Effect of Wet Milling and Purification Process on Yield and Color of Jackfruit Seed Starch

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Abstract. Jackfruit is a common fruit, which grows in Asian areas, including Thailand, India, China and Vietnam. Jackfruit seeds are a by-product of the food industry, and are typically ignored as a source of nutrients in the human diet given their possible application. Starch production is a conventional way to take advantage of this by-product source. The wet milling and purification process are the two main processes for extracting and improving the color of starch. Therefore, this research aims to determine the impact of wet milling and purification process on yield and color of starch as well as evaluate some parameters of physicochemical properties in jackfruit seed starch. In this study, the ratio of jackfruit seed : water (1:1, 1:2, 1:3, 1:4 and 1:5) and the time of wet milling (1, 2, 3, 4 and 5 min) are investigated. Three main factors observed in the purification process were the type of solvent (water and ethanol), solvent ratio (1:2, 1:4, 1:6 and 1:8) and the number of purification (1, 2, 3, 4 and 5 times). The result revealed that the ratio of jackfruit seed : water reached the highest starch yield was 1:3 (32.11±1.06%, db) and the time achieved 3 min (40.77±1.25%, db). The selected solvent for purification of starch was ethanol with the ratio of sample : water was 1:4 and the number of purification was two times for the best color. Some of characteristic nutrition values was evaluated, including %protein: 3.8, %ash: 0.08, %carbohydrate 86.99 and %lipid 0.20. The water solubility index and viscosity of starch were contradictory. When the temperature increased, the viscosity of starch decreased but the solubility index increased. Jackfruit seed starch was spherical or semi-oval which was presented by SEM (Scanning Electron Microscope) method.

1. Introduction
Jackfruit is one of the most famous cultivated fruits that is widely found in tropical and subtropical area, such as Malaysia, China, Thailand, Vietnam [1]. Jackfruit contains edible yellow flesh and seeds. The edible flesh are consumed fresh or processed into some of industrial product such as dried jackfruit, canned jackfruit or pie filler. Jackfruit seeds account for approximately 8-15% of fruit weight and have a short shelf-life due to their high carbohydrate content (starch content above 60%) [2]. However, seeds are normally disposed as waste in the processing of jackfruit or steamed to eat as a snack or used in some local food [3]. Due to their high starch concentration, the isolation of starch in jackfruit seed can
increase the value of jackfruit, reduce costs for environmental treatment and economic improvement [4].

In much work on starch separation from new sources as well as the production of new starch based products, the value of starch in the food and pharmaceutical industries has been reported [5-12]. 60% of the world’s starch are used in food industries and the 40% remaining starch are used in other industries as thickening, membrane forming, gel forming, microencapsulation and emulsifying agent [13]. Different natural origin of starch would create the unique properties, functions and applications. For example, the starch extracted from pineapple stem by grinning has high amyllose content and low viscosity, as compared to commercial starch [14]. Thus, the research for new starches are very important.

A number of studies have been conducted on jackfruit seed starch. For example, starch seeds from Brazilian jackfruit (Artocarpus heterophyllus L.) can be used as food product with similar morphological properties as cereal starches, with the soft seed had first and final gelatinisation temperature of 36 °C and 56 °C, respectively and the hard seed had initial and final gelatinisation at 40 °C and 61 °C [15]. Taewee Tongdang (2008) investigated some properties of starch derived from three Thai aromatic fruit seeds and found that seed starches from chempedak and jackfruit were identical in swelling flour, solubility, gelatinization temperature and enthalpy [16].

Milling or grinding and purification are important processes in starch productions. Milling is essential to damage protein and cell-wall matrices for the release of starch granules [17]. However, grinding was not selected as it causes structural damage to the starch [18;19]. Meanwhile, wet milling is a common methods to extract starch from raw material with minimal damage, cost efficiency and simplicity. Optimization work is required to reduce the amount of starch being lost during the process [20]. Purifications is the process of removing some of the non-starch components and improving the color of jackfruit seed starch. Overall, the conditions of wet milling and purification processes affect mainly the yield and quality of starch. Thus, this research aimed to investigate some effects of wet milling and purification on yield and color of starch. From that, we can determine suitable parameters for jackfruit seed starch production process.

