Effect of ball milling time on structural properties and colour analysis of calamansi (*Citrus microcarpa*) peel powder

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ABSTRACT Calamansi (*Citrus microcarpa*) is a common lime widely used in food flavouring, herbal medicines and juice processing. The peel, which is the primary by-products of the juice processing industry has the potential to be utilized in powder form. This resulting powder offers versatility for use in both food and non-food applications. Therefore, drying and milling is essential unit operations for food powder production. In this study, the effect of ball milling time (30 to 120 minutes) on the morphology, structure and colour of freeze dried calamansi peel powder was investigated. The results demonstrate that extending the ball milling time from 30 to 120 minutes transforms large fragments into smaller particles, while maintaining structural integrity and enhancing the green colour of freeze-dried calamansi peel powder. These findings indicate the effective change in particle size and the resulting changes in the material's physical properties, allowing for tailored utilisation and application across a wide range of applications.

KEYWORDS: Calamansi Peel; Powder; Ball Milling; SEM; XRD.
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INTRODUCTION

Calamansi (*Citrus microcarpa*) is the hybrid species of Citrus reticulata and Citrus japonica and its small round shape fruit is well-known for various health benefits (Venkatachalam *et al*., 2023). In Malaysia, calamansi production is about 12,757 metric tonnes in 2019 and this figure indicates a rising pattern from the previous year (Department of Agriculture Malaysia, 2021). Calamansi has been a source of herbal medicine and is also commonly used for culinary purposes. Meanwhile in production sector, it is widely used in juice extraction, which mainly utilized the pulp and juice, leaving the peels as waste. The waste disposal is detrimental to the land and the environment (Girotto *et al*., 2015). Nevertheless, the peels still contain valuable compounds and nutrients in them that can be further utilized (Singh *et al*., 2020).

Transforming by-products into powdered form represents a proactive strategy to address environmental concerns while also establishing new revenue streams in the food industry (Chitrakar *et al*., 2023, Lazăr *et al*., 2022). In recent study, Thumwong *et al*. (2023) emphasized the potential benefits of peel powder as a source of natural antioxidants and a reinforcing bio-filler, owing to its substantial lignocellulosic biomass content. The peel is also an excellent candidate for enhancing the structural integrity and functional qualities of biofilms due to its high cellulose, soluble carbohydrate, and pectin contents. Previous research has demonstrated the usefulness of by-product peel as a filler, particularly suitable for coating applications and food packaging (Rojas-Bravo *et al*., 2019, Hanani *et al*., 2019). Peel powder is also widely employed as a functional food ingredient, especially in the flour, bakery, and confectionery industries (Mai *et al*., 2022, Belose *et al*., 2021). It is also reported to serve as a food seasoning, food colourant, flavouring agent and food additives (Espinosa-Garza *et al*., 2018, Wedamulla *et al*., 2022).

Ball milling is an efficient food processing technology capable of producing peel powders. This mechanical technique has been widely used in various industries worldwide, as it is deemed environmentally-friendly and cost-effective nature (Piras *et al*., 2019). During the milling process, the centrifugal force propels the balls to collide forcefully with both the container walls and the sample, initiating a dynamic interaction that leads to collisions and attrition (Chitrakar *et al*., 2023). In many cases, the reduction of particle size by ball milling results in an increased surface area, leading to enhanced physicochemical properties such as colour, structural attributes, solubility, water-holding capacity, bulk density, and chemical reactivity (Chitrakar *et al*., 2020, Wang *et al*., 2020).

The purpose of the present study was to study the effect of milling time on the morphology, structure and colour of calamansi peel powder, thereby contributing to a more comprehensive understanding of the changes occurring at both the structural and visual levels.

METHODOLOGY

Sample Preparation

Fresh calamansi fruits were sourced from a local market in Kuala Nerus, Terengganu, Malaysia. The peel was separated from the rest of the fruit and thoroughly washed using tap water to remove any dirt, debris, or surface contaminants that might be present. Afterward, the peel was dried in a freeze dryer (Labconco, FreeZone, Kansas City, MO, USA) for 24 hours.

Milling Process

The freeze-dried calamansi peel was milled using a planetary ball mill (XQM-0.4 L, Kexi, China). The milling process was conducted at room temperature for four different durations: 30, 60, 90, and 120 minutes, which are considered short-time ball milling (Nasrullah et al., 2021). The speed of the milling was performed at 800 rpm using 15 stainless steel milling balls with a diameter of 10 mm, which were selected based on a preliminary investigation.

