Optimization of mannan polysaccharide extraction from palm kernel meal using ultrasound-assisted extraction (UAE) method

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Abstract. Research on the extraction of mannan polysaccharides from palm kernel meal (PKM) remains limited. This study aimed to optimize the ultrasound-assisted extraction (UAE) method, focusing on varying extraction times to maximize the yield of mannan polysaccharides from PKM. A completely randomized design (CRD) was utilized, involving seven treatments and five replications. The treatments included: T0 (untreated PKM; control); T1 (PKM+UAE for 30 minutes+E1 [single extraction]); T2 (PKM+UAE for 60 minutes+E1); T3 (PKM+UAE for 90 minutes+E1); T4 (PKM+UAE for 30 minutes+E2 [sequential extraction]); T5 (PKM+UAE for 60 minutes+E2) and T6 (PKM+UAE for 90 minutes+E2). Yield data were analyzed descriptively, while fiber composition and total sugar content were statistically evaluated using ANOVA and Duncan's multiple range test. Results indicated that the T2 and T5 treatments produced the highest yields (E1:4.62%; E2:9.20%) compared to other methods. Total sugar content in UAE-treated samples (3.96%-10.79%) was significantly higher ($p<0.05$) than in the control (T0). However, no significant differences ($p>0.05$) were observed between T2-T3 and T5-T6. In conclusion, the 60-minute UAE method (T5) was the most effective and efficient, achieving the highest yields of mannan polysaccharides and total sugar content. This study underscores the potential of UAE to enhance bioactive polysaccharide extraction from PKM.

1 Introduction

In 2023, Indonesia's palm oil production reached 48,235,405 tons [1]. The expansion of the palm oil industry generates by-products, including palm kernel meal (PKM), which accounts for approximately 45–46% of the processed palm kernel [2]. PKM, a by-product of palm oil extraction, holds significant potential as a source of animal feed [3]. However, its application

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has been limited primarily to poultry feed because of its high crude fiber (12–20%), which is predominantly composed of non-starch polysaccharides (NSPs), especially mannan. These NSPs may impair digestion in livestock [4].

Recent advances in waste processing have demonstrated that mannan can be converted into prebiotics by hydrolyzing it into mannan oligosaccharides (MOS), which has shown promising effects in enhancing feed efficiency for livestock [5]. One effective technique for extracting mannan from PKM is ultrasound-assisted extraction (UAE). This method is widely used for the extraction of bioactive compounds from plant materials [6], but its application in PKM has not been thoroughly explored. UAE is known for enhancing solvent penetration into cell walls and facilitating the release of intracellular components, making it a potential technique for extracting mannan polysaccharides from PKM [5].

This study investigates the use of UAE with a probe-type sonicator for extracting mannan polysaccharides from PKM. Given the lack of research in this area, the study aims to optimize the extraction solvent and duration to determine the most efficient conditions for maximizing the yield of mannan polysaccharides from PKM.

This study aims to evaluate the UAE method by optimizing extraction times for both single and sequential processes to maximize mannan polysaccharide yield from PKM.

2 Materials and methods

2.1 Materials

Materials for PKM extraction included a digital balance, magnetic stirrer, washing machine, spray dryer, and probe-type sonicator. PKM was sourced from CV. Nuansa Baru, Bogor, Indonesia. Chemicals used included acetic acid 0,1 M, NaOH 1 M, and other reagents for subsequent analyses.

2.2 PKM extraction process

The ultrasound-assisted extraction (UAE) process involved 100 g of PKM mixed with an acetic acid solution in a beaker and homogenized using a magnetic stirrer. The sample was subjected to extraction using a probe-type sonicator set at 40% amplitude for durations of 30, 60, and 90 minutes. After extraction, the sample was filtered using a washing machine (35 minutes) to separate the supernatant, which was collected, from the solid residue. Sequential extraction was performed by soaking the residue in a NaOH solution. The supernatant was neutralized with HCl, dried using a spray dryer, and prepared for analysis.

2.3 Yield

The yield of the final extract, after drying, was calculated using the method described by AOAC [7].

2.4 Chemical analysis

Palm kernel meal extract was chemically analyzed, including the determination of Neutral Detergent Fiber (NDF), Acid Detergent Fiber (ADF), hemicellulose, cellulose, and lignin content, were conducted according to the methods described by AOAC [7].

