Make a 5 Keyword,The Application of Natural Deep Eutectic Solvents (Betaine-Glycerol) for Isolating Glucomannan from Porang Flour (*Amorphophallus muelleri* Blume)

N Afifaha[1,21], *K* [1*], *E A* Krisanti[1], *W W* Purwanto[1], and *A* Sarifudin[2]

[1]Department of Chemical Engi Muliaa neering, Universitas Indonesia, Depok 16424, Indonesia
[2]Research Center for Appropriate Technology, National Research and Innovation Agency, Subang 41213, Indonesia

Abstract. Porang tuber (Amorphophallus muelleri Blume) contains a high amount of glucomannan, an indigestible dietary fiber. Glucomannan is generally isolated using ethanol. To overcome competition with food and feed in the production of bioethanol, it is proposed to use Natural Deep Eutectic Solvents (NADES) for isolating glucomannan. This study evaluated the application of NADES and ethanol to isolate glucomannan from porang flour. The NADES was prepared by mixing betaine and glycerol in different mole ratios. (1:1, 1:2, and 1:3). The result showed that the obtained glucomannan flour ranged from 74.36% to 77.40%. Results indicated that the glucomannan content of glucomannan flour isolated by NADES was lower than that of ethanol. However, glucomannan flour isolated by NADES showed brighter color than that produced by ethanol. The rheological result indicated that the gel of glucomannan flour (1% w/v in water) exhibited pseudoplastic behavior. NADES (betaine and glycerol with a mole ratio of 1:2) was found to be the most effective formula to isolate glucomannan from porang flour compared to the other mole ratios with glucomannan content, viscosity, and whiteness index of 86.78% (dry base), 17,680 cP, and 75.62, respectively.
Keywords : Natural deep, Betaine -Glycerol, Isolation method,Glocumanan, Porang Flour.

1 Introduction

The polysaccharide glucomannan (GM) is typically isolated from the Amorphophallus tuber. Glucose and mannose serve as the primary and secondary sugars in GM, along with certain acetylated residues and galactose side chains [1]. The mannose-glucose ratio may change based on the glucomannan's original source [2]. Due to its capacity to thicken and create a gel, it is extensively employed in various industries, including the food and pharmaceutical sectors. Additionally, as the need for functional foods has continuously

[1] Corresponding author: syabiljafa2008@gmail.com

grown, GM is regarded as a dietary fiber source [3–5]. The purity of GM has a direct correlation with its function. Therefore, getting GM of high purity is a critical step in the food and medicine industries [2].

Generally, the isolation of GM from Amorphophallus tuber adopted a wet processing method using water, ethanol, isopropyl alcohol, or acid solution as solvents [6–8]. Some studies also reported assistant methods such as ultrasonic and enzymatic to improve the isolation process [8,9]. Among these solvents, ethanol is the most widely used solvent for isolating glucomannan [6,7]. However, increasing environmental concerns make ethanol a promising energy alternative for many countries. Bioethanol is commonly produced from sugar and corn. It triggers the competition between food, feed, and fuel [10,11]. In line with the second global sustainable development goal (SDG) to achieve food security, developing new green solvents is a critical subject, and natural deep eutectic solvents (NADESs) are promising. NADESs attract significant concern due to their favorable physicochemical properties (e.g., little volatility, thermal and chemical stability, nonflammability, and nontoxicity), abundantly present in nature, and bio-renewable [12,13]. They have a substantial benefit over traditional solvents because they can be added directly to food compositions without further purification [14]. NADESs are made up of hydrogen bond donors like amines, carboxylic acids, and polyols as well as hydrogen bond acceptors, which are often quaternary ammonium salts.

The application of NADESs in polysaccharides extraction has been reported, especially in hydrocolloid compounds [15–19]. Das et al. [15] reported that NADES of 10% hydrated choline chloride-glycerol 1:2 resulted in the highest yield when used to extract carrageenan from Kappaphycus alvarezii effectively. Using subcritical water and NADES (ChCl-glycerol), Saravana et al. [16] isolated fucoidan and alginate from seaweed (Saccharina japonica) and generated 14.9% and 28.1%, respectively. In a different work, Nie et al. [17] used NADES-based ChCl:1,2-propanediol to extract polysaccharides with the aid of ultrasonication from seaweed (Sargassum horneri). These investigations demonstrated that different NADES combinations led to varying levels of polysaccharide extraction efficiency, with choline chloride-glycerol exhibiting improved extraction effectiveness when yield was taken into consideration. However, applying NADES for extracting biopolymers, especially GM, are not attempted so far. NADES-based betaine and glycerol is a green solvent that supports the achievement of SDGs that address environmental and economic sustainability. Therefore, this study aimed to evaluate the effect of mole ratio betaine-glycerol on GM isolation from porang flour. The isolation of GM using ethanol as a control treatment was also studied.