Morphologies Observation

The morphologies of the calamansi peel powder were observed using a scanning electron microscope (SEM) (JEOL, JSM 6360 LA, Japan). Prior to SEM analysis, the samples were coated with gold particles using a gold coater (JEOL, JEE 420, Japan) to form a thin layer that enhanced thermal conduction and secondary electron detection. Images of the samples were captured at a magnification of 3000×.

Structure Determination

The structure and phase formation of the calamansi peel powder at different milling times were characterized using X-ray diffraction (XRD) (Rigaku, Miniflex II, Japan) with Cu Kα radiation (wavelength of 1.54 Å). The sample was placed on a sample holder with a step size of 0.02°, and a 2θ angle range of 5° to 80° was scanned directly at a rate of 2° per minute

Colour Measurement

The colour of both the fresh peel and the milled samples was measured using a colourimeter. (FRU WR-10QC, Shenzhen Wave Optoelectronics Technology Co Ltd, Shenzhen, China). The CIELAB parameters L^*, a^*, and b^* were used to describe the colour for each sample. In this context, L^* represents the lightness, ranging from 0 for black to 100 for white, while a^* represents the green-red coordinate, being green when negative and red when positive. Similarly, b^* signifies the blue-yellow coordinate, appearing blue when negative and yellow when positive. The total colour change (ΔE^*) were determined by using the following (Equation 1):

$$\Delta E^* = \sqrt{(a^* - a_0{}^*)^2 + (b^* - b_0{}^*)^2 + (L^* - L_0{}^*)^2} \quad (1)$$

where subscript $_0$ represent to the colour of fresh sample.

RESULT AND DISCUSSION

Microstructure Observation

The morphology of calamansi peel powder milled at different times is presented in Figure 1. It can be observed that the microstructure of the peel powder exhibited an irregular shape with large fragments after 30 minutes. Meanwhile, extending the milling duration resulted in smaller particle sizes. The increase in ball milling time not only elevated the prevalence of smaller particle fractions due to particle breakage but also resulted in a diverse range of calamansi peel particle shapes. The analysis indicates that after 120 minutes of milling, the particle size reached a minimum of d_{min} = 0.511 µm. This reduction in particle size with increased milling time can be attributed to increased shear stress during the milling process. This observation aligns with previous findings by Hong *et al*. (2021) and Ramachandraiah and Chin (2016). Generally, smaller particle size perform better in terms of solubility, mixing, dispersibility, and flowability (Aman *et al*., 2021), contributing to more efficient processes and favourable outcomes across diverse applications.