2.5 Total sugar content

The total sugar content was measured using the method of Dubois *et al* [8] using concentrated sulfuric acid, 5% phenol reagent, and a wavelength of 490 nm, with D-glucose as the standard. Measurements were conducted using a spectrophotometer.

2.6 Experimental design

Palm kernel meal extract processes employed a completely randomized design (CRD) with seven treatments and five replications per treatment. The extraction was performed at a ratio of 1:8 (w/v). The treatments were as follows:
T0: PKM without extraction (Control)
T1: PKM + UAE for 30 minutes + E1
T2: PKM + UAE for 60 minutes + E1
T3: PKM + UAE for 90 minutes + E1
T4: PKM + UAE for 30 minutes + E2
T5: PKM + UAE for 60 minutes + E2
T6: PKM + UAE for 90 minutes + E2

2.7 Data analysis

Yield data were analyzed descriptively. Fiber components and total sugar content were subjected to ANOVA, followed by Duncan's multiple range test, using IBM SPSS version 26.

3 Results and discussion

3.1 Yield of palm kernel meal extract

The ultrasound-assisted extraction (UAE) method, using a sonicator (*Cole Parmer*), was employed to optimize the extraction of mannan components from PKM. This technique, which represents a novel approach for PKM, utilizes an amplitude of 40%, based on prior studies indicating superior results at this level compared to 20% [9]. However, the optimal extraction time for PKM remains to be fully established, requiring evaluation at different time points to maximize the yield of mannan components. The yield (%) of PKM extract obtained using UAE is illustrated in Figure 1.

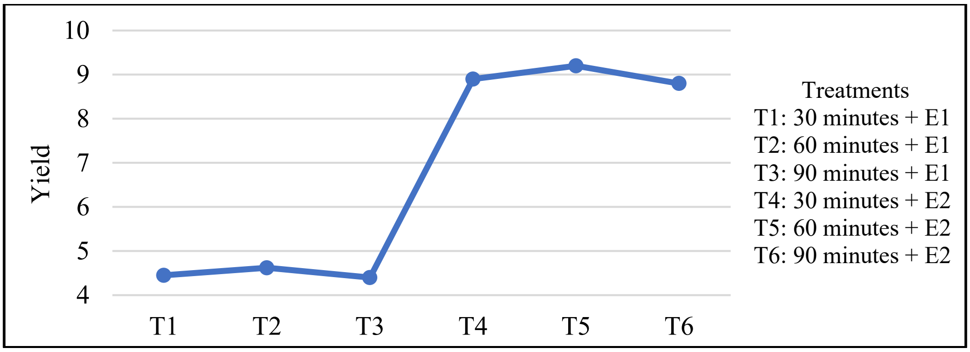

Fig. 1. The yield of palm kernel meal extraction using the ultrasound-assisted extraction (UAE) method, comparing single extraction (E1) and sequential extraction (E2) at varying extraction durations.

The yield of PKM extract obtained via UAE is shown in Figure 1. Single extraction (E1) involves a one-time extraction process, while sequential extraction (E2) is performed by continuing the single extraction process. The results suggest that the sequential extraction process (T4, T5, T6) achieved higher yields compared to the single-step extraction (E1). This can be attributed to the differing solvent capacities over the 30, 60, and 90-minute extraction periods, which facilitated the release of bioactive compounds. These findings align with the work of Chalif and Alauhdin [10], where solvent properties were shown to significantly influence extraction efficiency. The longer extraction times, particularly at 90 minutes (T3, T6), resulted in reduced yields, likely due to the degradation of bioactive compounds [6]. The highest yield was observed at 60 minutes for both extraction methods (T2 and T5).

3.2 Fiber components in palm kernel meal extract

The fiber composition of PKM and UAE-extracted samples is presented in Table 1, with analyses of NDF and ADF. Statistical analysis revealed significant differences ($p<0.05$) in NDF and ADF between the control and UAE-extracted samples. The control exhibited high NDF and ADF levels, indicative of strong structural carbohydrates in the cell wall [11]. Significant differences were also noted between the single (T1, T2, T3) and sequential extractions (T4, T5, T6), suggesting that the use of two solvents in E2 and a larger solvent volume enhanced fiber solubilization [12].

Hemicellulose analysis revealed significant differences ($p<0.05$) between the extraction times, with 60 minutes yielding the most efficient hydrolysis. This aligns with the study by [13], where amorphous regions of hemicellulose were more readily hydrolyzed. Similar trends were observed for cellulose content, with E2 demonstrating higher efficiency. For lignin, extraction at 30 minutes showed the highest solubilization, with prolonged extraction times leading to a decline in lignin extraction, consistent with prior research [14].

