

# Optimization of ready-to-drink Chrysanthemum flower tea formulation based on bioactive compounds and sensory properties

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**Abstract.** This study aims to optimize the formulation of ready-to-drink Chrysanthemum flower tea using Response Surface Methodology-Box Behnken Design (RSM-BBD) to meet modern consumer preferences for health-oriented and convenient beverages. The optimal formulation was determined based on maximizing bioactive compounds, including total phenolics, flavonoids, and anthocyanins, as well as antioxidant activity and sensory acceptability. The best formulation involved brewing 2 g of Chrysanthemum flower powder in water at 90°C with 14.281% sugar. This formulation yielded a total phenolic content of 535.752 mg GAE/g, total flavonoid content of 49.909 mg QE/g, and anthocyanin content of 5.929 mg/L, indicating significant health-promoting properties. Sensory evaluations highlighted the formulation's excellent taste, aroma, and color, making it highly appealing to consumers. The findings have practical implications for the beverage industry, providing a scientific basis for developing functional and consumer-friendly herbal drinks. This optimized Chrysanthemum tea formulation aligns with the growing demand for health-conscious products that combine nutritional benefits with sensory satisfaction.

## 1 Introduction

*Chrysanthemum morifolium* is a flowering plant, primarily grown in China, Japan, Europe, and the United States, and is often cultivated for medicinal uses [1]. It is known for its aromatic and flavorful qualities, making it popular as a beverage. *Chrysanthemum* flowers offer numerous health benefits, including anti-inflammatory, immune-regulating, anti-allergic, antioxidant, antimicrobial, anticancer, anti-obesity, liver-protective, and kidney-protective effects. The extracts of these flowers contain valuable phytochemicals such as cyanidin-3-O-(6''-O-malonyl) glucoside, delphinidin 3-O-(6''-O-malonyl) glucoside-3', rutin, quercetin, isorhamnetin, rutinoid, which contribute to their use in supplementary food products and pharmaceuticals [2,3].

In Indonesia, North Sumatera ranks as the fourth-largest producer of Chrysanthemum flowers, making it a primary region for flower production [4]. Specifically

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Tanah Karo Regency, notably in Raya Village, Berastagi District, cultivates over 11 *Chrysanthemum* cultivars. However, waste management becomes a significant issue, as *Chrysanthemums* are predominantly used for ornamental purposes, particularly in traditional ceremonies. This creates a supply-demand mismatch, leading to excess flowers and waste. Yellow and white *Chrysanthemum* varieties are primarily favored for decorations, while other cultivars, such as the white-purple *Batik* *Chrysanthemum*, receive less attention and are mainly used as complementary elements in bouquets. *Batik* refers to a *Chrysanthemum* cultivar distinguished by its unique white-purple petal pattern, resembling the intricate designs of traditional Indonesian batik textiles. This underutilization of certain cultivars underscores the need for diversification of *Chrysanthemum* applications to manage oversupply and add market value.

*Chrysanthemum* tea, an increasingly popular product, is consumed in various forms, including infusions, individual brews, and ready-to-drink formats [5]. Research by Han et al. [6] highlights that purple *Chrysanthemum* tea, derived from cultivars such as *Batik*, contains anthocyanins and linarin, which are absent in yellow *Chrysanthemum* tea, and exhibits higher antioxidant activity compared to yellow varieties. This suggests a potential functional advantage for using underutilized purple *Chrysanthemum* cultivars in beverage production.

Ready-to-drink tea products have gained immense popularity due to their convenience and ease of consumption [7,8]. However, creating a ready-to-drink formulation presents challenges in maintaining both flavor and functional benefits. Current studies predominantly focus on yellow *Chrysanthemum* tea, with limited exploration of purple cultivars like *Batik*, despite their distinct phytochemical profiles and health benefits. *Batik* *Chrysanthemum*, is known for its antioxidant, anti-inflammatory, and immune-boosting properties, offers potential as a health-conscious beverage option [2]. This study addresses the research gap by optimizing the formulation of a ready-to-drink *Chrysanthemum* tea using *Batik* *Chrysanthemum*. By employing Response Surface Methodology (RSM) with Box-Behnken Design (BBD), the study aims to maximize total phenolic, flavonoid, and anthocyanin content while ensuring superior sensory quality. This research addresses waste management challenges and aligns with consumer trends for health-conscious, convenient beverage options.

