

## Optimizing Konjac Glucomannan Hydrolysate (KGMH) as a Natural Binder in Foam Mat Drying for Functional Herbal Powder Drink

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### ABSTRACT

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Phytochemical-rich herbal beverages are typically perishable due to their liquid form. Powder conversion via foam mat drying offers improved shelf stability but requires effective natural binders. Konjac glucomannan (KGM) is a promising candidate, though its high viscosity necessitates enzymatic hydrolysis to produce functional konjac glucomannan hydrolysate (KGMH). This study investigated konjac glucomannan hydrolysate (KGMH) as a natural binder for foam mat drying of a herbal blend (Java tea, turmeric, and seed-under-leaf). The research optimized KGMH functionality by evaluating KGM source (Thailand vs. Indonesia), enzyme concentration (100–200 IU/g), and KGM concentration (10–30% w/v) on KGMH properties (viscosity, reducing sugar, total carbohydrate, and degree of polymerization (DP)), herbal drink powder characteristics (physical, phenolic, and flavonoid content, also antioxidant activity using DPPH and FRAP methods). The herbal powder was produced by mixing the herbal extract with KGMH to form a stable foam, followed by drying at 60°C for 12 hours and grinding into powder. Results demonstrated that Enzymatic hydrolysis significantly reduced KGMH viscosity and DP, while increasing reducing sugars, with Indonesian konjac yielding higher viscosity than Thai konjac, and higher KGM concentrations increasing viscosity but requiring greater enzyme levels for effective depolymerization. The herbal drink using binder from Thai KGMH at 200 IU/g enzyme and 20% KGM concentration produced the highest retention of total phenolic content (2.83 mg GAE/g), total flavonoids (3266.52 mg QE/g), and antioxidant activity (93.53 mM TE/g by FRAP) in the herbal drink powder.

**Keywords:** Konjac glucomannan, foam mat drying, binder, antioxidant, functional foods, mannanase enzyme.

### 1. INTRODUCTION

Herbal beverages, derived from plants such as turmeric, Java tea, and seed-under-leaf, are rich in phytochemicals, particularly phenolic compounds and flavonoids, which exhibit potent antioxidant activity beneficial for human health [1]. However, their liquid form poses significant challenges, including limited shelf life due to microbial growth, enzymatic degradation, and oxidative instability [2]. Converting

these herbal extracts into powder form through dehydration techniques offers a viable solution to enhance stability, ease of storage, and transportation. Among drying methods, foam mat drying stands out due to its efficiency in preserving heat-sensitive bioactive compounds, rapid drying kinetics, and scalability. This technique involves whipping the liquid extract into stable foam before drying, which increases the surface area and accelerates moisture removal. However, the process requires binders to

stabilize the foam structure and maintain powder integrity during drying and storage [3].

Konjac glucomannan (KGM), a natural polysaccharide derived from *Amorphophallus konjac* tubers, has emerged as a promising binder due to its unique physicochemical properties, including high water-binding capacity, film-forming ability, and biocompatibility [4]. As a soluble dietary fiber, KGM also offers additional health benefits, such as cholesterol reduction and prebiotic effects, aligning with consumer demand for functional food ingredients. However, native KGM's high viscosity and poor solubility limit its direct application in foam mat drying[5], [6], [7]. Enzymatic hydrolysis using mannanase offers a safe and food-grade solution to reduce KGM's molecular weight, yielding konjac glucomannan hydrolysate (KGMH) with improved functionality while retaining its dietary fiber properties. Unlike chemical hydrolysis, enzymatic treatment avoids toxic residues and preserves the nutritional integrity of the final product, making it ideal for food applications[8], [9].

The efficiency of enzymatic hydrolysis is highly dependent on critical parameters, including the konjac source (which differs in glucomannan purity and molecular structure), enzyme concentration (mannanase activity), and substrate concentration. These factors collectively influence the degree of polymerization, solubility, and binding capacity of KGMH, which in turn affect foam stability, powder reconstitution, and bioactive retention. For instance, insufficient hydrolysis may result in inadequate foam expansion, while excessive hydrolysis could compromise the powder's mechanical strength [5], [8], [10]. Despite KGM's potential, systematic studies optimizing these parameters for herbal powder production are scarce.

This study aims to bridge this gap by optimizing the enzymatic hydrolysis of KGM for foam mat drying of a multicomponent herbal blend (turmeric, Java tea, and seed-under-leaf). We evaluate the effects of konjac source (Thailand vs Indonesia), enzyme concentration (100–200 IU/g), and konjac concentration (10–30% w/v) on KGMH functionality, foam properties, and the final powder's physicochemical and antioxidant characteristics. The findings will provide actionable insights for the food industry to produce shelf-stable, nutrient-dense herbal powders using a natural, health-promoting binder, thereby meeting the growing demand for clean-label functional foods.