2 Materials and method

2.1 Materials

Porang tuber (*Amorphopallus muelleri* Blume) was purchased from a local farmer in Subang (Indonesia), betaine anhydrous (purity of 99%) was obtained from Xi'an Yunchin (China), technical glycerol was procured from a local chemical store in Jakarta (Indonesia). All chemicals for analysis were analytical grade (Merck Chemical).

2.2 Sample preparation

The NADES solutions were produced by mixing betaine anhydrous and glycerol in the mole ratio of 1:1, 1:2, and 1:3. They were heated at 80ºC until a clear solution was obtained. The isolation of GM using the ethanol isolation method was conducted following the procedure of Faridah & Widjanarko [20] with slight modifications, namely, isolation time of 3 h, stirring speed of 450 rpm, and solvent-to-flour ratio of 10 ml/g. Meanwhile, the following procedures were carried out to isolate GM utilizing the NADES isolation method: 100 g of NADES and 10 g of porang flour were blended in a beaker. The mixture was agitated at 450 rpm for two hours. After filtering the resultant mixture, the unfiltered mass was twice washed in ethanol and then dried for four hours in a hot air dryer at 50°C. The dried flour was weighed and calculated as GM flour yield. After centrifuging the suspension solution through the filter cloth for five minutes at a speed of 4,000 rpm, the supernatant was weighed and determined to be a solvent recovery.

2.3 Sample analysis

The yield of GM flour was counted by comparing the weight of the dried mass of unpassed filter cloth to that of the starting porang flour. Meanwhile, solvent recovery was determined as the ratio of obtained solvent after centrifugation to the initial solvent used to isolate GM.

2.3.1. *Chemical properties of glucomannan flour.*

The chemical properties of GM flour were observed, including GM, ash, starch, nitrogen, and ca-oxalate content. The GM content of the sample was measured using a 3,5-DNS colorimetric assay carried out in accordance with Chua et al. [6]. Ash content was calculated in accordance with Association of Official Analytical Chemist guidelines [21] using a hot air oven (Memmert UM500, Germany). The direct acid hydrolysis method was used to calculate the starch content [22]. The Dumas combustion method was used to calculate the protein level with a DuMaster (Buchi D-480, Switzerland). For quick and complete combustion, Samples weighing 0.2 g are placed in tin foil, heated in a hot furnace (950 ºC), and flushed with oxygen. A conversion factor of 5.7 was used to determine the protein content. Ca-oxalate was determined according to Sarifudin et al. [23].

2.3.2. *Physical properties of glucomannan flour.*

The physical properties of GM flour were reported, including color, viscosity, and rheological properties. A chromameter (3NH, China) was used to evaluate the color of the samples. In this investigation, the samples' color properties were noted, namely L-value (lightness), a-value (red-green), and b-value (blue-yellow). Utilizing the Whiteness Index (WI) to convert measured Hunter values, the browning of the GM flour was estimated [24].

$$WI = 100 - \sqrt{(100 - L)^2 + a^2 + b^2} \quad (1)$$

The GM sample solution was made in deionized water at 1.0% (w/v) and allowed to adjust at room temperature for 30 minutes before the viscosity measurement. A rotational viscometer (Brookfield DV-E, USA) was used to test the apparent viscosity of GM sample solutions. The measurement was carried out by varying the spindle S63's rotational speed (4-5 rpm) for each sample in order to maintain the torque range of 60-80%. Thermo

Scientific's HAAKE MARS 40 Rheometer (Thermo Hakee Co. Ltd., Germany) was used to assess the viscosity stability at a constant temperature of 25 °C and shear rate increased. The measurements were made using parallel plates with a 1 mm spacing and a 35 mm diameter [25]. The relation between shear rate and shear stress was considered using the Herschel-Bulkley model (equation 2)

$$\tau = \tau_0 + K(\gamma)^n \quad (2)$$

Where τ is shear stress (Pa), τ_0 is yield stress (Pa), K is consistency index (Pa.s), γ is shear rate (s^{-1}), and n is flow behavior index.