## 2 Materials and methods

*Batik* cultivar of *Chrysanthemum* (*Chrysanthemum morifolium* Ramat) was collected from Raya Village, Berastagi District, Tanah Karo Regency, North Sumatra Province. Fresh flowers used in this study had a uniform shape, size, and color.

### 2.1 Making process of ready-to-drink *Chrysanthemum* flower tea drink in bottle packaging

*Chrysanthemum* flowers were washed under tap water, withered by letting stand at room temperature for 24 hours, and dried using an oven at 60 °C for 24 hours. The dried *Chrysanthemum* flowers are mashed by blending and sifted through a 10-mesh sieve. The RTDT formulation was optimized using the RSM-BDD program. The independent variables included *Chrysanthemum* powder in tea bags (1g, 1.5g, 2g), brewing water temperature (30 °C, 60 °C, 90 °C), and sugar concentration (0%, 7.5%, 15%). Each tea bag was brewed in 100 mL water and stirred for 5 minutes. Sugar and citric acid (0.25%) were added, and the tea was packaged in plastic bottles and stored at 10 °C.

The formulation optimization for *Chrysanthemum* tea preparation was conducted using the Design Expert 13.0® software with the Response Surface Methodology (RSM) Box-Behnken Design program. The independent variables in this study included the weight of tea

powder, brewing temperature, and sugar concentration. The optimized formulations of the Chrysanthemum tea ingredients as determined by the Design Expert 13.0® RSM Box-Behnken Design (BBD) program are presented in Table 1.

## 2.2 Packaging and storage of Chrysanthemum flower tea beverages in bottles

The Chrysanthemum flower tea drink was packaged into a polyethylene terephthalate plastic bottle using the hot filling method. The Chrysanthemum flower tea drinks were then stored for 3 days before analysis of its physicochemical and sensory properties.

## 2.3 Selection of the best Chrysanthemum tea drink formulation

The best Chrysanthemum tea drink was selected based on the highest desirability level in the optimization results using Response Surface Methodology (RSM) with the Box-Behnken Design (BBD) in the Design Expert 13.0® program. This was conducted on samples to obtain products with optimal values for parameters such as total phenolic content, total flavonoid content, total anthocyanin content, and sensory test values. The best formulation of Chrysanthemum flower tea was then tested for its antioxidant activity using the DPPH method.

**Table 1.** Optimization design of ready-to-drink Chrysanthemum flower tea formulation in bottles based on Chrysanthemum flower tea powder weight, brewing water temperature, and sugar concentration using Design Expert 13.0® RSM Box-Behnken Design

Std	Run	Chrysanthemum flower powder weight (g)	Brewing water temperature (°C)	Sugar concentration (%)
16	1	1.5	60	7.5
5	2	1	60	0
6	3	2	60	0
10	4	1.5	90	0
3	5	1	90	7.5
8	6	2	60	15
17	7	1.5	60	7.5
4	8	2	90	7.5
11	9	1.5	30	15
2	10	2	30	7.5
7	11	1	60	15
15	12	1.5	60	7.5
12	13	1.5	90	15
13	14	1.5	60	7.5
1	15	1	30	7.5
14	16	1.5	60	7.5
9	17	1.5	30	0

## 2.4 Determination of total phenolic content

First, the maximum wavelength of gallic acid was determined, followed by establishing the standard curves to obtain the calibration curve and the linear regression equation  $y = ax+by = ax+b$  for gallic acid. For the total phenolic assay, a 0.5 mL sample was pipetted into test tubes, and an additional 1.25 mL was added, then shaken for 5 minutes. Subsequently, 1 mL of sodium bicarbonate was added and the mixture was shaken again. The sample was then incubated, and its absorbance was measured using a visible spectrophotometer at the determined wavelength from the blank's maximum wavelength.