## 2. MATERIALS AND METHODS

### 2.1 Materials

The materials required to produce herbal powder using the foam mat drying method included konjac glucomannan (food grade) (it was purchased from *Chemipan*, Thailand and the local market in Bantul, Yogyakarta, Indonesia) as the primary binder, along with three medicinal herbs: Java tea (*Orthosiphon aristatus*), turmeric (*Curcuma longa*), and seed-under leaf (*Phyllanthus niruri*), from WKT Tegal, Indonesia. Fresh egg white was utilized as a natural foaming agent, while carboxymethyl cellulose (CMC) served as a foam stabilizer, and deionized water was used as the solvent throughout the process. For the analytical procedures, the following reagents were employed: deionized water, 98% ethanol for extraction purposes, sodium chloride (NaCl), aluminum chloride (AlCl<sub>3</sub>) for flavonoid quantification, sodium acetate (CH<sub>3</sub>COONa), DPPH (2,2-diphenyl-1-picrylhydrazyl) for antioxidant capacity assessment, acetate buffer (CH<sub>3</sub>COONa buffer, pH 3.6), TPTZ (2,4,6-tripyridyl-s-triazine) for the FRAP assay, hydrochloric acid (HCl), and iron(III) chloride (FeCl<sub>3</sub>). All chemicals used were of analytical grade and obtained from Sigma-Aldrich and Merck.

### 2.1 Enzymatic Hydrolysis of Konjac Glucomannan

The hydrolysis process was initiated by heating distilled water to 70°C in a water bath. Mannanase enzyme was then added according to the experimental variables (100-200 IU/g KGM) with constant stirring for 15 minutes. Konjac glucomannan powder was gradually incorporated into the solution using a spoon, followed by continuous stirring for 30 minutes to ensure complete hydrolysis [5]. The final concentration of konjac was added according to the variable (10, 20, and 30% (w/v)). The enzymatic reaction was terminated by boiling the mixture for 15 minutes. The resulting konjac glucomannan hydrolysate (KGMH) was cooled to room temperature[5], [8].

### 2.2 Preparation of Herbal Powder via Foam Mat Drying

A herbal blend was prepared by combining Java tea (40%), turmeric (5%), and seed-under-leaf (55%) in a 1:20 (w/v) ratio with distilled water [11]. The mixture was boiled for 15 minutes and filtered through muslin cloth to obtain a clear extract. For formation, 50 g of the herbal extract was mixed with 50 g of KGMH and homogenized, followed by the addition of 0.25 g CMC.

Foaming was achieved by incorporating 10 g of fresh egg white and whipping the mixture at 20,000 rpm for 5 minutes using an Ultra-Turrax homogenizer, with careful attention to homogenizer positioning for uniform foam formation. The foam was then spread uniformly (0.8 mm thickness) on drying trays and dehydrated at 60°C for 12 hours. The dried product was ground (particle size <150 µm), packaged in airtight bags, and stored in desiccators prior to analysis [7], [12].

### 2.3 Characterization of Konjac glucomannan hydrolysate (KGMH)

#### 2.3.1 Analysis of the viscosity of KGMH solution

The viscosity of 1% KGMH solutions was measured using a rotational viscometer (Brookfield DV-II+ Pro) equipped with an LV-3 spindle at 28±0.5°C. Before measurement, samples were prepared by dissolving KGMH powder in distilled water under gentle stirring at 60°C for 2 hours, followed by cooling to room temperature and degassing to remove air bubbles. The viscometer was calibrated with standard silicone oil, and measurements were conducted at 100 rpm shear rate, with readings taken after 60 seconds of stabilization to ensure equilibrium. Three replicate measurements were performed for each sample, and the viscosity was recorded in centistokes (cSt),

#### 2.3.2 Analytical Methods for Carbohydrate Characterization of KGMH (Reducing Sugar, Total Carbohydrate, and Degree of Polymerization (DP))

Reducing sugar content was determined using the 3,5-dinitrosalicylic acid (DNS) colorimetric method. A standard calibration curve was first prepared using glucose solutions (0-100 µg/mL) in distilled water. For sample analysis, 2 mL of the test solution was mixed with 2 mL of DNS reagent in a test tube. The mixture was heated in boiling water for exactly 10 minutes to develop the red-brown color characteristic of reduced DNS. After immediate cooling in an ice-water bath, the absorbance was measured at 570 nm using a UV-Vis spectrophotometer. The reducing sugar concentration was calculated from the standard curve and expressed as glucose equivalents (mg/g)[8], [13].

