

## CHAPTER 6

### **To develop functional food product by incorporating the encapsulated phytochemical extract**

#### **6.1. Introduction**

The growing interest in functional food products has paved a way for development of innovative food products with the aim to provide health benefits beyond basic nutritional attributes. Consumers generally seek functional food comprising enhanced sensory attributes such as aroma, flavor, texture, and visual appeal and also having similarities to traditional foods found in the market (Augustin, 2001, Klahorst, 2006, Kwak & Jukes, 2001, Klont, 1999).

Functional foods are natural or processed foods that contain biologically active compounds. When it is consumed (nontoxic or adequate amount) provides clinically proven health benefits. The promotion of better health benefits, reduction of the risk of diseases (chronic, viral etc.) and management of their symptoms can be clinically evidenced by using biomarkers (Martirosyan et al., 2021). Functional foods bridge the gap between traditional or general food system and nutrition by delivering nutritional and bioactive compounds above normal nutrition, mitigating disease, promoting health and reducing health care costs etc. One of the promising approaches involves incorporating bioactive compounds rich extracts for the development of food formulation which can provide better nutrients and various health benefits (Xiao et al., 2019). In contrast, direct use of phytochemicals may face various challenges as these substances are sensitive such as instability under some environmental fact or such as heat, light, and oxygen, as well as limited bioavailability and may affect the color and taste of the developed food. In this case, encapsulation of the flower extract is an effective approach to address these limitations prior to addition in food formulations.

Encapsulation is a technique which encloses a core i.e. bioactive compounds within a wall material, enhancing their stability and controlled release while maintaining the extract's properties. This technique helps the development of food products with a better shelf life, higher nutritional value, and better sensory attributes etc. Red brick colored Nongmangkha flowers belong to acanthaceae family and an important medicinal

plant that blooms from February to April. These flowers were reported to deliver beneficial health effects on hyperlipidemia (Chakravarty et al., 2014), as were showed antioxidant and radical scavenging activities (Nongthombam et al., 2018), and possessed hypoglycemic and hypolipidemic properties (Ahmed et al., 2016). Nongmangkha flowers were believed to cure pox; prevent skin diseases like sores, scabies, have anti-allergic effects, treat wounds and tumors, act as a blood purifier (Koushik et al., 2020), kidney stones, and liver disorders (Das et al., 2017). The flowers contain steroids, terpenoids, flavonoids, phenols, etc. (Nongthombam et al., 2018). Functional food products are described as offering additional health benefits beyond their conventional nutritional value (Khan et al., 2013). Importantly, while developing functional food products, they must be standardized to ensure the safety of administering bioactive compounds as a health optimization tool. Indeed, it is crucial to set up a standardized process for the development of functional food products to guarantee the safe delivery of bioactive compounds to support improvement of health (Martirosyan et al., 2021). As functional food products demands are increasing which might be due to the rising costs of health care, the steady increase in life expectancy and the interest of the elderly in the improvement of life quality etc. (Kraus et al., 2015). Edible flowers can be termed as functional food due to its nutritional properties, antioxidant activity, antimicrobial activity, color, flavor, mood and stress reduction capability etc. Gummy jelly candy is highly favored by all age circles. According to Burey et al. (2009) gummy jellies are produced using concentrated sugar solutions, gelling agents, and other components. Gummies are generally low of required nutritional value and may lead to obesity, hyperglycemia and tooth problem etc. (Khawaja et al., 2019 and Rippe et al., 2016).

This chapter focuses on the development of two functional food products gummies and ready-to-serve (RTS) beverages incorporated with encapsulated phytochemical extracts from Nongmangkha flower and explored their physicochemical properties, sensory characteristics, and overall acceptability.

## **6.2. Materials and Methods**

### **6.2.1. Materials**

Nongmangkha flower were collected from Sivasagar, Assam, India. Watermelon and Ash gourd were collected from local market Tezpur, Assam, India. All the materials

or ingredients used for development of gummy and RTS beverage are pectin, agar-agar, citric acid etc. are food grades. All chemicals utilized for experimental analysis were analytical grade.

### **6.2.2. Extraction of Nongmangkha flower extract**

Nongmangkha flower extract was obtained by using ultrasound pretreated microwave assisted extraction technique. Based on our previous study (Chapter 4), here ultrasound pre-treatment at 250 W for 15 min was given first in sample solution (1 g of shade dried powdered Nongmangkha flower in 10 mL of 80 % ethanol) in a ratio of 1:10. After ultrasound pretreated the mixer was undergone microwave assisted extraction at 700 W for 5 min. The obtained extract was filtered and dried in rotary evaporator. Finally, the dried extract was kept in a refrigerator for further use.

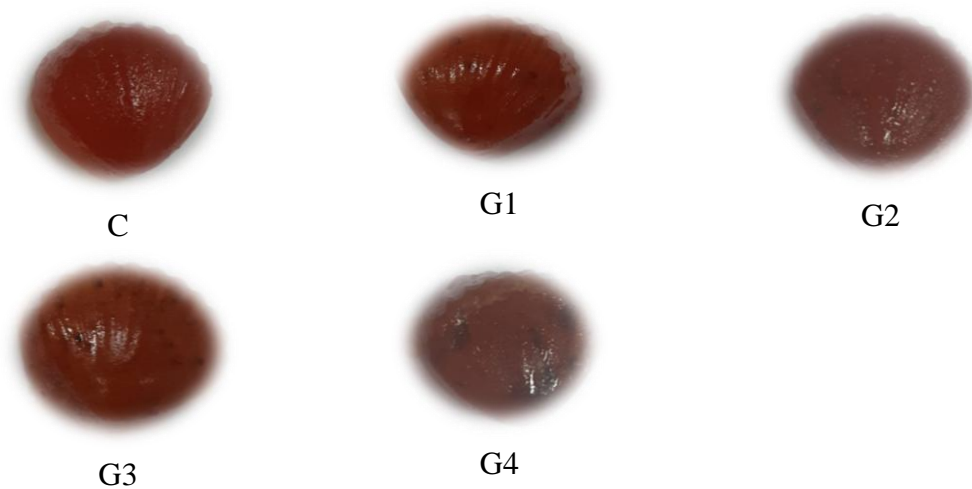
### **6.2.3. Preparation of encapsulates**

