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JAWABAN DAN KLARIFIKASI TERHADAP ALASAN DITOLAKNYA USULAN KENAIKAN JABATAN GURU BESAR

Yang bertanda tangan di bawah ini:

Nama : Dr. Ir. Beni Hidayat, M.Si
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Dengan ini memberikan jawaban dan klarifikasi terhadap hasil penilaian kenaikan jabatan fungsional guru besar saya pada tanggal 3 September 2024, sebagaimana diuraikan di bawah ini

1. Syarat administrasi berupa dokumen rekomendasi senat tidak memenuhi syarat dengan catatan "Berita Acara Senat jumlah yang hadir 21 orang akan tetapi total anggota senat tidak diketahui sehingga tidak bisa dihitung apakah yang hadir sudah mencapai kuorum (lebih dari 60%)"
2. Syarat khusus tambahan paper 2 tidak memenuhi dengan catatan "Paper 2 sebagai syarat tambahan karena kurang dari 3 tahun setelah S3 tidak bisa diterima karena co-promotor terlibat sebagai co-author (artinya paper ini masih terkait dengan tugas S3"
3. Bukti korespodensi syarat khusus tidak lengkap dengan catatan "Data bukti korespodensi tidak ditemukan respon author terhadap komentar reviewer, sebaiknya dilampirkan manuscript sebelum dan sesudah revisi"
4. Syarat tambahan berupa hibah penelitian kurang lengkap, dengan catatan 1) tidak menyertakan SK Ketua Peneliti, 2) tidak menyertakan dampak dari hibah penelitian tersebut

Jawaban dan klarifikasiyaitu

1. Jumlah anggota senat Politeknik Negeri Lampung sebanyak 31 orang, dengan jumlah yang hadir 21 orang, maka telah tercapai quorum (67%). Bukti quorum terlampir di Dokumen Rekomendasi/Berita Acara Senat.
2. Syarat khusus tambahan Paper 2 terbit dan disusun setelah penulis menyelesaikan pendidikan S3. Saya setuju dengan pendapat reviewer bahwa paper ini masih terkait dengan tugas S3. Oleh karena pada saat pengajuan revisi ini (Februari 2025) penulis telah lebih dari 3 tahun (lulus 21 November 2021, tanggal ijazah 22 Januari 2022) maka syarat khusus tambahan paper 2 ini tidak saya ajukan/dihilangkan.
3. Bukti korespodensi syarat khusus telah dilengkapi dengan manuscript sebelum dan sesudah revisi. Terlampir di syarat khusus/korepodensi karya ilmiah yang sudah dilengkapi.

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4. SK Ketua Peneliti dan Dampak Hibah Penelitian pada syarat tambahan hibah penelitian telah dilengkapi. Terlampir di syarat tambahan hibah penelitian/SK, Kontrak, Laporan Hibah Penelitian.

Demikian jawaban dan klarifikasi ini saya buat, semoga bermanfaat untuk kelengkapan usulan jabatan fungsional Guru Besar. Atas perhatian dan kerjasama Bapak/Ibu reviewer saya ucapkan terima kasih.

Bandar Lampung, 11 Pebruari 2025

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dan Pengabdian Kepada Masyarakat

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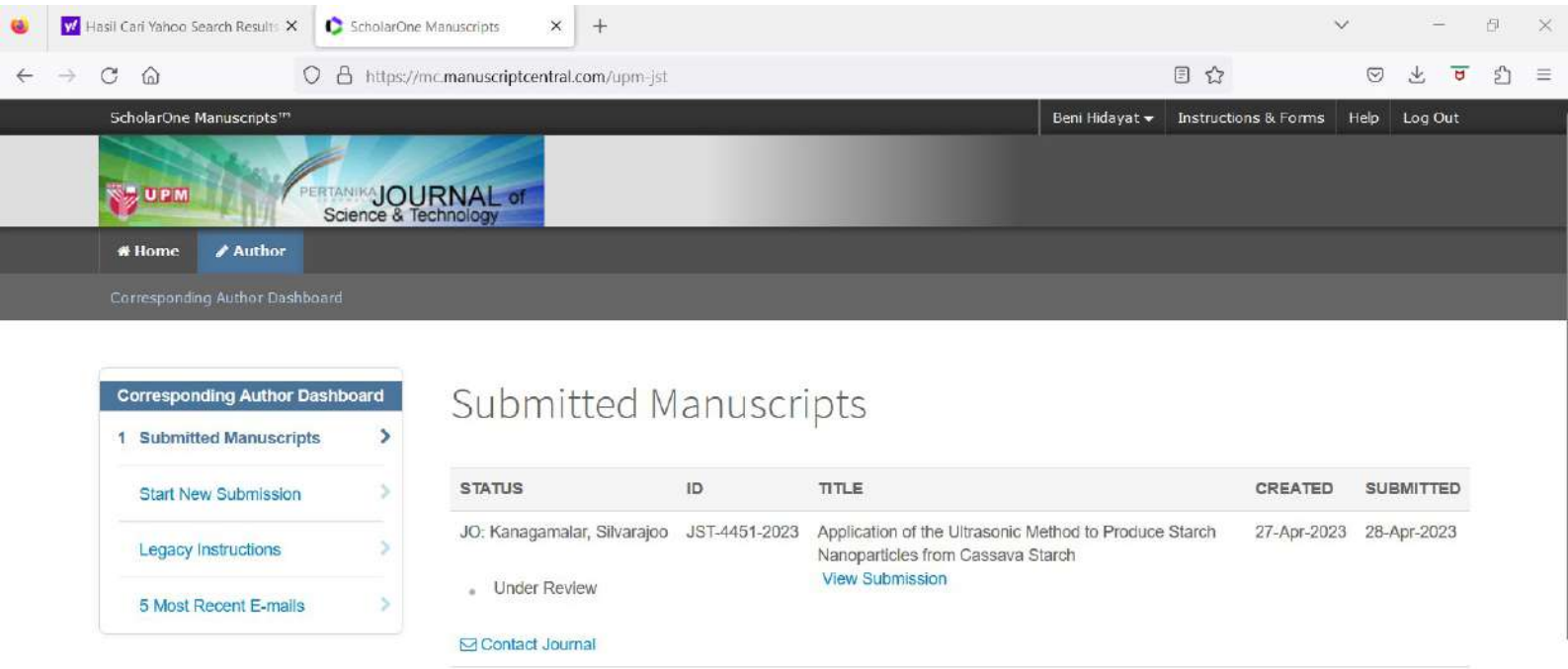
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17-Aug-2023

mailto:vida@polinela.ac.id,

Dear Dr. Hidayat,

Manuscript ID JST-4451-2023 entitled "Application of the Ultrasonic Method to Produce Starch Nanoparticles from Cassava Starch" which you submitted to the Journal of Science and Technology, has been reviewed. The comments of the reviewer(s) are included at the bottom of this letter. I invite you to respond to the reviewer(s)' comments and revise your manuscript.

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Once again, thank you for submitting your manuscript to the Journal of Science and Technology and I look forward to receiving your revision.

Sincerely,
Chief Executive Editor, Journal of Science and Technology

Reviewer(s)' Comments to Author:

Reviewer: 1 (with attachment)

Comments to the Corresponding Author
1. Pg. 3: line 6-24 - The first and second paragraph should be combined, so that it provides a more comprehensive overview about the research topic.
2. Page 3: line 36-49 - The whole paragraph lacks the limitations associated with the approaches mentioned. It might be useful to add a statement highlighting the specific limitations or challenges faces in the development of SNPs based on tapioca.
3.The last paragraph of the introduction section which highlights the objective lacks specific information about the 'best characteristics' - in terms of what?
4. Pg. 4:line 22 - include the units for starch concentration
5. Page 4:line 40 - Equation for yield analysis should be numbered.
6. There are several inconsistency in tense used in the methods section (authors switch between past tense and future tense without a clear reason for the change). It would be more appropriate to maintain consistent tense throughout the entire section, preferably using past tense since it describes the methods and procedures used in the study.
7. Page 4 (SNP Formation) - need elaboration or citation on the specific steps involved in preparing the cassava starch solution.
8. For the SNP yield - How many replications were conducted for each combination of ultrasonication process time and starch concentration?
9. Page 5 (Yield of starch nanoparticles) - Authors should discussed why the yields for the processing time of 90 minutes with a starch concentration of 2% and the processing time of 90 minutes with a starch concentration of 3% are quite close?
10. Page 18 : Conclusion - highlight the potential implications of the study's findings in the context of nanoparticle production. Also, add future research directions based on the study's outcomes

Reviewer: 2

Comments to the Corresponding Author
-The manuscript presents routine work and its present form does not bring new relevant information to the field. The findings were not comprehensively discussed. It seems that the paper is a report instead of a scientific paper.
-Introduction is too short and should be rewritten. It should be expanded to include a more detailed discussion of current problems.
-Would you explicitly specify the novelty of your work? What progress against the most recent state-of-the-art similar studies was made?
-The literature review section should be improved. It should be dedicated to present critical analysis of state-of-the-art related work to justify the objective of the study. Also, critical comments should be made on the results of the cited works.
Please revise the uncertainty analysis to include explicit consideration of all sources of uncertainty in all measured quantities, and then propagate each contribution into a combined, expanded uncertainty whenever possible. This MUST include uncertainty in the calibration. For example, simply stating the specified tolerance of a balance, given by a manufacturer, is inadequate. Simply stating the correlation coefficient of a fit, absent an analysis of the uncertainty of the data that are fit, is insufficient. Simply stating that a particular ASTM or other standard method was used is insufficient. Simply providing the repeatability of replicates with no discussion of uncertainty in calibration is insufficient. These four items are cited only as examples; all sources of uncertainty must be addressed. Uncertainty may be discussed in the text and also added to the figures (in the form of uncertainty bars), but at a minimum uncertainty must be discussed in the text. The number of significant figures provided in tables or text must be derived from and consistent with the uncertainty analysis. Any measured quantity that is presented must be accompanied by an analysis of the uncertainty of that measurement.

-The discussion statements are speculations than supported substantial description and extension from the experimental data.
-It would be excellent if the importance of this issue was validated by detailed research and thoroughly documented data.
-Conclusions should be amended to incorporate a broader discussion of the significance and potential application of this specific study.

