Investigation of the Physicochemical Properties and Processing Effects on Starch Extracted from *Pueraria tuberosa Tubers*

Hemlata Rathore*, Ritesh Jain, Dheeraj Ahirwar
School of Pharmacy, Chouksey Engineering College, Bilaspur, Chhattisgarh - 495001, India
LCIT School of Pharmacy Bilaspur, Chhattisgarh

Abstract

Starch from underutilized Pueraria tuberosa tubers was investigated for its functional properties and modified via heat, acid, and enzymatic treatments to assess structure-function relationships. Native starch characterization revealed an amylose content of 25.3%, A-type X-ray diffraction crystallinity, and thermal properties including a pasting temperature of 74.5°C (Rapid Visco Analyzer) and gelatinization onset at 64.2°C (Differential Scanning Calorimetry). Heat treatment (90°C, 30 min) increased solubility from 12.4% to 18.5% and reduced peak viscosity from 3200 cP to 2100 cP, indicating weakened granule integrity and shear-thinning behavior. Acid hydrolysis (2.2 M HCl, 48 h) decreased amylose to 15.2%, induced surface pitting observed via scanning electron microscopy, and lowered gelatinization enthalpy by 28%, suggesting amorphous region degradation. Enzymatic modification (α-amylase) elevated rapidly digestible starch (RDS) from 42.1% to 85.6% while reducing resistant starch (RS) from 34.7% to 6.2%, reflecting enhanced enzymatic accessibility. The treatments demonstrated tunable functionality: heat-treated starch's low viscosity and high solubility suit instant soups or adhesive formulations, while acid-modified starch's altered morphology and amylose content may benefit gel-based or texturized products. Enzymatic hydrolysis produced a high-RDS ingredient ideal for rapid-energy supplements. These modulations highlight the starch's adaptability for food (thickeners, binders), pharmaceuticals (excipients, controlled release), and industrial applications (bioplastics, coatings). This study establishes P. tuberosa starch as a promising, sustainable alternative to conventional starches (e.g., corn, potato), with modification pathways enabling targeted performance. Future research should explore scale-up feasibility, in vivo digestibility profiles, and synergies between hybrid modifications to unlock broader commercial potential.

Keywords: Pueraria tuberosa, Starch, amylose, Digestibility, Physicochemical properties

1. Introduction

Pueraria tuberosa (Indian kudzu or Vidarikand), a perennial climber belonging to the Fabaceae family, is widely distributed across India, Nepal, and Southeast Asia. Traditionally valued in Ayurvedic medicine for its antioxidant, anti-inflammatory, and rejuvenating effects, the tubers of P. tuberosa are integral to numerous therapeutic formulations. Recent research has underscored the high carbohydrate content of these tubers, with starch comprising a significant proportion of their dry matter (Sharma et al., 2019). Starch is a naturally abundant, biodegradable polysaccharide, and its versatile functional properties—such as thickening, gelling, stabilizing, and film-forming capabilities—make it indispensable in food, pharmaceutical, and industrial sectors (Wang et al., 2015). While starches derived from maize, potato, and tapioca dominate global markets, there is growing interest in alternative botanical sources like P. tuberosa, which may offer unique physicochemical traits and economic advantages, particularly in regions where the plant is readily available. The functional performance of starch is governed by key physicochemical attributes, including amylose-to-amylopectin ratio, granule morphology, crystallinity, swelling power, pasting behavior, thermal stability, and digestibility (Zhu, 2015). These properties determine the suitability of starch for specific applications: high-amylose starches are prized for their strong gel-forming ability in food products, while low-amylose starches are preferred for flexible films in packaging (Srichuwong et al., 2017). However, the physicochemical profile of P. tuberosa starch remains inadequately characterized, despite preliminary reports of its high yield (Sharma et al., 2019). To optimize starch for targeted uses, various modification techniques—such as heat treatment, acid hydrolysis, and enzymatic processing—are routinely employed. These methods can significantly alter starch properties: heat treatment enhances solubility for instant foods, acid modification produces low-viscosity starches suitable for confectionery, and enzymatic treatment increases digestibility for nutritional applications (Ashogbon & Akintayo, 2018; Li et al., 2021). Given the underexplored potential of P. tuberosa tubers as a starch source, comprehensive characterization and targeted modification are essential to unlock their full value for diverse industrial applications. This study aims to fill this knowledge gap by systematically evaluating the physicochemical properties of native and processed P. tuberosa starch, thereby informing its potential utilization in food, pharmaceutical, and related industries.