2. Materials and methods

2.1 Materials

Jackfruit seeds (Artocarpus heterophyllus) at healthy state were received at Tien Giang province (Vietnam). The jackfruit seeds were washed and stored in refrigerator for starch production.

![Figure 1. Jackfruit seed used in this study](image)

2.2 Chemicals

Distilled water (pH between 6.5 and 8), diethyl ether (99.5% purity), Na₂CO₃ (99.5% purity), ascorbic acid (99.7% purity), NaHCO₃ (99.5% purity), FeSO₄.7H₂O (99.0-101.0% purity), CuSO₄.4H₂O (99.5%), K₂SO₄ (99.5% purity), H₂SO₄, NaOH.
2.3 Extraction of starch
Starch was extracted with some modifications from the jackfruit seeds according to the Luciano et al process [21]. The seeds were washed and dried in 24h at 45-50 °C to remove the white aril. Then, the thin brown skins covers were peeled and washed with water. The jackfruit seed starch was extracted by wet-milling method. Next, the ground slurry was screened and settled for about 16 hrs to remove protein in starch, and then removed the supernatant from the upper layers. Pick the starch from settle and centrifuged for 10 min at 5000 rpm to extract the starch water. After that, the starch was purified appropriated solvent and dried at 40 °C for 18 hrs then stored in a desiccators.

\[
Yield (\%) = \frac{weight\ of\ initial\ sample}{weight\ of\ jackfruit\ seed\ (db)}
\]  

(1)

2.4 Analytical method
2.4.1 Compositional analysis
The total protein was estimated according to the Kjeldahl system (Protein content = Nitrogen content x 6.25) [22]. Fat was determined by Soxhlet and Ash content was determined by calcination at 600 °C. Moisture was determined by an Infrared Moisture Determination Blace (Ohaus MB45) by weight 0.5 g of starch and measured at 104-105 °C for 2 min. Total carbohydrate content was calculated by following formula [23]:

\[
%\ Carbohydrate = 100 - (\%\ moisture + \%\ ash + \%\ protein + \%\ fat)
\]  

(2)

2.4.2 Physicochemical properties
Surface morphology of starch granule was observed on Scanning Electronic Microscope (SEM)
Swelling and solubility was determined according to Nisit and CS (2011) [24]. 2 g of starch was dispersed with 100 mL distilled water and heated at 30, 40, 50, 60, 70 and 80 °C. Then, it was centrifuged at speed of 5000 rpm for 10 min. The supernatant was dried at 105 °C to constant weight. The solubility was ratio of weigh of soluble starch and starch sample. After centrifugation on weight of dried starch, the swelling strength was starch weight. Viscosity was calculated with a several acceptable modifications according to Szychaj et al [25]. 10 g of starch was dispersed with 200 mL distilled water and heated to completely gelatinize. Viscosity was measured from 30 to 70 °C with NDJ-8S Viscosimeter (China).

2.4.3 Determination of Color
Color evaluation was conducted by using a Konica Minolta CR-400 and expressed in term of L* (brightness ranges from 0-100), a* (from green to red) and b* (from blue to yellow). The value of whiteness was obtained by following equation [26]

\[
Whiteness = 100 - [(100 - L*)^2 + a*^2 + b*^2]^{1/2}
\]  

(3)

2.4.4 Analysis of data
The results were expressed as mean ± standard deviation (S.D.) determined using Microsoft Excel (Microsoft Inc., Redmond, WA, USA) software. The experimental results were evaluated using One-way Analysis of Variance (ANOVA) study in the Statgraphics system with a significance interval of 95%.
3. Results and discussion

3.1 Some of approximate composition of fresh and starch jackfruit seed

Some of composition of fresh and starch jackfruit seeds in this study were determined. These results were presented in table 1.