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Application of the Ultrasonic Method to Produce Starch Nanoparticles from Cassava Starch

Journal:	*Journal of Science and Technology*
Manuscript ID	JST-4451-2023
Manuscript Type:	Regular Article
Scope of the Journal:	Resource-based industry < APPLIED SCIENCES AND TECHNOLOGIES, Food industry < Resource-based industry < APPLIED SCIENCES AND TECHNOLOGIES, APPLIED SCIENCES AND TECHNOLOGIES, Manufacturing and process technologies and engineering < APPLIED SCIENCES AND TECHNOLOGIES, Material processing technology < Manufacturing and process technologies and engineering < APPLIED SCIENCES AND TECHNOLOGIES
Keywords:	cassava starch, starch nanoparticles, ultrasonic
Abstract:	Starch nanoparticles have the potential to be developed as a cassava starch derivative. The research aims to obtain the optimal process conditions (ultrasonic process time and starch concentration) to produce starch nanoparticles with the best characteristics. The treatment variables used in this study were the duration of the ultrasonication process (30, 60, and 90 minutes) and the starch concentration (1, 2, and 3%). The results showed that the ultrasonication process time and starch concentration affected the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and SNP clarity score. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce SNP products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27%, and a solution clarity score of 3.80 (not clear). Abstract SNP Cassava Starch.docx

Running Title

Cassava Starch-based Starch Nanoparticles

Application of the Ultrasonic Method to Produce Starch Nanoparticles from Cassava Starch

ABSTRACT

Starch nanoparticles have the potential to be developed as a cassava starch derivative. The research aims to obtain the optimal process conditions (ultrasonic process time and starch concentration) to produce starch nanoparticles with the best characteristics. The treatment variables used in this study were the duration of the ultrasonication process (30, 60, and 90 minutes) and the starch concentration (1, 2, and 3%). The results showed that the ultrasonication process time and starch concentration affected the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and SNP clarity score. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce SNP products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27%, and a solution clarity score of 3.80 (not clear).

-what are the units?

-define SNP

Keywords : cassava starch, starch nanoparticles, ultrasonic

INTRODUCTION

Starch is a natural, renewable, biodegradable polymer that many plants use as energy storage. Starch is the second most abundant biomass in nature and is found in staple crop commodities such as rice, corn, wheat, cassava, and potatoes (BeMiller and Whistler, 2009).

The primary potential source of starch in Indonesia is cassava starch obtained from the cassava extraction process (Zukryandry et al., 2022). Based on data from the Food and Agriculture Organization (FAO) in 2012, Indonesia is the world's third exporter of tapioca, followed by Thailand and Vietnam (Hidayat et al., 2021). According to BPS (2022), Indonesia's cassava production in 2021 will be 19,341,233 tons, and Lampung Province, with a production of 6,683,758 tons, is the main producer of cassava in Indonesia (34.5%).

-suggest to combine the first and second paragraph to provide a more comprehensive overview

Starch nanoparticles (SNPs) have the potential to be developed as a tapioca derivative product. SNPs are nano-sized starch derivative products (one billionth of a meter, 10-9 meters) with a size range of 1–100 nm (EFSA, 2011). The process of modifying starch into starch nanoparticles products has many advantages, including increasing stability, chemical reactivity, flowability, opacity, and mechanical strength (Zhu et al., 2007), improving the sensory characteristics of the product (Sharma et al., 2013), and enhancing encapsulation ability for bioactive components (Ezhilarasi et al., 2013).

Despite their potential, the development of SNPs based on tapioca is relatively limited and is mostly developed from corn starch (Le-Corre et al., 2010; Kim et al., 2013; Kumari et al., 2019) and rice starch (Zuo et al., 2009). The manufacture of SNPs can be carried out using various methods, namely, acid hydrolysis (Le-Corre et al., 2010), enzymatic hydrolysis (Le-Corre et al., 2012), high-pressure homogenization (Liu et al., 2009), gamma irradiation (García et al., 2012; Lamanna et al., 2013), combination of acid hydrolysis and ultrasonication (Kim et al., 2013; Goncalves et al., 2014), and ultrasonication (Bel Haaj et al., 2013). The research results by Bel Haaj et al. (2013) showed that SNP products can be prepared solely with the ultrasonic method.

-this paragraph lacks a clear mention of the limitations associated with these approaches.
- might be useful to add a statement highlighting the specific limitations or challenges faced in the development of SNPs based on tapioca.

According to Jambrak et al. (2010), the ultrasonication process to produce SNPs can be carried out using an ultrasonic probe or a bath system. Compared to an ultrasonic system bath, the use of an ultrasonic system probe will be more effective with a shorter processing time (Bonto et al., 2020) and produce SNP products with better characteristics (Bel Haaj et al., 2013). This study aims to obtain optimal process conditions (ultrasonic process time and starch

concentration) to produce cassava starch-based starch nanoparticles with the best characteristics. - in terms of what property?

MATERIALS AND METHODS

Equipment

The main tools used are Ultrasonication probe Biomaisen type MSUCD 650, UV-Vis single beam spectrophotometer Aelab type AE-S60-4U, and Particle Size Analyzer (PSA) Malvern Zetasizer Nano ZS type.

Starch Nanoparticle (SNP) Formation

The formation of SNPs from cassava starch was initiated by preparing 50 ml of cassava starch solution with concentrations according to treatment (1%, 2%, and 3%). -include units used The probe temperature is set below 40°C, kept constant by adding ice, and the process frequency is set at 20 kHz. The probe used has a diameter of 6 cm with an ultrasonic power of 650 W. The ultrasonication process is then carried out with the duration of the ultrasonication process according to the treatment (30, 60, and 90 min). The solution resulting from the sonification process was then filtered using 1-micron Whatman filter paper and tested for yield and characteristics.

Yield Analysis

The yield is the percentage of the dry weight of the SNP product divided by the initial weight of the starch raw material, with the following formula:

$$\text{Yield (\%)} = \frac{mass\ of\ SNP\ (gram)}{mass\ of\ initial\ starch\ (gram)} \text{ x } 100\%$$

Analysis of Particle Size

The distribution and size of SNPs were analyzed using a particle size analyzer (PSA) with the dynamic light scattering (DLS) method that utilizes infrared scattering. The SNP solution sample was put into the PSA cuvette. Infrared scattering is fired at the sample so that the sample will react to produce Brownian motion (random motion of the particles). This random motion is then analyzed by the tool, where the smaller the particle size, the faster the movement.

In addition to the distribution and size of SNPs, the polydispersity Index (PI) value will also be obtained, a measure of molecular mass distribution in the sample.

The PI value indicates the level of confidence in the size of the particles dispersed in a solution. The smaller the polydispersity value, the better the confidence level of the particle size distribution in the starch solution. Conversely, if the polydispersity value is higher, then the particles present in the sample are not uniform and unstable and will quickly flocculate.

Transmittance Analysis

Samples of SNP solution resulting from the sonication process of various treatments were put into the spectrophotometer cuvette. Analysis was conducted by placing a cuvette into a UV-Vis spectrophotometer with a 450–800 nm wavelength range. The results obtained were then recorded in the form of transmittance percentage values.

Clarity Analysis

Observation of the clarity of the SNP solution was carried out after being left for 2 hours (Bel Haaj et al., 2013). The test was carried out sensory using 10 panelists using a hedonic score of 1–5 (score 1 = very unclear; score 2 = not clear; score 3 = not clear enough; score 4 = clear; score 5 = very clear).

RESULT AND DISCUSSION

Yield of starch nanoparticles

The yield of SNPs at various ultrasonic process times and starch concentrations is presented in Table 1. The test results in Table 1 show that the ultrasonication process duration of 90 minutes and 3% starch concentration will produce SNP products with higher yields (13.68%) than that of other treatments. The higher yield of this SNP indicates that increasing the ultrasonication process time to 90 minutes and increasing the starch concentration to 3% will cause the breakdown of starch molecules into nano-sized to become more intensive. The yield of SNPs using the ultrasonic method (13.68%) is relatively the same as the acid hydrolysis method (15%) but lower than the combined acid and ultrasonic hydrolysis method, which can reach 78% (Kim et al., 2013).

The longer the ultrasonic process, the more intensive the degradation process of starch molecules. According to Czechowska-Biskup et al. (2005), the ultrasonic application will cause the degradation of starch molecules caused by mechanochemical

effects. The more intensive the starch degradation process, the smaller the starch granule size.

The increase in SNP yields up to 3% starch concentration indicated that up to 3% starch solution concentration, the starch degradation process was still occurring intensively. A different opinion was conveyed by Bel Haaj et al. (2013), which stated that the ultrasonication process without chemical treatment was effective at low concentrations (1-2%). The conditioning of starch in the form of an aqueous solution, not a suspension is based on the results of Czechowska-Biskup et al. (2005), which showed that the process of degradation of starch molecules was more effective in aqueous/solution conditions.

Table 1. Yield of SNP at various ultrasonication process times and starch concentrations

Treatment	SNP yield (%)
Processing time 30 minutes, starch concentration 1%	11.94
Processing time 30 minutes, starch concentration 2%	13.18
Processing time 30 minutes, starch concentration 3%	13.33
Processing time 60 minutes, starch concentration 1%	12.02
Processing time 60 minutes, starch concentration 2%	13.37
Processing time 60 minutes, starch concentration 3%	13.56
Processing time 90 minutes, starch concentration 1%	12.32
Processing time 90 minutes, starch concentration 2%	13.66
Processing time 90 minutes, starch concentration 3%	13.68

Distribution and Particle Size of SNP

Distribution, particle size, and PI of SNPs at various ultrasonic process times and starch concentrations are presented in Table 2 and Figure 1. The test results in Figure 1 show the percentage of SNP particle size at various particle sizes continuously using a particle size analyzer (PSA), while the test results in Table 2 show the particle size in various particle size groups (≤100 nm, 101-1000 nm, and > 1000 nm).

The test results in Figure 1 show that most of the SNPs are 101 to 1000 nm in size. This shows that the sonication process is quite effective in reducing the size of starch particles. According to Boufi et al. (2018) and Zuo et al. (2012), the ultrasonic method was able to damage and reduce the size of starch granules. The research results in Table 2 also show the presence of particles with a diameter of more than 1000 nm with a small intensity. Particles with a size of more than 1000 nm are thought to be starch particles that have agglomerated into a larger size. According to Jambrak et al. (2010), with changes in temperature and longer storage time, nanoparticles can agglomerate into larger sizes.

The test results in Table 2 show that the ultrasonic process of starch with a concentration of 1–3% for 30–90 minutes will produce SNP products with a diameter range of 230.80 nm to 501.50 nm and a PI value range of 0.34–0.58 nm. The lowest PI was shown in the sonication time of 60 minutes with a starch concentration of 3% with a PI of 0.34 and an average particle size of 333.70 nm. The low PI indicates that the particle size dispersion of SNP is homogeneous and evenly distributed. A PI value greater than 0.70 indicates a very wide distribution of particle sizes so that sedimentation is likely to occur.