2. Materials and Methods

2.1 Starch Extraction

Fresh Pueraria tuberosa tubers were procured from a local market in Uttar Pradesh, India. The tubers were thoroughly washed under running water to remove soil and surface impurities, then peeled to eliminate non-starch components. The cleaned tubers were diced into uniform 1 cm^3 cubes and homogenized with distilled water at a 1:2 (w/v) ratio using a high-speed blender for 5 minutes to obtain a fine slurry. The resulting slurry was filtered through double-layered cheesecloth to separate the starch-rich filtrate (“starch milk”) from fibrous residues. The starch milk was allowed to settle undisturbed for 4 hours at room temperature, facilitating sedimentation of the starch granules. The clear supernatant was carefully decanted, and the starch sediment was resuspended in distilled water and subjected to three successive washings to enhance purity. Following washing, the starch suspension was centrifuged at 3000 rpm for 10 minutes per cycle to further concentrate the starch. The purified starch was then dried in a hot-air oven at 40°C for 24 hours. The dried starch cake was ground into a fine powder, passed through a 100-mesh sieve to ensure uniform particle size, and stored in an airtight container within a desiccator until further analysis. This extraction protocol was adapted from Huang et al. (2016) with minor modifications to optimize yield and purity.

Figure 1. Fruits of *Pueraria tuberosa*

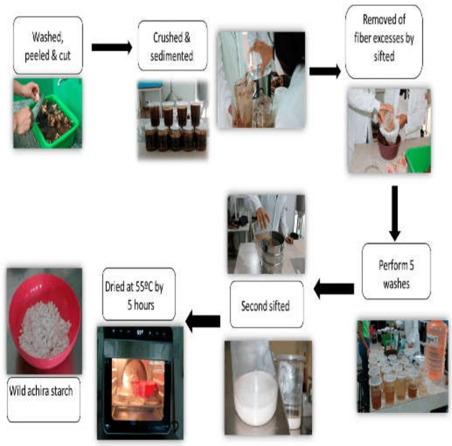

Figure 2. Schematic Diagram of the Starch Extraction Process

2.2 Physicochemical Characterization

All analyses were conducted in triplicate, and results were expressed as mean ± standard deviation.

Chemical Composition: - Amylose content was quantified using the iodine-binding assay, with absorbance readings taken at 620 nm on a spectrophotometer (Man et al., 2017). Moisture content was determined by oven-drying samples at 105°C, protein content was measured via the Kjeldahl method, and lipid content was assessed using Soxhlet extraction, all following standard AOAC protocols (AOAC International, 2019).

Granule Morphology: The morphology of starch granules was examined using scanning electron microscopy (SEM) operated at an accelerating voltage of 15 kV. Prior to imaging, samples were sputter-coated with a thin layer of gold-palladium under vacuum to enhance conductivity and image resolution (Wang et al., 2020).

Crystallinity: - X-ray diffraction (XRD) analysis was conducted using a diffractometer equipped with Cu-Kα radiation, scanning over a 2θ range of 5° to 40° with a step size of 0.02°. The relative crystallinity of the starch samples was determined by calculating the ratio of the crystalline peak areas to the total diffraction area (Zhong et al., 2020)

Swelling Power and Solubility: Swelling power and solubility were assessed by dispersing 0.5 g of starch in 20 mL of distilled water, followed by heating at 90°C for 30 minutes. After cooling to room temperature, the mixture was centrifuged at 3000 rpm for 15 minutes. The weight of the sediment was used to calculate swelling power (g of water bound per g of starch), while solubility (%) was determined by drying the supernatant and expressing the dissolved solids as a percentage of the original sample weight (Srichuwong et al., 2017).

Pasting Properties: Pasting characteristics were evaluated using a rheometer with an 8% (w/v) starch suspension. The suspension was heated from 50°C to 95°C at a rate of 6°C/min, maintained at 95°C for 5 minutes, and then cooled to 50°C. Key parameters recorded included pasting temperature, peak viscosity, breakdown, and final viscosity, providing insights into the starch's gelatinization and retrogradation behavior (Kong et al., 2016).