The Nongmangkha flower extract obtained by using ultrasound pretreated microwave assisted extraction technique was used for encapsulation. The extract was dried by using rotary evaporator and this dry extract was encapsulated by ion gelation technique using sodium alginate (3 %) and calcium chloride solution (5 %). Based on our earlier study, 3 % of dried flower extract was added in sodium alginate solution (3 %) and mixed properly and separately, 5 % calcium chloride solution was also prepared. By using Encapsulator machine (model: B390, Buchi, Switzerland) the encapsulates were obtained, where the solution of sodium alginate (mixed with flower extract) was dropped into calcium chloride solution with mild stirring. After 30 min of immersed in calcium chloride solution, encapsulates were washed with distilled water and kept in a glass petri plates and let it air dried. The dried encapsulates were put in a zip lock pouch and kept inside an air tight container at 4 °C for further use.

### **6.2.4. Gummy preparation**

The composition of gummy was finalized by underwent various trials to process the gummy development. Initially, watermelon juice (50 mL) was taken and small amount of water is also added to mix well with other ingredients. With juice sugar (52.75 %) was added and heated while stirring until it dissolved properly. Pectin (2.53 %), agar-

agar (2.11 %) and citric acid (0.41 %) was also added later into the mixture. The whole mixture was mixed properly until it reached to TSS 72 °Brix. Except control (C) set of gummy, in other set of gummies 0.5 %, 1 %, 1.5 % and 2 % encapsulates were added and they are coded as G1, G2, G3 and G4 accordingly. The gummy mixture was poured into moulds immediately and kept in a refrigerator for setting properly. After the formation of gummies, they were kept in air tight container for further analysis. (Fig. 6.1.).



**Fig. 6.1.** Gummies prepared C, G1, G2, G3 and G4 with encapsulated beads.

#### **6.2.5. RTS beverage preparation**

In case of RTS beverage, ash guard juice (50 %) was taken and it was added to pre-prepared sugar syrup solution. In this sugar syrup solution sugar (11.5 %), citric acid (0.22 %) and water (38.29 %) was mixed properly. The whole mixture was heated until it reached TSS 13 °Brix and homogenized with crude phytochemical extract (CERTS) and separately, another set was prepared where encapsulates were added (BRTS) after that in sterilized glass bottles the RTS beverage were filled hot and pasteurized it at 90 °C for 25 min. There were 3 different sets of beverages were prepared such as control (CRTS), with 1 % encapsulates (BRTS) and with 0.5 % of flower extract (CERTS). They were kept in refrigerator for further analysis.

#### **6.2.6. Proximate analysis**

The freshly prepared 5 sets of gummies which were coded as C, G1, G2, G3 and G4 analysed for moisture, total protein, total fiber and ash by adopting AOAC (2000)

method. Moisture content was determined by using drying oven (NDO-710W, Eyela) at 105 °C. Ash was analyzed by using muffle furnace. For fiber was analysed by employing Fibro plus (FES06) apparatus (Pelican Equipment, Chennai, India).

#### 6.2.7. Color analysis

The color parameter of all the gummies and RTS beverages were analyzed through hunter colorimeter (Hunter Lab, Reston, Virginia, USA). The color readings of L\*, a\* and b\* parameters were recorded in triplicate for gummies (G1, G2, G3 and G4) and RTS beverages (CRTS, BRTS & CERTS).

#### 6.2.8. Texture profile analysis (TPA)

The textural properties of gummies were determined by employing texture analyser (TA-XT plus, Stable Micro System, UK) using 0.5 R probe and the load cell was 30 kg. The texture parameters such as hardness, adhesiveness, cohesiveness and chewiness were analyzed for developed gummies (C, G1, G2, G3 and G4).

#### 6.2.9. Titratable acidity

Titration acidity of gummies and RTS beverage was determined by adopting volumetric method (Mohd et al., 2024), where 1 g of gummy was dissolved in 9 mL of Milli-Q water. In case of RTS beverage, it was also mixed with Milli-Q water. After that in that solution few drops of phenolphthalein were mixed. This homogenized solution was titrated with sodium hydroxide (0.1 N) and continue until there was purple color observed. With the help of Eq. 6.1 the Titratable acidity (%) was measured.

$$\text{Titration Acidity (\%)} = \frac{\text{Molarity of NaOH} \times \text{mL of NaOH} \times \text{equivalent weight of acid}}{\text{Weight of sample}} \times 100 \quad (6.1)$$

#### 6.2.10. TSS and pH

Total soluble solids were determined in gummies and RTS beverage. In a hand-held Atago refractometer, 2 mm thick slice sample (in case of gummies) were placed in the visor for the measurement (Delgado et al., 2015). In case of RTS beverage few drops of beverage were placed in the visor of refractometer and check the readings.

pH was measured by using a pH meter where it was equipped with a glass combined electrode. The gummies were cut into thin slices and mixed with hot water (1:10 w/v) at 27 °C and it was homogenized (Mohd et al., 2024). In case of RTS beverage, it was taken to check the pH by using the pH meter.

#### **6.2.11. Water activity**

The water activity of gummies were measured by using water activity meter (Aqualab 4TE Decagon Devices Inc. Washington, USA) at 25 °C by placing 2 g of sample (Mohd et al., 2024).

#### **6.2.12. Preparation of gummy extract for TPC, TFC and DPPH radical scavenging activity**

The extraction of gummy was done by adopting the method of Mohd et al., (2024) with a slight modification, by taking 5 g of Gummy dissolved in 50 mL of distilled water. It was kept for stirring in a magnetic stirrer until it was dissolved well. After that it was centrifuged at 6000 rpm for 15 min and the supernatant was filtered through Whatman no 1 filter paper. The extract was kept in refrigerated condition for further analysis.

##### **6.2.12.1. Total phenolic content**

The phenolic content of gummies and RTS beverage was determined by using Folin-Ciocalteu reagent (FCR) method (Panhekar et al., 2019). 0.2 mL of extract of gummies and same amount of RTS beverage was taken in separate test tubes. It was mixed with 0.5mL of FCR (diluted with water 1:10) and after 5 minutes 2 mL sodium carbonate (20 %) was added. The whole mixture was incubated for 1h. The absorbance was measured at 650 nm by using UV-Vis spectrophotometer. Gallic acid used as a standard for the calculation of the calibration curve and the results were represented as mg gallic acid equivalent per gram (gummies) or mL (RTS beverage).

##### **6.2.12.2. Total flavonoid content**

Total flavonoid content of gummies and RTS beverage was done by following the method of Panhekar et al. (2019). 1 mL extract was taken in a test tube and 4 mL

distilled water was added into it. After that 0.3 mL of sodium nitrite (5 %) was added into it and after 5 min 0.3 mL of aluminium chloride ( $\text{AlCl}_3$ ) (10 %) was added. The whole mixture was shaken and kept for 5 min and after that 2 mL of 1 M sodium hydroxide was added in to the mixture and finally it was diluted up to 10 mL by using distilled water and mixed well. The absorbance of the whole mixture was taken at 510 nm and results were expressed in terms of quercetin equivalent (mg QE/g).

#### **6.2.12.3. DPPH radical scavenging activity**

DPPH radical scavenging of gummies and RTS beverage was measured by adopting the procedure of Tundis et al. (2015) with slight modifications. 0.5 mL extract was taken and it was mixed with 2.5 mL of DPPH solution (0.1 mM DPPH). This mixture was incubating for 30 min in dark at room temperature and wavelength was read at 517 nm by using spectrophotometer. DPPH radical scavenging activity was done by using the following Equation 6.2.

$$\text{DPPH radical scavenging activity (\%)} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of control}} \times 100 \quad (6.2.)$$

#### **6.2.13. Sensory evaluation**

The developed gummies such were taken for sensory analysis by some semi-trained and untrained panel members at Department of Food Engineering and Technology, Tezpur University. 9 point hedonic scale was adopted where, a scale of extremely dislike (1) to extremely like (9) were to evaluate sensory parameters such as appearance, color, taste, mouth feel, after taste, texture and overall acceptability.

#### **6.2.13. Statistical analysis**

The data values were calculated for analysis of variance analysis (ANOVA) having level of significance at 95 % ( $p < 0.05$ ) by using IBM SPSS Statistics v.20 (SPSS, Chicago, IL, USA). All the experiments for gummies and RTS beverage were carried out in triplicates. Significance differences among the data were achieved by choosing Duncan's multiple range test in the software.

## 6.3. Results and Discussion

### 6.3.1. Composition of gummies

The addition of encapsulated Nongmangkha flower extract showed a significant enhancement in nutritional and functional properties of gummies as compared to the control gummy. Moisture (wet basis), ash, total fiber and total protein were analyzed for gummies and data is mentioned in Table 6.1. It was observed that the moisture content of all these gummies exhibited no significant differences ( $p > 0.05$ ). This can be assumed that incorporation of encapsulates might not considerably alter the water retention capacity of the product. This consistency is desirable for maintaining textural properties and shelf stability. The lowest moisture content was observed in G1 gummy i.e. 35.16 % and highest was found in G3 gummy (37.18 %). Burrey et al., (2009) found moisture content of 55.08 % in gummies containing sucralose was used as the substitute sugars and 29.44 % in gummies where brown sugar was utilized. Teixeira-Lemos et al. (2021) found the moisture content of 18.2 (g water/g sample) in Orange and honey gummy and 20.82 (g water/g sample) in Berries mix gummy. It can be understood that the moisture content of gummies varies based on their composition of the ingredients. The gummies showed to have intermediate level of moisture content due to its composition of high sugars along with other hygroscopic substances.

Ash content increased significantly in all functional gummies compared to the control gummy (0.1 %). Encapsulates incorporated gummies showed to have ash content ranged from 0.66 % (G4) to 0.76 % (G1). This enhancement is likely due to the mineral content of the encapsulated phytochemical matrix and as well as overall encapsulates composition especially sodium alginate, demonstrating the nutritional enrichment of the gummies. The control recorded the lowest ash content (0.1 %), whereas G1 exhibited the highest value (0.76 %). Viswanathan et al. (2014) studied compared ash content of sodium alginate from various brown seaweeds and they found that highest ash content of 23.01 % was observed in *Padina gymnospora*. Others such as *Chnoospora implexa*, *Lobophora variegata* had showed ash content of 21.53 %, 12.78 % respectively. As encapsulates were made by using polymer sodium alginate, so it can be seen that sodium alginate also helped in the increment of ash content in gummies.



Total fiber content also exhibited a gradual increase from the control (0.29 %) to encapsulates incorporated gummies. G4 (0.45 %) showed a comparable fiber level to G3. The enhanced fiber levels are beneficial for functional food claims, as dietary fiber contributes to improved gut health. The crude fiber content showed a notable increase from the control (0.26) to G4 (0.45).

The Control had the lowest protein content (0.79 %), while G3 recorded the highest value (1.32 %). A slight decline in protein content was observed in G3 and G4 compared to G2, suggesting that the treatment process might affect protein stability or interaction with other components and also possibly due to variability in ingredient dispersion at lower encapsulates concentrations.

**Table 6.1.** Physicochemical properties of gummies

Parameters	Control	G1	G2	G3	G4
Moisture (% wet basis)	36.24±0.94 <sup>a</sup>	37.26±1.83 <sup>a</sup>	37.97±1.32 <sup>a</sup>	37.85±0.69 <sup>a</sup>	37.04±1.73 <sup>a</sup>
Water activity	0.74±0.00 <sup>a</sup>	0.78±0.00 <sup>b</sup>	0.78±0.00 <sup>b</sup>	0.79±0.03 <sup>b</sup>	0.83±0.03 <sup>c</sup>
Ash (%)	0.15±0.05 <sup>a</sup>	0.68±0.5 <sup>b</sup>	0.77±0.12 <sup>b</sup>	0.77±0.08 <sup>b</sup>	0.79±0.07 <sup>b</sup>
Crude fiber (%)	0.26±0.04 <sup>a</sup>	0.37±0.04 <sup>b</sup>	0.37±0.07 <sup>b</sup>	0.53±0.03 <sup>c</sup>	0.57±0.02 <sup>c</sup>
Protein (%)	0.74±0.06 <sup>a</sup>	1.08±0.09 <sup>b</sup>	1.38±0.16 <sup>c</sup>	1.20±0.2 <sup>bc</sup>	1.01±0.17 <sup>bc</sup>
Titrateable acidity (%)	0.05±0.03 <sup>a</sup>	0.05±0.09 <sup>a</sup>	0.06±0.02 <sup>a</sup>	0.06±0.05 <sup>a</sup>	0.05±0.05 <sup>a</sup>
pH	4.33±0.25 <sup>a</sup>	4.3±0.2 <sup>a</sup>	4.6±0.15 <sup>a</sup>	4.2±0.25 <sup>a</sup>	4.3±0.25 <sup>a</sup>
Total soluble solids (°Brix)	72.17±0.29 <sup>a</sup>	71.83±0.29 <sup>a</sup>	72±0.0 <sup>a</sup>	72.17±0.29 <sup>a</sup>	72.17±0.29 <sup>a</sup>
TPC (mg GAE/g)	0.078±0.01 <sup>a</sup>	0.089±0.009 <sup>a</sup>	1.14±0.056 <sup>b</sup>	1.20±0.031 <sup>b</sup>	1.35±0.020 <sup>c</sup>
TFC (mg QE/g)	0.015±0.002 <sup>a</sup>	0.03±0.004 <sup>b</sup>	0.041±0.007 <sup>bc</sup>	0.048±0.003 <sup>cd</sup>	0.053±0.03 <sup>d</sup>
DPPH activity (%)	12.016±1.32 <sup>a</sup>	12.92±0.33 <sup>ab</sup>	13.83±0.27 <sup>b</sup>	15.91±0.23 <sup>c</sup>	16.35±0.40 <sup>c</sup>

All data are the mean ± SD of three replicates. Mean followed by different letters in the same row differs significantly ( $p \leq 0.05$ )

### 6.3.2. Water activity

Water activity was also in the range from 0.67<sub>aw</sub> (C) to 0.78<sub>aw</sub> (G3). Water activity showed a noticeable increase in bead-containing samples compared to the control. G1 (0.75), G2 (0.77), and G3 (0.78) exhibited significantly higher values ( $p < 0.05$ ). This increase might be attributed to the moisture-binding capacity of the encapsulates. The slightly elevated water activity in of gummies compared to control gummy aligns with their increased bead concentration, which can influence microbial stability and shelf life. Water activity showed significant variation across the groups. The increase in water activity from G1 to G4 suggests a possible correlation with addition of

encapsulates. Moisture of Gummy in generally is intermediate-level and as it is high in sugars and other hygroscopic substances, resulting in a low water activity.

### **6.3.3. Titratable Acidity and pH**

Both titratable acidity and pH remained stable across the variants of gummies, with no significant differences ( $p > 0.05$ ). The titratable acidity ranged between 0.05–0.06 %, and the pH remained mildly acidic (4.2–4.6), ensuring flavor consistency and product stability. There were no significant differences were observed in titratable acidity across the all verities of gummies ( $p > 0.05$ ). The values ranged from 0.05 (control) to 0.06 (G3), indicating that the addition of encapsulates did not alter the acid content appreciably and stability of encapsulates during the process. The pH values were statistically similar across all groups, ranging from 4.2 (G3) to 4.6 (G2). This consistency suggests that encapsulates did not significantly affect the overall acidity of the gummies. As we can observe that there was no significant difference of titratable acid in all the gummies so there is no significance difference of pH. This indicates that the incorporation of encapsulates till 2% has no significant alteration of titratable acidity and pH.

### **6.3.4. Total Soluble Solids (TSS)**

All samples retained the same TSS value of 72 °Brix, confirming that the inclusion of encapsulated encapsulates did not alter the sugar content or sweetness levels of the gummies. The total soluble solids (°Brix) showed no significant differences among the gummies, with values ranging from 71.83 to 72.17 °Brix.

### **6.3.5. Total Phenolic Content (TPC) and Total Flavonoid Content (TFC)**

A significant increase in total phenolic content was observed from the Control (0.078 mg GAE/g) to G4 (1.35 mg GAE/g) (Table 6.1.). The progressive rise in TPC with increasing the amount of encapsulates highlights the potential enhancement of phenolic compounds in Gummies. This trend highlights the contribution of encapsulated phytochemicals to the functional properties of the gummies.

The total flavonoid content also increased significantly among these gummies. The control exhibited the lowest TFC (0.015 mg QE/g), while G4 recorded the highest

value (0.053 mg QE/g). This trend aligns with the observed increase in phenolic content, reinforcing the contribution of flavonoid to the antioxidant profile of the treated samples and indicating that higher amount of encapsulates are necessary to achieve significant flavonoid enrichment.

### **6.3.6. DPPH Radical Scavenging Activity (%)**

Antioxidant activity, measured as DPPH radical scavenging activity and it was observed to be increased significantly with increase in encapsulates concentration in Gummies from 12.016 % (control) to 16.35 % (G4) (Table 6.1.). These results demonstrate the ability of encapsulates to deliver the bioactive compounds or phenolic compounds with antioxidant properties into the food matrix effectively. The correlation between TPC, TFC, and DPPH activity further underscores the role of phenolic and flavonoid compounds in enhancing antioxidant property. Binding properties of Sodium alginate may aid in the retention or activation of phenolic and flavonoid compounds, as reported by Huang et al. (2019). The progressive increase in TPC and TFC, along with improved DPPH radical scavenging activity, reflects potential of sodium alginate in enhancing antioxidant property (Zhou et al., 2020).

### **6.3.7. Color**

The color parameters ( $L^*$ ,  $a^*$ ,  $b^*$ ) of the Gummies revealed significant differences among all the Gummies (Table 6.2). The lightness ( $L^*$ ) values decreased from 46.44 (Control) to 43.88 (G4), indicating a slight darkening of the Gummies. The redness ( $a^*$ ) values increased significantly in the gummies, with values ranging from 16.91 (Control) to 18.68 (G3), though there were no significant differences were observed among the Gummies incorporated with encapsulates. Yellowness ( $b^*$ ) showed a significant increase from 11.91 (Control) to 13.73 (G3), suggesting enhanced chromaticity in the treated samples. The observed changes in color parameters could be attributed to Maillard reactions, pigment incorporation due to incorporation of the colored encapsulates. The slight darkening and increased chromaticity observed are consistent with findings by Kim et al. (2019), who attributed these changes to Maillard reactions and pigment transformation during processing or incorporation of encapsulates.

**Table 6.2.** Color parameters of gummies

Sample	L*	a*	b*
Control	46.74±0.87 <sup>c</sup>	16.91±1.28 <sup>a</sup>	11.91±1.02 <sup>a</sup>
G1	45.82±0.48 <sup>bc</sup>	19.01±0.70 <sup>b</sup>	12.68±0.15 <sup>ab</sup>
G2	44.91±0.24 <sup>ab</sup>	19.74±0.17 <sup>b</sup>	13.40±0.27 <sup>bc</sup>
G3	44.87±0.11 <sup>ab</sup>	19.89±0.072 <sup>b</sup>	13.70±0.08 <sup>c</sup>
G4	43.88±1.08 <sup>a</sup>	18.68±0.88 <sup>b</sup>	13.73±0.35 <sup>c</sup>

All data are the mean ± SD of three replicates. Mean followed by different letters in the same column differs significantly ( $p \leq 0.05$ )

### 6.3.8. Texture profile analysis

Texture is an essential parameter to analyze for Gummies or any other confectionary items. Hardness, springiness, cohesiveness, gumminess and chewiness are measured for all the Gummies formulations and results are shown in Table 6.2. Hardness values varied significantly ( $p < 0.05$ ) across the Gummies. The Control Gummy exhibited a hardness of 36.21 g, which was higher than G3 (30.28 g) but lower than G1 (40.07 g). G4 had a value of 38.51 g, indicating that the addition of encapsulates affected the firmness of the samples. G2 showed intermediate hardness (34.84 g), suggesting a moderate effect of the treatment. Springiness decreased significantly from G1 (0.98) to G4 (0.56) Gummies, with the Control showing a value 0.93. G1 exhibited a relatively higher springiness, indicating potential variations in elasticity due to structural modifications from encapsulates. Cohesiveness helps to determine the strength, breakable range of inner bonds which hold the structure of Gummy and deformation (Bölük et al. 2021). Cohesiveness varied across the samples, with the highest value observed in G1 (0.08) and the lowest in G4 (0.029). The Control group displayed a moderate cohesiveness of 0.04. Chewiness and gumminess are closely related to gummy hardness. The firmer the structure of the gummy, the harder it becomes, resulting in increased chewiness and gumminess (Mahat et al., 2020). Gumminess values followed a similar trend, with control recording the highest gumminess (11.39) and G3 the lowest (0.69). Chewiness decreased significantly ( $p < 0.05$ ) than the Control (10.59) in all the gummies.

**Table 6.3.** Texture profile analysis of gummies

Sample	Hardness (N)	Springiness	Cohesiveness	Gumminess (N)	Chewiness (N)
Control	36.21±0.44 <sup>c</sup>	0.931± 0.05 <sup>c</sup>	0.315±0.04 <sup>c</sup>	11.39±1.34 <sup>c</sup>	10.59±1.18 <sup>c</sup>
G1	40.07±0.32 <sup>c</sup>	0.975± 0.05 <sup>c</sup>	0.083± 0.01 <sup>b</sup>	3.34± 0.48 <sup>b</sup>	3.27± 0.63 <sup>b</sup>
G2	34.84±1.16 <sup>b</sup>	0.800±0.07 <sup>b</sup>	0.037±0.01 <sup>a</sup>	1.31±0.24 <sup>a</sup>	1.04±0.23 <sup>a</sup>
G3	30.28±0.54 <sup>a</sup>	0.706±0.05 <sup>b</sup>	0.030±0.01 <sup>a</sup>	0.98±0.24 <sup>a</sup>	0.69±0.17 <sup>a</sup>
G4	38.51±0.62 <sup>d</sup>	0.560±0.09 <sup>a</sup>	0.029±0.01 <sup>a</sup>	1.62±0.14 <sup>a</sup>	0.90±0.11 <sup>a</sup>

All data are the mean ± SD of three replicates. Mean followed by different letters in the same row differs significantly ( $p \leq 0.05$ )

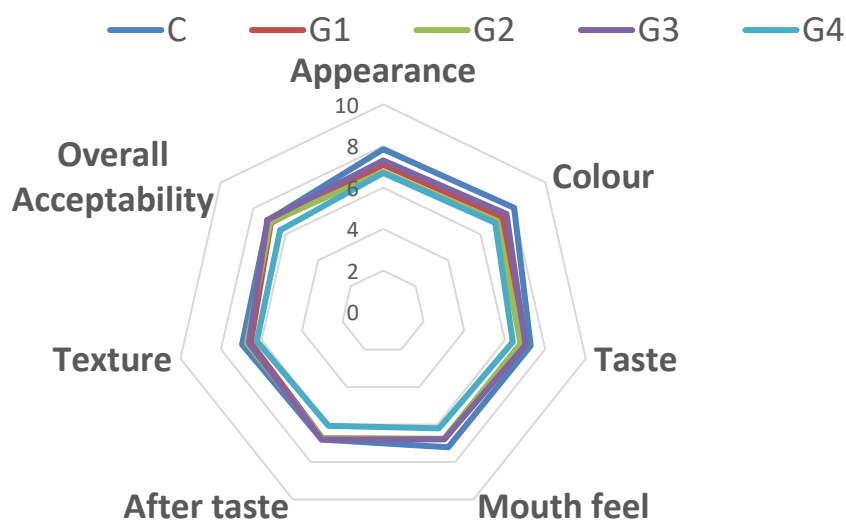
### 6.3.9. Sensory evaluation

Sensory evaluation is a scientific field that measures, examines, and interprets how people react or accept to certain food by their sense organs (Lohar et al., 2020). Sensory evaluation of food applies experimental design principles and statistical analysis. Sensory analysis of Gummies was done by within the 24 h of its preparation. As mentioned earlier 9 point hedonic scale was used with 1 being extremely disliked, 2 signified as slightly disliked, 3 was moderately disliked, 4 being slightly disliked, 5 being neither liked nor disliked, 6 was slightly liked, 7 was signified moderately liked, 8 was highly liked and 9 was extremely liked (Mahat et al., 2020). All the Gummies were evaluated by the parameters including appearance, color, taste, mouth feel, after taste, texture and overall acceptability with the help of semi trained panellist and results (Table 6.4.) were shown in Fig. 6.2. The responses from panellist help to understand more insight and inferences about the Gummies. In many ways, our perception of products is largely shaped by visual cues, especially color perception (Spencer et al., 2018). Since color significantly affects consumer appetite, it plays a crucial role in the overall palatability of food (Patras et al., 2019). From Table 6.4. it can be observed that the appearance was mostly appreciated by panellist was control Gummy and after that G3. In case of color also Control Gummy C was highly accepted followed by Gummy G3. Similarly, taste was also highly appreciated by control Gummy C and followed by Gummy G3. Addition of encapsulates effect on the taste of the Gummies. Mouthful was found mostly accepted in case of control Gummy C and followed by G1 and G3. The

after taste was found highly appreciated both in control Gummy C and G3. Texture was highly accepted in control Gummy C and followed by G2. Addition of encapsulates increase hardness of gummies which might not be like by the sensory panelists. The overall acceptability was observed in G3 and control Gummies. It can be assume that along with control Gummy also G3 Gummy was stood best consumer acceptability.

**Table 6.4.** Sensory evaluation scores of gummies

Sample Code	Appearance	Color	Taste	Mouth feel	After taste	Texture	Overall Acceptability
Control	7.84± 0.95	8.06±0.96	7.28±1.47	7.21±1.61	6.8±1.79	6.96±1.62	7.09±1.50
G1	7.09±0.83	7.34±1.11	6.84±1.16	6.78±1.40	6.73±1.51	6.53±1.23	6.9±1.28
G2	6.75±1.43	7.03±1.07	6.78±1.59	6.75±1.45	6.73±1.47	6.72±1.29	6.94±1.29
G3	7.3±1.43	7.59±1.41	7.03±1.49	6.78±1.45	6.8±1.48	6.62±1.4	7.12±1.38
G4	6.7±1.13	6.9±1.46	6.4±1.63	6.2±1.67	6.06±1.47	6.23±1.48	6.33±1.53



**Fig. 6.2.** Sensory score of Gummies

### 6.3.10. Properties of RTS beverage

The results of the study on Ash gourd Ready-to-Serve (RTS) beverage formulations are presented in Tables 6.5 and 6.6, which include physicochemical properties and color analysis of CRTS, CERTS, and BRTS. These parameters play a crucial role in determining the quality, stability, and consumer acceptability of RTS

beverages. The physicochemical characteristics of the RTS formulations indicate significant variations among CRTS, CERTS, and BRTS in terms of Titratable acidity, pH, TPC, TFC, and DPPH radical scavenging activity.

#### **6.3.10.1. Total Phenolic Content, Total Flavonoid Content and DPPH radical scavenging activity**

TPC content of CRTS, CERTS and BRTS were estimated and it was observed that addition of both encapsulates and crude significantly ( $p < 0.05$ ) increase the TPC content as well as TFC and DPPH radical scavenging activity. The phenolic content of CERTS (3.81 mg GAE/mL) was significantly higher than BRTS (1.19 mg GAE/mL) and CRTS (0.41 mg/mL). Similarly, the flavonoid content followed the same trend, with CERTS (0.64 mg QE/mL) exhibiting the highest TFC, followed by BRTS (0.49 mg QE/mL) and CRTS (0.07 mg QE/mL). Phenolic and flavonoid compounds contribute to antioxidant properties and have been linked to several health benefits, including reduced oxidative stress and improved cardiovascular health (Shahidi & Ambigaipalan, 2015). The higher phenolic and flavonoid content in CERTS could be attributed to the presence of certain bioactive compounds present in crude extract as well as in the encapsulates. The antioxidant activity, as measured by DPPH radical scavenging percentage, was highest in CERTS (57.17 %), followed by BRTS (51.43 %) and CRTS (37.61 %). The high antioxidant activity of CERTS may be directly related to its higher phenolic and flavonoid content. The high antioxidant activity of CERTS suggests that it may offer greater protection against oxidative stress, which is beneficial for health-conscious consumers. BRTS, with moderate antioxidant activity, remains a good alternative, whereas CRTS, with the lowest antioxidant capacity, is likely less effective in delivering health benefits. This suggests that CERTS might have better health-promoting properties, making it a more potent functional beverage.