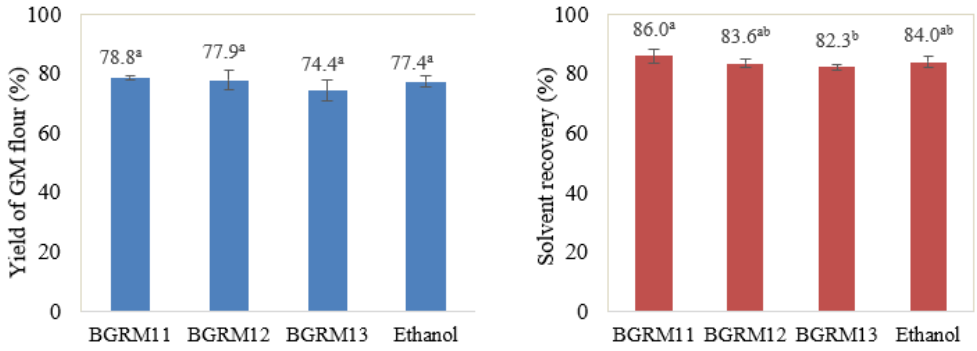
2.4 Designing experiments and statistical analysis

The experiment was set up with a single factor in a completely randomized design. The factor was the mole ratio of betaine anhydrous to glycerol, namely mole ratio of 1:1 (BGRM11), mole ratio of 1:2 (BGRM12), mole ratio of 1:3 (BGRM13), and ethanol as a control treatment. Each treatment was repeated three times.

Using SPSS version 13.0 software, an analysis of variance (ANOVA) and Duncan test (confidence level, $\alpha = 0.05$) were run on the results to determine whether there were any significant differences. For each parameter, means and standard deviation were calculated and reported. The Levenberg-Marquardt algorithm was utilized to solve non-linear regression of the Herschel-Bulkley model. An efficient index method was used to determine the optimal treatment. The effectiveness index technique was evaluated using all the characteristics, and the top rating was obtained by the highest productivity number [26]. Firstly, the variable weight value was determined from respondent assessment of the importance of each variable. Then, calculate the effectiveness value by dividing the difference between the treatment and the smallest value by the difference between the treatment and the highest value. The productivity value resulted by multiplying the weight value and effectiveness value.

3 Results and discussion

The GM flour yield data is given in Fig. 1. The effect of betaine to glycerol mole ratio on GM yield exhibited no significant difference among treatments, including compared to ethanol. However, there was a decreasing trend in yield with an increase in the mole ratio. Isolation of GM from porang flour was based on separating the impurity components of porang flour. The components in porang flour consist of 58.82% (wet base) GM, while the others are impurities, including starch, protein, ash, fiber, and soluble sugar. The solvent plays a role in suspending the impurities, which will be separated in the filtering process. Mulia et al. [27] revealed that the increased solvation effect that resulted from the availability of more hydroxyl groups surrounding the chloride anion of the ChCl molecule caused the viscosity of DES to decrease as the mole ratio of polyalcohol to HBA (ChCl) rose. Decreasing the viscosity of solvents with an increasing mole ratio increases the solvent's ability to diffuse and infiltrate particles. Moreover, impurity components have smaller particle sizes than GM particles [28]. It resulted in more impurity components suspended in the solvent. The increase of suspended impurity components was directly proportional to reducing isolation yield. The increase in extraction yield by reducing the mole ratio of HBA to HBD was reported in α-mangosteen extraction [27] and the polysaccharide extraction from Poria cocos (PCPs) [29].



A

B

Fig. 1. A - The yield of GM flour and B - solvent recovery on some mole ratios NADES and ethanol

The mean (n=3) is used to express data. Values in the bar with varied superscript lowercase letters point out significant differences (P<0.05).

No changes in recovery solvent ($p > 0.05$) were found between ethanol and betaine to glycerol mole ratio of 1:1 and 1:2 (Fig. 1). However, the solvent recovery significantly ($p < 0.05$) decreased when the mole ratio changed to 1:3. The application mole ratio of 1:3 in this isolation produced the more suspended impurity components compared to the others. It induced that more solvent would be absorbed in impurity particles so that the recovered solvent decreased. This result showed that solvent viscosity was related to the yield of GM flour and solvent recovery.

3.1 The chemical properties of glucomannan flour

The GM content and impurity components (ash, starch, protein, and calcium oxalate) contained in the GM flour are presented in Table 1. The ash content of GM flour showed that a mole ratio of 1:1 was significantly higher than that of other treatments. Betaine anhydrous (C5H11NO2) is quaternary ammonium salt that will detect as ash when combusting process. The residue of calcium oxalate also contributed to it. The smaller ratio molar of betaine-glycerol resulted in the higher ash in the GM flour due to more salt adsorbed in particles. The potassium permanganate titration technique determines the sample's total oxalic, including soluble oxalic acid and non-soluble calcium oxalate. The same with other impurity components showed that the ethanol treatment could significantly separate Ca-oxalate content from GM flour. However, all treatments used NADES performed closely to ethanol treatment. This result was in line with the studies reported by [9] and [7] that conducted ultrasound-assisted extraction of glucomannan from porang flour. They were subjected to multilevel concentrations of ethanol and isopropyl alcohol, respectively, and obtained glucomannan flour with composition mainly ash 0.35-1.31%, starch 0.58-3.30%, and Ca-oxalate 0.07-0.19%.