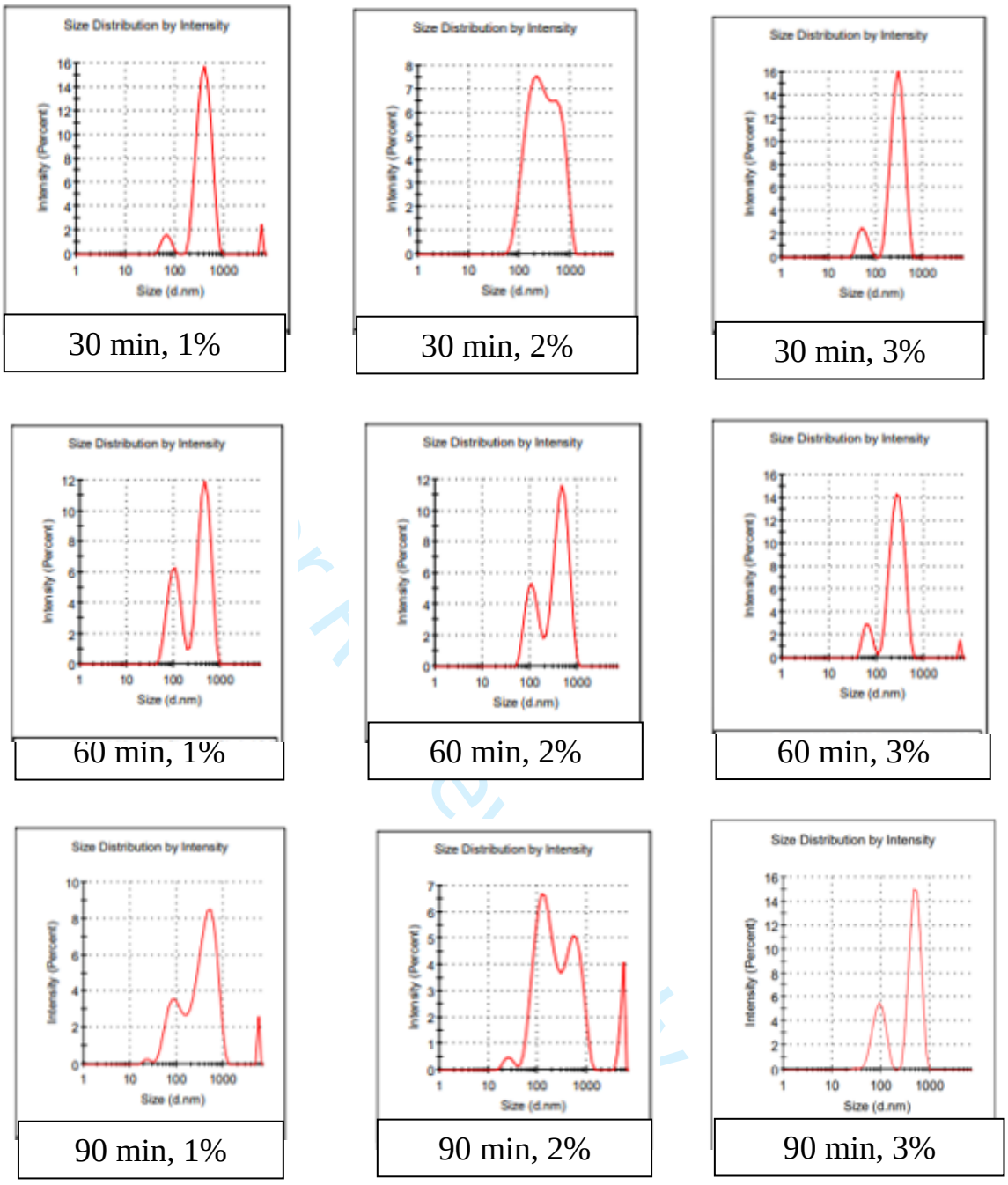

Figure 1. Distribution of various SNPs sizes at various ultrasonication process times and starch concentrations.

Table 2. Particle size distribution per size group and polydispersity index of SNPs at various ultrasonication process times and starch concentrations.

Treatment	SNP Particle Size				Polydispersity index
	$\leq 100\ nm$ (%)	101 – 1000 nm (%)	> 1000 nm (%)	Average (nm)	
Processing time 30 minutes, starch concentration 1%	6.30	93.70	0.00	501.50	0.47
Processing time 30 minutes, starch concentration 2%	7.60	91.50	0.90	419.90	0,47
Processing time 30 minutes, starch concentration 3%	11.00	89.00	0.00	470.20	0.46
Processing time 60 minutes, starch concentration 1%	12.00	86.40	1.60	429.60	0.51
Processing time 60 minutes, starch concentration 2%	16.70	83.30	0.00	355.00	0.47
Processing time 60 minutes, starch concentration 3%	22.90	77.10	0.00	333.70	0.34
Processing time 90 minutes, starch concentration 1%	20.10	76.80	3.10	430.30	0.50
Processing time 90 minutes, starch concentration 2%	22.30	69.70	8.00	422.90	0.58
Processing time 90 minutes, starch concentration 3%	23.60	76.40	0.00	230.80	0.58

The test results showed that the ultrasonication process duration of 90 minutes and 3% starch concentration would produce SNP products with a particle size of less than 100 nm, which was higher (23.6%) than that of the other treatments. The results also showed that the longer the sonification process and the higher the starch concentration, the higher the percentage of SNP particles less than 100 nanometers in size. This indicates that the ultrasonication process can break down starch granules into smaller sizes. The phenomenon of acoustic cavitation by ultrasonic waves causes starch particles to break into nano-sized pieces (Czechowska-Biskup et al., 2005). The increase in the percentage of SNP particle size in line with the increase in concentration up to 3% also shows that at a starch concentration of up to 3%; the cavitation process which causes the breakdown of starch granules into nano-sized still occurs effectively. The increase in the cavitation process in line with the increase in starch concentration in the formation of SNPs was also reported by Jambrak et al. (2010).

Starch Nanoparticles Transminttance Values

The results of testing the transmittance value of SNPs at various ultrasonic process times and starch concentrations are presented in Table 3 and Figure 2. The test results show that the ultrasonication process duration of 30 minutes and 1% starch concentration will produce SNP products with the highest transmittance values (86.38%). Conversely, the ultrasonication process time of 90 minutes and 3% starch concentration will produce SNP products with the lowest transmittance value (61.27%).

Table 3. Transmittance values of SNPs at various ultrasonic process times and starch concentrations.

Treatment	Transmittance (%) at wavelength (nm)					Average transmittance (%)
	450	500	600	700	800	
Processing time 30 minutes, starch concentration 1%	85.31	85.62	86.78	86.60	88.34	86.38
Processing time 30 minutes, starch concentration 2%	75.17	76.55	77.37	77.96	80.16	77.33
Processing time 30 minutes, starch concentration 3%	66.72	67.47	69.27	69.97	72.00	69.05
Processing time 60 minutes, starch concentration 1%	82.37	82.30	83.25	83.20	83.83	82.98
Processing time 60 minutes, starch concentration 2%	69.65	70.78	72.58	73.71	76.88	72.65
Processing time 60 minutes, starch concentration 3%	62.74	63.56	64.76	64.72	66.48	64.31
Processing time 90 minutes, starch concentration 1%	78,97	78,51	78,16	78,12	78,47	78.24
Processing time 90 minutes, starch concentration 2%	64.91	65.86	67.27	68.40	70.22	67.47
Processing time 90 minutes, starch concentration 3%	58.29	59.41	61.09	62.23	64.23	61.27

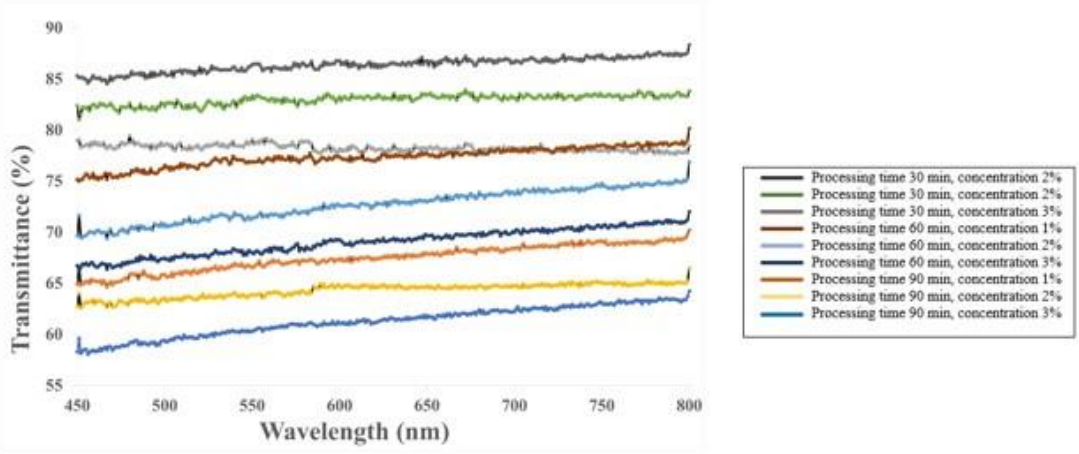

Figure 2. SNP transmittance curves for various ultrasonication process times and starch concentrations.

Ultrasonic process time of 90 minutes and concentration of 30% (Table 2, Figure 2) will produce SNPs with the lowest transmittance value compared to other treatments. The lower transmittance value of the SNP is strongly related to the size of the SNP particles. The smaller the SNP particle size is, the more difficult it is for the starch particles to precipitate and the lower the transmittance value is. On the other hand, the larger the SNP particle size is, the faster the particles settle and the greater the transmittance value is. Changes in the transmittance of SNPs and a decrease in particle size were also reported by Bel Haaj et al. (2013) on SNP formation in corn starch. According to Bel Haaj et al. (2013), SNPs with a size of more than 10 µm will precipitate quickly.

Starch Nanoparticles Clarity Score

The results of testing the clarity score of SNPs at various lengths of the ultrasonication process are presented in Table 4 and Figure 3. The test results show that the 90-minute ultrasonication process and 3% starch concentration will produce SNPs with the lowest level of clarity compared to other treatments. The lower clarity of the SNP is strongly related to the size of the SNP particles and their solubility. The smaller the SNP particle size, the lower the clarity of the SNP solution because the nano-sized SNP particles will dissolve and have difficulty settling even though they have been left for 2 hours. The increase in SNP solubility with the smaller particle size is mainly related to the increase in the porosity of starch granules (Sujka, 2017). Changes in the level of clarity of SNP solutions, along with a decrease in particle size, were also reported by Jambrak et al. (2010) and Kim et al. (2013) on SNP formation in corn starch.

Table 4. SNP clarity scores at various ultrasonic process times and starch concentrations

Treatment	SNP clarity score (%)
Processing time 30 minutes, starch concentration 1%	3.80
Processing time 30 minutes, starch concentration 2%	3.60
Processing time 30 minutes, starch concentration 3%	3.10
Processing time 60 minutes, starch concentration 1%	2.90
Processing time 60 minutes, starch concentration 2%	2.80
Processing time 60 minutes, starch concentration 3%	2.70
Processing time 90 minutes, starch concentration 1%	2.40
Processing time 90 minutes, starch concentration 2%	2.20
Processing time 90 minutes, starch concentration 3%	2.10

Score description: 1 = very unclear; 2 = not clear; 3 = not clear enough; 4 = clear; 5 = very clear.