Serial dilutions of gallic acid were used as standards, and the total phenolic content was expressed in terms of mg equivalents of gallic acid (mg GAE) [9].

## 2.5 Determination of total flavonoid content

For the total flavonoid content assay, 0.5 mL extract was transferred into a 10 mL volumetric flask and added with 0.1 mL aluminum chloride 10% (v/v), 0.1 mL potassium acetate, and the volume was adjusted with distilled water. The incubation was conducted at 25 °C for 30 minutes and absorbance was determined using maximum wavelength of the blank using a UV-Vis spectrophotometer [9].

## 2.6 Determination of anthocyanin content

The anthocyanin content of Chrysanthemum tea powder was measured with the pH differential methods [10]. The Chrysanthemum tea was mixed thoroughly with 20 mL buffer pH 1.0 (0.025 M potassium chloride), and pH 4.5 (0.4 M sodium acetate buffer). Incubation was conducted for 20 minutes at room temperature and filtered. The absorbance of the filtrate was read at 510 and 700 nm, and the anthocyanin content of Chrysanthemum tea powder was calculated by using the following equation:

$$\text{Total Anthocyanin} = \frac{A \times V}{M} \quad (1)$$

where A = (A<sub>520 nm</sub> – A<sub>700 nm</sub>) pH 1.0 – (A<sub>520 nm</sub> – A<sub>700 nm</sub>) pH 4.5

V = extract volume (mL), M = fresh mass of tea sample (g)

## 2.7 Organoleptic test

Organoleptic testing in this study was conducted using non-standard panellists, who had not been trained in carrying out organoleptic assessment and testing activities. The hedonic test, used to assess and measure product liking, was based on a rating sheet [11]. The panelists consisted of 70 people who were students of the Food Technology Study Program, Faculty of Agriculture, Universitas Sumatera Utara. The number of favorability levels depends on the specified quality range. was conducted using numerical values, which were then statistically analyzed. Organoleptic data in this study consisted of taste and aroma expressed on a numerical scale (1=strongly dislike, 2=dislike, 3=somewhat dislike, 4=neutral, 5=somewhat like, 6=like, and 7=strongly like).

## 2.8 Statistical analysis

Optimization of ready-to-drink Chrysanthemum tea beverage formulation in bottle packaging was conducted using the Design Expert v.13 Response Surface Methodology (RSM) Box-Behnken Design (BBD) program.

## 3 Results and discussion

The effect of the Chrysanthemum powder weight, brewing water temperature, and sugar concentration on total phenolic, total flavonoid, and anthocyanin content of Chrysanthemum RTDT is shown in Table 2.

**Table 2.** Response value of total phenolic content, total flavonoid content and anthocyanin content, and organoleptic value of taste and aroma

Run	B (g)	S (°C)	G (%)	Total Phenolic Content (mg GAE/g)	Total Flavonoid Content (mg QE/g)	Anthocyanin Content (mg/g)	Taste	Aroma
1	1.5	60	7.5	253.84	19.62	3.11	6.11	6.04
2	1	60	0	208.83	18.04	2.96	5.57	5.84
3	2	60	0	319.96	29.06	4.07	5.64	5.83
4	1.5	90	0	401.85	31.33	6.53	5.61	5.83
5	1	90	7.5	304.20	24.01	4.57	5.94	5.97
6	2	60	15	305.68	26.81	4.11	5.94	5.90
7	1.5	60	7.5	323.22	27.30	3.74	5.71	5.90
8	2	90	7.5	735.08	59.97	7.26	5.76	5.86
9	1.5	30	15	69.40	7.80	0.49	6.00	5.69
10	2	30	7.5	115.22	10.04	0.71	5.70	5.81
11	1	60	15	138.75	18.55	1.69	5.96	5.99
12	1.5	60	7.5	237.90	21.38	2.67	5.86	6.00
13	1.5	90	15	338.06	28.31	4.63	6.07	6.06
14	1.5	60	7.5	337.76	21.00	2.09	5.86	5.90
15	1	30	7.5	32.95	10.01	1.13	5.71	5.87
16	1.5	60	7.5	266.90	20.42	1.04	5.97	5.97
17	1.5	30	0	42.76	7.05	0.38	5.76	5.80