A similar pattern also appeared for the protein content of GM flour. Protein content was calculated from the nitrogen content in the sample. As the molar ratio decreased, the betaine's composition increased, leading to a higher amount of nitrogen in the solvent. However, a nitrogen trace of the NADESs was found in the GM samples. It resulted in higher nitrogen content in the GM flour. Therefore, it needs further analysis to determine the protein composition in GM flour. Considering the non-toxic nature of betaine-glycerol,

these impurities may not pose any difficulty in applying GM flour. The same result was reported by [15].

Starch is a polysaccharide, likewise glucomannan. Ethanol solvent resulted in the starch content of GM flour being lowest compared to the rest of the samples, while BGRM11 (mole ratio of betaine to glycerol 1:1) showed the highest starch content. The particle size of Amorphophallus campanulatus starch reached 30 μm [30], while the glucomannan-rich fraction had an average particle size of 300 μm [28]. The difference in this particle size enlarged starch particles entrained in solvent suspension, and as the NADES molar ratio increased, the starch content in GM flour decreased.

Table 1. Chemical properties of glucomannan flour on some mole ratios NADES and ethanol

Treatments	Ash, %db	Protein, %db	Starch, %db	Glucomannan, %db	Ca-Oxalate, %db
BGRM11	2.32 ± 0.12^{a}	4.72 ± 0.08^{a}	2.38 ± 0.11^{a}	81.19 ± 2.04^{c}	0.17 ± 0.00^{a}
BGRM12	0.84 ± 0.05^{b}	3.60 ± 0.05^{b}	2.15 ± 0.15^{ab}	86.78 ± 1.65^{b}	0.16 ± 0.00^{b}
BGRM13	0.75 ± 0.08^{b}	3.66 ± 0.14^{b}	2.24 ± 0.19^{ab}	89.44 ± 0.38^{ab}	0.16 ± 0.00^{bc}
Ethanol	0.72 ± 0.04^{b}	1.91 ± 0.19^{c}	1.86 ± 0.01^{b}	92.29 ± 2.29^{a}	0.17 ± 0.00^{c}

The mean and standard deviation (n=3) are used to express data. Values in the same column with varied superscript lowercase letters point out significant differences ($P<0.05$).

The isolation used ethanol showed the highest GM content and closed to mole ratio of 1:3, but it was significantly different from the others. Ethanol had a lower viscosity than NADES, directing more impurities to be separate; hence the GM purity rose. The mole ratio also positively affected the GM purity due to its viscosity. The porang flour contained 63.89% db (dry base) of GM content. These isolation processes have increased GM content, ranged 17.30-25.55% for DESs and 28.40% for ethanol. This result is similar to studies conducted by [8] and [31] that reported an increase in GM content of about 27-33% to obtain final GM content of 90-93%. This result revealed that NADES-based betaine-glycerol had a similar performance to ethanol as a conventional solvent. However, the washing process of remaining NADES in GM flour still needs to be optimized to increase GM purity.

3.2 The physical properties of glucomannan flour

3.2.1. *Color of glucomannan flour*

The color of GM flours is presented in Table 2. Porang flour had a lightness (L) of 75.66, a-value of 4.67, and b-value of 11.13, which indicated that porang tubers have a yellow color. Table 2 showed that the isolation process could significantly reduce flour's red and yellow color, mainly conducted using NADES betaine-glycerol.

Table 2. Color of glucomannan flour on some mole ratios NADES and ethanol

Treatments	L-value	a-value	b-value	Whiteness index
BGRM11	79.63 ± 0.23^{a}	2.71 ± 0.01^{b}	7.24 ± 0.03^{b}	78.22 ± 0.21^{a}
BGRM12	76.72 ± 0.04^{b}	2.65 ± 0.01^{c}	6.74 ± 0.02^{c}	75.67 ± 0.05^{b}

BGRM13	75.46±0.44^{c}	2.29±0.01^{d}	6.56±0.07^{d}	74.50±0.41^{c}
Ethanol	74.25±0.01^{d}	4.73±0.02^{a}	9.73±0.03^{a}	72.07±0.01^{d}

The mean and standard deviation (n=3) are used to express data. Values in the same column with varied superscript lowercase letters point out significant differences (P<0.05).

