi.org/10.5958/0974-360X.2018.00899.5>
4. Ayernor, G. S. T., Hammond, A., & Graffham, A. (2002). The combination of rice malt and Amyloglucosidase for the production of sugar syrup. *African Journal of Science and Technology*, 3(1), 10–17.
5. Cappa, C., Lavelli, V., & Mariotti, M. (2015). Fruit candies enriched with grape skin powders: Physicochemical properties. *LWT - Food Science and Technology*, 62, 569–575. <https://doi.org/10.1016/j.lwt.2014.07.039>
6. Cedeño-Pinos, C., Martínez-Tomé, M., Murcia, M. A., Jordán, M. J., & Bañón, S. (2020). Assessment of rosemary (*Rosmarinus officinalis* L.) extract as antioxidant in jelly candies made with fructan fibres and stevia. *Antioxidants*, 9(12), 1289. <https://doi.org/10.3390/antiox9121289>
7. Chibudike, H. O., Ndukwe, N. A., Lawal, A. K., Chibudike, E. C., & Adeyoju, O. A. (2021). Optimization of process condition for laboratory scale enzymatic conversion of cassava starch to glucose syrup.
8. Constantino, A. B. T., & Garcia-Rojas, E. E. (2023). Microencapsulation of beta-carotene by complex coacervation using amaranth carboxymethyl starch and lactoferrin for application in gummy candies. *Food Hydrocolloids*, 9, 108488. <https://doi.org/10.1016/j.foodhyd.2023.108488>

9. Delgado, P., & Bañón, S. (2018). Effects of replacing starch by inulin on the physicochemical, texture and sensory characteristics of gummy jellies. *CyTA - Journal of Food*, 16(1), 1–10. <https://doi.org/10.1080/19476337.2017.1327462>
10. Eke-Ejiofor, J. (2015). Functional properties of starches, physico-chemical and rheological properties of glucose syrup made from cassava and different potato varieties. *International Journal of Recent Scientific Research*, 6(6), 4400–4406.
11. Food and Agriculture Organization. (2012). Composition of functional foods: Guidelines for the assessment and labeling. Food and Agriculture Organization of the United Nations.
12. Gouveia, S. C., Castilho, P. C., & Silva, A. M. (2018). Nutritional and health-promoting compounds of gummy candies: A review. *International Journal of Food Sciences and Nutrition*, 69(5), 543–552. <https://doi.org/10.1080/09637486.2017.1374384>
13. Gouveia, T. I. A., Bastos, M., Rocha, F., & Agostinho, C. (2018). Development of vegan, sugar-free gummy candies—applicability to formulations with paracetamol.
14. Gunasekera, L. (2008). Sessile joyweed (*Alternanthera sessilis*): A popular leafy vegetable in Southeast Asia but federal noxious weed in USA. In *Proceedings of the 16th Australian Weeds Conference*, Queensland Weed Society, 347–348.
15. Ho, C. W., Aida, W. M. W., Maskat, M. Y., & Osman, H. (2008). Effect of thermal processing of palm sap on the physico-chemical composition of traditional palm sugar. *Pakistan Journal of Biological Sciences*, 11, 989–995.
16. Hossain, M. A., Shah, M. D., Gnanaraj, C., & Iqbal, M. (2011). In vitro total phenolics, flavonoids contents and antioxidant activity of essential oil, various organic extracts from the leaves of tropical medicinal plant 16. *Tetragonia* from Sabah. *Asian Pacific Journal of Tropical Medicine*, 4(9), 717–721.
17. Hull, P. (2010). *Glucose syrups: Technology and application*. Wiley-Blackwell.
18. Jain, S., Meena, M. L., & Meena, R. (2011). Dietary fiber content in traditional Indian leafy vegetables. *Food Research International*, 44(1), 350–355. <https://doi.org/10.1016/j.foodres.2010.10.002>
19. Jalalpure, S. S., Agrawal, N., Patil, M., Chimkode, R., & Tripathi, A. (2008). Antimicrobial and wound healing activities of leaves of *Alternanthera sessilis* Linn. *International Journal of Green Pharmacy*, 2(3).