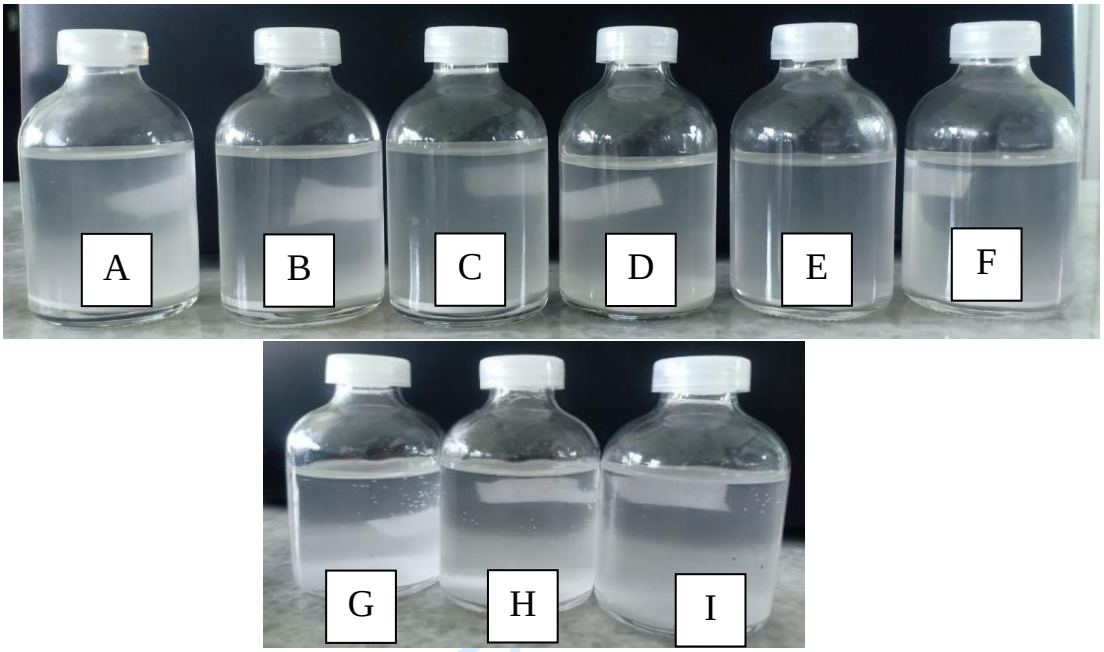

Figure 3. Clarity of SNP solutions at various ultrasonic process times and starch concentrations (A = 30 min, 1%; B = 30 min, 2%; C = 30 min, 3%; D = 60 min, 1%; E = 60 min, 2%; F = 60 min, 3%; G = 90 min, 1%; H = 90 min, 2%; I = 90 min, 3%)

The decrease in the clarity score is also directly proportional to the decline in the transmittance value. The smaller the particle size, the lower the transmittance value and the lower the clarity score. If a solution is passed by light, there will be a scattering of dissolved particles, which causes a reduction in transparency. This is closely related to the size of the particles dispersed in the solution. In solutions containing nano-sized granules, these granules are soluble so that the scattering effect becomes more significant, reducing the transmittance value of the solution and its clarity score.

CONCLUSION

Ultrasonic process time and starch concentration affect the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and SNP clarity score. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce SNP products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydipersity -spelling error index of 0.581, transmittance value of 61.27% , and a solution clarity score of 3.80 (not clear).

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Cassava Starch-based Starch Nanoparticles

Application of the Ultrasonic Method to Produce Starch Nanoparticles from Cassava Starch

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ABSTRACT

Starch nanoparticles have the potential to be developed as a cassava starch derivative. The research aims to obtain the optimal process conditions (ultrasonic process time and starch concentration) to produce starch nanoparticles with the best characteristics. The treatment variables used in this study were the duration of the ultrasonication process (30, 60, and 90 minutes) and the starch concentration (1%, 2%, and 3%). The results showed that the ultrasonication process time and starch concentration affected the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and clarity score of starch nanoparticles. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce starch nanoparticles products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27%, and a solution clarity score of 3.80 (not clear). To simplify the process, the development of SNPs based on tapioca can be prepared solely with the ultrasonic method.

Keywords: Cassava starch, starch nanoparticles, ultrasonic

INTRODUCTION

Starch is a natural, renewable, biodegradable polymer that many plants use as energy storage. Starch is the second most abundant biomass in nature and is found in staple crop commodities such as rice, corn, wheat, cassava, and potatoes (BeMiller & Whistler, 2009). The primary potential source of starch in Indonesia is cassava starch obtained from the cassava extraction process (Zukryandry et al., 2022). Based on data from the Food and Agriculture Organization (FAO) in 2012, Indonesia is the world's third exporter of tapioca, followed by Thailand and Vietnam (Hidayat et al., 2021). According to BPS (2022), Indonesia's cassava production in 2021 will be 19,341,233 tons, and Lampung Province, with a production of 6,683,758 tons, is the main producer of cassava in Indonesia (34.5%).

Starch nanoparticles (SNPs) have the potential to be developed as a tapioca derivative product. SNPs are nano-sized starch derivative products (one billionth of a meter, 10-9 meters) with a size range of 1–100 nm (EFSA Scientific Committee, 2011). The process of modifying starch into starch nanoparticles products has many advantages, including increasing stability, chemical reactivity, flowability, opacity, and mechanical strength (Zhu et al., 2007), improving the sensory characteristics of the product (Sharma et al., 2013), and enhancing encapsulation ability for bioactive components (Ezhilarasi et al., 2013).

Despite their potential, the development of SNPs based on tapioca is relatively limited and is mostly developed from corn starch (Le-Corre et al., 2010; Kim et al., 2013; Kumari et al., 2020) and rice starch (Zuo et al., 2012). Compared to corn starch and rice starch, cassava starch (tapioca) is a more economical source of starch in Indonesia. The development of SNPs based on tapioca will increase the added value of the tapioca industry.

The manufacture of SNPs can be carried out using various methods, namely, acid hydrolysis (Le-Corre et al., 2010), enzymatic hydrolysis (Le-Corre et al., 2012), high-pressure homogenization (Liu et al., 2016), gamma irradiation (Garcia et al., 2011; Lamanna et al., 2013), combination of acid hydrolysis and ultrasonication (Kim et al., 2013; Goncalves et al., 2014), and ultrasonication (Haaj et al., 2013). The research results by Haaj et al. (2013) showed that SNP products can be prepared solely with the ultrasonic method, so that it will simplify the manufacturing process.

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According to Jambrak et al. (2010), the ultrasonication process to produce SNPs can be carried out using an ultrasonic probe or a bath system. Compared to an ultrasonic system bath, the use of an ultrasonic system probe will be more effective with a shorter processing time (Bonto et al., 2021) and produce SNP products with better characteristics (Haaj et al., 2013). This study aims to obtain optimal process conditions (ultrasonic process time and starch concentration) to produce cassava starch-based starch nanoparticles with the best characteristics (yield, distribution and particle size, transmittance, and clarity).

MATERIALS AND METHODS

Equipment

The main tools used are Ultrasonication probe Biomaisen type MSUCD 650, UV-Vis single beam spectrophotometer Aelab type AE-S60-4U, and Particle Size Analyzer (PSA) Malvern Zetasizer Nano ZS type.

Starch Nanoparticle (SNP) Formation

The formation of SNPs from cassava starch was modified from method of Haaj et al. (2013) by preparing 50 ml of cassava starch solution with concentrations according to treatment (1%, 2%, and 3%). The probe temperature is set below 40^{o}C, kept constant by adding ice, and the process frequency is set at 20 kHz. The probe used has a diameter of 6 cm with an ultrasonic power of 650 W. The ultrasonication process is then carried out with the duration of the ultrasonication process according to the treatment (30, 60, and 90 min). The solution resulting from the sonification process was then filtered using 1-micron Whatman filter paper and tested for yield and characteristics.

Yield Analysis

The yield is the percentage of the dry weight of the SNP product divided by the initial weight of the starch raw material, with the following Equation 1:

$$\text{Yield (\%)} = \frac{mass\ of\ SNP\ (gram)}{mass\ of\ initial\ starch\ (gram)} \text{ x } 100\% \qquad (1)$$

Analysis of Particle Size

The distribution and size of SNPs were analyzed using a particle size analyzer (PSA) with the dynamic light scattering (DLS) method that utilizes infrared scattering. The SNP solution sample was put into the PSA cuvette. Infrared scattering was fired at the sample so that the sample would react to produce Brownian motion (random motion of the particles). This random motion then analyzed by the tool, where the smaller the particle size, the faster the movement.

In addition to the distribution and size of SNPs, the polydispersity Index (PI) value could also be obtained, a measure of molecular mass distribution in the sample. The PI value indicates the level of confidence in the size of the particles dispersed in a solution. The smaller the polydispersity value, the better the confidence level of the particle size distribution in the starch solution. Conversely, if the polydispersity value is higher, then the particles present in the sample are not uniform and unstable and would quickly flocculate.

Transmittance Analysis

Samples of SNP solution resulting from the sonication process of various treatments were put into the spectrophotometer cuvette. Analysis was conducted by placing a cuvette into a UV-Vis spectrophotometer with a 450–800 nm wavelength range. The results obtained were then recorded in the form of transmittance percentage values.

Clarity Analysis

Observation of the clarity of the SNP solution was carried out after being left for 2 hours (Haaj et al., 2013). The test was carried out sensory using 10 panelists using a hedonic score of 1–5 (score 1 = very unclear; score 2 = not clear; score 3 = not clear enough; score 4 = clear; score 5 = very clear).

RESULT AND DISCUSSION

Yield of starch nanoparticles

The yield of SNPs at various ultrasonic process times and starch concentrations is presented in Table 1. The test results in Table 1 show that the ultrasonication process duration of 90 minutes and 3% starch concentration will produce SNP products with higher yields (13.68%) than that of other treatments. The higher yield of this SNP

indicates that increasing the ultrasonication process time to 90 minutes and increasing the starch concentration to 3% will cause the breakdown of starch molecules into nano-sized to become more intensive. The yield of SNPs using the ultrasonic method (13.68%) is relatively the same as the acid hydrolysis method (15%) but lower than the combined acid and ultrasonic hydrolysis method, which can reach 78% (Kim et al., 2013).

The longer the ultrasonic process, the more intensive the degradation process of starch molecules. According to Czechowska-Biskup et al. (2005), the ultrasonic application will cause the degradation of starch molecules caused by mechanochemical effects. The more intensive the starch degradation process, the smaller the starch granule size.

The increase in SNP yields up to 3% starch concentration indicated that up to 3% starch solution concentration, the starch degradation process was still occurring intensively. A different opinion was conveyed by Haaj et al. (2013), which stated that the ultrasonication process without chemical treatment was effective at low concentrations (1-2%). The conditioning of starch in the form of an aqueous solution, not a suspension is based on the results of Czechowska-Biskup et al. (2005), which showed that the process of degradation of starch molecules was more effective in aqueous/solution conditions.