Table 1: Approximate composition of fresh seed and seed starch samples

| Parameters     | Fresh jackfruit seeds | Jackfruit seed starch |
|----------------|------------------------|-----------------------|
| Moisture       | 52.785 ±0.44           | 8.99 ± 0.24           |
| Ash            | 1.46 ±0.01             | 0.08 ± 0.01           |
| Lipid          | 0.935 ± 0.03           | 0.215 ± 0.02          |
| Total protein  | 27.21 ± 0.07           | 3.79 ± 0.18           |
| Carbohydrate   | 17.60 ± 0.04           | 86.99 ± 0.20          |

Table 1 showed that approximate composition of fresh and starch samples, including moisture, ash, lipid, total protein and carbohydrate. The carbohydrate in jackfruit seeds account for 17.60 ± 0.04% and increase to 86.99 ± 0.29 in Jackfruit seed starch. The quiet low ash, total protein and lipid content was due to starch extraction and purification process which removed partially the non-starch components. The starch had a weak moisture value (8.99±0.25%) which is acceptance for marketing and storage [27]. The different chemical composition of the sample was very specific, owing to techniques used to isolate and examine the starch and its botanical properties.

3.2 Investigating the effect of the ratio of samples : water in wet milling on yield

Figure 2 presented that the starch yield in wet milling at different ratio of sample and water. The result showed that the starch yield tended to increase from 1:1 (21.76 ± 0.27%) to 1:3 (32.11 ± 1.06%). However, there was no significant different of starch yield from 1:3 to 1:5 with ANOVA test at a level of significance of 0.05.

Figure 2. The effect of the ratio of sample and water in wet milling on starch yield

The yield of jackfruit starch was influenced by many factors, such as variety, planting environment, moisture content of materials and extraction methods. Wet milling is a popular methods in the extraction of starch and the ratio of solvent and sample were one of the factors which affected the milling ability. The high solvent ratio could increase the chance of starch in jackfruit seeds contact with water and could
increase a diffusion rate, resulting in an increase in starch yield. However, the yield starch would not continue to increase after reaching balance.

3.3 Investigating the effect of the time in wet milling on yield

Figure 3 showed that the starch yield in wet milling at different times. The result of Figure 2 noticed that the more the wet milling time increase, the more the yield starch increase. The starch yield was 32.08±0.56% at 1 min and increased to 41.11±1.96% at 5 min. Meanwhile, there were no significant difference between 1 min and 2 min as well as from 3 min to 5 min.

![Figure 3](image)

Figure 3. The effect of the time (min) in wet milling on yield starch

Starch yield improved during the milling process, which may be attributed to a higher breakdown of cellulosic materials, thereby producing more starch from the cells. However, the amount of starch increased to 40.77±1.25 at 3 min and then became saturated. This is probably because the particle size of the material could not be smaller. Moreover, amount of starch that was tightly bound to the sample, could not release so the yield starch did not change.

3.4 Investigating the effect of the solvent ratio on color starch

Figure 4 showed the effect of the type of solvent in purification process on the whiteness of starch through L, a* and b* parameter with L* value is the light (100) and dark (0) correlation in sample, a* value range from +a* (green) to –a* (red) and b* value range from +b* (yellow) to –b* (blue). Such findings revealed that the form of solvent influenced starch color in the purification process. The whiteness of starch jackfruit seed was 87.60±0.18, 88.68±0.16 and 89.73±0.23% with not purification, H2O and ethanol, respectively. The whiteness of starch may be affected by some coloring agent such as beta carotene pigment. Meanwhile, ethanol can extract colorants better than H2O so the whiteness of the starch purified with ethanol was the highest.

![Figure 4](image)

Figure 4. The effect of the type of solvent on the color of seed starch.
3.5 Investigating the effect of the solvent ratio on color starch

The effect of solvent ratio to whiteness of starch was presented in Figure 5. When the ethanol content increased, the whiteness of starch tended to increase. However, there was no significant different from 1:4 to 1:8 ratio. The lowest whiteness of starch was 89.41±0.06 and the highest whiteness of starch was 89.73±0.18. High solvent content increased the exposure of solvent and colorant, resulting in discoloration of the compound also increased.