20. Jenkins, D. J. A., Kendall, C. W. C., Augustin, L. S. A., Franceschi, S., Hamidi, M., Marchie, A., Jenkins, A. L., & Axelsen, M. (2002). Glycemic index: Overview of implications in health and disease. *American Journal of Clinical Nutrition*, 76(1), 266S–273S. <https://doi.org/10.1093/ajcn/76.1.266S>
21. Karim, A. A., & Bhat, R. (2009). Gelatin alternatives for the food industry: Recent developments, challenges and prospects. *Trends in Food Science & Technology*, 19(12), 644–656. <https://doi.org/10.1016/j.tifs.2008.08.001>
22. Karthikeyan, K., Shanthi, P., & Sachdanandam, P. (2009). Mineral composition of *Alternanthera sessilis* leaves. *Indian Journal of Nutritional Sciences*, 46(2), 114–118.
23. Kumar, Y. S., & Sanjib, D. (2013). Evaluation of anti-diarrhoeal property of crude aqueous extract of *Alternanthera sessilis* Linn. *International Journal of Pharmaceutical Innovations*, 3, 213–217.
24. Kumar, A., Chauhan, G. S., & Sharma, S. (2015). Water binding and textural properties of plant fibers. *Food Hydrocolloids*, 48, 37–46. <https://doi.org/10.1016/j.foodhyd.2015.01.035>
25. Kumosa, L. S., Zatterberg, V., & Schouenborg, J. (2018). Gelatin promotes rapid restoration of the blood brain barrier after acute brain injury. *Acta Biomaterialia*, 65, 137–149. <https://doi.org/10.1016/j.actbio.2017.10.020>
26. Lele, V., Ruzauskas, M., Zavistanaviciute, P., Laurusiene, R., Rimene, G., Kiudulaite, D., & Bartkiene, E. (2018). Development and characterization of the gummy-supplements enriched with probiotics and prebiotics. *CyTA - Journal of Food*, 16(1), 580–587. <https://doi.org/10.1080/19476337.2018.1433721>
27. Mariod, A. A., & Adam, H. F. (2013). Review: Gelatin, source, extraction and industrial applications. *Acta Scientiarum Polonorum Technologia Alimentaria*, 12(2), 135–147.
28. Moloughney, S. (2011). Functional confectionery: Finding the sweet spot. *Nutraceuticals World*, (2011), 2–3.
29. Morton, J. F. (1988). Notes on distribution, propagation, and products of *Borassus* palms (Arecaceae). *Economic Botany*, 42(3), 420–441. <https://doi.org/10.1007/BF02858927>
30. Mutlu, C., Tontul, S. A., & Erbaş, M. (2018). Production of a minimally processed jelly candy for children using honey instead of sugar. *LWT*, 93, 499–505. <https://doi.org/10.1016/j.lwt.2018.03.064>