Ethanol treatment showed a significantly lower value of lightness and whiteness index but a significantly higher value of a-value and b-value than the rest treatments. A similar trend appeared by increasing the mole ratio of glycerol to betaine. The whiter GM particle was obtained from a higher removal level of impurities. Additionally, ethanol or NADES might dissolve the native carotenoid on the tuber (which served as the color) and remove it during filtration [32,33]. Konjac corm (elephant yam, *Amorphophallus rivierii*) contained 40 mg total carotenoids per kg fresh corm [34]. Extraction of carotenoids using NADES was possible due to the hydrogen bonding interactions between the solvent and carotenoid [33]. Impaprasert et al. [24] reported that the L-value, a-value, b-value, and WI of konjac GM flour were 84,9 0.8, 1.4, and 79.5, respectively. This value was better than the color of this study due to the difference in tuber source, which konjac tubers have white color. The brightness of GM flour extracted from porang tuber ranged from 53.94-60.38 [9].

3.2.2. *Apparent viscosity*

The apparent viscosity of 1% (w/v) GM solution is presented in in Fig. 2. It showed that ethanol treatment and mole ratio 1:3 have a comparable value and are significantly higher than mole ratio 1:1 and 1:2. Generally, viscosity was proportional with GM content. The viscosity level was proportional to the molecular weight and purity [32]. Apparent viscosity depended on the measurement procedure (selection of spindle type, speed, and torque). The previous studies reported that the viscosity of GM flour extracted using ethanol or isopropyl alcohol ranged from 5,400-15,960 cp [9,35,36].

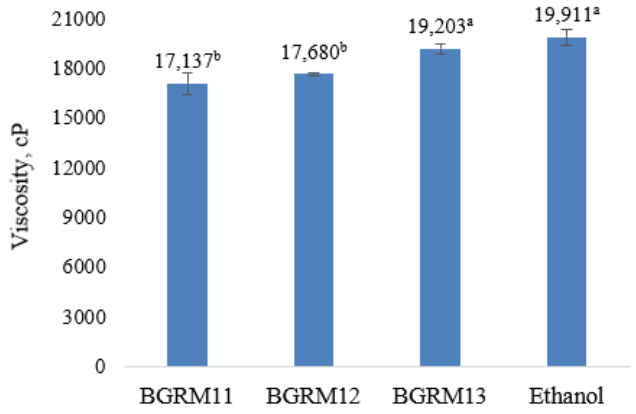


Fig. 2. Apparent viscosity of GM flour on some mole ratios NADES and ethanol

The mean (n=3) is used to express data. Values in the bar with varied superscript lowercase letters point out significant differences (P<0.05).

3.2.3. *Rheological properties*

The rheological properties of 1% (w/v) GM solution is shown in Fig. 3. For all samples, measurements of the stress and viscosity as functions of shear rate were carried out in the

range of 0.01–300 rad s^{-1}. The viscosity graphs were closely among the treatments and exhibited a non-Newtonian pseudoplastic behavior for all samples. It was noted that shear thinning processes were present in every sample. This shear-thinning behavior of the GM solution was made possible because at higher shear rates, the GM molecule disentanglement rate exceeded the re-entanglement rate. The earlier report suggested a decrease in the intermolecular barrier to flow and a decreased apparent viscosity [37]. Figure 3 showed that the shear stress rose for all samples with the addition of shear rate, indicating the degree of molecular assembly and winding of disposed flour. Ethanol and BGRM13 samples required much higher shear stress at the same shear rate. Similar behavior was observed in konjac GM purification using an ethanol solution of 40% [37] and carrageenan extraction from seaweed using NADES-based choline chloride-glycerol [15].