Table 1
Yield of SNP at various ultrasonication process times and starch concentrations (mean ± SD, n =3)

Treatment	SNP yield (%)
Processing time 30 minutes, starch concentration 1%	11.94 ± 0.02
Processing time 30 minutes, starch concentration 2%	13.18 ± 0.20
Processing time 30 minutes, starch concentration 3%	13.33 ± 0.18
Processing time 60 minutes, starch concentration 1%	12.02 ± 0.11
Processing time 60 minutes, starch concentration 2%	13.37 ± 0.17
Processing time 60 minutes, starch concentration 3%	13.56 ± 0.21
Processing time 90 minutes, starch concentration 1%	12.32 ± 0.23
Processing time 90 minutes, starch concentration 2%	13.66 ± 0.24
Processing time 90 minutes, starch concentration 3%	13.68 ± 0.05

Distribution and Particle Size of SNP

Distribution, particle size, and PI of SNPs at various ultrasonic process times and starch concentrations are presented in Table 2 and Figure 1. The results in Figure 1 show the percentage of SNP particle size at various particle sizes continuously using a particle size analyzer (PSA), while the results in Table 2 show the particle size in various particle size groups (≤100 nm, 101-1000 nm, and > 1000 nm).

The results in Figure 1 show that most of the SNPs are 101 to 1000 nm in size. This shows that the sonication process is quite effective in reducing the size of starch particles. According to Boufi et al. (2018) and Zuo et al. (2012), the ultrasonic method was able to damage and reduce the size of starch granules. The research results in Table 2 also show the presence of particles with a diameter of more than 1000 nm with a small intensity. Particles with a size of more than 1000 nm are thought to be starch particles that have agglomerated into a larger size. According to Jambrak et al. (2010), with changes in temperature and longer storage time, nanoparticles can agglomerate into larger sizes.

The results in Table 2 show that the ultrasonic process of starch with a concentration of 1–3% for 30–90 minutes will produce SNP products with a diameter range of 230.80 nm to 501.50 nm and a PI value range of 0.34–0.58 nm. The lowest PI was shown in the sonication time of 60 minutes with a starch concentration of 3% with a PI of 0.34 and an average particle size of 333.70 nm. The low PI indicates that the particle size dispersion of SNP is homogeneous and evenly distributed. A PI value greater than 0.70 indicates a very wide distribution of particle sizes so that sedimentation is likely to occur.

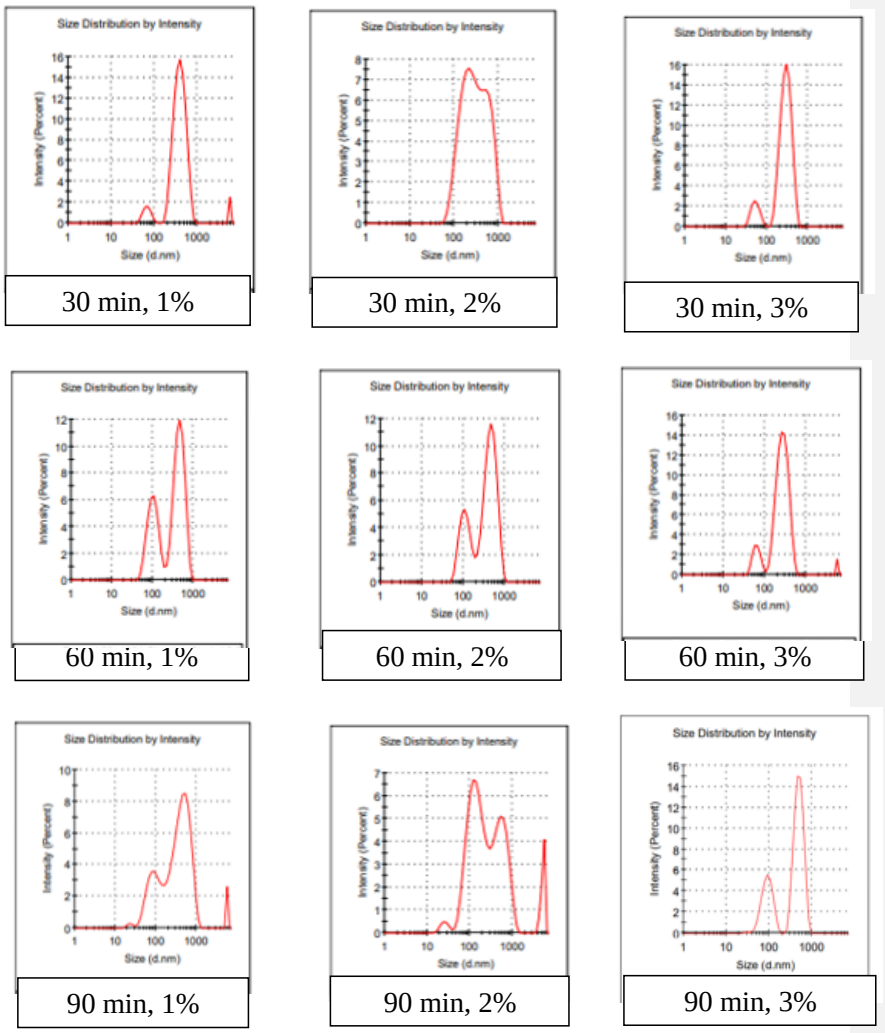

Figure 1. Distribution of various SNPs sizes at various ultrasonication process times and starch concentrations.

Table 2

Particle size distribution per size group and polydispersity index of SNPs at various ultrasonication process times and starch concentrations

Treatment	SNP Particle Size				Polydispersity index
	≤ 100 *nm* (%)	101 – 1000 nm (%)	> 1000 nm (%)	Average (nm)	
Processing time 30 minutes, starch concentration 1%	6.30	93.70	0.00	501.50	0.47
Processing time 30 minutes, starch concentration 2%	7.60	91.50	0.90	419.90	0,47
Processing time 30 minutes, starch concentration 3%	11.00	89.00	0.00	470.20	0.46
Processing time 60 minutes, starch concentration 1%	12.00	86.40	1.60	429.60	0.51
Processing time 60 minutes, starch concentration 2%	16.70	83.30	0.00	355.00	0.47
Processing time 60 minutes, starch concentration 3%	22.90	77.10	0.00	333.70	0.34
Processing time 90 minutes, starch concentration 1%	20.10	76.80	3.10	430.30	0.50
Processing time 90 minutes, starch concentration 2%	22.30	69.70	8.00	422.90	0.58
Processing time 90 minutes, starch concentration 3%	23.60	76.40	0.00	230.80	0.58

The results showed that the ultrasonication process duration of 90 minutes and 3% starch concentration would produce SNP products with a particle size of less than 100 nm, which was higher (23.6%) than that of the other treatments. The results also showed that the longer the sonification process and the higher the starch concentration, the higher the percentage of SNP particles less than 100 nanometers in size. This indicates that the ultrasonication process can break down starch granules into smaller sizes. The phenomenon of acoustic cavitation by ultrasonic waves causes starch particles to break into nano-sized pieces (Czechowska-Biskup et al., 2005). The increase in the percentage of SNP particle size in line with the increase in concentration up to 3% also shows that at a starch concentration of up to 3%; the cavitation process which causes the breakdown of starch granules into nano-sized still occurs effectively. The increase in the cavitation process in line with the increase in starch concentration in the formation of SNPs was also reported by Jambrak et al. (2010).

Starch Nanoparticles Transminttance Values

The transmittance value of SNPs at various ultrasonic process times and starch concentrations are presented in Table 3 and Figure 2. The results show that the ultrasonication process duration of 30 minutes and 1% starch concentration will produce SNP products with the highest transmittance values (86.38%). Conversely, the ultrasonication process time of 90 minutes and 3% starch concentration will produce SNP products with the lowest transmittance value (61.27%).

Table 3

Transmittance values of SNPs at various ultrasonic process times and starch concentrations (mean ± SD, n =3)

Treatment	Transmittance (%) at wavelength (nm)					Average transmittance (%)
	450	500	600	700	800	
Processing time 30 minutes, starch concentration 1%	85.31 ± 0.08	85.62 ± 0.04	86.78 ± 0.13	86.60 ± 0.65	88.34 ± 0.16	86.53 ± 0.20
Processing time 30 minutes, starch concentration 2%	75.17 ± 0.11	76.55 ± 0.13	77.37 ±0.44	77.96 ±0.42	80.16 ± 0.05	77.44 ± 0.14

Processing time 30 minutes, starch concentration 3%	66.72 ± 0.32	67.47 ± 0.64	69.27 ± 0.23	69.97 ± 0.34	72.00 ± 1.63	69.09 ± 0.52
Processing time 60 minutes, starch concentration 1%	82.37 ± 0.64	82.30 ± 0.36	83.25 ± 0.43	83.20 ± 0.19	83.83 ± 0.55	82.99 ± 0.34
Processing time 60 minutes, starch concentration 2%	69.65 ± 1.06	70.78 ± 0.40	72.58 ± 0.64	73.71 ± 0.18	76.88 ± 0.40	72.72 ± 0.30
Processing time 60 minutes, starch concentration 3%	62.74 ± 0.38	63.56 ± 0.51	64.76 ± 0.40	64.72 ± 0.13	66.48 ± 0.27	64.45 ± 0.15
Processing time 90 minutes, starch concentration 1%	78.97 ± 0.48	78.51 ± 0.30	78.16 ± 0.12	78.12 ± 0.43	78.47 ± 0.27	78.45 ± 0.29
Processing time 90 minutes, starch concentration 2%	64.91 ± 0.48	65.86 ± 0.65	67.27 ± 0.19	68.40 ± 0.28	70.22 ± 0.24	67.33 ± 0.31
Processing time 90 minutes, starch concentration 3%	58.29 ± 0.25	59.41 ± 0.41	61.09 ± 0.11	62.23 ± 0.23	64.23 ± 0.10	61.05 ± 0.22

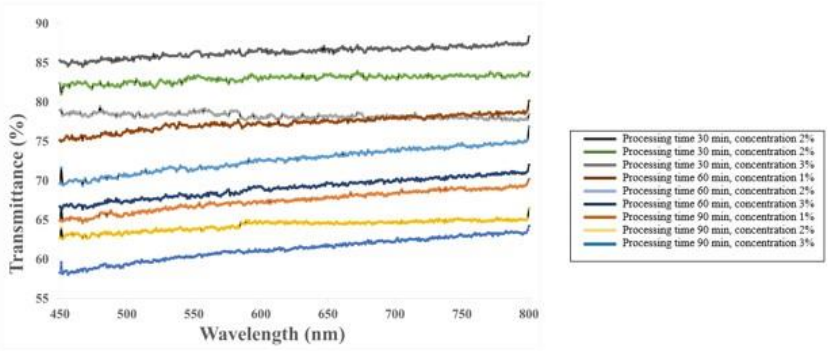

Figure 2. SNP transmittance curves for various ultrasonication process times and starch concentrations.