Total carbohydrate content was quantified through the phenol-sulfuric acid assay. A reference standard curve was generated using serial dilutions of glucose (0-100 µg/mL) in distilled water. In the

analytical procedure, 1 mL of sample solution was combined with 1 mL of 5% (w/w) aqueous phenol solution, followed by rapid addition of 5 mL concentrated sulfuric acid. After 20 minutes of incubation at room temperature (25±1°C), the characteristic yellow-orange color was measured at 490 nm. Results were calculated based on the standard curve and reported as total carbohydrate content in glucose equivalents (mg/g)[14].

The degree of polymerization was mathematically determined as the ratio of total carbohydrate content to reducing sugar content:  $DP = (\text{Total Carbohydrate content}) / (\text{Reducing Sugar content})$ . This dimensionless parameter provides insight into the average chain length of polysaccharides in the sample, where higher values indicate greater polymerization[10].

### 2.4 Physical Characterization of Herbal Drink Powder

The powder's physical properties were evaluated through three key parameters: moisture content, hygroscopicity, and water solubility index. Moisture content was determined following the AOAC standard method [15], where samples were dehydrated in a precision oven at 105 ± 1°C until constant weight was achieved.

Hygroscopicity assessment was conducted using an adapted Cai and Corke protocol [16], [17]. Approximately 1 g of powder was exposed to 75% relative humidity (maintained by NaCl saturated solution) in a sealed desiccator at 25 ± 1°C for seven days. The moisture absorption capacity was calculated from the weight difference and expressed as grams of absorbed moisture per 100 grams of dry powder.

Water solubility was quantified using the water solubility index method [17], [18]. This involved measuring the soluble fraction of the powder after aqueous dispersion and centrifugation, providing insight into the product's reconstitution properties. All measurements were performed in triplicate to ensure analytical precision.

### 2.5 Analysis of Total Phenolic Compounds (TPC) and Total Flavonoid Content (TFC)

The total phenolic content was quantified using the Folin-Ciocalteu method[11], [19]. Briefly, 1000 µL of each test sample and standard solution were pipetted into separate test tubes. Then, 5000 µL of

7.5% Folin-Ciocalteu reagent was added to each tube and allowed to react for 8 minutes at room temperature. Subsequently, 4000  $\mu$ L of 1% sodium hydroxide (NaOH) solution was added, and the mixture was incubated for 1 hour in the dark. The absorbance of the resulting, blue-colored complex was measured at 730 nm using a UV-Vis spectrophotometer. Gallic acid was used as the standard reference, and results were expressed as mg gallic acid equivalents (GAE) per gram of sample[11], [19].

The total flavonoid content was analyzed using the aluminum chloride colorimetric method[18]. In this procedure, 1000  $\mu$ L of sample or standard solution was mixed with 1500  $\mu$ L of ethanol, followed by the addition of 100  $\mu$ L of 10% aluminum chloride ( $\text{AlCl}_3$ ) and 100  $\mu$ L of 1M sodium acetate ( $\text{CH}_3\text{COONa}$ ). The mixture was then diluted with 2800  $\mu$ L of distilled water and vortexed thoroughly. After 30 minutes of incubation at room temperature in the dark, the absorbance of the yellow-colored complex was measured at 370 nm. Quercetin was used as the standard reference, and results were expressed as mg quercetin equivalents (QE) per gram of sample [20], [21].

## 2.6 Methods for Antioxidant Activity Analysis using DPPH Radical Scavenging Assay and Ferric Reducing Antioxidant Power (FRAP) Assay

The antioxidant capacity was evaluated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging method with modifications[11], [22]. A 0.1 mM DPPH solution was prepared in methanol and stored protected from light. For analysis, 100  $\mu$ L of sample extract (1 mg/mL in methanol) was mixed with 3.9 mL of DPPH solution and incubated in the dark at room temperature for 30 minutes. The absorbance was measured at 517 nm against ethanol blank. A standard curve was prepared using Trolox solutions (0–500  $\mu$ M). The antioxidant activity was expressed as micromolar Trolox equivalents per gram of dry weight powder ( $\mu$ M TE/g DW).

The FRAP assay was performed according to the standard method using FRAP reagents [21], [22], [23]. The FRAP reagent was prepared by mixing 300 mM acetate buffer (pH 3.6), 10 mM TPTZ (2,4,6-tripyridyl-s-triazine) in 40 mM HCl, and 20 mM  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  in a 10:1:1 ratio. Fresh FRAP reagent (3 mL) was mixed with 100  $\mu$ L of sample extract and incubated at 37°C for 30 minutes. The absorbance was read at 593 nm against a reagent blank. A standard curve was

generated using  $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$  solutions (0–1000  $\mu$ M). Results were expressed as micromolar Trolox equivalents per gram of dry weight powder ( $\mu$ M TE/g DW).