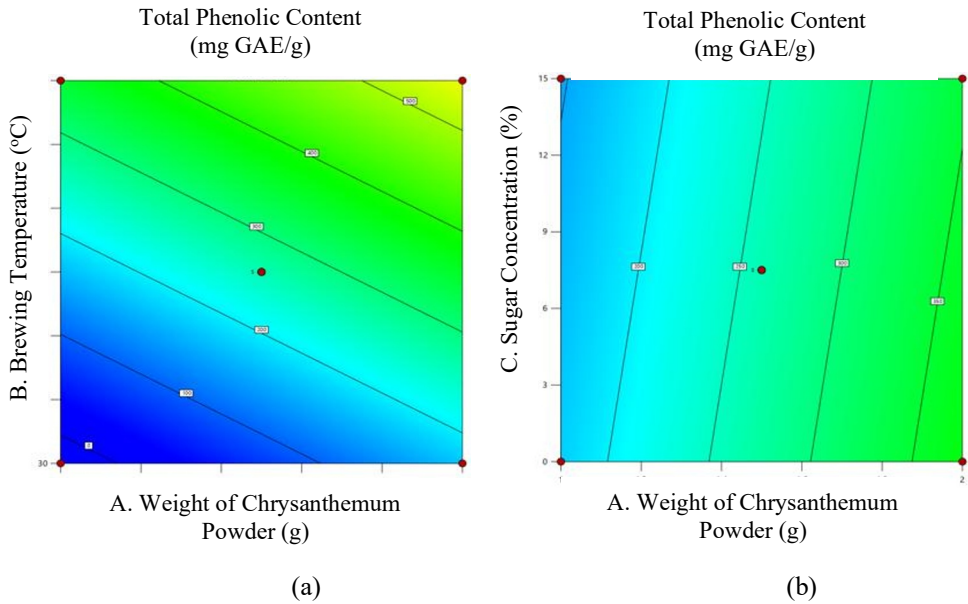
B=weight of Chrysanthemum powder in tea bags, S=temperature of brewing water, G=sugar concentration

### 3.1 Total phenolic content response

The total phenolic content in Chrysanthemum tea drinks ranged from 32.95 to 735.08 mg GAE/g. The contour plot in Figure 1(a) shows that the weight of the tea powder and the temperature of the brewing water have increased and significantly influence the total phenolic content value. However, the contour plot Figure 1(b) shows that sugar concentration has a decreased and nonsignificant influence on the total phenolic value. According to Christina et al. [12], the use of high temperatures will cause the total phenol content to be higher because high temperatures can increase the release of phenolic compounds on the cell wall. This finding highlights the importance of optimizing these parameters to maximize the extraction of phenolic compounds. Phenolic compounds are well-documented for their antioxidant properties, which contribute to various health benefits such as reducing oxidative stress and lowering the risk of chronic diseases. Therefore, enhancing their extraction aligns with the consumer demand for functional beverages that support health and well-being.

This result is significant, as it provides practical guidance for consumers and the beverage industry to optimize brewing conditions for functional and health-focused product formulations. It also emphasizes the potential of Chrysanthemum tea as a high-antioxidant beverage when prepared under specific conditions.

Conversely, the results in Figure 1(b) indicate that sugar concentration has a decreasing and nonsignificant effect on the total phenolic content. This finding suggests that while sugar can enhance sensory appeal, it does not substantially interfere with the phenolic compound extraction. This is significant for the beverage industry, as it highlights that sweetness can be adjusted to meet consumer preferences without compromising the functional benefits of the tea.