![Figure 5](image)

**Figure 5.** The effect of the solvent ratio on color of seed starch

3.6 Investigating the effect of the time purification on color starch

Figure 6 showed the effect of the time purification on whiteness of starch. The result noticed that the number of the purification increased, the color of jackfruit seed starch became whiter. Whiteness of starch were 88.82±0.23 at 1 time purification and increased to 89.76±0.14 at 5 time purifications. According to One-way ANOVA and LSD comparison, there was no significant different at the number of purification from 2 time to 5 time. The more number of purification the starch were, the more whiteness index were. However, the whiteness index of jackfruit seed starch could not be increased anymore when it reached the critical point. Differences in color parameter of starches could be due to genetic, climate and agronomic factors. The whiteness index of tapioca starch was 87%, pigeon pea starch was 91.39. To meet consumer preference, more whiteness is the desired feature of starch as an ingredient in food products such as cake and puddings.

![Figure 6](image)

**Figure 6.** The effect of the time purification on color starch

3.7 Surface morphology and particle size distribution

Results from scanning electron microscopy (SEM) were presented in Figure 7. The grain shape and size are the basic properties of starches. Different particle sizes will result in different physicochemical properties such as gelatinization temperature, solubility, swelling powders, viscosity, and methylene blue absorption capacity. The jackfruit seed starch had half-oval or spherical shape that was free
flowable, without stomata on the starch granule. The particle size of jackfruit seed starch was not uniform. The maximum diameter of starch granules was between 8 and 10 µm. The result of this experiment are similar to some of the previous report [16, 20].

![Figure 7](image1.png)

**Figure 7.** Scanning electron microscopy (SEM) of starch from native jackfruit seed at (A) x500, (B) x1000, (C) x3000 and (D) x5000 magnification level.

### 3.8 Swelling powder, solubility and viscosity

Figure 8 revealed that jackfruit starch has viscosity, swelling powder, and solubility. The result showed that the higher the starch temperature decreased the starch viscosity. Meanwhile, the swelling powder and solubility increased.

![Figure 8](image2.png)

**Figure 8.** Viscosity, swelling powder and solubility of native jackfruit seed starch

Viscosity is a very critical parameter that governs starch applicability in both food industries and other field [9]. High viscosity is an inherent advantage of starch in different applications but it also make disadvantage in some cases. The viscosity of jackfruit seed starch was around 3 Pas at 30 °C and decreased about 2 Pas at 70 °C. The solubility and swelling powder of starch were affected by particle size and temperature. A temperature increase has created intermolecular bonding disruptions (hydrogen interaction), and the opening of the chains allows water substances to penetrate. When the starch gelatinization temperature has been reached, the starch granule only has limited swelling which is solubilized by a quantity of carbohydrate. However, there was an increase swelling powder at the temperature gelatinization.
4.0 Conclusion
Wet milling and purification process of jackfruit seed starch was investigated. This result showed that the ratio of jackfruit seed: water reached the highest starch yield was 1:3 (32.11 ± 1.06%, db) and the time achieved 3 min (40.77 ± 1.25%, db) in wet milling. All investigated factors in the purification affected the color of jackfruit seed starch strongly. The selected solvent for purification of starch was solvent with the ratio of carbohydrate: water was 1:4, and the number of purification was two times for the best color. Jackfruit seed starch was spherical or semi-oval and starch granules had an average diameter of between 8 and 10 μm. The water solubility index and viscosity of starch were contradictory. When the temperature increased, the viscosity of starch decreased but the solubility index increased. The result of this work would promote the processing of starches from jackfruit seeds to increase both the value of jackfruit and economic growth.