## 2.7 Data Analysis

The data were analyzed using three-way ANOVA to evaluate the combined effects of konjac source (Thailand vs Indonesia), enzyme concentration (100 IU vs 200 IU), and konjac concentration (10%, 20%, 30%) on all response variables, including hydrolysis characteristics and powder quality parameters. For significant factors identified by ANOVA ( $p < 0.05$ ), post-hoc Duncan's HSD tests were performed to determine specific differences between treatment groups. All statistical analyses were performed using SPSS 17 with a designated significance level of  $p < 0.05$ , and all experiments were conducted in triplicate to ensure data reliability.

# 2 RESULTS AND DISCUSSION

## 3.1 Characterization of KGMH

### 3.1.1 The Viscosity of KGMH Solution.

The viscosity of konjac glucomannan hydrolysate (KGMH) was significantly influenced by konjac source, enzyme concentration, and konjac concentration ( $p < 0.05$ ) (Table 1). Thai konjac produced lower viscosity solutions (809–3,829 cStokes at 100 IU) compared to Indonesian konjac (3,367–33,520 cStokes), likely due to differences in molecular weight and purity[9]. Increasing enzyme concentration from 100 IU to 200 IU drastically reduced viscosity, particularly for Indonesian konjac (>99% reduction at 10% concentration). Higher konjac concentrations (10% to 30%) exponentially increased viscosity, especially in Indonesian samples (100-fold increase at 100 IU).

The optimal conditions for foam mat drying were achieved with 200 IU enzyme and 10% Thai konjac, yielding a viscosity of 66.5 cStokes—ideal for foam formation without excessive thickness. In contrast, Indonesian konjac at 100 IU resulted in impractical viscosities (>33,000 cStokes), emphasizing proper enzyme dosing and raw material selection. These findings demonstrate that enzymatic hydrolysis effectively tailors KGMH functionality for food processing, balancing binding capacity and processability [4].

### 3.1.2 Reducing sugar, Total Carbohydrate, and Degree of Polymerization (DP) of KGMH

The analysis of reducing sugar, total carbohydrate, and degree of polymerization (DP) of KGMH produced from Indonesian konjac revealed significant variations depending on enzyme concentration (mannanase: 100 and 200 IU/g) and konjac concentration (10%, 20%, and 30%) (**Table 2**). Native konjac glucomannan (KGM) had the lowest reducing sugar content and highest DP (1.82), confirming its high molecular weight and minimal hydrolysis. Enzymatic treatment increased reducing sugar levels, with the highest value observed in sample B 200-20, indicating extensive hydrolysis at 200 IU/g enzyme and 20% konjac. Total carbohydrate content remained relatively stable (67.90–93.15 mg/g), except for sample B 200-20, which showed a slight reduction, possibly due to partial degradation into smaller sugars. The DP decreased significantly with hydrolysis, reaching the lowest value in sample B

200-20 (0.29), confirming effective depolymerization at higher enzyme levels[24].

The increase in reducing sugar content with higher enzyme concentration (200 IU/g) demonstrates that mannanase effectively breaks down KGM into smaller oligosaccharides. The highest reducing sugar in sample B 200-20 suggests that 20% konjac concentration optimizes enzymatic hydrolysis, whereas further increases (e.g., 30%) may hinder enzyme accessibility. The stable total carbohydrate content in most samples implies that hydrolysis primarily affects molecular size rather than total sugar content. The drastic reduction in DP (from 1.82 in KGM to 0.29–0.80 in KGMH) confirms successful depolymerization, with sample B 200-20 exhibiting the most significant reduction, likely due to optimal enzyme-substrate interaction. These findings indicate that enzymatic hydrolysis effectively modifies KGM into low-DP KGMH, enhancing solubility and potential functional properties for food applications [8].

**Tabel 1** The viscosity of KGMH and KGM solution 1% (w/v) in different sources of konjac and the concentration of KGM and mannanase enzyme.

Sample code	KGM Source	Enzyme concentration (IU/g konjac)	KGM concentration (%)	Viscosity of (w/v) (CStokes)	KGMH	Viscosity of KGM 1% (w/v) (CStokes)
T 100-10	Thailand	100	10	809.500±0.00 <sup>βAc</sup>	239.60±0.00 <sup>αAc</sup>	
T 100-20			20	1723.00±0.00 <sup>βAb</sup>	505.50±0.00 <sup>αAb</sup>	
T 100-30			30	3829.00±0.00 <sup>βAa</sup>	1106.67±19.03 <sup>αAa</sup>	
T 200-10		200	10	66.53±12.62 <sup>βBc</sup>	207.53±5.15 <sup>αBc</sup>	
T 200-20			20	2252.00±12.00 <sup>βBb</sup>	532.63±0.95 <sup>αBb</sup>	
T 200-30			30	2252.00±32.69 <sup>βB<sup>a</sup></sup>	644.53±2.73 <sup>αB<sup>a</sup></sup>	
B 100-10	Indonesia	100	10	3366.67±152.04 <sup>αAc</sup>	129.60±0.72 <sup>βAc</sup>	
B 100-20			20	20486.67±324,113 <sup>αAb</sup>	239.13±0.61 <sup>βAb</sup>	
B 100-30			30	33520.00±2986.99 <sup>αAa</sup>	478.67±3.60 <sup>βAa</sup>	
B 200-10		200	10	185.50±9.68 <sup>αB<sup>a</sup></sup>	29.84±3.53 <sup>βB<sup>c</sup></sup>	
B 200-20			20	2733.33±142.34 <sup>αB<sup>b</sup></sup>	71.62±0.96 <sup>βB<sup>b</sup></sup>	
B 200-30			30	26130.33±345.72 <sup>αB<sup>c</sup></sup>	252.10±1.22 <sup>βB<sup>a</sup></sup>	