Ultrasonic process time of 90 minutes and concentration of 30% (Table 2, Figure 2) will produce SNPs with the lowest transmittance value compared to other treatments. The lower transmittance value of the SNP is strongly related to the size of

the SNP particles. The smaller the SNP particle size is, the more difficult it is for the starch particles to precipitate and the lower the transmittance value is. On the other hand, the larger the SNP particle size is, the faster the particles settle and the greater the transmittance value is. Changes in the transmittance of SNPs and a decrease in particle size were also reported by Bel Haaj et al. (2013) on SNP formation in corn starch. According to Haaj et al. (2013), SNPs with a size of more than 10 µm will precipitate quickly.

Starch Nanoparticles Clarity Score

The clarity score of SNPs at various lengths of the ultrasonication process are presented in Table 4 and Figure 3. The results show that the 90-minute ultrasonication process and 3% starch concentration will produce SNPs with the lowest level of clarity compared to other treatments. The lower clarity of the SNP is strongly related to the size of the SNP particles and their solubility. The smaller the SNP particle size, the lower the clarity of the SNP solution because the nano-sized SNP particles will dissolve and have difficulty settling even though they have been left for 2 hours. The increase in SNP solubility with the smaller particle size is mainly related to the increase in the porosity of starch granules (Sujka, 2017). Changes in the level of clarity of SNP solutions, along with a decrease in particle size, were also reported by Jambrak et al. (2010) and Kim et al. (2013) on SNP formation in corn starch.

Table 4
SNP clarity scores at various ultrasonic process times and starch concentrations (mean ± SD, n =10)

Treatment	SNP clarity score (%)
Processing time 30 minutes, starch concentration 1%	3.80 ± 0.13
Processing time 30 minutes, starch concentration 2%	3.60 ± 0.20
Processing time 30 minutes, starch concentration 3%	3.10 ± 0.27
Processing time 60 minutes, starch concentration 1%	2.90 ± 0.22
Processing time 60 minutes, starch concentration 2%	2.80 ± 0.08
Processing time 60 minutes, starch concentration 3%	2.70 ± 0.07
Processing time 90 minutes, starch concentration 1%	2.40 ± 0.13
Processing time 90 minutes, starch concentration 2%	2.20 ± 0.09
Processing time 90 minutes, starch concentration 3%	2.10 ± 0.21

Score description: 1 = very unclear; 2 = not clear; 3 = not clear enough; 4 = clear; 5 = very clear.

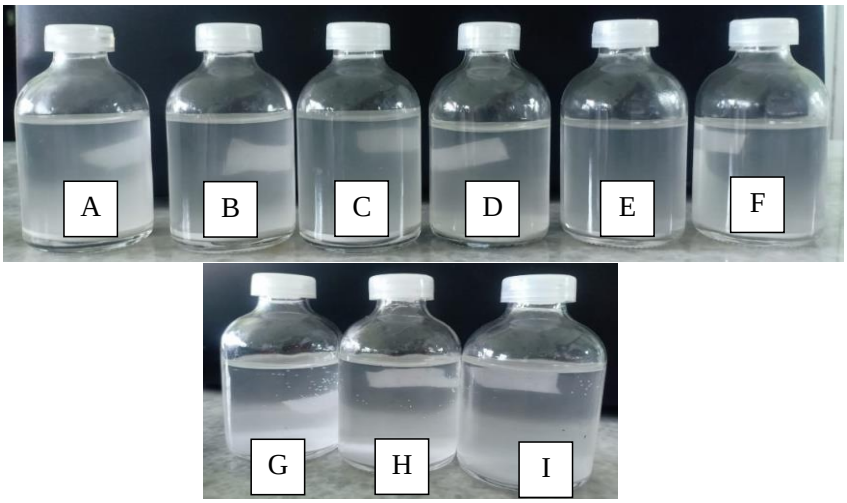

Figure 3. Clarity of SNP solutions at various ultrasonic process times and starch concentrations (A = 30 min, 1%; B = 30 min, 2%; C = 30 min, 3%; D = 60 min, 1%; E = 60 min, 2%; F = 60 min, 3%; G = 90 min, 1%; H = 90 min, 2%; I = 90 min, 3%)

The decrease in the clarity score is also directly proportional to the decline in the transmittance value. The smaller the particle size, the lower the transmittance value and the lower the clarity score. If a solution is passed by light, there will be a scattering of dissolved particles, which causes a reduction in transparency. This is closely related to the size of the particles dispersed in the solution. In solutions containing nano-sized granules, these granules are soluble so that the scattering effect becomes more significant, reducing the transmittance value of the solution and its clarity score.

CONCLUSION

Ultrasonic process time and starch concentration affect the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and SNP clarity score. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce SNP products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27% , and a solution clarity score of 3.80 (not clear).

To simplify the process, the development of SNPs based on tapioca can be prepared solely with the ultrasonic method. Further research is needed to improve the yield of SNPs based on tapioca.

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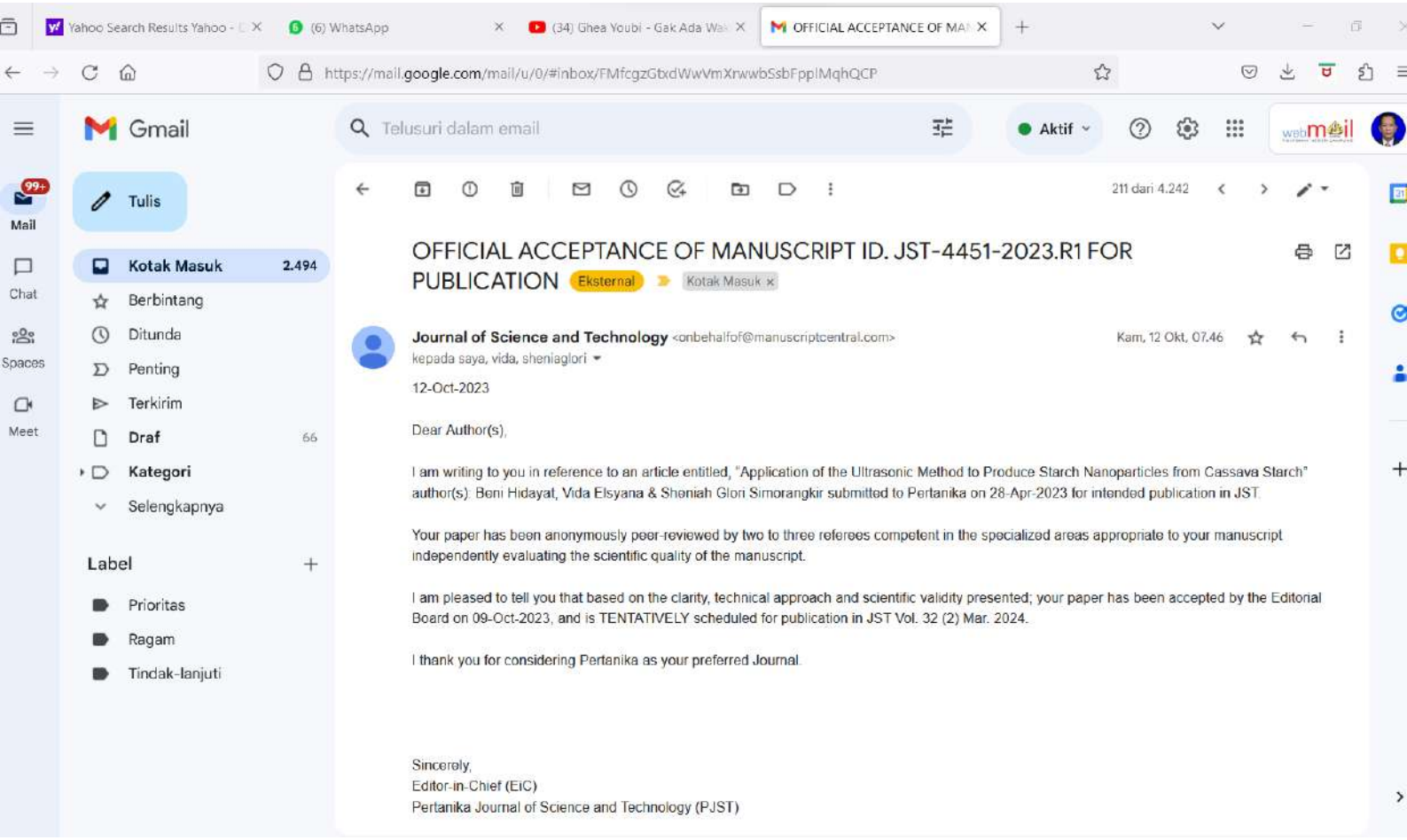

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Application of the Ultrasonic Method to Produce Starch Nanoparticles from Cassava Starch

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ABSTRACT

Starch nanoparticles have the potential to be developed as a cassava starch derivative. The research aims to obtain the optimal process conditions (ultrasonic process time and starch concentration) to produce starch nanoparticles with the best characteristics. The treatment variables used in this study were the duration of the ultrasonication process (30, 60, and 90 minutes) and the starch concentration (1%, 2%, and 3%). The results showed that the ultrasonication process time and starch concentration affected the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and clarity score of starch nanoparticles. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce starch nanoparticle products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27%, and a solution clarity score of 3.80 (not clear). Tapioca-based SNPs can be developed solely with ultrasonic method to simplify the process.

Keywords: Cassava starch, starch nanoparticles, ultrasonic

INTRODUCTION

Starch is a natural, renewable, biodegradable polymer many plants use to store energy. Starch is the second most abundant biomass in nature and is found in staple crop commodities such as rice, corn, wheat, cassava, and potatoes (BeMiller & Whistler, 2009). The primary potential source of starch in Indonesia is cassava starch obtained from cassava extraction (Zukryandry et al., 2022). Based on data

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from the Food and Agriculture Organization (FAO) in 2012, Indonesia is the world's third exporter of tapioca, followed by Thailand and Vietnam (Hidayat et al., 2021). According to BPS-Statistics of Lampung Province (2022), Indonesia's cassava production in 2021 will be 19,341,233 tons, and Lampung Province, with a production of 6,683,758 tons, is the main producer of cassava in Indonesia (34.5%).

Starch nanoparticles (SNPs) have the potential to be developed as a tapioca derivative product. SNPs are nano-sized starch derivative products (one billionth of a meter, 10-9 meters) with a size range of 1–100 nm (EFSA Scientific Committee, 2011). The process of modifying starch into starch nanoparticle products has many advantages, including increasing stability, chemical reactivity, flowability, opacity, and mechanical strength (Zhu et al., 2007), improving the sensory characteristics of the product (Sharma et al., 2013), and enhancing encapsulation ability for bioactive components (Ezhilarasi et al., 2013).