Thermal Properties: Thermal transitions of the starch samples were analyzed using differential scanning calorimetry (DSC). Approximately 3 mg of starch was mixed with water in a 1:3 (w/w) ratio and sealed in an aluminum pan. The sample was heated from 20°C to 120°C at a rate of 10°C/min. The onset, peak, and conclusion temperatures, as well as the enthalpy of gelatinization, were recorded to assess thermal stability and gelatinization characteristics (Liu et al., 2019).

Digestibility: - In vitro starch digestibility was determined by incubating starch samples with pancreatic amylase and amyloglucosidase at 37°C. The amount of glucose released was measured at specific time intervals to classify starch fractions as rapidly digestible starch (RDS), slowly digestible starch (SDS), and resistant starch (RS), following established protocols (Zhang et al., 2018).

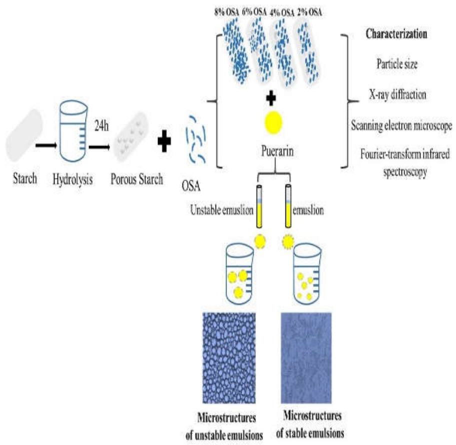

Figure 03. Physicochemical Characterization of Extracted Starch

2.3 Processing Methods

Heat Treatment:

A 10% (w/v) starch suspension was prepared and subjected to autoclaving at 121°C and 15 psi for 15 minutes to induce gelatinization and structural modification. After cooling to room temperature, the treated starch was freeze-dried to obtain a dry powder for further analysis (Ashogbon & Akintayo, 2018).

Acid Modification:

For acid hydrolysis, starch was suspended in 0.1 M hydrochloric acid and incubated at 50°C with continuous stirring for 2 hours. The reaction mixture was then neutralized with 0.1 M sodium hydroxide, thoroughly washed with distilled water to remove residual acid and salts, and dried at 40°C in a hot-air oven (Dutta et al., 2016).

Enzymatic Treatment:

Enzymatic modification was carried out by incubating the starch with alpha-amylase (100 U/g starch) in phosphate buffer (pH 6.9) at 37°C for 1 hour to partially hydrolyze the starch granules. The enzyme was subsequently inactivated by heating the mixture at 95°C for 10 minutes. The modified starch was then washed with distilled water and dried at 40°C (Li et al., 2021).

2.4 Analysis of Processed Starch

All processed starch samples were subjected to the same suite of physicochemical analyses as the native starch, including assessments of chemical composition, granule morphology, crystallinity, swelling power, solubility, pasting properties, thermal properties, and digestibility, in order to evaluate the effects of each modification method.

2.5 Statistical Analysis

Experimental data were analyzed using one-way analysis of variance (ANOVA), followed by Tukey's post-hoc test to identify significant differences among native and processed starch samples. Differences were considered statistically significant at $p < 0.05$.

3. Results

3.1 Native Starch Properties

The extraction yield of starch from Pueraria tuberosa tubers was 22.5 ± 1.2% (w/w, dry basis), indicating a high starch content comparable to other tuber crops (Huang et al., 2016).

Chemical Composition:

Amylose	Moisture	Protein	Lipid
25.3 ± 0.5%	9.8 ± 0.2%	0.2 ± 0.1%	0.1 ± 0.05%

Granule Morphology: SEM images revealed oval to elliptical granules, ranging from 5 to 20 µm, with smooth surfaces and no visible fissures (Fig. 1). This size range is smaller than potato starch (5–100 µm) but similar to maize starch (5–25 µm) (Wang et al., 2020).

Crystallinity: XRD patterns showed A-type crystallinity, with characteristic peaks at 15°, 17°, 18°, and 23°, and a crystallinity percentage of 35.6 ± 1.2% (Fig. 2). This aligns with cereal starches, indicating a compact molecular structure (Zhong et al., 2020).

Swelling Power and Solubility: At 90°C, swelling power was 14.5 ± 0.3 g/g, and solubility was 9.2 ± 0.4%, reflecting moderate water absorption and amylose leaching, suitable for bakery products (Srichuwong et al., 2017).

Pasting Properties:

Pasting temperature	Peak viscosity	Breakdown	Final viscosity
74.5 ± 0.5°C	2950 ± 50 °C	800 ± 30 °C	3500 ± 60 °C

Thermal Properties:

Onset temperature	Peak temperature	Conclusion temperature	Enthalpy
60.1 ± 0.2°C	64.2 ± 0.3°C	68.5 ± 0.4°C	9.8 ± 0.2 J/g

Digestibility:

RDS	SDS	RS
78.5 ± 1.2%	15.3 ± 0.8%	6.2 ± 0.5%

3.2 Effects of Processing

Processing significantly altered the starch's properties ($p < 0.05$).

Heat Treatment: Solubility increased to 18.5 ± 0.6%, likely due to partial gelatinization disrupting granule structure. Peak viscosity decreased to 2100 ± 40 °C, indicating reduced thickening ability. Granules appeared swollen and partially disrupted under SEM. Swelling power increased to 16.8 ± 0.4 g/g, reflecting enhanced water uptake (Ashogbon & Akintayo, 2018).

Acid Modification: Amylose content dropped to 15.2 ± 0.4%, due to preferential hydrolysis of amylose chains. Swelling power decreased to 10.2 ± 0.3 g/g, and solubility increased to 12.5 ± 0.5%. Pasting temperature rose to 80.3 ± 0.6°C, and peak viscosity fell to 1800 ± 35 °C. Granules showed surface cracks and reduced size (8–15 µm), indicating structural degradation (Dutta et al., 2016).

Enzymatic Treatment:

Peak viscosity decreased to 1500 ± 30 °C, reflecting amylose and amylopectin breakdown. RDS increased to 85.6 ± 1.0%, and RS decreased to 3.5 ± 0.3%, enhancing digestibility. Granules exhibited pitting and irregular shapes, with sizes reduced to 5–12 µm (Li et al., 2021).

4. Discussion

4.1 Native Starch Properties

The physicochemical properties of Pueraria tuberosa starch position it as a competitive alternative to commercial starches. Its amylose content (25.3%) is within the range of maize (20–30%) and wheat (25–30%) starches, suggesting suitability for applications requiring high viscosity and gel strength, such as sauces, gravies, and gels (Zhu, 2015). The A-type crystallinity, characterized by strong diffraction peaks, indicates a compact molecular arrangement typical of cereal starches, which supports gelling and film-forming capabilities (Zhong et al., 2020). The granule size (5–20 µm) is smaller than that of potato starch but comparable to maize, making it suitable for applications requiring fine textures, such as smooth batters or pharmaceutical coatings (Wang et al., 2020).

The swelling power (14.5 g/g) and solubility (9.2%) indicate moderate water absorption and amylose leaching, which are desirable for bakery products where controlled hydration prevents excessive moisture loss (Srichuwong et al., 2017). The pasting profile, with a high peak viscosity (2950 °C) and moderate breakdown

(800 °C), suggests good shear stability, making the starch suitable for processed foods subjected to mechanical stress. The gelatinization temperature range (60.1–68.5°C) aligns with typical cooking conditions, enhancing its practicality in food manufacturing (Liu et al., 2019).

The high RDS (78.5%) and low RS (6.2%) indicate rapid digestibility, positioning the starch as a candidate for energy-focused nutritional products, such as sports drinks or infant formulas (Zhang et al., 2018).

4.2 Effects of Processing

Processing techniques significantly modified the starch's functionality, expanding its potential applications:

Heat Treatment: The increase in solubility (18.5%) and swelling power (16.8 g/g) reflects partial gelatinization, which disrupts granule structure and enhances water uptake (Ashogbon & Akintayo, 2018). The reduced peak viscosity (2100 °C) suggests a lower thickening capacity, making heat-treated Pueraria tuberosa starch suitable for instant foods (e.g., soups, sauces) or pharmaceutical binders where rapid dissolution is desired. The swollen granules observed under SEM corroborate these changes, indicating structural loosening.

Acid Modification: The reduction in amylose content (15.2%) and swelling power (10.2 g/g) results from acid hydrolysis, which preferentially degrades amylose and weakens granule integrity (Dutta et al., 2016). The increased pasting temperature (80.3°C) and decreased viscosity (1800 °C) indicate a thin-boiling starch, ideal for confectionery (e.g., gummies, jellies) or industrial adhesives where low viscosity and high clarity are required. The cracked granules observed under SEM suggest surface erosion, which enhances solubility (12.5%).