Note: The results above show the mean value ± standard deviation from three replicate measurements. In the post-hoc statistical analysis, Greek letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM source, uppercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the enzyme concentration, and lowercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM concentration."

**Tabel 2** Reducing Sugar, Total Carbohydrate, and Degree of Polymerization (DP) of KGMH of KGM from Indonesia

Sample code	Enzyme concentration (IU/g)	KGM concentration (%)	Reducing Sugar (mg/g sampel)	Total Carbohydrate (mg/g sampel)	Degree of Polymerization (DP)
KGM	0	0	46.64±6.17 <sup>cc</sup>	83.93±9.79 <sup>cd</sup>	1.82±0.29 <sup>Aa</sup>
B 100-20	100	20	114.73±12.03 <sup>Bb</sup>	91.41±2.84 <sup>bc</sup>	0.80±0.06 <sup>cc</sup>
B 200-10		10	165.04±17.13 <sup>Ab</sup>	85.60±15.50 <sup>Ab</sup>	0.52±0.13 <sup>bd</sup>
B 200-20	200	20	236.42±13.84 <sup>Ac</sup>	67.90±11.86 <sup>Ac</sup>	0.29±0.06 <sup>Bc</sup>
B 200-30		30	215.22±16.36 <sup>Aa</sup>	93.15±21.57 <sup>Aa</sup>	0.44±0.14 <sup>Bb</sup>

Note: The data represent mean values ± standard deviation from three replicate experiments. In post hoc analysis, uppercase letters denote significant differences ( $p < 0.05$ ) across enzyme concentrations, while lowercase letters indicate significant differences ( $p < 0.05$ ) across konjac concentrations.

### 3.2 Physical Properties of Herbal Drink Powder using binder of KGM in different enzymatic hydrolysis parameters

The physical characteristics of the herbal powder drink were evaluated based on a blend of turmeric, Java tea, and seed-under-leaf, using hydrolyzed konjac glucomannan (KGMH) as a binder. The KGMH was produced through enzymatic hydrolysis using mannanase at different concentrations (100 and 200 IU/g konjac), with konjac flour sourced from Thailand and Indonesia, and varying konjac concentrations (10%, 20%, and 30%). These processing conditions resulted in different KGMH properties, which were then applied as binders in the herbal powder formulation (Table 2).

The moisture content of the herbal powder varied significantly depending on the konjac source, enzyme concentration, and konjac concentration. Samples with Indonesian konjac generally exhibited higher moisture content compared to those with Thai konjac, particularly at lower enzyme concentrations (100 IU/g). This suggests that the origin of konjac influences water retention in the powder. Additionally, increasing the konjac concentration from 10% to 30% led to a decrease in moisture content, possibly due to the higher molecular interaction of KGMH, reducing free water availability.

Hygroscopicity was significantly affected by the konjac source, with Thai konjac-based samples showing higher values than those from Indonesia. This may be attributed to differences in the polysaccharide structure and residual enzyme activity post-hydrolysis. Higher enzyme concentration (200 IU/g) generally

increased hygroscopicity, likely due to greater depolymerization of KGMH, exposing more hydrophilic groups. However, increasing konjac concentration did not consistently affect hygroscopicity, indicating a complex interaction between hydrolysis degree and binder concentration[18].

Bulk density was higher in samples containing Indonesian konjac, suggesting that the native konjac properties influence particle packing. Enzyme concentration also played a role, as samples hydrolyzed with 200 IU/g exhibited lower bulk density, possibly due to finer particle size from more extensive hydrolysis. Interestingly, konjac concentration had varying effects—higher concentrations (20-30%) sometimes increased bulk density, likely due to improved binding and agglomeration[18].

Solubility was significantly higher in Thai konjac samples, particularly at 200 IU/g enzyme concentration, indicating that hydrolysis efficiency depends on konjac origin. The increased solubility at higher enzyme levels suggests that more extensive hydrolysis enhances KGMH dispersibility. However, increasing konjac concentration (especially to 30%) reduced solubility in some cases, possibly due to excessive cross-linking or incomplete solubilization of higher-molecular-weight fractions[25].