Despite their potential, the development of SNPs based on tapioca is relatively limited and is mostly developed from corn starch (Le-Corre et al., 2010; Kim et al., 2013; Kumari et al., 2020) and rice starch (Zuo et al., 2012). Compared to corn and rice starch, cassava starch (tapioca) is a more economical source in Indonesia. The development of SNPs based on tapioca will increase the added value of the tapioca industry.

The manufacture of SNPs can be carried out using various methods, namely, acid hydrolysis (Le-Corre et al., 2010), enzymatic hydrolysis (Le-Corre et al., 2010), high-pressure homogenization (Liu et al., 2016), gamma irradiation (Garcia et al., 2011; Lamanna et al., 2013), a combination of acid hydrolysis and ultrasonication (Kim et al., 2013; Goncalves et al., 2014), and ultrasonication (Haaj et al., 2013). The research results by Haaj et al. (2013) showed that SNP products could be prepared solely with the ultrasonic method, simplifying the manufacturing process.

According to Jambrak et al. (2010), the ultrasonication process to produce SNPs can be carried out using an ultrasonic probe or a bath system. Compared to an ultrasonic system bath, using an ultrasonic system probe will be more effective with a shorter processing time (Bonto et al., 2021) and produce SNP products with better characteristics (Haaj et al., 2013). This study aims to obtain optimal process conditions (ultrasonic process time and starch concentration) to produce cassava starch-based starch nanoparticles with the best characteristics (yield, distribution and particle size, transmittance, and clarity).

MATERIALS AND METHODS

Equipment

The main tools used are Ultrasonication probe Biomaisen type MSUCD 650, UV-Vis single beam spectrophotometer Aelab type AE-S60-4U, and Particle Size Analyzer (PSA) Malvern Zetasizer Nano ZS type.

Starch Nanoparticle (SNP) Formation

The formation of SNPs from cassava starch was modified from the method of Haaj et al. (2013) by preparing 50 ml of cassava starch solution with concentrations according to treatment (1%, 2%, and 3%). The probe temperature is set below 40°C, kept constant by adding ice, and the process frequency is set at 20 kHz. The probe used has a diameter of 6 cm with an ultrasonic power of 650 W. The ultrasonication process is then carried out with the duration of the ultrasonication process according to the treatment (30, 60, and 90 min). The solution resulting from the sonification process was then filtered using 1-micron Whatman filter paper and tested for yield and characteristics.

Yield Analysis

The yield is the percentage of the dry weight of the SNP product divided by the initial weight of the starch raw material, with the following Equation 1:

$$Yield\ (\%) = \frac{mass\ of\ SNP\ (gram)}{mass\ of\ initial\ \ starch\ (gram)} \times 100\% \qquad (1)$$

Analysis of Particle Size

The distribution and size of SNPs were analyzed using a particle size analyzer (PSA) with the dynamic light scattering (DLS) method that utilizes infrared scattering. The SNP solution sample was put into the PSA cuvette. Infrared scattering was fired at the sample so that the sample would react to produce Brownian motion (random motion of the particles). The tool then analyzes this random motion, where the smaller the particle size, the faster the movement.

In addition to the distribution and size of SNPs, the polydispersity Index (PI) value, a measure of molecular mass distribution in the sample, could also be obtained. The PI value indicates the level of confidence in the size of the particles dispersed in a solution. The smaller the polydispersity value, the better the particle size distribution confidence level in the starch solution. Conversely, if the polydispersity value is higher, then the particles present in the sample are not uniform and unstable and would quickly flocculate.

Transmittance Analysis

Samples of SNP solution resulting from the sonication process of various treatments were put into the spectrophotometer cuvette. Analysis was conducted by placing a cuvette into a UV-Vis spectrophotometer with a 450–800 nm wavelength range. The results obtained were then recorded in the form of transmittance percentage values.

Clarity Analysis

Observation of the clarity of the SNP solution was carried out after being left for 2 hours (Haaj et al., 2013). The sensory test was carried out using 10 panelists using a hedonic score of 1–5 (score 1 = very unclear; score 2 = not clear; score 3 = not clear enough; score 4 = clear; score 5 = very clear).

RESULT AND DISCUSSION

Yield of Starch Nanoparticles

The yield of SNPs at various ultrasonic process times and starch concentrations is presented in Table 1. The test results in Table 1 show that the ultrasonication process, with a duration of 90 minutes and 3% starch concentration, will produce SNP products with higher yields (13.68%) than other treatments. The higher yield of this SNP indicates that increasing the ultrasonication process time to 90 minutes and increasing the starch concentration to 3% will cause the breakdown of starch molecules into nano-sized to become more intensive. The yield of SNPs using the ultrasonic method (13.68%) is relatively the same as the acid hydrolysis method (15%) but lower than the combined acid and ultrasonic hydrolysis method, which can reach 78% (Kim et al., 2013).

The longer the ultrasonic process, the more intensive the degradation process of starch molecules. According to Czechowska-Biskup et al. (2005), the ultrasonic application will cause the degradation of starch molecules caused by mechanochemical effects. The more intensive the starch degradation process, the smaller the granule size.

The increase in SNP yields up to 3% starch concentration, indicating that up to 3% starch solution concentration, the starch degradation process was still occurring intensively. A different opinion was conveyed by Haaj et al. (2013), which stated that the ultrasonication process without chemical treatment was effective at low concentrations (1%–2%). The conditioning of starch in the form of an aqueous solution, not a suspension, is based on the

Table 1
Yield of SNP at various ultrasonication process times and starch concentrations (mean ± SD, n =3)

Treatment	SNP yield (%)
Processing time 30 minutes, starch concentration 1%	11.94 ± 0.02
Processing time 30 minutes, starch concentration 2%	13.18 ± 0.20
Processing time 30 minutes, starch concentration 3%	13.33 ± 0.18
Processing time 60 minutes, starch concentration 1%	12.02 ± 0.11
Processing time 60 minutes, starch concentration 2%	13.37 ± 0.17
Processing time 60 minutes, starch concentration 3%	13.56 ± 0.21
Processing time 90 minutes, starch concentration 1%	12.32 ± 0.23
Processing time 90 minutes, starch concentration 2%	13.66 ± 0.24
Processing time 90 minutes, starch concentration 3%	13.68 ± 0.05

results of Czechowska-Biskup et al. (2005), which showed that the process of degradation of starch molecules was more effective in aqueous/solution conditions.

Distribution and Particle Size of SNP

Distribution, particle size, and PI of SNPs at various ultrasonic process times and starch concentrations are presented in Table 2 and Figure 1. The results in Figure 1 show the percentage of SNP particle size at various particle sizes continuously using a particle size analyzer (PSA). In contrast, the results in Table 2 show the particle size in various particle size groups (≤100 nm, 101-1000 nm, and > 1000 nm).

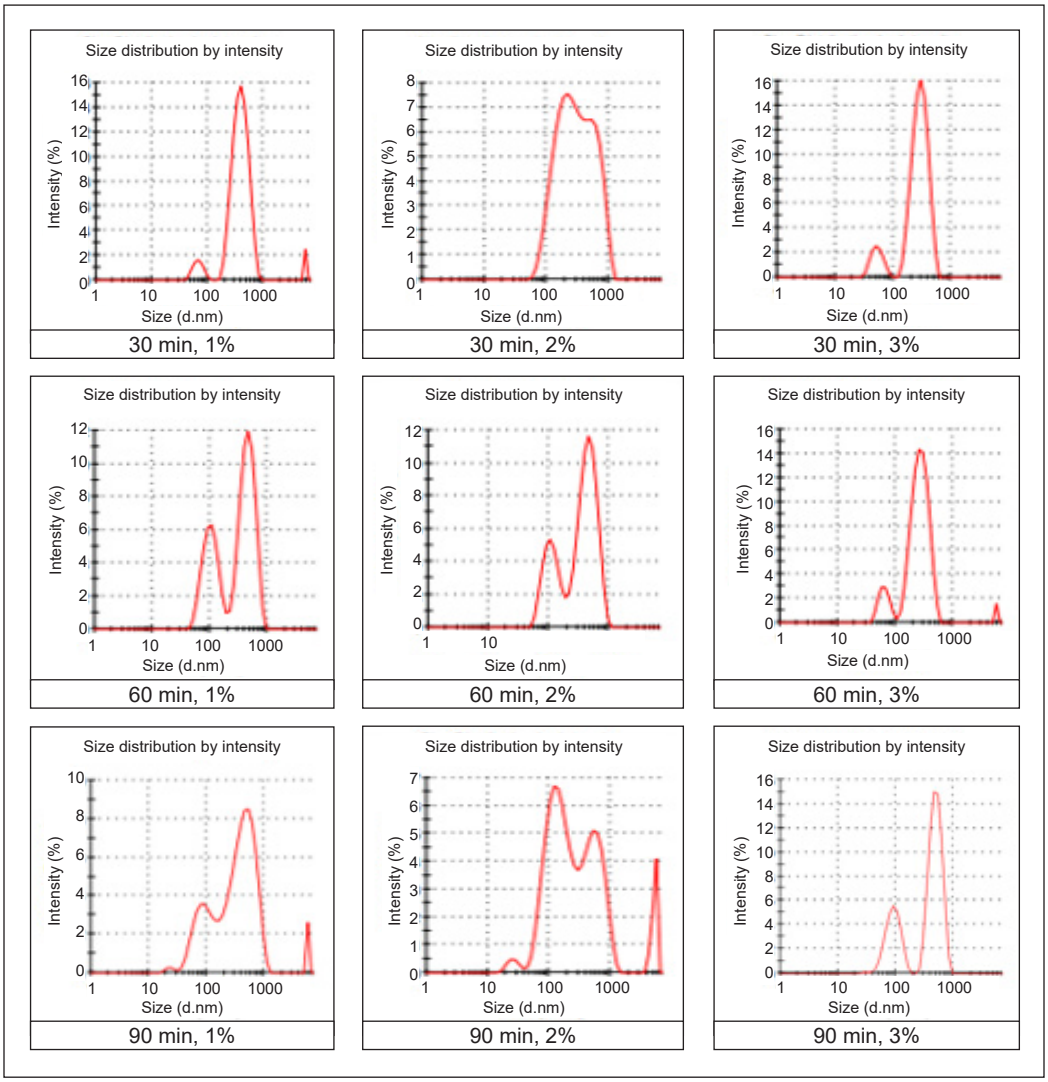

Figure 1. Distribution of various SNP sizes at various ultrasonication process times and starch concentrations

Table 2
Particle size distribution per size group and polydispersity index of SNPs at various ultrasonication process times and starch concentrations

Treatment	SNP Particle Size				Polydispersity index
	≤ 100 nm (%)	101 – 1000 nm (%)	> 1000 nm (%)	Average (nm)	
Processing time 30 minutes, starch concentration 1%	6.30	93.70	0.00	501.50	0.47
Processing time 30 minutes, starch concentration 2%	7.60	91.50	0.90	419.90	0,47
Processing time 30 minutes, starch concentration 3%	11.00	89.00	0.00	470.20	0.46
Processing time 60 minutes, starch concentration 1%	12.00	86.40	1.60	429.60	0.51
Processing time 60 minutes, starch concentration 2%	16.70	83.30	0.00	355.00	0.47
Processing time 60 minutes, starch concentration 3%	22.90	77.10	0.00	333.70	0.34
Processing time 90 minutes, starch concentration 1%	20.10	76.80	3.10	430.30	0.50
Processing time 90 minutes, starch concentration 2%	22.30	69.70	8.00	422.90	0.58
Processing time 90 minutes, starch concentration 3%	23.60	76.40	0.00	230.80	0.58

Most SNPs are 101 to 1000 nm in size showing, that the sonication process is quite effective in reducing the size of starch particles (Figure 1). According to Boufi et al. (2018) and Zuo et al. (2012), the ultrasonic method was able to damage and reduce the size of starch granules. The research results in Table 2 also show the presence of particles with a diameter of more than 1000 nm with a small intensity. Particles with a size of more than 1000 nm are thought to be starch particles that have agglomerated into a larger size. According to Jambrak et al. (2010), with changes in temperature and longer storage time, nanoparticles can agglomerate into larger sizes.

