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Enhanced Food Formulation Potential of Amylase-Gallic Acid Modified Sweet Potato and Cassava Starches

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ABSTRACT

Background: Starches from sweet potato (*Ipomoea batatas*) and cassava (*Manihot esculenta*) are widely used in food applications but often exhibit limitations in hydration, solubility, and flow properties. This study aimed to evaluate whether Amylase-Gallic acid modification and starch blending can enhance the functional properties of native and composite starches. **Methods:** Ten starch samples, including single-source and blends (75:25, 50:50, 25:75), were prepared and analyzed for swelling power, solubility, water absorption rate (WAR), water and oil absorption capacities (WAC and OAC), and bulk and tapped densities (BD and TD). Comparisons were made between native and amylase–Gallic acid modified starches. **Results:** Native starches showed high variability in swelling power (0.02–0.27 g/g) and solubility (2–10%). Amylase-Gallic acid modification also significantly increased uniformity and improved hydration and binding properties (P<0.05). The 50:50 SP-CS composite exhibited the highest WAR (10.4 g/g), while WAC and OAC increased substantially across modified starches. Bulk and tapped densities also rose, indicating enhanced flowability and compaction. These improvements are attributed to partial enzymatic hydrolysis, exposure of hydrophilic sites, and polyphenol-starch interactions. **Conclusions:** Amylase–Gallic acid modification optimally improves the functional properties of the native and composite SP-CS starches, enhancing their water and oil retention, hydration, and flow characteristics. These findings support the potential application of such modified starches in food formulation, stabilization, and development of value-added products.

Introduction

Starches derived from tuber crops such as sweet potato (SP) (*Ipomoea batatas*) and cassava (CS) (*Manihot esculenta*) are vital carbohydrate sources in many developing countries, playing a significant role in human diets and contributing to food and nutrition security. Their wide availability, affordability, and renewable nature also make them suitable candidates for use in diverse food applications (Moorthy, 2004, Oke *et al.*, 1998). However, native starches often display functional limitations, including poor solubility, low resistance to heat and shear, and restricted swelling capacity, which reduce their effectiveness in food processing and formulation (Tester *et al.*, 2004). To address these challenges,

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various modification techniques-chemical, physical, and enzymatic-have been investigated to enhance starch functionality and expand their potential for developing value-added food products (Bemiller, 1997, Singh *et al.*, 2007). Several studies have demonstrated improvements in the functional quality of starch through such modifications (Hoover, 2001). Among enzymatic methods, α-amylase is frequently employed to alter starch granule structure, reduce crystallinity, and increase surface reactivity, thereby enhancing solubility and swelling capacity (Zhang *et al.*, 2023). Gallic acid, a naturally occurring polyphenolic compound abundant in many plant sources, contributes not only to antioxidant potential (Rahmawati *et al.*, 2024) but also interacts with starch molecules to induce cross-linking and stabilize granule architecture, ultimately improving thermal and physicochemical characteristics (Wiącek and Sujka, 2024). The incorporation of bioactive compounds such as Gallic acid into starch systems has gained attention for its dual role in functional improvement and nutritional enhancement.

In addition to biochemical modification, blending starches from different botanical sources has emerged as an effective strategy to combine complementary functional attributes. SP starch is known for its high amylopectin content and desirable pasting behavior, while CS starch provides excellent clarity, viscosity, and freeze-thaw stability (Wang *et al.*, 2024)). Their combination offers opportunities to optimize functional properties, improve processing performance, and design customized starch- based formulations suited for diverse food applications. Blended and modified starches not only provide technological benefits for food texture, encapsulation, and stability but also contribute to developing cost-effective, regionally available alternatives to imported starches, thereby supporting food and nutrition security.