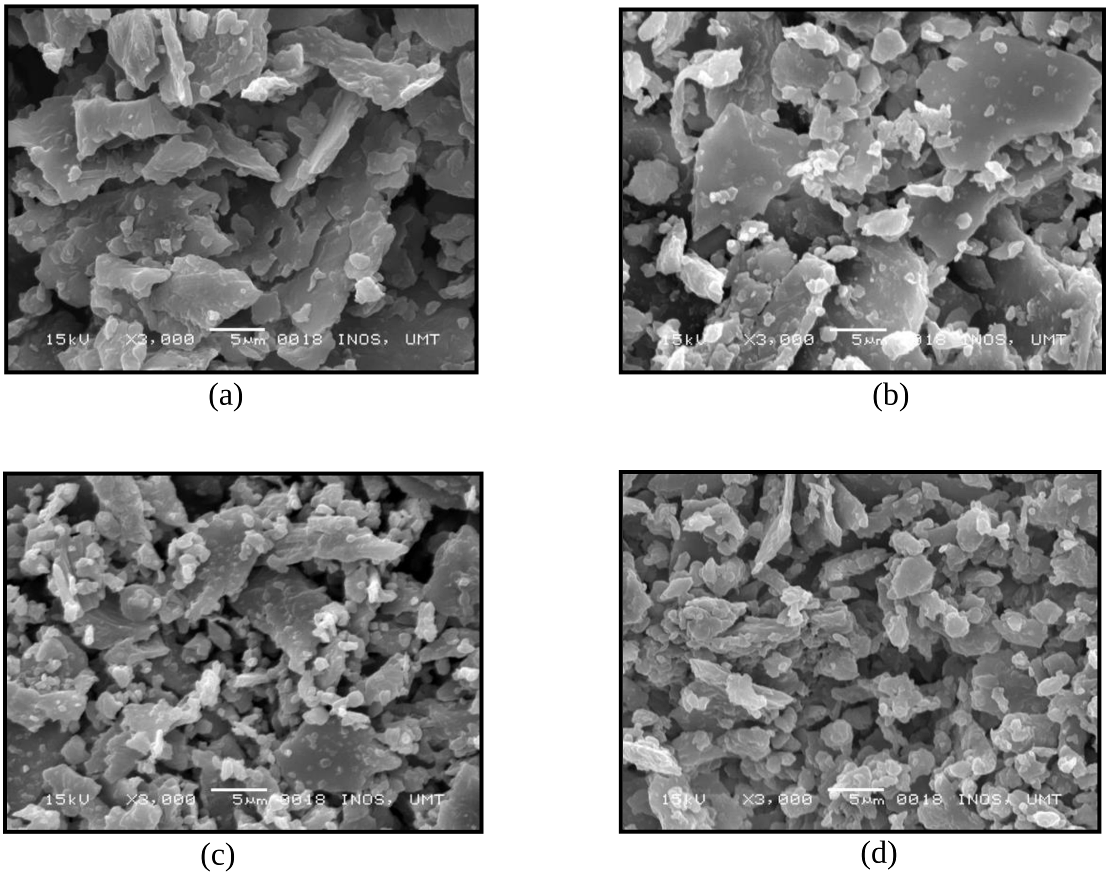

Figure 1. Scanning electron microscope micrograph (SEM) of calamansi peel powder treated for different milling times. (a) 30 minutes; (b) 60 minutes; (c) 90 minutes; and (d) 120 minutes.

Structural Properties

Figure 2 depicts the diffraction spectra of freeze-dried calamansi peel, both before and after milling. It is evident that amorphous humps are present in the XRD patterns. Additionally, the peak intensity did not undergo significant change. These results suggest that the milling process applied to calamansi peel for durations ranging from 30 to 120 minutes did not induce significant damage to the crystalline region. Similar findings regarding the preservation of structural integrity under the influence of milling were reported by Aman *et al*. (2021) for ginger powder and ElBeltagy *et al*. (2022) for pomegranate peel powder.

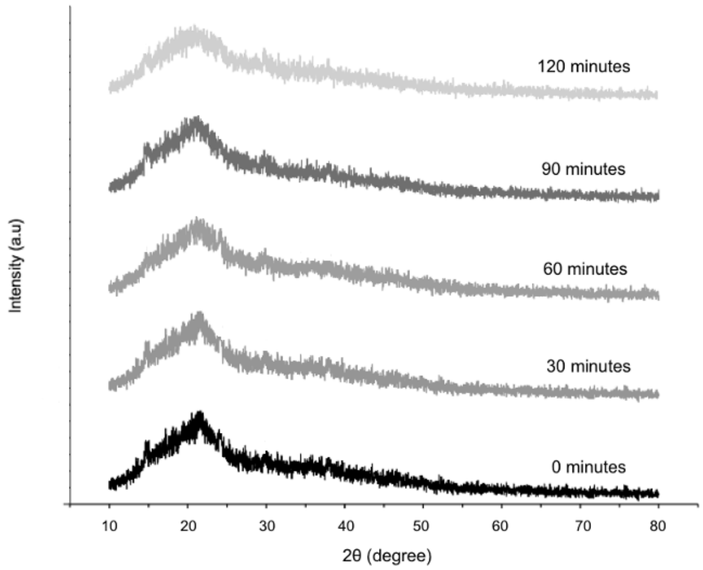

Figure 2. X-ray diffraction spectrum of calamansi peel powder treated for different milling times.

Colour Analysis

The results of both fresh samples and the freeze-dried powder samples of various milling times are presented in Table 1. When comparing the freeze-dried powder to the fresh sample, it is notable that the freeze-drying process results in a more vibrantly coloured product. This phenomenon has also been previously reported by Guiné and Barroca (2012), Ishwarya and Anandharamakrishnan (2015) and Jakubczyk and Jaskulska (2021). Freeze drying removes moisture from the product through sublimation, a process conducted at extremely low temperatures. This method effectively inhibits browning reactions and leads to enhanced colour stability in the final product. It also can be observed that the lightness of the milled sample increased as the particle size decreased. The reduction in particle size might lead to improved light reflection and lightness, a finding noted by Wang *et al*. (2020) as well.

Table 1. The colour values for calamansi fresh peel and peel powder.