**Table 3.** Physical properties of herbal drink powder with binder of KGMH at different sources and concentrations of enzyme and KGM.

Sample code	Hydrolysis parameters			Physical Properties of Herbal Drink Powder			
	KGM Source	Enzyme conc. (IU/g)	KGM Concentration (w/v)	Moisture Content (%)	Hygroscopicity (g/100 g)	Bulk Density (gr/cm <sup>3</sup> )	Solubility (%)
T 100-10	Thailand	100	10	5.95±0.31 <sup>aAa</sup>	15.83±0.48 <sup>aBc</sup>	0.74±0.03 <sup>βAb</sup>	75.52±3.35 <sup>αBa</sup>
T 100-20			20	5.79±0.51 <sup>aAb</sup>	17.34±0.46 <sup>aBa</sup>	0.68±0.03 <sup>βAc</sup>	89.67±2.91 <sup>αBb</sup>
T 100-30			30	5.25±1.05 <sup>aAc</sup>	18.09±0.28 <sup>aBb</sup>	0.74±0.02 <sup>βAa</sup>	96.04±10.35 <sup>αBc</sup>
T 200-10		200	10	5.45±0.30 <sup>αBa</sup>	18.97±0.27 <sup>αAc</sup>	0.80±0.04 <sup>βBb</sup>	118.85±3.99 <sup>αAa</sup>
T 200-20		200	20	4.63±0.39 <sup>αBb</sup>	21.64±0.65 <sup>αAa</sup>	0.72±0.02 <sup>βBc</sup>	92.77±15.30 <sup>αAb</sup>
T 200-30		200	30	4.78±0.04 <sup>αBc</sup>	19.45±0.13 <sup>αAb</sup>	0.73±0.06 <sup>βBa</sup>	108.85±2.10 <sup>αAc</sup>
B 100-10	Indonesia	100	10	6.11±0.24 <sup>αAa</sup>	8.86±0.32 <sup>βBc</sup>	0.91±0.03 <sup>αAb</sup>	65.89±2.81 <sup>βBa</sup>
B 100-20			20	5.20±0.13 <sup>αAb</sup>	9.45±0.19 <sup>βBa</sup>	0.96±0.01 <sup>αAc</sup>	61.48±7.38 <sup>βBb</sup>
B 100-30			30	4.39±0.38 <sup>αAc</sup>	10.31±0.08 <sup>βBb</sup>	0.97±0.02 <sup>αAa</sup>	46.73±1.86 <sup>βBc</sup>
B 200-10		200	10	5.53±0.24 <sup>αBa</sup>	11.51±2.49 <sup>βAc</sup>	0.87±0.04 <sup>βBb</sup>	64.56±2.12 <sup>βAa</sup>
B 200-20		200	20	5.83±0.34 <sup>αBb</sup>	12.19±0.21 <sup>βAa</sup>	0.86±0.02 <sup>βBc</sup>	67.60±9.39 <sup>βAb</sup>
B 200-30		200	30	4.76±0.25 <sup>αBc</sup>	10.81±0.46 <sup>βAb</sup>	0.90±0.02 <sup>αBa</sup>	57.47±2.25 <sup>βAc</sup>

Note: The results above show the mean value ± standard deviation from three replicate measurements. In the post-hoc statistical analysis, Greek letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM source, uppercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the enzyme concentration, and lowercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM concentration.

### 3.3 Total Phenolic Content (TPC) and Total Flavonoid Content (TFC) of Herbal Drink Powder

The results demonstrated that the source of KGM, enzyme concentration, and KGM concentration significantly influenced the total phenolic content (TPC) and total flavonoid content (TFC) of the herbal drink powder (Table 4). Thai KGMH generally yielded higher TPC and TFC values compared to Indonesian KGMH. Higher enzyme concentration (200 IU/g) led to increased TPC and TFC in Thai KGMH samples, while the opposite trend was observed in Indonesian KGMH, suggesting source-dependent enzymatic effects. Additionally, KGM concentration played a role, with 20% KGM often producing the highest TPC and TFC, followed by 10% and 30%, indicating an optimal mid-range concentration for phenolic and flavonoid retention. These variations may be attributed to differences in polysaccharide structure and hydrolysis efficiency between KGM sources, affecting bioactive

compound release. Overall, Thai KGMH at 200 IU/g enzyme and 20% KGM provided the best preservation of antioxidants, highlighting the importance of optimizing binder selection and processing conditions for enhanced functional properties in herbal powders.