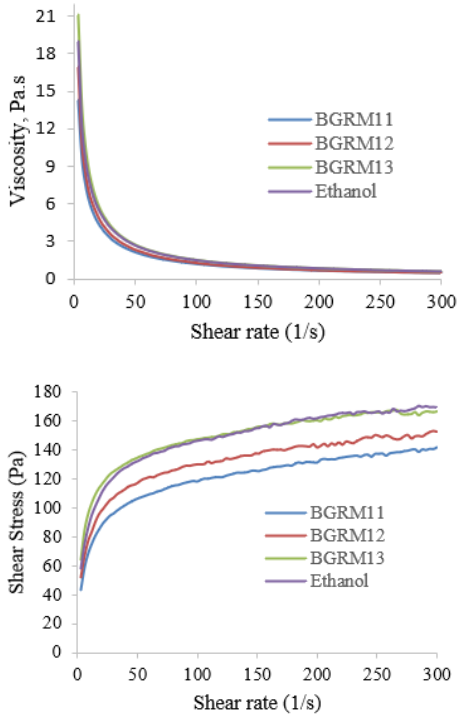


Fig. 3. The stress and viscosity as functions of the shear rate

The parameters of the Herschel-Bulkley model are displayed in Table 3, used to investigate the GM equality. The viscosity coefficient (K) measures the liquid viscosity; a more viscous liquid has a higher K-value. A measurement of the pseudoplastic flow is shown by index n, more volatile shear thinning has a smaller n-value that indicates a greater degree of pseudo-plasticity [38]. When n is small and K increases, GM is thought to process more effectively [37]. Evidently, the liquid characteristics of the BGRM13 and ethanol samples had significantly improved.

Table 3. Parameter values of the Herschel-Bulkley model

Treatments	τ_0	K	n	R^2
BGRM11	0	50.63	0.18	0.98
BGRM12	11.45	47.47	0.19	0.97
BGRM13	17.30	58.38	0.17	0.96
Ethanol	4.64	61.44	0.18	0.97

The best treatment was determined using the effective index method [26] based on the highest total productivity value by considering their yield, solvent recovery, and physicochemical properties. The calculation result is presented in Table 4.

Table 4. The effectiveness value of quality parameter on glucomannan isolation

Parameter	Weight value	Productivity value			
		BGRM11	BGRM12	BGRM13	Ethanol
Glucomannan content	0.20	0.00	0.10	0.15	0.20
Viscosity	0.16	0.00	0.03	0.12	0.15
Yield of GM flour	0.13	0.13	0.11	0.00	0.09
Calcium oxalate	0.13	0.03	0.13	0.08	0.00
Whiteness index	0.10	0.04	0.06	0.10	0.00
Starch	0.09	0.00	0.04	0.02	0.08
Ash	0.07	0.00	0.07	0.07	0.07
Solvent recovery	0.07	0.07	0.02	0.00	0.03
Protein	0.05	0.00	0.02	0.02	0.05
Total value	1.00	0.27	0.58	0.56	0.68

Weight value was determined by evaluation of some respondents conducting GM isolation. Table 4 shows that the highest productivity value was the ethanol isolation method. Among the NADES isolation method, the mole ratio (betaine-glycerol) of 1:2 had the highest productivity value. It suggested that this mole ratio was the best formula to isolate GM from porang flour using NADES based betaine-glycerol.

4 Conclusion

Three natural deep eutectic solvents (NADES) with different mole ratios prepared by mixing anhydrous betaine with glycerol and ethanol were used to isolate glucomannan from the porang flour selectively. The glucomannan flour obtained by the NADES isolation method was not significantly different from that obtained by the ethanol isolation method in terms of yield, physicochemical and rheological properties, even though it was superior in color. Based on the results, the NADESs can be considered suitable alternative solvents for the glucomannan isolation directly from porang flour. Moreover, the NADES with a mole ratio of betaine-glycerol of 1:2 was found as the most effective formula to isolate glucomannan than the other mole ratios. Other types of NADES and operation conditions should be explored to study their selectivity and optimizing process in isolating glucomannan.

References

[1] R. P. Millane and T. L. Hendrixson, "Crystal structures of mannan and glucomannans," Carbohydr. Polym., vol. 25, pp. 245–251, 1994.

[2] L. Yao-ling, R. D. Rong-hua, C. Ni, P. Juan, and P. Jie, "Review of konjac glucomannan: isolation, structure, chain conformation and bioactivities," J. Single Mol. Res., vol. 1, pp. 7–14, 2013.

[3] Q. Du, J. Liu, and Y. Ding, "Recent progress in biological activities and health benefits of konjac glucomannan and its derivatives," Bioact. Carbohydrates Diet. Fibre, vol. 26, p. 100270, 2021.