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References
[1] Trinh P T N, Giang B L, Danh T T, Hung Q T, Du C V, Tuan N T and Dung L T 2020 IOP Conf. Ser. Mater. Sci. Eng. 736 022079
[2] Tran N Y T, Nhan N P T, Thanh V T, Chinh N D, Tri D L, Nguyen D V, Vy T A, Truc T T and Thin P V 2020 IOP Conf. Ser. Mater. Sci. Eng. 736 022065
[3] Trinh P T N, Nguyen T Q, Hau N V, Hung Q T, Du C V, Tuan N T, Thuy N T L, Dung L T 2020 IOP Conf. Ser. Mater. Sci. Eng 736 022080
[4] Nguyen M T, Nguyen V T, Minh L V, Trieu L H, Cang M H, Bui L M, Le X T and Danh V T 2020 IOP Conf. Ser. Mater. Sci. Eng 736 062011.
[5] Nguyen V T, Nguyen M T, Tran Q T, Thin P V, Bui L M, Le T H N, Le V M and Linh H T K 2020 IOP Conf. Ser. Mater. Sci. Eng 736 022063.
[6] Prado H J, Matulewicz M C, Bonelli P R and Cukierman A L 2009 Carbohydr. Res. 344 1325–31
[7] Carmona-garcia R, Sanchez-rivera M M and Méndez-montealvo G 2009 Carbohydr. Polym. 76 117–22
[8] Tran Q T, Le T T T, Pham M Q, Do T L, Vu M H, Nguyen D C, Bach L G, Bui L M and Pham Q L 2019 Molecules 24 895
[9] Pham T N, Tran B P, Tran T H, Nguyen D C, Nguyen T N P, Nguyen T Q, Vo D V N, Le X T, Nguyen D T and Bach L G 2019 IOP Conf. Ser. Mater. Sci. Eng. 479 012012
[10] Minh N P, Trang T H P, Trang N T T and Bach L G 2019 Res. Crops 20 1 180-186
[11] Mai H C, Nguyen T S V, Le T H N, Nguyen D C and Bach L G 2019 Processes 7 90
[12] Thanh V T, Tran N Y T, Linh N T V, Vy T A, Truc T T 2020 IOP Conf. Ser. Mater. Sci. Eng. 736 062014
[13] Jobling S 2004 I Curr. Opin. Plant Biol. 7 210–8
[14] Nakthong N, Wongsaongsup R and Amornsakchai T 2017 Characteristics and potential utilizations of starch from pineapple stem waste Industrial Crops and Products 105 74–82
[15] Suely M, Samara F, Albuquerque M De, Rafaela I, Silva A, Silva D, Magnani M and Queiroga V 2014 Food Chem. 143 440-5
[16] Tongdang T 2008 Starch - Stärke 60 199–207
[17] Gidley M J, Hanashiro I, Mohd N, Hill S E, Huber A, Jane J, Liu Q, Morris G A, Rolland-sabaté A, Striegel A M and Gilbert R G 2010 Carbohydr. Polym. 79 255–61
[18] Hasjim J and Jane J 2009 J. Food Sci. 74 556–62
[19] Kumar C S, Malleshi N G, Bhattacharya S, Kumar C S, Malleshi N G and Bhattacharya S 2008 Int. J. Food Prop. 11 845–57
[20] Syahariza Z A, Li E and Hasjim J 2010 Carbohydr. Polym. 82 14–20
[21] Sobral A, Cristina I and Moraes F 2017 Starch - Stärke 69 11–2
[22] Nguyen V T, Le V M, Vo T S, Bui L M, Anh H L T, Danh V T D 2020 Preliminary phytochemical screening and determination of total polyphenols and flavonoids content in the leaves of Houttuynia cordata Thunb, IOP Conf. Ser. Mater. Sci. Eng. 736 062013
[23] Mariotti F, Tomé D and Mirand P P 2008 Converting Nitrogen into Protein—Beyond 6.25 and Jones’ Factors Critical Reviews in Food Science and Nutrition 48 177–84.
[24] Kittipongpatana N, Janta S and Kittipongpatana O 2014 Pharm. Sci 24 415-420.
[25] Lvi T, Spychaj T, Zdanowicz M, Kujawa J and Schmidt B 2013 Polimery 58 501–630
[26] Naknaen P 2014 Food Biophys. 9 249–59
[27] Nwokocha L M, Nwokocha K E and Williams P A 2012 Starch - Stärke 64 246–54