31. Nithya, V., Chidambaram, R., & Dhanasekaran, D. (2020). Protein profile of edible leafy greens. *Journal of Food Science and Technology*, 57(5), 1800–1806. <https://doi.org/10.1007/s13197-019-04198-0>
32. Naknean, P., & Meenune, M. (2015). Impact of clarification of palm sap and processing method on the quality of palm sugar syrup (*Borassus flabellifer* Linn.). *Sugar Tech*, 17(2), 195–203. <https://doi.org/10.1007/s12355-014-0331-7>
33. Radhakrishnan, S., Saravana Bhavan, P., Seenivasan, C., Muralisankar, T., & Shanthi, R. (2015). Effects of native medicinal herbs (*Alternanthera sessilis*, *Eclipta alba*, and *Cissus quadrangularis*) on growth performance, digestive enzymes, and biochemical constituents of the monsoon river prawn *Macrobrachium malcolmsonii*. *Aquaculture Nutrition*, 21(4), 496–506.
34. Sarkar, T., Mukherjee, M., Roy, S., & Chakraborty, R. (2023). Palm sap sugar: An unconventional source of sugar exploration for bioactive compounds and its role on functional food development. *Heliyon*, 9(4).
35. Sia, J., Yee, H. B., Santos, J. H., & Abdurrahman, M. K. A. (2010). Cyclic voltammetric analysis of antioxidant activity in cane sugars and palm sugars from Southeast Asia. *Food Chemistry*, 118, 840–846.
36. Simamora, L., Zebua, D. D. N., Handoko, Y. A., & Widyawati, N. (2021). The continuity of palm sugar production: A literature review. *Jurnal Ilmiah Membangun Desa Dan Pertanian*, 6(2), 37.
37. Singh, A., Kandasamy, T., & Odhav, B. (2009). In vitro propagation of *Alternanthera sessilis* (sessile joyweed), a famine food plant. *African Journal of Biotechnology*, 8(21).
38. Srikaeo, K., Sangkhiaw, J., & Likittrakulwong, W. (2019). Productions and functional properties of palm sugars. *Walailak Journal of Science and Technology*, 16(11), 897–907. <https://doi.org/10.14456/vol16iss11pp897>
39. Sravani, V. L., Abbas, Z., & Surya, P. (2017). A review on *Alternanthera sessilis*. *Indo American Journal of Pharmaceutical Sciences*, 4(9), 2845–2852. <https://doi.org/10.5281/zenodo.888221>
40. Sultana, S., Hossain, M. A. M., Zaidul, I. S. M., & Ali, M. E. (2018). Multiplex PCR to discriminate bovine, porcine, and fish DNA in gelatin and confectionery products. *LWT - Food Science and Technology*, 92, 169–176. <https://doi.org/10.1016/j.lwt.2018.02.019>

41. Swain, M. R., Ray, R. C., & Patra, J. K. (2012). Citric Acid: Microbial Production and Applications in Food and Pharmaceutical Industries. In D. A. Vargas (Ed.), Citric Acid (pp. 1–22). Nova Science Publishers. <https://www.researchgate.net/publication/236894915>
42. Unni, B., Borah, A., Wann, S., Singh, H., Devi, B., & Bhattacharjee, M. (2009). Phytochemical and anti-bacterial study of traditional medicinal plants of North East India on *Escherichia coli*. Asian Journal of Experimental Sciences, 23(1), 103–108.

## CHAPTER 7

### APPENDIX 1: INGREDIENTS USED FOR PREPARATION OF GUMMY



**FIG 1** *Alternanthera sessilis*



**FIG 4** GELATIN



**FIG 2** WATER



**FIG 5** PALM SUGAR



**FIG 3** GLUCOSE SYRUP



**FIG 6** CITRIC ACID

## **APPENDIX 2: STAGES IN THE PREPARATION OF GUMMY**



**FIG 7 BLANCHED LEAVES**



**FIG 10 ADDED ALL INGREDIENTS**



**FIG 8 EXTRACT OF LEAF**



**FIG 11 COOKED UNTIL DESIRED  
CONSISTENCY**



**FIG 9 MELTED PALM SUGAR**



**FIG 12 MOULDING AND COOLING**



**FIG 13 FINAL PRODUCT**

### APPENDIX 3: HEDONIC SCALE SCORE CARD

Name:

Product:

Date:

The samples are provided. Taste the sample and check how much you like or dislike each of the characters.

Sample	Appearance	Aroma	Texture	Mouthfeel	Taste	Overall acceptance
S0						
S1						
S2						
S3						
S4						

#### 5 point hedonic scale

5	Like a lot
4	Like a little
3	Neither like or dislike
2	Dislike a little
1	Dislike a lot

Comments:

Signature: