

## FORMULATION AND QUALITY EVALUATION OF *SPIROGYRA*-BASED STARCH CRACKERS

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### Abstract

This study aimed to determine the formulation and the combination level of *Spirogyra* in starch-based crackers and to evaluate product qualities through sensory acceptance, moisture content and water activity ( $a_w$ ) in accordance with the Thai Community Product Standard (TCPS 107/2554). Different starch ratios were first examined to establish formulation. Sensory evaluation using a 9-point hedonic scale ( $n = 50$ ) indicated the starch ratio of tapioca starch and all-purpose flour of 130:70 achieved the highest overall acceptability ( $p < 0.05$ ) and was selected as the optimal formulation. *Spirogyra* puree was subsequently incorporated at 10%, 15%, and 20% (w/w). The 15% formulation obtained the highest overall liking score ( $8.2 \pm 0.4$ ) and demonstrated balanced sensory attributes confirmed by just-about-right (JAR) analysis. Increasing *Spirogyra* concentration significantly increased moisture content ( $p < 0.05$ ); however, all samples complied with TCPS 107/2554, with moisture content below 12% and  $a_w$  below 0.60, indicating the microbial stability. These findings suggested that incorporation of 15% *Spirogyra* in a 130:70 starch ratio provides optimal sensory quality while meeting regulatory standards for commercial snack production.

**Keywords:** *Spirogyra*, Starch Crackers, Sensory Evaluation, Formulation

### Introduction

*Spirogyra sp.* is a freshwater green macroalga traditionally consumed in some regions of Thailand. Although it is locally available and has been used as a seasonal food ingredient, its application in processed snack products remains limited. In particular, the incorporation of freshwater macroalgae into starch-based cracker systems requires careful formulation to maintain product quality, safety and sensory acceptance.

Thai starch crackers (Khao Kriap) are produced through mixing, steaming, slicing, drying, and expansion during frying. Product characteristics such as crispiness and overall acceptability depend largely on starch composition and moisture control. The addition of high-protein or high-fiber ingredients may influence dough structure and final texture. Therefore, determining an appropriate level of incorporation is necessary to ensure that product stability and consumer acceptance are maintained.

In addition to technological considerations, market trends also support the development of plant-based snack products. The Thai healthy snack market was valued at approximately 28,314 million baht in 2023, with continued annual growth (Krungthep Turakij, 2023). *Spirogyra* can now be produced year-round through greenhouse cultivation, resulting in more consistent raw materials for food applications (Donsujit et al., 2025).

Despite its potential, limited research has systematically determined the appropriate incorporation level of freshwater *Spirogyra* in starch-based cracker formulations, particularly with respect to sensory acceptability and moisture content and  $a_w$  parameters required for commercial production. Therefore, this study aimed to determine a standardized formulation for *Spirogyra*-based crackers by investigating the optimal incorporation level and evaluating product quality through sensory analysis, moisture content determination and  $a_w$  measurement.

## Research Objectives

1. To determine the formulation for *Spirogyra*-based crackers
2. To investigate the concentration of *Spirogyra* for production
3. To analyze the moisture content and  $a_w$  to ensure commercial production

## Literature Review

*Spirogyra* is a filamentous green macroalga belonging to the family Zygnemataceae. Previous studies have reported protein contents ranging from 12.0% to 36.96% on a dry weight basis (Sriwattana et al., 2024; Yongkhamcha & Buddhakala, 2023). It also contains substantial amounts of carbohydrates, dietary fiber, and phenolic compounds. Due to this composition, *Spirogyra* has been considered a potential functional ingredient in food systems.

Thai crackers (Khao Kriap) are starch-based snacks produced by steaming to achieve starch gelatinization, followed by drying and expansion during frying (Tipnee et al., 2015). The quality of these products depends largely on starch composition and moisture distribution. Tapioca starch, which is rich in amylopectin, promotes expansion, while wheat flour contributes protein that strengthens the dough matrix. Previous studies have shown that moderate substitution of tapioca starch with wheat flour can improve structural integrity and sensory properties. In contrast, excessive substitution may reduce expansion and produce a dense texture.

According to the Thai Community Product Standard (TCPS 107/2554), the moisture content of uncooked crackers must not exceed 12.0% (w/w). In addition,  $a_w$  below 0.60 is generally required to inhibit microbial growth in low-moisture foods. These parameters are essential for ensuring efficient expansion during frying and preventing mold growth under tropical storage conditions. The incorporation of high-protein or high-fiber ingredients, such as macroalgae, may affect moisture retention and structural properties, necessitating formulation optimization.

## Research Methodology

### 1. Preparation of *Spirogyra* Puree

Fresh *Spirogyra* was obtained from a commercial supplier, which is cultivated in Ban Na Khuha, Phrae Province, Thailand. The alga were washed thoroughly to remove impurities and then blanched in boiling water for 1 min to reduce microbial load and inactivate enzymes. After blanching, excess water was removed by manual pressing to obtain a consistent base weight. The sample was then homogenized using a high-speed blender for 2 min to produce a fine puree. The puree was prepared fresh prior to incorporation into the cracker formulations.

### 2. Production of Crackers

All dry powdered ingredients were sieved and mixed thoroughly. Water was added and the mixture was kneaded using a high-speed mixer for 5 min until a uniform dough was obtained. The dough was divided into 100 g portions and shaped into cylindrical logs with a diameter of approximately 5 cm. The dough was steamed in boiling water for 6 min to allow starch gelatinization. After cooling,

the samples were frozen at -18°C for 60 min to facilitate slicing. The frozen dough was sliced into thin pieces with a thickness of 2-3 mm. The slices were deep-fried for 4 min until fully expanded, then placed in a hot air oven for 5 min to remove excess oil.

### 3 Determination of the Standard Starch Formulation

Three starch ratios were evaluated while maintaining a constant total flour weight of 200 g. Formulation 1 consisted of tapioca starch only. Partial replacement with all-purpose flour was tested at two levels: 35% and 50%. All other ingredients were kept constant to ensure that differences in product characteristics were attributed to starch composition (Table 1).

**Table 1:** Formulations of *Spirogyra*-Based Crackers with Varying Starch Ratios

Ingredients (grams)	Formula 1	Formula 2	Formula 3
Tapioca starch	200	130	100
All-purpose flour	-	70	100
Baking powder	3	3	3
Sugar	15	15	15
Salt	2.5	2.5	2.5
Pepper	1	1	1
Seasoning powder	15	15	15
Water	50	50	50
Total Flour Weight	200	200	200

### 4. Study of the concentration of *Spirogyra* for production

The formulation was fortified with blanched *Spirogyra* puree at three levels: 10%, 15% and 20% (w/w). The effect of alga incorporation on sensory properties, moisture content and  $a_w$  was evaluated.

### 5. Analysis of Moisture Content and $a_w$

The physicochemical quality was analyzed in accordance with TCPS 107/2554 standards. Moisture content was determined using the AOAC gravimetric method, ensuring levels remained below 12.0% to facilitate optimal puffing and prevent microbial spoilage. Additionally,  $a_w$  was measured at 25±1°C, maintaining values below 0.60 to inhibit the growth of pathogenic bacteria, yeasts and molds, thereby ensuring the product's shelf-stability without preservatives.

### 6. Sensory Evaluation

Sensory acceptance was evaluated by 50 panelists using a 9-point hedonic scale. The attributes evaluated included color, aroma, saltiness, sweetness, oiliness, crispiness and overall liking. Moreover, the just-about-right (JAR) scale was used to optimize the intensity of the *Spirogyra* puree and seasonings. The formulation was considered optimal and required no further adjustment if the percentage difference between “Too Much” and “Too Little” remained within the range of -20% to 20%. Each panelist received two pieces of coded crackers per formulation. Drinking water was provided for palate cleansing between samples. The presentation order was randomized.

### 7. Statistical Analysis

Physicochemical analyses were performed in triplicate. Sensory evaluation was conducted once with 50 panelists, and individual responses were treated as independent observations for statistical analysis. Data were subjected to a One-Way Analysis of Variance (ANOVA) to determine significant differences among formulations. Mean differences were further analyzed using Duncan’s Multiple Range Test at a confidence level of 95% ( $p < 0.05$ ).

## Results

### Study of the formulation for *Spirogyra*-based crackers

The sensory acceptance of *Spirogyra*-based crackers formulated with different starch ratios was evaluated by 50 panelists using a 9-point hedonic scale. Attributes including color, aroma, saltiness, sweetness, oiliness, crispiness, and overall liking are summarized in Table 2.

**Table 2:** Sensory Evaluation Scores of *Spirogyra*-Based Crackers with Varying Starch Ratios

Formulation	Color	Aroma	Saltiness	Sweetness	Oiliness	Crispiness	Overall Liking
Formula 1	7.3 ± 0.5 <sup>b</sup>	7.2 ± 0.4 <sup>b</sup>	6.8 ± 0.6 <sup>a</sup>	6.8 ± 0.5 <sup>a</sup>	6.4 ± 0.4 <sup>b</sup>	6.1 ± 0.7 <sup>b</sup>	6.6 ± 0.5 <sup>b</sup>
Formula 2	7.8 ± 0.4 <sup>a</sup>	7.9 ± 0.3 <sup>a</sup>	7.0 ± 0.5 <sup>a</sup>	7.1 ± 0.4 <sup>a</sup>	7.4 ± 0.5 <sup>a</sup>	7.8 ± 0.4 <sup>a</sup>	7.2 ± 0.4 <sup>a</sup>
Formula 3	7.6 ± 0.6 <sup>a</sup>	7.7 ± 0.5 <sup>a</sup>	6.4 ± 0.7 <sup>b</sup>	6.3 ± 0.6 <sup>b</sup>	5.8 ± 0.6 <sup>c</sup>	5.2 ± 0.8 <sup>c</sup>	5.7 ± 0.6 <sup>c</sup>

**Source:** Values are expressed as Mean ± SD (n=50). Different superscript letters (a, b, c) within the same column indicate significant differences (p<0.05) according to Duncan's New Multiple Range Test (DMRT)

Starch composition significantly affected sensory attributes (p<0.05). Formula 2 (tapioca starch: all-purpose flour, 130:70) achieved the highest scores across most attributes, particularly crispiness (7.8 ± 0.4<sup>a</sup>) and overall liking (7.2 ± 0.4<sup>a</sup>). Formulas 1 and 3 showed significantly lower scores in texture-related attributes, especially oiliness and crispiness (p<0.05).

### Study of the concentration of *Spirogyra* for production

Sensory preference and just-about-right (JAR) analysis were conducted to determine the *Spirogyra* puree concentration (10%, 15%, and 20% w/w) (Table 3). The 15% formulation obtained the highest overall liking (8.2 ± 0.4<sup>a</sup>) and showed significantly higher color and aroma intensities (p<0.05). The 10% formulation received the highest crispiness score (8.1 ± 0.4<sup>a</sup>) but lower overall liking due to weak algae identity. The 20% formulation showed significantly lower scores in texture-related attributes. JAR analysis confirmed that the 15% formulation fell within the optimal ±20% range for all attributes, indicating no further adjustment was required (Table 5). In contrast, the 10% formulation showed negative deviations in color (-32%) and aroma (-35%), suggesting insufficient algae intensity (Table 4). The 20% formulation exhibited excessive color (+35%) and aroma (+34%) and a marked negative deviation in crispiness (-45%), indicating an overly hard texture (Table 6)

**Table 3:** Sensory Evaluation Scores of Crackers with Different *Spirogyra* Concentrations

<i>Spirogyra</i> (%)	Color	Aroma	Saltiness	Sweetness	Oiliness	Crispiness	Overall Liking
10%	7.1 ± 0.6 <sup>b</sup>	7.0 ± 0.5 <sup>b</sup>	7.0 ± 0.4 <sup>a</sup>	6.9 ± 0.5 <sup>a</sup>	7.2 ± 0.6 <sup>a</sup>	8.1 ± 0.4 <sup>a</sup>	7.4 ± 0.5 <sup>b</sup>
15%	7.9 ± 0.4 <sup>a</sup>	8.1 ± 0.3 <sup>a</sup>	7.2 ± 0.5 <sup>a</sup>	7.1 ± 0.4 <sup>a</sup>	7.4 ± 0.5 <sup>a</sup>	7.8 ± 0.5 <sup>a</sup>	8.2 ± 0.4 <sup>a</sup>
20%	6.8 ± 0.7 <sup>c</sup>	6.5 ± 0.8 <sup>c</sup>	6.4 ± 0.6 <sup>b</sup>	6.3 ± 0.7 <sup>b</sup>	6.1 ± 0.8 <sup>b</sup>	5.9 ± 0.9 <sup>b</sup>	6.2 ± 0.7 <sup>c</sup>

**Sauce:** Values are Mean ± SD (n=50). Different superscripts (a, b, c) in the same column indicate significant differences (p<0.05) by DMRT

**Table 4:** JAR Relative Percentages and Improvement Guidelines for 10% *Spirogyra* Formula

Attributes	% Too Little	% JAR	% Too Much	Difference (%Too much - %Too little)	Improvement Guidelines
Color	40%	52%	8%	-32%	Increase color intensity
Aroma	45%	45%	10%	-35%	Increase alga aroma
Saltiness	10%	80%	10%	0%	None
Sweetness	10%	85%	5%	-5%	None
Oiliness	5%	90%	5%	0%	None
Crispiness	5%	88%	7%	2%	None

**Table 5:** JAR Relative Percentages and Improvement Guidelines for 15% *Spirogyra* Formula

Attributes	% Too Little	% JAR	% Too Much	Difference (%Too much - %Too little)	Improvement Guidelines
Color	12%	75%	13%	1%	None
Aroma	15%	70%	15%	0%	None
Saltiness	10%	80%	10%	0%	None
Sweetness	12%	78%	10%	-2%	None
Oiliness	15%	75%	10%	-5%	None
Crispiness	18%	72%	10%	-8%	None

**Table 6:** JAR Relative Percentages and Improvement Guidelines for 20% *Spirogyra* Formula

Attributes	% Too Little	% JAR	% Too Much	Difference (%Too much - %Too little)	Improvement Guidelines
Color	5%	55%	40%	35%	Decrease Color Intensity
Aroma	8%	50%	42%	34%	Decrease Alga Aroma
Saltiness	15%	75%	10%	-5%	None
Sweetness	10%	70%	20%	10%	None
Oiliness	10%	60%	30%	20%	None
Crispiness	55%	35%	10%	-45%	Increase Crispiness

### Study of Moisture Content of Optimal Formulations

Moisture contents of the three starch bases complied with the Thai Community Product Standard (TCPS 107/2554), which specifies a maximum of 12.0%. The 130:70 ratio (Formula 2) was selected as the optimal baseline for fortification (Table 7).

**Table 7:** Moisture Content of Standard Cracker Starch Formulations

Formulation	Starch Ratio (Tapioca: All-purpose)	Moisture Content (%)
Formula 1	200 : 0	8.42 ± 0.15
Formula 2	130: 70	9.15 ± 0.22
Formula 3	100: 100	10.38 ± 0.18

### Moisture and $a_w$ of *Spirogyra*-Fortified Crackers

Increasing *Spirogyra* concentration from 10% to 20% significantly increased moisture content ( $p < 0.05$ ), reflecting the water-holding capacity of algal protein and fiber (Table 8). All samples maintained  $a_w$  below 0.60, ensuring microbial stability. The 15% formulation provided the optimal balance between product quality and safety, with a moisture content of  $9.56 \pm 0.08\%$  and an  $a_w$  of  $0.51 \pm 0.01$ .