The results in Table 2 show that the ultrasonic process of starch with a concentration of 1%–3% for 30–90 minutes will produce SNP products with a diameter range of 230.80 nm to 501.50 nm and a PI value range of 0.34–0.58 nm. The lowest PI was shown in the sonication time of 60 minutes with a starch concentration of 3% with a PI of 0.34 and an average particle size of 333.70 nm. The low PI indicates that the particle size dispersion of SNP is homogeneous and evenly distributed. A PI value greater than 0.70 indicates a very wide distribution of particle sizes so that sedimentation is likely to occur.

The results showed that the ultrasonication process, with a duration of 90 minutes and 3% starch concentration, would produce SNP products with a particle size of less than 100 nm, which was higher (23.6%) than the other treatments. The results also showed that

the longer the sonification process and the higher the starch concentration, the higher the percentage of SNP particles less than 100 nanometers in size. It indicates that ultrasonication can break down starch granules into smaller sizes. The phenomenon of acoustic cavitation by ultrasonic waves causes starch particles to break into nano-sized pieces (Czechowska-Biskup et al., 2005). The increase in the percentage of SNP particle size in line with the increase in concentration up to 3% also shows that at a starch concentration of up to 3%, the cavitation process, which causes the breakdown of starch granules into nano-sized, still occurs effectively. The increase in the cavitation process, in line with the increase in starch concentration in the formation of SNPs, was also reported by Jambrak et al. (2010).

Starch Nanoparticles Transminttance Values

The transmittance value of SNPs at various ultrasonic process times and starch concentrations are presented in Table 3 and Figure 2. The results show that the ultrasonication process, with a duration of 30 minutes and 1% starch concentration, will produce SNP products with the highest transmittance values (86.38%). Conversely, the ultrasonication process time of 90 minutes and 3% starch concentration will produce SNP products with the lowest transmittance value (61.27%).

Ultrasonic process time of 90 minutes and concentration of 30% (Table 2, Figure 2) will produce SNPs with the lowest transmittance value compared to other treatments. The

Table 3
Transmittance values of SNPs at various ultrasonic process times and starch concentrations (mean ± SD, n =3)

Treatment	Transmittance (%) at wavelength (nm)					Average transmittance (%)
	450	500	600	700	800	
Processing time 30 minutes, starch concentration 1%	85.31 ± 0.08	85.62 ± 0.04	86.78 ± 0.13	86.60 ± 0.65	88.34 ± 0.16	86.53 ± 0.20
Processing time 30 minutes, starch concentration 2%	75.17 ± 0.11	76.55 ± 0.13	77.37 ±0.44	77.96 ±0.42	80.16 ± 0.05	77.44 ± 0.14
Processing time 30 minutes, starch concentration 3%	66.72 ± 0.32	67.47 ± 0.64	69.27 ± 0.23	69.97 ± 0.34	72.00 ± 1.63	69.09 ± 0.52
Processing time 60 minutes, starch concentration 1%	82.37 ± 0.64	82.30 ± 0.36	83.25 ± 0.43	83.20 ± 0.19	83.83 ± 0.55	82.99 ± 0.34
Processing time 60 minutes, starch concentration 2%	69.65 ± 1.06	70.78 ± 0.40	72.58 ± 0.64	73.71 ± 0.18	76.88 ± 0.40	72.72 ± 0.30
Processing time 60 minutes, starch concentration 3%	62.74 ± 0.38	63.56 ± 0.51	64.76 ± 0.40	64.72 ± 0.13	66.48 ± 0.27	64.45 ± 0.15
Processing time 90 minutes, starch concentration 1%	78.97 ± 0.48	78.51 ± 0.30	78.16 ± 0.12	78.12 ± 0.43	78.47 ± 0.27	78.45 ± 0.29
Processing time 90 minutes, starch concentration 2%	64.91 ± 0.48	65.86 ± 0.65	67.27 ± 0.19	68.40 ± 0.28	70.22 ± 0.24	67.33 ± 0.31
Processing time 90 minutes, starch concentration 3%	58.29 ± 0.25	59.41 ± 0.41	61.09 ± 0.11	62.23 ± 0.23	64.23 ± 0.10	61.05 ± 0.22

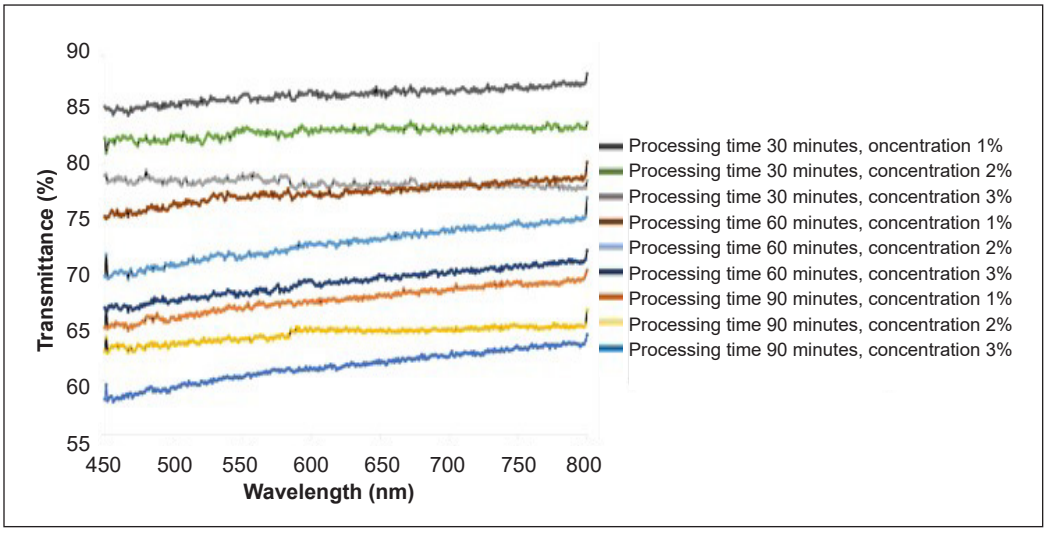

Figure 2. SNP transmittance curves for various ultrasonication process times and starch concentrations

lower transmittance value of the SNP is strongly related to the size of the SNP particles. The smaller the SNP particle size is, the more difficult it is for the starch particles to precipitate and the lower the transmittance value is. On the other hand, the larger the SNP particle size is, the faster the particles settle and the greater the transmittance value is. Changes in the transmittance of SNPs and a decrease in particle size were also reported by Bel Haaj et al. (2013) on SNP formation in corn starch. According to Haaj et al. (2013), SNPs with a size of more than 10 µm will precipitate quickly.

Starch Nanoparticles Clarity Score

The clarity scores of SNPs at various lengths of the ultrasonication process are presented in Table 4 and Figure 3. The results show that the 90-minute ultrasonication process and 3% starch concentration will produce SNPs with the lowest level of clarity compared to other treatments. The lower clarity of the SNP is strongly related to the size of the SNP particles and their solubility. The smaller the SNP particle size, the lower the clarity of the SNP solution because the nano-sized SNP particles will dissolve and have difficulty settling even though they have been left for 2 hours. The increase in SNP solubility with the smaller particle size is mainly related to the increase in the porosity of starch granules (Sujka, 2017). Changes in the level of clarity of SNP solutions, along with a decrease in particle size, were also reported by Jambrak et al. (2010) and Kim et al. (2013) on SNP formation in corn starch.

The decrease in the clarity score is also directly proportional to the decline in the transmittance value. The smaller the particle size, the lower the transmittance value and the clarity score. If a solution is passed by light, there will be a scattering of dissolved

particles, which causes a reduction in transparency. It is closely related to the size of the particles dispersed in the solution. In solutions containing nano-sized granules, these are soluble so that the scattering effect becomes more significant, reducing the transmittance value of the solution and its clarity score.

Table 4
SNP clarity scores at various ultrasonic process times and starch concentrations (mean ± SD, n =10)

Treatment	SNP clarity score (%)
Processing time 30 minutes, starch concentration 1%	3.80 ± 0.13
Processing time 30 minutes, starch concentration 2%	3.60 ± 0.20
Processing time 30 minutes, starch concentration 3%	3.10 ± 0.27
Processing time 60 minutes, starch concentration 1%	2.90 ± 0.22
Processing time 60 minutes, starch concentration 2%	2.80 ± 0.08
Processing time 60 minutes, starch concentration 3%	2.70 ± 0.07
Processing time 90 minutes, starch concentration 1%	2.40 ± 0.13
Processing time 90 minutes, starch concentration 2%	2.20 ± 0.09
Processing time 90 minutes, starch concentration 3%	2.10 ± 0.21

Score description: 1 = very unclear; 2 = not clear; 3 = not clear enough; 4 = clear; 5 = very clear

Figure 3. Clarity of SNP solutions at various ultrasonic process times and starch concentrations (A = 30 min, 1%; B = 30 min, 2%; C = 30 min, 3%; D = 60 min, 1%; E = 60 min, 2%; F = 60 min, 3%; G = 90 min, 1%; H = 90 min, 2%; I = 90 min, 3%)

CONCLUSION

Ultrasonic process time and starch concentration affect the yield, particle size and distribution, polydispersity index, optical characteristics (transmittance), and SNP clarity score. Ultrasonic process time of 90 minutes and starch concentration of 3% will produce SNP products with a yield of 13.68%, particle size ≤ 100 nm of 23.6%, average particle size of 230.8 nm with polydispersity index of 0.581, transmittance value of 61.27%, and a solution clarity score of 3.80 (not clear).

Tapioca-based SNPs can be developed solely with ultrasonic method to simplify the process. Further research is needed to improve the yield of SNPs based on tapioca.

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