The study revealed that konjac glucomannan hydrolysate (KGMH), influenced by its source, enzyme concentration, and KGM concentration, played a crucial role in preserving total phenolic content (TPC) and total flavonoid content (TFC) in the herbal drink powder. The Thai KGMH consistently exhibited higher TPC and TFC values compared to Indonesian KGMH, likely due to differences in molecular structure, degree of polymerization, and hydrolysis efficiency, which may enhance the encapsulation and stabilization of bioactive compounds. Higher enzyme concentration (200 IU/g) improved TPC and TFC in Thai KGMH, suggesting that controlled enzymatic hydrolysis breaks down KGM into smaller, more

soluble fractions that better interact with and protect phenolic and flavonoid compounds. Conversely, the lower performance of Indonesian KGMH at higher enzyme levels may indicate over-hydrolysis, leading to reduced molecular interactions with antioxidants [18], [26].

The optimal KGM concentration (20%) provided the highest TPC and TFC, likely because it formed an effective polysaccharide matrix that physically entrapped and shielded phenolic and flavonoid molecules from degradation, while still allowing

sufficient solubility. Lower concentrations (10%) may have offered insufficient protection, whereas higher concentrations (30%) could have led to excessive viscosity, hindering compound release during analysis. These findings suggest that KGMH acts as a protective carrier, with its hydrolysis-dependent structural properties influencing antioxidant retention. The superior performance of Thai KGMH at moderate enzyme and KGM levels highlights its potential as a functional binder for enhancing the stability of bioactive compounds in herbal powders.

**Table 4.** Total Phenolic Content (TPC) and Total Flavonoid Content (TFC) of herbal drink powder with KGMH as a binder in different sources and concentrations of enzyme and KGM.

Sample code	Hydrolysis parameters			Chemical Properties of Herbal Drink Powder	
	KGM Sources	Enzyme concentrations (IU/g)	KGM Concentrations (%)	TPC (mgGAE/g)	TFC (mgQE/g)
T 100-10	Thailand	100	10	1.83±0.34 <sup>aBb</sup>	1826.60±351.51 <sup>aBb</sup>
T 100-20			20	2.75±0.15 <sup>aBa</sup>	2668.27±216.05 <sup>aBa</sup>
T 100-30			30	1.71±0.14 <sup>aBc</sup>	1702.78±93.70 <sup>aBc</sup>
T 200-10		200	10	2.83±0.18 <sup>aAb</sup>	3266.52±233.34 <sup>aAb</sup>
T 200-20			20	2.34±0.12 <sup>aAa</sup>	2473.54±210.94 <sup>aAa</sup>
T 200-30			30	2.12±0.09 <sup>aAc</sup>	2052.92±153.48 <sup>aAc</sup>
B 100-10	Indonesia	100	10	0.96±0.28 <sup>BBb</sup>	1201.61±434.73 <sup>BBb</sup>
B 100-20			20	0.75±0.19 <sup>BBa</sup>	981.87±62.17 <sup>BBa</sup>
B 100-30			30	0.59±0.07 <sup>BBc</sup>	840.64±155.47 <sup>BBc</sup>
B 200-10		200	10	0.45±0.06 <sup>BAb</sup>	507.31±132.12 <sup>BAb</sup>
B 200-20			20	1.08±0.17 <sup>BAa</sup>	1416.96±101.33 <sup>BAa</sup>
B 200-30			30	1.04±0.08 <sup>BAc</sup>	1641.52±137.43 <sup>BAc</sup>

Note: The results above show the mean value ± standard deviation from three replicate measurements. In the post-hoc statistical analysis, Greek letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM source, uppercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the enzyme concentration, and lowercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM concentration.

### 3.4 Antioxidant Activity of Herbal Drink Powder

The antioxidant activity of the herbal drink powder, as measured by DPPH free radical scavenging and FRAP assays, was significantly influenced by the source of KGMH, enzyme concentration, and KGM concentration (Table 5). Notably, Thai KGMH consistently exhibited superior antioxidant activity compared to Indonesian KGMH. This difference may stem from variations in

the molecular weight distribution, degree of branching, and monosaccharide composition of KGM derived from different sources. Thai KGMH likely possesses a more favorable structural configuration that enhances its ability to encapsulate and stabilize antioxidant compounds (e.g., polyphenols and flavonoids from Java tea, curcuminoids from turmeric, and phenolic acids from seed-under-leaf) during foam-mat drying. The hydrophilic and film-

forming properties of KGMH may have created a protective matrix around these bioactive compounds, reducing their exposure to oxidative degradation and thermal damage during dehydration.

The enzyme concentration also played a critical role in modulating antioxidant retention. Higher enzyme levels (200 IU/g) generally improved antioxidant activity in Thai KGMH samples likely because controlled hydrolysis produced smaller KGM fragments with increased solubility and better interaction with antioxidants. These hydrolyzed fractions may have formed a more cohesive protective network during drying, preventing the loss of volatile and heat-sensitive compounds [27]. Conversely, Indonesian KGMH showed reduced antioxidant activity at higher enzyme concentrations, possibly due to excessive depolymerization, which weakened its structural integrity and ability to entrap antioxidants effectively.