#### **6.3.10.2. Titratable acidity and pH**

The acidity levels varied slightly, with CERTS showing the highest titratable acidity (0.31 %), followed by CRTS (0.27 %) and BRTS (0.27 %). The pH values were also within a narrow range (3.23–3.7), with CERTS exhibiting the highest pH (3.7), suggesting a relatively lower acidity compared to CRTS and BRTS. The pH and acidity levels significantly affect the beverage's microbial stability and sensory properties (Saha

et al., 2020). A higher acidity level typically enhances the shelf life and contributes to a tart flavor, which can be desirable in fruit-based beverages. CERTS had slightly higher acidity, making it more stable and tangy.

### 6.3.10.3. Color analysis

Color is an essential quality attribute of RTS beverages, influencing consumer preference and perception. CRTS exhibited the highest lightness value (31.57), while CERTS had the lowest (27.05) (Table 6.5). BRTS was intermediate at 29.06. Lower  $L^*$  values suggest a darker appearance, which may be due to the presence of more intense pigments from the crude extract of Nongmangkha flower. A lower  $L^*$  value in CERTS suggests a more intense color, potentially due to the higher concentration of anthocyanins or carotenoids, which also contribute to its superior antioxidant properties. The  $a^*$  values showed that CRTS (-0.42) and BRTS (-0.27) were more towards the green spectrum, whereas CERTS (0.9) had a slight red hue. A positive  $a^*$  value in CERTS suggests that it contains higher anthocyanin or carotenoid pigments, which could also be linked to its higher phenolic content. The  $b^*$  values revealed significant differences in the yellow-blue color range. BRTS (8.19) exhibited the highest  $b^*$  value, indicating a more intense yellowish appearance, whereas CRTS had the lowest (2.86). CERTS was in between (3.98). The variations in  $b^*$  values suggest differences in ingredient composition, effect of processing conditions on encapsulates and crude extract of Nongmangkha flower, and also pigment stability.

**Table 6.5.** Physicochemical properties of RTS

Sl. no	Parameters	CRTS	CERTS	BRTS
1	Titrateable acidity (%)	0.27±0.02 <sup>a</sup>	0.31±0.08 <sup>a</sup>	0.27±0.09 <sup>b</sup>
2	pH	3.37±1.84 <sup>a</sup>	3.7±1.47 <sup>a</sup>	3.23±0.82 <sup>a</sup>
3	TPC (mg/mL)	0.41±0.07 <sup>a</sup>	3.81±0.15 <sup>c</sup>	1.19±0.17 <sup>b</sup>
4	TFC (mg/mL)	0.07±0.005 <sup>a</sup>	0.64±0.008 <sup>c</sup>	0.49±0.004 <sup>b</sup>
5	DPPH activity (%)	37.61±1.85 <sup>a</sup>	57.17±1.48 <sup>c</sup>	51.43±3.2 <sup>b</sup>
6	$L^*$	31.57±2.45 <sup>a</sup>	27.05±3.52 <sup>c</sup>	29.06±2.85 <sup>b</sup>
7	$a^*$	-0.42±0.03 <sup>a</sup>	0.9±0.07 <sup>c</sup>	-0.27±0.02 <sup>b</sup>
8	$b^*$	2.86±0.24 <sup>a</sup>	3.98±1.18 <sup>b</sup>	8.19±2.09 <sup>c</sup>



All data are the mean  $\pm$  SD of three replicates. Mean followed by different letters in the same row differs significantly ( $p \leq 0.05$ )

#### **6.4. Conclusions**

The results indicate that the incorporation of encapsulates effectively enhanced the nutritional and functional properties of the gummies, particularly in terms of ash content, crude fiber, protein content, and antioxidant activity. The significant increases in TPC and TFC, along with improved DPPH activity, highlight the potential health benefits associated with the gummies. On the other hand, it can be said that the results confirm that incorporating encapsulates of Nongmangkha flower extract was significantly improved the nutritional and functional properties of the gummies. Higher encapsulates concentrations led to an increased phytochemical content and antioxidant activity, aligning with the goals of developing functional food products. The lack of significant changes in moisture content, titratable acidity, pH, and TSS suggests that the incorporation of encapsulates did not compromise the sensory and compositional stability of the Gummies. Variations in hardness, springiness, cohesiveness, gumminess, and chewiness suggest that addition of encapsulates in Gummy preparation influenced the textural quality and consumer acceptability of the products. Furthermore, while the increased protein, fiber, and mineral content enhances the health profile of the gummies.

The analysis of physicochemical properties and color characteristics of RTS formulations indicates that CERTS has superior bioactive properties due to its higher total phenolic and flavonoid content, leading to enhanced antioxidant activity. These characteristics make CERTS a potentially more health-beneficial beverage compared to CRTS and BRTS. Overall, the study highlights the importance of ingredient selection and processing techniques in enhancing the physicochemical properties and bioactivity of RTS beverages. Future studies should focus on optimizing formulation parameters to further enhance the functional and sensory properties of these beverages. These results provide valuable insights into the potential applications of Nongmangkha flower extract encapsulates in functional food development, offering enhanced nutritional, antioxidant, sensory, and textural profiles. Future studies should focus on bioavailability of enriched components.