Table 1. Fiber components of palm kernel meal and the extract obtained using the ultrasound-assisted extraction (UAE) method (%)

Treatments	NDF	ADF	Hemicellulose	Cellulose	Lignin
T0	60.20 ± 0.03^{a}	31.08 ± 0.05^{a}	29.12 ± 0.08^{a}	23.13 ± 0.08^{a}	6.92 ± 0.06^{a}
T1	2.73 ± 0.06^{c}	2.05 ± 0.04^{b}	0.68 ± 0.03^{c}	0.89 ± 0.25^{c}	0.93 ± 0.02^{b}
T2	3.01 ± 0.67^{c}	1.56 ± 0.09^{c}	1.45 ± 0.58^{b}	1.13 ± 0.05^{c}	0.34 ± 0.04^{d}
T3	2.54 ± 0.10^{c}	1.47 ± 0.04^{c}	1.07 ± 0.10^{c}	1.08 ± 0.03^{c}	0.33 ± 0.03^{d}
T4	4.50 ± 0.11^{b}	2.57 ± 0.53^{b}	1.93 ± 0.64^{b}	1.61 ± 0.27^{b}	1.14 ± 0.01^{b}
T5	4.92 ± 0.89^{b}	2.13 ± 0.40^{b}	2.79 ± 1.28^{b}	1.82 ± 0.02^{b}	0.58 ± 0.03^{c}
T6	3.85 ± 0.21^{b}	1.98 ± 0.03^{b}	1.87 ± 0.24^{b}	1.47 ± 0.04^{b}	0.56 ± 0.03^{c}

Notes: $^{a-d}$) Different superscripts within the same column indicate significant differences ($p< 0.05$) in the fiber components of palm kernel meal (PKM). E1 (Single extraction); E2 (Sequential extraction). T0 (PKM without extraction); T1 (PKM + UAE for 30 minutes + E1); T2 (PKM + UAE for 60 minutes + E1); T3 (PKM + UAE for 90 minutes + E1); T4 (PKM + UAE for 30 minutes + E2); T5 (PKM + UAE for 60 minutes + E2); T6 (PKM + UAE for 90 minutes + E2). ADF (Acid Detergent Fiber); NDF (Neutral Detergent Fiber).

3.3 Total sugar content in palm kernel meal extract

Total sugar content serves as an indicator of the polysaccharide fraction extracted from PKM and quantified using a spectrophotometer. The hydrolysis of mannan fractions into mannose and MOS further enhances its potential as a prebiotic [3]. The results of total sugar analysis are shown in Table 2.

Table 2. Total sugar components of palm kernel meal and the extract obtained using the ultrasound-assisted extraction (UAE) method (%)

Treatments	Total Sugar (%)
T0	3.73 ± 0.04^{d}
T1	3.96 ± 0.03^{d}
T2	6.34 ± 0.24^{c}
T3	6.61 ± 0.03^{c}
T4	8.46 ± 0.05^{b}
T5	10.79 ± 0.19^{a}
T6	10.76 ± 0.03^{a}

Notes: $^{a-d}$) Superscripts within the same column indicate significant differences (p<0.05) in total sugar content of palm kernel meal (PKM). E1 (Single extraction); E2 (Sequential extraction). T0 (PKM without extraction); T1 (PKM + UAE for 30 minutes + E1); T2 (PKM + UAE for 60 minutes + E1); T3 (PKM + UAE for 90 minutes + E1); T4 (PKM + UAE for 30 minutes + E2); T5 (PKM + UAE for 60 minutes + E2); T6 (PKM + UAE for 90 minutes + E2).

Total sugar content analysis, as shown in Table 2, revealed that extraction time significantly impacted ($p<0.05$) the total sugar content, with the sequential extraction (E2) yielding significantly higher amounts (T4, T5, T6) compared to the single extraction (E1). This observation is consistent with findings by Yulianti *et al* [15], where longer extraction times facilitated higher sugar yields. Prolonged solute-solvent interaction enhances solubility and accelerates extraction rates, but yields diminish once the optimal extraction time is surpassed.

4 Conclusion

The ultrasound-assisted extraction (UAE) method for extracting mannan polysaccharides from palm kernel meal was optimized at 60 minutes. The sequential extraction approach (E2) yielded higher extract recovery, total sugar content, and fiber components, indicating its superiority in maximizing mannan polysaccharide yield.