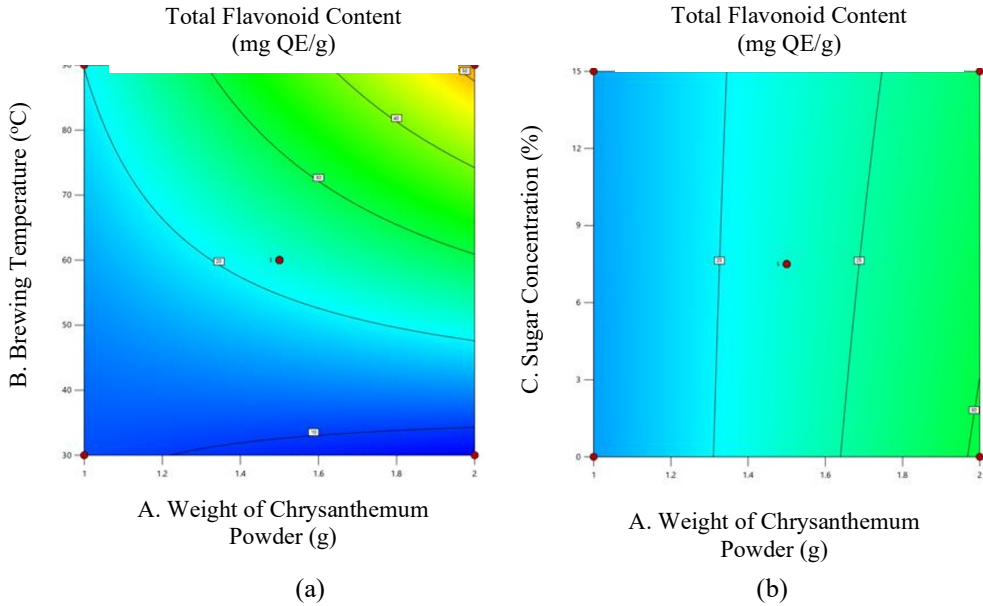


**Fig. 1.** (a) Contour plot of Chrysanthemum flower powder weight and brewing water temperature to total phenol content response. (b) Contour plot of Chrysanthemum flower powder weight and sugar concentration to total phenol content response

### 3.2 Total flavonoid content response

The total flavonoid content in the Chrysanthemum tea beverages ranged from 7.05 to 59.97 mg QE/g, with variations significantly influenced by brewing parameters, particularly the weight of tea powder and brewing water temperature (Figure 2(a)). This aligns with prior studies by Antony and Farid [13], which emphasize that temperature plays a crucial role in enhancing the extraction of flavonoids. These bioactive compounds are known for their antioxidant, anti-inflammatory, and anti-carcinogenic properties, making them a vital component of functional beverages targeting health-conscious consumers. The observation that higher brewing temperatures significantly increase flavonoid content can be attributed to their effect on the cell walls and intracellular matrices of plant material. Higher temperatures likely facilitate the breakdown of cell structures, enhancing the release of bound flavonoids into the brewing medium.

Figure 2(b) shows that sugar concentration had a decreased and nonsignificant impact on flavonoid content. This result indicates that the addition of sugar, while important for flavor and consumer acceptability, does not interfere with the extraction or stability of flavonoids. This is significant for product development in the beverage industry, as it allows for flexibility in adjusting sweetness levels without compromising the functional value of the tea. The sensitivity of flavonoids to temperature, as noted by Antony and Farid [13], also highlights a potential challenge in balancing optimal extraction with the preservation of sensory properties. Excessively high temperatures could risk over-extraction of certain compounds, potentially leading to undesirable bitterness or astringency. This underscores the need for fine-tuning brewing parameters to strike a balance between functionality and sensory acceptance.