Sample	*L**	*a**	*b**	*ΔE*
Fresh peel	45.32 (0.16)^{a}	-4.59 (0.17)^{a}	19.23 (0.09)^{a}	-
Milled 30 minutes	79.89 (0.09)^{b}	-2.44 (0.14)^{b}	21.21 (0.25)^{b}	34.70 (0.17)^{a}
Milled 60 minutes	83.17 (0.25)^{c}	-3.00 (0.11)^{c}	20.45 (0.04)^{c}	37.90 (0.10)^{b}
Milled 90 minutes	83.48 (0.16)^{c}	-3.05 (0.07)^{c}	18.08 (0.01)^{d}	38.21 (0.16)^{c}
Milled 120 minutes	83.52 (0.06)^{c}	-3.18 (0.04)^{c}	15.79 (0.02)^{e}	38.38 (0.06)^{c,d}

Note: Note: The values in parentheses indicate the values of standard deviation. Values with different letters (a-e) within the same column indicate significant differences at $p < 0.05$ by Duncan's multiple range test.

Meanwhile, the results show that as ball milling time is increased, the greenness becomes more enhanced at 60 minutes and remains consistent at 90 and 120 minutes. Chitrakar *et al*. (2020) reported that the improved green colour of the milled product corresponded to an increased chlorophylls content, facilitated by a reduction in particle size that resulted in a larger surface area. This, in turn, led to extensive release of chlorophylls during quantification; which are the major pigments of calamansi peel. Overall, the results show that less pronounced colour changes in the powder samples were observed during a 30 minutes milling duration due to short milling time. Conversely, milling durations of 90 and 120 minutes resulted in comparable colour changes.

CONCLUSION

This study investigates the effect of ball milling time (ranging from 30 to 120 minutes) on the structure and colour of freeze-dried calamansi peel powder. SEM micrographs reveal that extending the milling duration transforms large fragments into smaller particle sizes while maintaining the structural integrity. Notably, the milling process results in a noticeable enhancement in the green colour of freeze-dried sample. In future studies, undertaking thorough particle size analysis of calamansi peel powder will be crucial to gain deeper insights into milling effects, including their influence on chlorophyll distribution. Additionally, exploring optimal milling times could lead to valuable advancements in process refinement.

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REFERENCES

[1] Aman, A. K., Singh, R. K., Kr, N. & Jabeen, A. 2021. Effect of superfine grinding on structural, morphological and antioxidant properties of ginger (*Zingiberofficinale*) nano crystalline food powder. *Materials Today: Proceedings,* 43**,** 3397-3403.

[2] Belose, B., Kotecha, P., Godase, S. & Chavan, U. 2021. Studies on utilization of orange peel powder in the preparation of cookies. *International Journal of Chemical Studies,* 9**,** 1600-1602.

[3] Chitrakar, B., Zhang, M., Zhang, X. & Bhandari, B. 2023. Valorization of asparagus-leaf by-product through nutritionally enriched chips to evaluate the effect of powder particle size on functional properties and rutin contents. *Drying Technology,* 41**,** 34-45.

[4] Chitrakar, B., Zhang, M., Zhang, X. & Devahastin, S. 2020. Bioactive dietary Fiber powder from asparagus leaf by-product: Effect of low-temperature ball milling on physico-chemical, functional and microstructural characteristics. *Powder Technology,* 366**,** 275-282.

[5] Department of Agriculture Malaysia. 2021. *Booklet Statistik Tanaman (Sub-sektor Tanaman Makanan). Putrajaya, Malaysia: Department of Agriculture Malaysia.* (http://www.doa.gov.my/index/resources/aktiviti_sumber/sumber_awam/maklumat_pertanian/perangkaan_tanaman/booklet_statistik_tanaman_2022.pdf). Last accessed on 20 August 2023.

[6] ElBeltagy, A. E., Elsayed, M., Khalil, S., Kishk, Y. F., Fattah, A. F. A. A. & Alharthi, S. S. 2022. Physical, Chemical, and Antioxidant Characterization of Nano-Pomegranate Peel and Its Impact on Lipid Oxidation of Refrigerated Meat Ball. *Journal of Food Quality,* 2022.

[7] Espinosa-Garza, G., Antonyan, N. & Loera-Hernández, I. 2018. Orange peel dehydration and creation of new edible products. *International Journal of Food Engineering,* 4**,** 327-331.

[8] Girotto, F., Alibardi, L. & Cossu, R. 2015. Food waste generation and industrial uses: a review. *Waste Management,* 45**,** 32-41.