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References

[1] Direktorat Jendral Perkebunan, Statistik Perkebunan Unggulan Nasional 2020-2023, (Sekretariat Direktorat Jendral Perkebunan, Jakarta, 2023)

[2] B. M. W. T. Gading, N. Ali, R. Irsan, Pengaruh pemberian bungkil inti sawit fermentasi dengan level berbeda terhadap pertambahan bobot badan kambing cross boer jantan lepas sapih. J. Ilm. Agrotani. **3**, 2 (2021).

[3] T. Pasaribu, Upaya meningkatkan kualitas bungkil inti sawit melalui teknologi fermentasi dan penambahan enzim untuk unggas. Wartazoa. **28**, 3 (2018) 10.14334/wartazoa.v28i3.1820.

[4] D. Wicaksana, S. Hidanah, W. P. Lokapirnasari, M. A. Al-Arif, M. Lamid, K. Suprianondo, Penggunaan bungkil inti sawit dan ß-mannanase pada produktivitas ayam petelur. Mediaka Vet. **4**, 1 (2021) 10.20473/jmv.vol4.iss1.2021.72-77

[5] D. T. Wahyuni, S. B. Widjanarko, Pengaruh jenis pelarut dan lama ekstraksi terhadap ekstrak karotenoid labu kuning dengan metode gelombang ultrasonik. J. Pangan dan Agroindustri. **3**, 2 (2015)

[6] M. T. Adiwibowo, Herayati, K. Erlangga, D. A. Fitria, Pengaruh metode dan waktu ekstraksi terhadap kualitas dan kuantitas saponin dalam ekstrak buah, daun, dan tangkai daun belimbing wuluh (*Avverhoa bilimbi L.*) untuk aplikasi detergen. J. Integr. Proses. **9**, 2 (2020) 10.36055/jip.v9i2.9262

[7] AOAC, Official method of analysis (Association Official Agriculture Chemist, Washington DC, 1999)

[8] M. Dubois, K. A. Gilles, J. K. Hamilton, P. A. Rebers, F. Smith, Colorimetric method for determination of sugars and related substances. Anal. Chem. **28**, 3 (1956) 10.1021/ac60111a017.

[9] E. Prihatini, R. Ismail, I. S. Rahayu, G. D. Laksono, D. Khairunissa, Compatibility testing of synthesized TIO2 nanoparticles on the fast-growing wood physical properties. J. Sains Nat. **14**, 2 (2024)

[10] C. Chalif, M. Alauhdin, Analisis pengaruh massa sampel, volume pelarut, dan waktu sonikasi pada ekstraksi bawang merah (*Allium ascalonicum L.*) terhadap kandungan Quercetin. Indones. J. Math. Nat. Sci. **47**, 1 (2024)

[11] M. Rodiallah, A. E. Harahap, A. Ali, T. Adelina, D. A. Mucra, B. Solfan, R. Misrianti, J. Juliantoni, E. Irawati, B. N. Ramadhan. Profil nutrisi dan fraksi serat pakan silase komplit berbahan ampas tebu dengan penambahan legume Indigofera dan molases. J. Trit. **14**, 1 (2023) 10.47687/jt.v14i1.377

[12] A. Yulianingtyas, B. Kusmartono, Optimasi volume pelarut dan waktu maserasi pengambilan flavonoid daun belimbing wuluh optimization of solvent volume and maceration time on extraction of flavonoids from averrhoa bilimbi leaves. J. Tek. Kim. **10**, 2 (2016)

[13] S. C. Yuansah, Potensi pembuatan gula non-digestible dari selulosa dan hemiselulosa menggunakan hidrolisis enzimatis. Canrea J. Food Technol. Nutr. Culin. **2**, 2 (2019) 10.20956/canrea.v2i2.116.

[14] G. Yoricya, S. A. P. Dalimunthe, R. Manurung, N. Bangun, Hidrolisis hasil delignifikasi tandan kosong kelapa sawit dalam sistem cairan ionik choline chloride. J. Tek. Kim. USU. **5**, 1 (2016)

[15] D. Yulianti, B. Susilo, R. Yulianingsih, Pengaruh lama ekstraksi dan konsentrasi pelarut etanol terhadap sifat fisika-kimia ekstrak daun Stevia (Stevia ebaudiana Bertoni M.) dengan metode microwave assisted extraction (MAE). J. Bioproses Komod. Trop. **2**, 1 (2014)