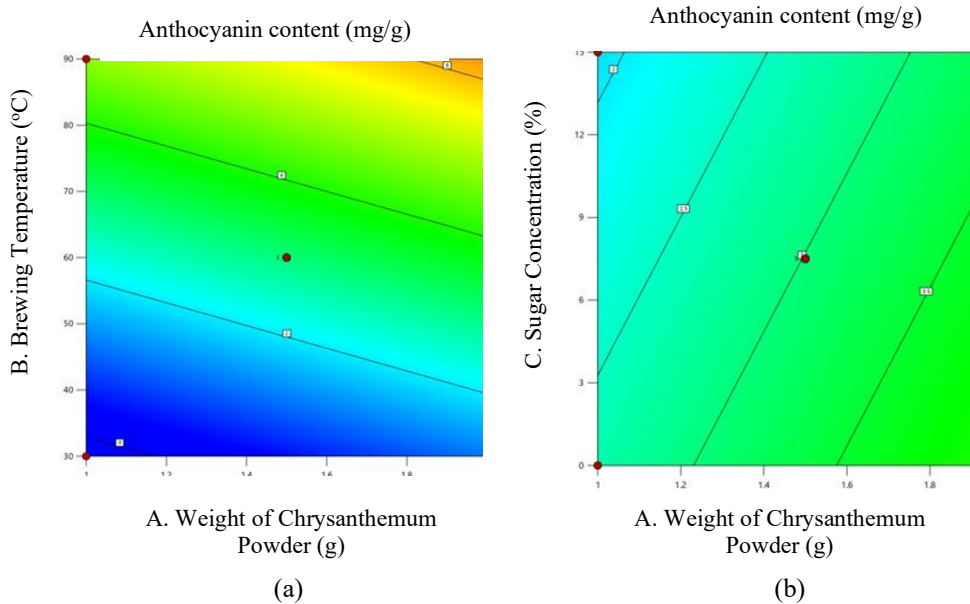


**Fig. 2.** (a) Contour plot of Chrysanthemum flower powder weight and brewing water temperature to total flavonoid content response. (b) Contour plot of Chrysanthemum flower powder weight and sugar concentration to total flavonoid content response

### 3.3 Anthocyanin content response

The anthocyanin content in Chrysanthemum tea beverages ranged from 0.38 to 7.26 mg/g, with brewing parameters playing a significant role in determining the observed values. As depicted in the contour plot (Figure 3(a)), the weight of tea powder and the temperature of the brewing water showed a strong and positive influence on anthocyanin extraction. This finding corroborates the observations of Sahin [15], who reported that higher water temperatures enhance the solubility and release of anthocyanins from plant matrices. This phenomenon occurs because elevated temperatures break down cellular structures, making anthocyanins more readily available for extraction into the liquid medium. By optimizing brewing parameters, manufacturers can maximize anthocyanin content in ready-to-drink Chrysanthemum tea, thus aligning the product with consumer preferences for health-enhancing functional beverages.

Figure 3(b) shows that sugar concentration demonstrated a decreased and nonsignificant influence on anthocyanin content. This suggests that sugar does not interfere with the extraction or stability of anthocyanins during the brewing process, providing flexibility for product formulations to cater to varying sweetness preferences without compromising functional benefits. This finding is advantageous for manufacturers seeking to develop low-sugar or sugar-free alternatives to meet the demands of health-conscious consumers. However, the sensitivity of anthocyanins to environmental factors, such as light and temperature during storage, emphasizes the importance of careful packaging and processing to preserve these compounds [15].