**Table 8:** Moisture Content and  $A_w$  of Crackers with Varying *Spirogyra* Concentrations

<i>Spirogyra</i> (%)	Moisture Content (%)	$a_w$
10%	$8.24 \pm 0.12^c$	$0.45 \pm 0.02^b$
15%	$9.56 \pm 0.08^b$	$0.51 \pm 0.01^{ab}$
20%	$11.45 \pm 0.25^a$	$0.58 \pm 0.03^a$

**Source:** Values are Mean  $\pm$  SD (n=3). Different superscripts (a, b, c) in the same column indicate significant differences ( $p < 0.05$ )

### Discussion

Formula 2 (130 g tapioca starch: 70 g all-purpose flour) was identified as the most suitable starch base. This could be explained by the fact that tapioca starch provides high amylopectin content, which supports expansion during frying, while all-purpose flour contributes gluten that reinforces the starch matrix (Mohamed et al., 1989; Kengkhetkit & Mopoung, 2019). A moderate substitution level improved sensory attributes, particularly color and crispiness, consistent with previous findings (Kengkhetkit & Mopoung, 2019). However, excessive wheat flour substitution (50%) resulted in a denser texture and might reduce expansion. Excessive protein content may increase matrix rigidity and limit stretching during frying (Mohamed et al., 1989). All formulations complied with the Thai Community Product Standard (TCPS 107/2554), with moisture content below 12.0% (Thai Industrial Standards Institute, 2011). Maintaining moisture below this level is important to ensure proper puffing and reduce the risk of mold development (Sammart & Sompoowech, 2016).

Sensory evaluation and just-about-right (JAR) analysis indicated that a 15% (w/w) *Spirogyra* puree incorporation level was the most appropriate, achieving the highest overall liking score ( $8.2 \pm 0.4$ ). The 10% formulation provided limited alga characteristics, while the 20% formulation showed significantly lower crispiness scores. The decrease in crispiness at 20% may be related to the high protein and fiber content of *Spirogyra*, which increases water-holding capacity and can interfere with moisture evaporation during frying (Surayot et al., 2015; Yongkhamcha & Buddhakala, 2023; Tipnee et al., 2015). Similar effects of high-fiber ingredients on texture have also been reported in starch-based snack systems (Maisont et al., 2021).

An increase in *Spirogyra* concentration corresponded with higher moisture content, rising from 8.24% to 11.45%. This trend is likely related to the hydrophilic nature of algal polysaccharides and proteins (Surayot et al., 2015). Despite this increase,  $a_w$  remained below 0.60 in all formulations. The value of  $a_w$  below 0.60 is widely recognized as the threshold that inhibits microbial growth in low-moisture foods (AOAC, 2000; Thai Industrial Standards Institute, 2011). These results indicate that *Spirogyra* can be incorporated at appropriate levels without compromising product stability or safety.

### Conclusion

The optimal formulation for *Spirogyra*-based starch crackers was a 130:70 tapioca starch-to-all-purpose flour ratio fortified with 15% (w/w) *Spirogyra* puree. This formulation achieved the highest overall sensory acceptance and exhibited balanced attributes according to JAR analysis. All

samples complied with the Thai Community Product Standard (TCPS 107/2554), with moisture content below 12% and  $a_w$  below 0.60. These results indicate that *Spirogyra* can be incorporated into starch crackers without compromising product quality or safety.

## Recommendations

This study did not include a comprehensive nutritional analysis of the final product. In addition, long-term shelf-life evaluation under storage conditions was not conducted and should be investigated in future studies.

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