The KGM concentration further influenced antioxidant preservation, with 20% KGM yielding the highest activity in most cases. This suggests that intermediate concentration provides an optimal balance between viscosity and film formation, ensuring sufficient encapsulation without hindering the release of antioxidants during analysis [28]. Lower concentrations (10%) may have offered inadequate protection, while higher concentrations (30%) could have led to excessive gelation, trapping antioxidants too tightly and reducing their measurable activity.

The foam-mat drying process itself likely contributed to the stabilization of antioxidants, as the porous structure created by KGMH may have facilitated rapid moisture removal while minimizing prolonged heat exposure [17], [20]. The interaction between KGMH and herbal bioactives could have also mitigated oxidative reactions by forming hydrogen bonds and hydrophobic associations, thereby preserving their radical-scavenging capacity [29].

**Table 5.** Antioxidant activity using DPPH free radical scavenging and FRAP method of herbal drink powder with KGMH as a binder in different sources and concentrations of enzyme and KGM.

Sample code	Hydrolysis parameters			Antioxidant activity of herbal drink powder	
	KGM Sources	Enzyme concentrations (IU/g)	KGM Concentrations % (w/v)	DPPH free radical scavenging (mMTE/g)	FRAP method (mMTE/g)
T 100-10	Thailand	100	10	31.37±13.99 <sup>aBb</sup>	55.43±11.99 <sup>aBb</sup>
T 100-20			20	46.30±3.47 <sup>aBa</sup>	89.22±4.00 <sup>aBa</sup>
T 100-30			30	34.26±6.27 <sup>aBc</sup>	56.44±4.64 <sup>aBc</sup>
T 200-10		200	10	50.35±10.26 <sup>aAb</sup>	93.53±9.82 <sup>aAb</sup>
T 200-20			20	46.05±11.58 <sup>aAa</sup>	75.71±5.89 <sup>aAa</sup>
T 200-30			30	42.28±12.08 <sup>aAc</sup>	67.29±1.34 <sup>aAc</sup>
B 100-10	Indonesia	100	10	5.48±0.72 <sup>βBb</sup>	25.34±6.27 <sup>βBb</sup>
B 100-20			20	4.15±1.54 <sup>βBa</sup>	20.09±2.95 <sup>βBa</sup>
B 100-30			30	2.83±0.28 <sup>βBc</sup>	14.50±0.97 <sup>βBc</sup>
B 200-10		200	10	3.55±0.21 <sup>βAb</sup>	14.20±1.97 <sup>βAb</sup>
B 200-20			20	8.19±1.23 <sup>βAa</sup>	29.32±4.48 <sup>βAa</sup>
B 200-30			30	2.17±0.18 <sup>βAc</sup>	21.99±2.74 <sup>βAc</sup>

Note: The results above show the mean value ± standard deviation from three replicate measurements. In the post-hoc statistical analysis, Greek letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM source, uppercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the enzyme concentration, and lowercase letters indicate values that are significantly different ( $p < 0.05$ ) based on the KGM concentration.

#### 4 CONCLUSION

This study successfully optimized the enzymatic hydrolysis of konjac glucomannan (KGM) for foam mat drying of a multicomponent herbal blend (turmeric, Java tea, and seed-under-leaf), addressing the need for natural, functional binders in clean-label food production. The findings demonstrated that KGM source, enzyme concentration, and KGM concentration significantly influenced the physicochemical and antioxidant properties of the resulting herbal powder. Thai KGMH, particularly at 200 IU/g enzyme concentration and 20% KGM, exhibited superior performance in preserving total phenolic content (TPC), total flavonoid content (TFC), and antioxidant activity (DPPH and FRAP) compared to Indonesian KGMH. This suggests that variations in KGM molecular structure and hydrolysis efficiency between sources play a critical role in bioactive compound retention.

The enzyme concentration was a key determinant of KGMH functionality, with moderate hydrolysis (200 IU/g) enhancing the protective effects of Thai KGMH by producing optimally sized polysaccharide fragments that effectively encapsulated antioxidants. Conversely, Indonesian KGMH showed reduced efficacy at higher enzyme levels, likely due to excessive breakdown of its polysaccharide matrix. Additionally, KGM concentration had a dose-dependent effect, with 20% KGM providing the best balance between viscosity and bioactive stabilization, ensuring optimal foam formation and drying efficiency while minimizing antioxidant degradation.

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#### CONFLICT OF INTEREST

No potential conflict of interest was reported by the author(s).

#### CREDIT AUTHORSHIP CONTRIBUTION STATEMENT

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