[9] Guiné, R. P. & Barroca, M. J. 2012. Effect of drying treatments on texture and color of vegetables (pumpkin and green pepper). *Food and Bioproducts Processing,* 90**,** 58-63.

[10] Hanani, Z. N., Yee, F. C. & Nor-Khaizura, M. 2019. Effect of pomegranate (*Punica granatum* L.) peel powder on the antioxidant and antimicrobial properties of fish gelatin films as active packaging. *Food Hydrocolloids,* 89**,** 253-259.

[11] Hong, S. J., Das, P. R. & Eun, J. B. 2021. Effects of superfine grinding using ball-milling on the physical properties, chemical composition, and antioxidant properties of *Quercus salicina* (Blume) leaf powders. *Journal of the Science of Food and Agriculture,* 101**,** 3123-3131.

[12] Ishwarya, S. P. & Anandharamakrishnan, C. 2015. Spray-freeze-drying approach for soluble coffee processing and its effect on quality characteristics. *Journal of Food Engineering,* 149**,** 171-180.

[13] Jakubczyk, E. & Jaskulska, A. 2021. The effect of freeze-drying on the properties of Polish vegetable soups. *Applied Sciences,* 11**,** 654.

[14] Lazăr, S., Constantin, O. E., Horincar, G., Andronoiu, D. G., Stănciuc, N., Muresan, C. & Râpeanu, G. 2022. Beetroot by-product as a functional ingredient for obtaining value-added mayonnaise. *Processes,* 10**,** 227.

[15] Mai, T. H. A., Tran, T. T. T. & Le, V. V. M. 2022. Use of pitaya peel powder for partial replacement of wheat flour in cookie making: Effects of particle size of pitaya peel powder on the product quality. *Journal of Food Processing and Preservation,* 46**,** e16214.

[16] Nasrullah, A., Khan, A. S., Bhat, A., Din, I. U., Inayat, A., Muhammad, N., Bakhsh, E. M. & Khan, S. B. 2021. Effect of short time ball milling on physicochemical and adsorption performance of activated carbon prepared from mangosteen peel waste. *Renewable Energy,* 168**,** 723-733.

[17] Piras, C. C., Fernández-Prieto, S. & De Borggraeve, W. M. 2019. Ball milling: a green technology for the preparation and functionalisation of nanocellulose derivatives. *Nanoscale Advances,* 1**,** 937-947.

[18] Ramachandraiah, K. & Chin, K. B. 2016. Evaluation of ball-milling time on the physicochemical and antioxidant properties of persimmon by-products powder. *Innovative Food Science & Emerging Technologies,* 37**,** 115-124.

[19] Rojas-Bravo, M., Rojas-Zenteno, E., Hernández-Carranza, P., Ávila-Sosa, R., Aguilar-Sánchez, R., Ruiz-López, I. & Ochoa-Velasco, C. 2019. A potential application of mango (*Mangifera indica* L. cv Manila) peel powder to increase the total phenolic compounds and antioxidant capacity of edible films and coatings. *Food and Bioprocess Technology,* 12**,** 1584-1592.

[20] Singh, B., Singh, J. P., Kaur, A. & Singh, N. 2020. Phenolic composition, antioxidant potential and health benefits of citrus peel. *Food Research International***,** 109114.

[21] Thumwong, A., Darachai, J., Thamrongsiripak, N., Tokonami, S., Ishikawa, T. & Saenboonruang, K. 2023. Fruit peel powder as natural antioxidant and reinforcing bio-filler in natural rubber latex gloves: Cases of mangosteen, pomelo and durian. *Antioxidants,* 12**,** 1119.

[22] Venkatachalam, K., Charoenphun, N., Srean, P., Yuvanatemiya, V., Pipatpanukul, C., Pakeechai, K., Parametthanuwat, T. & Wongsa, J. 2023. Phytochemicals, bioactive properties and commercial potential of calamondin (*Citrofortunella microcarpa*) fruits: A review. *Molecules,* 28**,** 3401.

[23] Wang, J., Zhang, M., Devahastin, S. & Liu, Y. 2020. Influence of low-temperature ball milling time on physicochemical properties, flavor, bioactive compounds contents and antioxidant activity of horseradish powder. *Advanced Powder Technology,* 31**,** 914-921.

[24] Wedamulla, N. E., Fan, M., Choi, Y.-J. & Kim, E.-K. 2022. Citrus peel as a renewable bioresource: Transforming waste to food additives. *Journal of Functional Foods,* 95**,** 105163.