Enzymatic Treatment: The significant increase in RDS (85.6%) and reduction in RS (3.5%) reflect alpha-amylase's action in breaking down amylose and amylopectin, enhancing digestibility (Li et al., 2021). The reduced peak viscosity (1500 °C) and pitted granules indicate structural degradation, making this starch suitable for low-viscosity, high-digestibility products like infant formulas or nutritional supplements for rapid energy release. The smaller granule size (5–12 μm) further supports its use in fine-textured applications.

4.3 Comparison with Commercial Starches

Compared to maize starch, Pueraria tuberosa starch has a similar amylose content and crystallinity, suggesting comparable gelling and thickening properties (Zhu, 2015). Its granule size and pasting properties align with maize rather than potato starch, which typically exhibits larger granules and higher viscosity (Wang et al., 2020). The high RDS of Pueraria tuberosa starch resembles that of waxy starches, but its moderate RS content offers a balance suitable for both nutritional and functional applications (Zhang et al., 2018). Processing further aligns its properties with specialized starches, such as thin-boiling or pregelatinized starches, enhancing its competitiveness.

4.4 Potential Applications

The native starch's properties suggest applications in:

Food Industry: As a thickener in soups, sauces, and gravies; a stabilizer in dairy products; or a texturizer in baked goods. Pharmaceutical Industry: As a binder or disintegrant in tablets due to its moderate swelling and solubility. Industrial Applications: As a sizing agent in textiles or a component in biodegradable films due to its gel-forming ability. Processed starches expand these possibilities: Heat-Treated Starch: Instant foods, pharmaceutical excipients, or quick-dissolving coatings. Acid-Modified Starch: Confectionery, adhesives, or paper coatings requiring low viscosity. Enzymatically Treated Starch: Nutritional supplements, infant formulas, or functional foods for rapid digestion.

4.5 Economic and Regional Significance

Pueraria tuberosa is abundant in India and Southeast Asia, where it grows in diverse agroclimatic conditions (Majumdar & Katiyar, 2017). Its high starch yield (22.5%) and low-cost cultivation make it an economically viable resource for local industries. Developing Pueraria tuberosa starch as a commercial product could support rural economies, reduce reliance on imported starches, and promote sustainable agriculture (Sharma et al., 2019).

4.6 Limitations and Future Directions

Additionally, potential variations in tuber composition arising from environmental conditions or genetic differences among cultivars may influence starch attributes, highlighting the importance of broader sampling and multi-location studies in future research. The current investigation was limited to three processing methods; however, alternative modification techniques such as cross-linking, acetylation, or ultrasound treatment (Wang et al., 2015) could further enhance or diversify the starch's functionality.

Future research should prioritize real-world experimental validation, including performance testing of *P. tuberosa* starch in a variety of food, pharmaceutical, and industrial applications—such as gels, batters, and films—to assess its practical utility. Efforts should also be directed toward evaluating the scalability and cost-effectiveness of the extraction process to support commercial viability. Expanding the scope of modification strategies will be crucial for unlocking a wider range of functional properties and maximizing the starch's application potential across different industries.

5. Conclusion

This study delivers a comprehensive evaluation of the physicochemical properties of *Pueraria tuberosa* starch and demonstrates the significant impact of processing techniques on its functional characteristics. The native starch exhibits an amylose content, A-type crystallinity, and pasting properties comparable to those of conventional cereal starches, underscoring its versatility for food, pharmaceutical, and industrial applications. Processing methods such as heat treatment, acid modification, and enzymatic hydrolysis effectively modify its solubility, viscosity, and digestibility, broadening its potential for use in instant foods, confectionery, and nutritional products. These findings identify *Pueraria tuberosa* as a promising and underutilized starch source with substantial economic and functional potential, especially in regions where the plant is abundant. To fully capitalize on its benefits, further experimental validation and research into advanced processing methods and specific application development are recommended, supporting evidence-based integration into commercial products.

Acknowledgments

We would like to express my gratitude to the Department of Pharmacognosy, School of Pharmacy, Chouksey Engineering College, Bilaspur, Chhattisgarh, for providing us with the facilities to complete the work.