## References

- Ahmed, M., Khan, T., & Yousaf, R. (2022). Influence of fortification on the texture and rheology of functional foods. *Journal of Food Science and Technology*, 59(4), 345-356.
- Akhtar, M., Dickinson, E., & Mazoyer, J. (2018). Protein-stabilizing effects of hydrocolloids in food emulsions. *Food Hydrocolloids*, 87, 326–336.
- Augustin, M. A. (2001). Functional foods: an adventure in food formulation. *Food Australia*, 53(10), 428-432.
- Bölük, E., Atik, D. S., Kolaylı, S., Demirci, A. Ş., & Palabiyik, I. (2021). Delivery of phenolics and caffeic acid phenethyl ester by propolis resin: Chewing gum system. *Food Bioscience*, 41, 101090.
- Burey, P., Bhandari, B. R., Rutgers, R. P. G., Halley, P. J., & Torley, P. J. (2009). Confectionery gels: A review on formulation, rheological and structural aspects. *International Journal of Food Properties*, 12(1), 176-210.
- Gupta, R., & Singh, P. (2020). Impact of protein enrichment and thermal treatments on food properties. *Food Chemistry*, 325, 126975.
- Huang, S., Zhao, J., & Liu, Y. (2021). Textural attributes of fiber-enhanced food products. *International Journal of Food Properties*, 24(7), 652-669.
- Huang, Y., Zhao, L., & Liu, S. (2019). Role of sodium alginate in preserving bioactive compounds during food processing. *Journal of Agricultural and Food Chemistry*, 67, 542-550.
- Jones, D., & Smith, L. (2021). Dietary fibers and gastrointestinal health: A systematic review. *Nutrition Research Reviews*, 34(3), 456-478.
- Khawaja, A. H., Qassim, S., Hassan, N. A., & Arafa, E. S. A. (2019). Added sugar: Nutritional knowledge and consumption pattern of a principal driver of obesity and diabetes among undergraduates in UAE. *Diabetes & Metabolic Syndrome: Clinical Research & Reviews*, 13(4), 2579-2584.
- Kim, H., Park, J., & Lee, K. (2019). Effects of thermal processing on color and antioxidant properties of food products. *Food Quality and Preference*, 27(2), 112-119.
- Klahorst, S.J. (2006). Flavour and innovation meet. *World of Food Ingredients*, June, 26–30.
- Klont, R. (1999). Healthy ingredients driving innovation. *World of Food Ingredients*, March/April, 43– 23.
- Kwak, N. S., & Jukes, D. J. (2001). Functional foods. Part 2: the impact on current regulatory terminology. *Food control*, 12(2), 109-117.

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- Lohar, P. B., Shrivastav, P. M., & Kulavoor, S. S. (2020). Development of fiber rich gummies.
- Mahat, M. M., Sabere, A. S. M., Nawawi, M. A., Hamzah, H. H., Jamil, M. A. F. M., Roslan, N. C., & Safian, M. F. (2020). The sensory evaluation and mechanical properties of functional gummy in the Malaysian market. *Preprints*.
- Martirosyan, D., Kanya, H., & Nadalet, C. (2021). Can functional foods reduce the risk of disease? Advancement of functional food definition and steps to create functional food products. *Functional Foods in Health and Disease*, 11(5), 213-221.
- Mohd Isa, N. S., Cheok, H. Y., Mohsin, A. Z., & Mohd Maidin, N. M. (2024). The Effect of Different Citric Acid Concentrations on Physicochemical and Antioxidant Properties of Red Pitaya Peel Gummy Candies. *Tropical Journal of Natural Product Research*, 8(12).
- Panhekar, D., Sawant, T., & Gogle, D. P. (2019). Phytochemicals extraction, separation and analysis. Global Education Limited.
- Pathare, P. B., Opara, U. L., & Al-Said, F. A. J. (2013). Colour measurement and analysis in fresh and processed foods: A review. *Food and bioprocess technology*, 6, 36-60.
- Patras, A. (2019). Stability and colour evaluation of red cabbage waste hydroethanolic extract in presence of different food additives or ingredients. *Food chemistry*, 275, 539-548.
- Prior, R. L., Wu, X., & Schaich, K. (2005). Standardized methods for the determination of antioxidant capacity and phenolics in foods and dietary supplements. *Journal of agricultural and food chemistry*, 53(10), 4290-4302.
- Rippe, J. M., & Angelopoulos, T. J. (2016). Relationship between added sugars consumption and chronic disease risk factors: current understanding. *Nutrients*, 8(11), 697.
- Saha, D., Shinde, J., & Mishra, S. (2020). Effect of pH and acidity on microbial stability of fruit-based beverages. *Journal of Food Science and Technology*, 57(3), 921-930.
- Shahidi, F., & Ambigaipalan, P. (2015). Phenolics and polyphenolics in foods, beverages and spices: Antioxidant activity and health effects—A review. *Journal of functional foods*, 18, 820-897.
- Singh, R., Kumar, P., & Sharma, K. (2021). Influence of hydrocolloids on the textural and sensory properties of functional foods. *International Journal of Food Science & Technology*, 56, 873-885.
- Smith, A., Brown, C., & Miller, D. (2019). Mineral and fiber fortification in processed foods. *Advances in Food Science and Nutrition*, 6(3), 213-225.
-

- Smith, L., Jones, D., & Brown, M. (2020). Mineral fortification using sodium alginate in food products. *Food Chemistry*, 128, 1120–1128.
- Spencer, A. R., Primbetova, A., Koppes, A. N., Koppes, R. A., Fenniri, H., & Annabi, N. (2018). Electroconductive gelatin methacryloyl-PEDOT: PSS composite hydrogels: Design, synthesis, and properties. *ACS biomaterials science & engineering*, 4(5), 1558-1567.
- Teixeira-Lemos, E., Almeida, A. R., Vouga, B., Morais, C., Correia, I., Pereira, P., & Guiné, R. P. (2021). Development and characterization of healthy gummy jellies containing natural fruits. *Open Agriculture*, 6(1), 466-478.
- Viswanathan, S., & Nallamuthu, T. (2014). Extraction of sodium alginate from selected seaweeds and their physiochemical and biochemical properties. *Extraction*, 3(4).
- Xiao, J., & Bai, W. (2019). Bioactive phytochemicals. *Critical reviews in food science and nutrition*, 59(6), 827-829.
- Zhang, W., Wang, X., & Chen, L. (2018). Phenolic compound activation and antioxidant capacity in thermally treated foods. *Journal of Agricultural and Food Chemistry*, 66(1), 678-685.
- Zhou, W., Wang, X., & Cheng, L. (2020). Antioxidant enhancement in sodium alginate-based food coatings. *Food Research International*, 131, 108980.