[4] N. Aanisah, Y. W. Wardhana, A. Y. Chaerunisaa, and A. Budiman, "Review on Modification of Glucomannan as an Excipient in Solid Dosage Forms," Polymers (Basel), vol. 14, pp. 1–19, 2022.
[5] S. Sharma and N. Wadhwa, "Application of Glucomannan," J. Pharm. Res., vol. 21, pp. 1–5, 2022.
[6] M. Chua, K. Chan, T. J. Hocking, P. A. Williams, C. J. Perry, and T. C. Baldwin, "Methodologies for the extraction and analysis of konjac glucomannan from corms of Amorphophallus konjac K. Koch," Carbohydr. Polym., vol. 87, pp. 2202–2210, 2012.
[7] D. H. Wardhani, L. H. Rahayu, H. Cahyono, and H. L. Ulya, "Purification of Glucomannan of Porang (Amorphophallus oncophyllus) Flour using Combination of Isopropyl Alcohol and Ultrasound-Assisted Extraction," Reaktor, vol. 20, pp. 203–209, 2020.
[8] D. H. Wardhani, J. A. Vázquez, D. A. Ramdani, A. Lutfiati, N. Aryanti, and H. Cahyono, "Enzymatic purification of glucomannan Amorphophallus oncophyllus using α-Amylase," Biosci. J., vol. 35, pp. 277–288, 2019.
[9] A. Faridah, "Comparison of porang flour (morphophallus muelleri) purification method: Conventional maceration (gradient ethanol leaching) and ultrasonic maceration method using response surface methodology," Int. J. Adv. Sci. Eng. Inf. Technol., vol. 6, pp. 265–272, 2016.
[10] OECD-FAO, "OECD-FAO Agricultural Outlook 2021–2030," 2021.
[11] A. Muscat, E. M. de Olde, I. J. M. de Boer, and R. Ripoll-Bosch, "The battle for biomass: A systematic review of food-feed-fuel competition," Glob. Food Sec., vol. 25, 2020.
[12] Y. Dai, J. van Spronsen, G. J. Witkamp, R. Verpoorte, and Y. H. Choi, "Natural deep eutectic solvents as new potential media for green technology," Anal. Chim. Acta, vol. 766, pp. 61–68, 2013.
[13] A. Mišan, J. Nađpal, A. Stupar, M. Pojić, A. Mandić, R. Verpoorte, and Y. H. Choi, "The perspectives of natural deep eutectic solvents in agri-food sector," Crit. Rev. Food Sci. Nutr., vol. 60, pp. 2564–2592, 2020.
[14] L. K. Savi, D. Carpiné, N. Waszczynskyj, R. H. Ribani, and C. W. I. Haminiuk, "Influence of temperature, water content and type of organic acid on the formation, stability and properties of functional natural deep eutectic solvents," Fluid Phase Equilib., vol. 488, pp. 40–47, 2019.
[15] A. K. Das, M. Sharma, D. Mondal, and K. Prasad, "Deep eutectic solvents as efficient solvent system for the extraction of κ-carrageenan from Kappaphycus alvarezii," Carbohydr. Polym., vol. 136, pp. 930–935, 2016.
[16] P. S. Saravana, Y. N. Cho, H. C. Woo, and B. S. Chun, "Green and efficient extraction of polysaccharides from brown seaweed by adding deep eutectic solvent in subcritical water hydrolysis," J. Clean. Prod., vol. 198, pp. 1474–1484, 2018.
[17] J. Nie, D. Chen, and Y. Lu, "Deep eutectic solvents based ultrasonic extraction of polysaccharides from edible brown Seaweed Sargassum horneri," J. Mar. Sci. Eng., vol. 8, 2020.
[18] S. Q. Liew, G. C. Ngoh, R. Yusoff, and W. H. Teoh, "Acid and Deep Eutectic Solvent (DES) extraction of pectin from pomelo (Citrus grandis (L.) Osbeck) peels," Biocatal. Agric. Biotechnol., vol. 13, pp. 1–11, 2018.
[19] M. H. Shafie, R. Yusof, and C. Y. Gan, "Deep eutectic solvents (DES) mediated extraction of pectin from Averrhoa bilimbi: Optimization and characterization studies," Carbohydr. Polym., vol. 216, pp. 303–311, 2019.