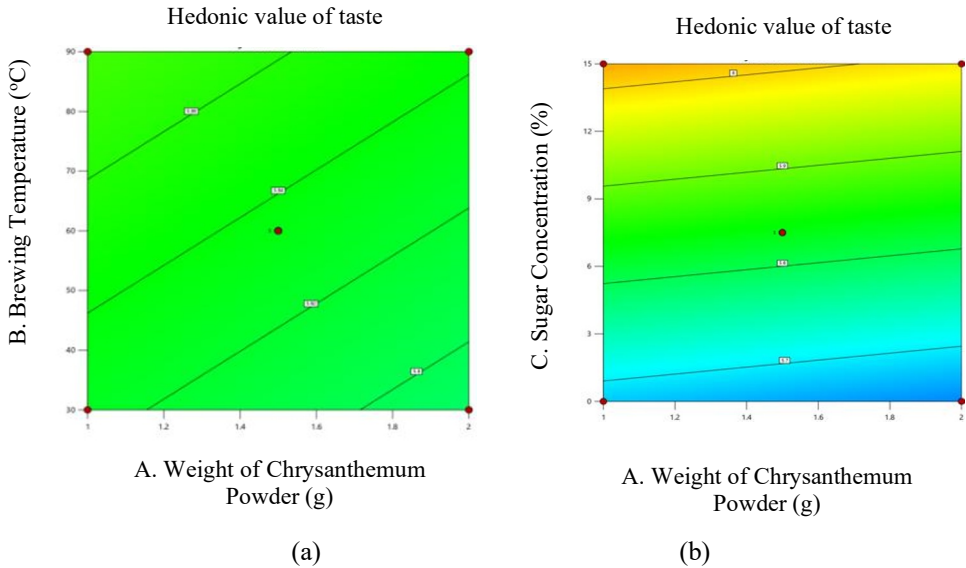
**Fig. 3.** (a) Contour plot of Chrysanthemum flower powder weight and brewing water temperature to anthocyanin content response. (b) Contour plot of Chrysanthemum flower powder weight and sugar concentration to anthocyanin content response

### 3.4 Hedonic organoleptic response (taste)

The sensory taste test results for Chrysanthemum tea ranged from 5.57 to 6.11, indicating a generally favorable response across the different formulations. The contour plot in Figure 4(a) illustrates that the weight of tea powder had a decreased but nonsignificant influence on taste scores. This suggests that variations in tea powder concentration within the tested range did not substantially alter the taste perception, likely because the sensory profile of the tea remains balanced despite changes in the concentration of tea compounds.

The temperature of the brewing water exhibited an increased but nonsignificant influence on taste scores. Although higher temperatures can enhance the release of phenolic and flavonoid compounds, contributing to the tea's bitterness or astringency, these effects did not significantly impact the overall taste perception. This implies that brewing temperature variations, within the tested range, are unlikely to disrupt the flavor balance of the tea, offering flexibility in production conditions without compromising sensory appeal.

Sugar concentration had a significant and positive influence on taste scores, as depicted in Figure 4(b). This aligns with the findings of Vickers et al. [16], who reported that sweetness perception directly correlates with sugar concentration. Sweetness is a critical attribute in ready-to-drink beverages, as it significantly enhances consumer acceptance by covering bitterness and improving flavor harmonization. The significant influence of sugar concentration highlights its pivotal role in achieving an optimal sensory balance in Chrysanthemum tea.



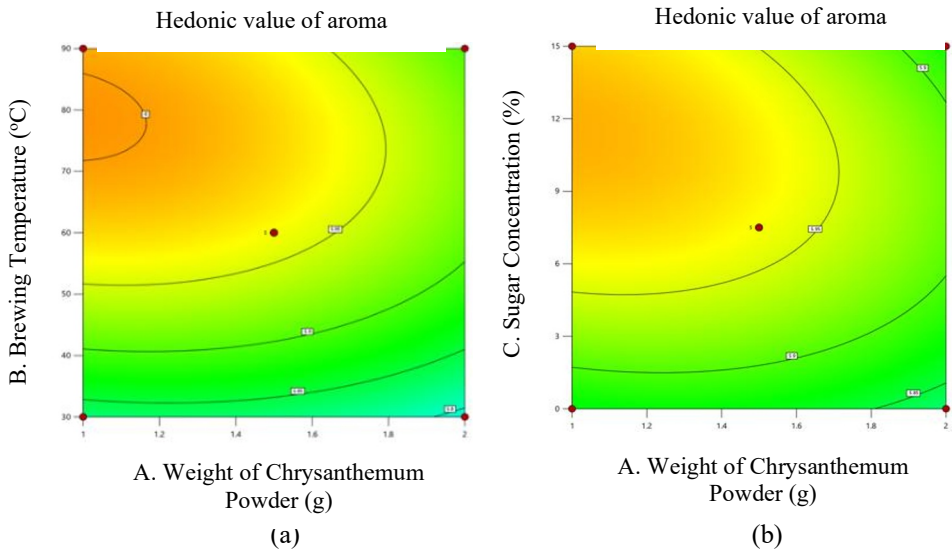
**Fig. 4.** (a) Contour plot of Chrysanthemum flower powder weight and brewing water temperature to hedonic organoleptic response (taste). (b) Contour plot of Chrysanthemum flower powder weight and sugar concentration to hedonic organoleptic response (taste)