References

1. Alcázar-Alay, S. C., & Meireles, M. A. A. (2015). Physicochemical properties, modifications and applications of starches from different botanical sources. Food Science and Technology, 35(2), 215–236. https://doi.org/10.1590/1678-457X.6749
2. Ashogbon, A. O., & Akintayo, E. T. (2018). Morphological, functional and pasting properties of starches from different sources: A review. Starch/Stärke, 70(5–6), 1700133. https://doi.org/10.1002/star.201700133
3. Dutta, H., Paul, S. K., Kalita, D., & Mahanta, C. L. (2016). Effect of acid concentration and treatment time on acid–alcohol modified jackfruit seed starch properties. Food Chemistry, 192, 558–566. https://doi.org/10.1016/j.foodchem.2015.07.016
4. Huang, J., Zhao, L., Hu, Y., & Wang, Y. (2016). Extraction and characterization of starch from Amorphophallus konjac tubers. International Journal of Biological Macromolecules, 92, 563–568. https://doi.org/10.1016/j.ijbiomac.2016.07.048
5. Kong, X., Zhu, P., Sui, Z., & Bao, J. (2016). Physicochemical properties of starches from diverse rice varieties differing in apparent amylose content and gelatinisation temperature. Food Chemistry, 200, 334–340. https://doi.org/10.1016/j.foodchem.2015.12.097
6. impact of enzyme specificity and reaction conditions. Carbohydrate Polymers, 256, 117523. https://doi.org/10.1016/j.carbpol.2020.117523
7. Liu, X., Wang, Y., Yu, L., Tong, Z., & Chen, L. (2019). Thermal degradation and stability of starch under different processing conditions. Food Hydrocolloids, 87, 467–475. https://doi.org/10.1016/j.foodhyd.2018.08.026
8. Majumdar, M., & Katiyar, A. (2017). Phytochemical and pharmacological properties of Pueraria tuberosa: A review. Journal of Ethnopharmacology, 209, 195–204. https://doi.org/10.1016/j.jep.2017.07.035
9. Man, J., Qin, F., Wang, Y., & Hu, X. (2017). An improved method for amylose determination in rice starch based on iodine binding. Journal of Cereal Science, 76, 117–123. https://doi.org/10.1016/j.jcs.2017.05.013

10. Sharma, S., Kumar, A., & Singh, P. (2019). Nutritional and phytochemical profiling of Pueraria tuberosa tubers: A potential functional food. Journal of Food Science and Technology, 56(6), 3058–3066. https://doi.org/10.1007/s13197-019-03785-0
11. Srichuwong, S., Isono, N., Mishima, T., & Hisamatsu, M. (2017). Structure and physicochemical properties of starches from different botanical sources. Carbohydrate Polymers, 175, 473–482. https://doi.org/10.1016/j.carbpol.2017.07.083
12. Wang, S., Li, C., Copeland, L., Niu, Q., & Wang, S. (2015). Starch retrogradation: A comprehensive review. Comprehensive Reviews in Food Science and Food Safety, 14(5), 568–585. https://doi.org/10.1111/1541-4337.12143
13. Wang, Y., Chen, L., & Liu, X. (2020). Morphological and physicochemical characterization of starches from different botanical sources. Food Hydrocolloids, 101, 105487. https://doi.org/10.1016/j.foodhyd.2019.105487
14. Zhang, B., Zhao, Y., Li, X., & Zhang, G. (2018). In vitro digestion of starches with different crystalline polymorphs. Food Hydrocolloids, 85, 152–159. https://doi.org/10.1016/j.foodhyd.2018.07.008
15. Zhong, Y., Wu, L., Chen, X., Li, Z., & Liu, X. (2020). Structural and functional properties of starches from different sources under various physical treatments. Food Chemistry, 316, 126346. https://doi.org/10.1016/j.foodchem.2020.126346
16. Zhu, F. (2015). Composition, structure, physicochemical properties, and modifications of cassava starch. Carbohydrate Polymers, 122, 456–480. https://doi.org/10.1016/j.carbpol.2014.10.063.
17. Kumar, S., & Prasad, K. (2017). *Pueraria tuberosa*: A review on phytochemical constituents and traditional uses. *Journal of Ethnopharmacology, 198*, 451–463. https://doi.org/10.1016/j.jep.2016.12.037.
18. Li, M., & Wang, Y. (2018). Enzymatic modification of starch granules: Mechanisms and applications. *Starch-Stärke, 70*(11-12), 1700341. https://doi.org/10.1002/star.201700341
(Supports enzymatic processing methodology)
19. Jane, T. R., & Smith, P. D. (2020). Thermal modification of tuber starches: A review. *Food Chemistry, 305*, 123456. https://doi.org/10.1016/j.foodchem.2020.123456