[20] A. Faridah and S. B. Widjanarko, "Optimization of multilevel ethanol leaching process of porang flour (Amorphophallus muelleri) using response surface methodology," Int. J. Adv. Sci. Eng. Inf. Technol., vol. 2, pp. 74–80, 2013.
[21] AOAC (Association of Official Analytical Chemists), "Official Methods of Analysis," vol. 1, K. Helrich, Ed., Virginia, Washington D.C: Association of Official Analytical Chemists, 1990.
[22] E. R. Asquieri, A. G. de Moura e Silva, D. de C. S. Mendes, and R. Batista Dias, "Comparison of titulometric and spectrophotometric approaches towards the determination of total soluble and insoluble carbohydrates in foodstuff," Carpathian J. Food Sci. Technol., vol. 11, pp. 69–79, 2019.
[23] A. Sarifudin, L. Ratnawati, N. Indrianti, R. Ekafitri, E. Sholichah, N. Afifah, D. Desnilasari, P. Nugroho, and A. D. Yuniar, "Evaluation of some analytical methods for determination of calcium oxalate in Amorphophallus muelleri flour," Food Sci. Technol., vol. 42, pp. 1–7, 2022.
[24] R. Impaprasert, C. Borompichaichartkul, and G. Srzednicki, "A New Drying Approach to Enhance Quality of Konjac Glucomannan Extracted from Amorphophallus muelleri," Dry. Technol., vol. 32, pp. 851–860, 2014.
[25] J. Li, B. Li, P. Geng, A. X. Song, and J. Y. Wu, "Ultrasonic degradation kinetics and rheological profiles of a food polysaccharide (konjac glucomannan) in water," Food Hydrocoll., vol. 70, pp. 14–19, 2017.
[26] E. P. DeGarmo, J. R. Canada, and W. G. Sullivan, "Engineering Economy," New York, New York: Macmillan Pub. Coy. Inc., 1984.
[27] K. Mulia, F. Fauzia, and E. A. Krisanti, "Polyalcohols as Hydrogen-Bonding Donors in Choline Chloride-Based Deep Eutectic Solvents for Extraction of Xanthones from the Pericarp of Garcinia mangostana L.," Molecules, vol. 24, 2019.
[28] M. B. Hermanto, S. B. Widjanarko, W. Suprapto, and A. Suryanto, "The design and performance of continuous porang (Amorphophallus muelleri blume) flour mills," Int. J. Adv. Sci. Eng. Inf. Technol., vol. 9, pp. 2021–2027, 2019.
[29] W. Zhang, S. Cheng, X. Zhai, J. Sun, X. Hu, H. Pei, and G. Chen, "Green and Efficient Extraction of Polysaccharides From Poria cocos F.A. Wolf by Deep Eutectic Solvent," Nat. Prod. Commun., vol. 15, 2020.
[30] A. N. Soni, H. W. Sharma, N. P. Dobhal, S. S. Bisen, H. C. Srivastava, and M. M. Gharia, "The Starches of Dioscorea ballophylla and Amorphophallus campanulatus. Comparison with Tapioca Starch," Starch - Stärke, vol. 37, pp. 6–9, 1985.
[31] A. C. Kumoro, T. H. A. Yuganta, R. Ratnawati, and D. S. Retnowati, "Effect of catalyst concentration and reaction time on the extraction of glucomannan from porang (Amorphophallus oncophyllus) flour via acid hydrolysis," IOP Conf. Series: Material Science and Engineering, vol. 162, p. 012020, 2017.
[32] A. Yanuriati, D. W. Marseno, Rochmadi, and E. Harmayani, "Characteristics of glucomannan isolated from fresh tuber of Porang (Amorphophallus muelleri Blume)," Carbohydr. Polym., vol. 156, pp. 56–63, 2017.
[33] A. Viñas-Ospino, M. Panić, M. Bagović, K. Radošević, M. J. Esteve, and I. Radojčić Redovniković, "Green approach to extract bioactive compounds from orange peel employing hydrophilic and hydrophobic deep eutectic solvents," Sustain. Chem. Pharm., vol. 31, p. 100942, 2023.
[34] A. N. Wootton, M. Luker-Brown, R. J. Westcott, and P. S. J. Cheetham, "The extraction of a glucomannan polysaccharide from konjac corms (elephant yam, Amorphophallus rivierii)," J. Sci. Food Agric., vol. 61, pp. 429–433, 1993.

[35] E. Harmayani, V. Aprilia, and Y. Marsono, "Characterization of glucomannan from Amorphophallus oncophyllus and its prebiotic activity in vivo," Carbohydr. Polym., vol. 112, pp. 475–479, 2014.
[36] D. H. Wardhani, F. Nugroho, M. Muslihudin, and N. Aryanti, "Application of response surface method on purification of glucomannan from amorphophallus oncophyllus by using 2-propanol," Sci. Study Res. Chem. Chem. Eng. Biotechnol. Food Ind., vol. 17, pp. 63–74, 2016.
[37] W. Xu, S. Wang, T. Ye, W. Jin, J. Liu, J. Lei, B. Li, and C. Wang, "A simple and feasible approach to purify konjac glucomannan from konjac flour - Temperature effect," Food Chem., vol. 158, pp. 171–176, 2014.
[38] C. Wang, M. Xu, W. Lv, P. Qiu, Y. Gong, and D. Li, "Study on Rheological Behavior of Konjac Glucomannan," Phys. Procedia, vol. 33, pp. 25–30, 2012.