### 3.5 Hedonic organoleptic response (aroma)

The sensory aroma test values for Chrysanthemum tea ranged from 5.69 to 6.06, indicating a generally favorable perception of the aroma by the participants. As shown in Figure 5(a), the weight of tea powder had a decreased and nonsignificant influence on the aroma scoring. This suggests that variations in tea powder weight within the tested range did not significantly impact the release or perception of aroma compounds. While the concentration of tea powder might influence the strength of the aroma, it seems that this effect was not large enough to reach statistical significance, possibly because the aroma profile of Chrysanthemum tea is inherently strong and not heavily dependent on powder quantity at the levels tested.

The temperature of the brewing water had a significant and increased influence on the aroma scoring. This result aligns with the findings of Fibrianto et al. [17], who noted that higher brewing temperatures lead to increased release of volatile aroma compounds. Hot water is more effective at extracting the essential oils and other volatile compounds responsible for the characteristic floral and aromatic notes of Chrysanthemum tea. The significant effect of water temperature on aroma indicates that consumers may perceive a more intense and pleasing fragrance when the tea is brewed at higher temperatures, which enhances the sensory appeal of the product.

The contour plot in Figure 5(b) indicates that sugar concentration had an increased but nonsignificant influence on aroma scores. This finding suggests that while the addition of sugar might slightly alter the aroma profile by interacting with certain compounds, it does not significantly affect the overall aroma perception. The lack of significance of this factor could be attributed to the fact that sugar primarily affects the sweetness and taste of the tea, rather than its aromatic compounds. Therefore, sugar concentration may not play a significant role in shaping the aroma experience for consumers.



**Fig. 5.** (a) Contour plot of Chrysanthemum flower powder weight and brewing water temperature to hedonic organoleptic response (aroma). (b) Contour plot of Chrysanthemum flower powder weight and sugar concentration to hedonic organoleptic response (aroma)

### 3.6 Optimization of Chrysanthemum flower tea

Based on the results of factor optimization and the experimental responses, a new formula was obtained with different levels of desirability. The formula with the highest desirability, closest to 1, was selected for antioxidant activity testing. The optimization results that have been carried out produce a new formula with a weight value of 2 g of Chrysanthemum flower powder, a brewing water temperature of 90°C, and a sugar concentration of 14.281% with a desirability value of 0.749.

### 3.7 Antioxidant activity of ready-to-drink Chrysanthemum tea

Optimization of the ready-to-drink Chrysanthemum flower tea formulation in new bottles was then tested for its antioxidant activity. This test was conducted using the DPPH method. The average  $IC_{50}$  value obtained from testing the antioxidant activity of this new formula is 68.31. This result indicates that the sample has a strong antioxidant activity value [18].

## 4 Conclusion

The optimization of the response variables resulted in a new ready-to-drink Chrysanthemum tea formulation with a weight of 2 g of Chrysanthemum flower tea powder, brewing water temperature of 90°C, and sugar concentration of 14.281%. This formulation, which achieved a desirability level of 0.749, was validated through Response Surface Methodology-Box Behnken Design (RSM-BBD) based on its physicochemical and sensory characteristics. The optimized formulation does not only demonstrate significant improvements in bioactive compound content but also aligns with current consumer trends toward health-conscious and convenient beverage options. Given its appealing sensory attributes, including taste and aroma, as well as its functional benefits (e.g., antioxidant properties), this product shows strong potential for commercialization in the health-oriented market. Furthermore, its cost-

effectiveness, through the optimized use of ingredients and production conditions, enhances its suitability for large-scale production, making it a promising candidate for the ready-to-market beverage industry.

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