This study, therefore, aims to investigate the effect of dual modification using α-amylase and Gallic acid on starches extracted from SP and CS, with a particular focus on functional properties such as swelling power, solubility, water-holding capacity, bulk density, and pasting characteristics. In addition, the influence of starch blending on these attributes is evaluated to determine synergistic effects that may enhance the performance of composite starch systems. By integrating enzymatic modification, polyphenol interaction, and starch blending, this research seeks to elucidate how such dual-modified starches can improve hydration, binding, and flow behaviors-thereby enhancing their potential as functional ingredients in food formulation and product development. Overall, the study underscores the relevance of SP–CS composites as sustainable and value-added starch sources contributing to improved food quality and nutritional security.

Materials and Methods

Materials

Fresh SP and CS tubers were procured locally. Analytical grade Gallic acid and α-amylase (Porcine α-amylase Sigma Aldrich,USA) were purchased from standard suppliers.

Starch extraction

Starch was extracted using the wet method by Dorantes-Fuertes (Dorantes-Fuertes *et al.*, 2024), involving washing, peeling, grating, and settling in water, followed by filtration and drying. Fresh tubers were washed, peeled, chopped into small cubes and then ground in a high-speed blender for 5 min. The pulp was suspended in ten times its volume of water, stirred for 5 minutes and filtered using double fold cheese cloth. The filtrate was allowed to stand for 2 hr. for the starch to settle, and the top liquid was decanted and discarded. Water was added to the sediment, and the mixture was stirred again for 5 minutes. Then, filtration was repeated as before, and the starch from filtrate was allowed to settle for overnight. After decanting the top liquid, the sediment (starch) was sun dried for 24 h and stored in air tight container till further use.

Modification procedure

The method applied by Matute *et al*. (Matute *et al.*, 2012), was used for the preparation of enzyme modified starch. To modify the starch, slurry was prepared by dispersing starch at a concentration of 40 g/l. To this, 1% (w/v) of α-amylase (porcine pancreatic α-amylase,Sigma-Aldrich, USA) was added. The mixture was maintained under

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continuous agitation at 90 °C and pH 5.5, adjusted using 0.1 mol/l sodium citrate buffer (Sigma-Aldrich, USA), for 60 minutes to facilitate enzymatic hydrolysis. Subsequently, 3% (w/v) Gallic acid (Sigma-Aldrich, USA) was added dropwise to the enzymatically modified starch slurry (Villanova and Lin, 2022) and the reaction was allowed to proceed for an additional 60 minutes. Upon completion, the pH of the slurry was readjusted to 5.5 using 1 M hydrochloric acid. The reaction mixture was then filtered, and the resulting starch cake was mixed with 2 liters of distilled water and re-filtered. The residue was washed three times with distilled water to remove any residual acid, and the modified starch was dried overnight in an oven (Labline Instruments, India) at 40 °C.

Preparation of starch samples

Ten starch samples were prepared based on the grouping shown in **Table 1**. For composite starches, native SP and CS starches were blended on a dry weight basis in the following proportions: 100:0, 75:25, 50:50, 25:75, and 0:100 (w/w), yielding five native formulations. The modified starch samples (α-amylase and Gallic acid modified) were prepared using the same compositional ratios as the native starches. Thus, a total of ten starch samples-five native (SP100, CS100, SP75:CS25, SP50:CS50, and SP25:CS75) and five modified counterparts-were obtained for comparative evaluation of their functional properties.

Table 1. Native and modified starch composites used in the study.

No	Starch Type	Composition(SP:CS)
1	Native	Sweet potato 100%
2	Native	Cassava 100%
3	Native pomposite	Sweet potato 75% : Cassava 25%
4	Native pomposite	Sweet potato 50% : Cassava 50%
5	Native pomposite	Sweet potato 25% : Cassava 75%
6	Modified	Sweet potato 100%
7	Modified	Cassava 100%
8	Modified pomposite	Sweet potato 75% : Cassava 25%
9	Modified pomposite	Sweet potato 50% : Cassava 50%
10	Modified pomposite	Sweet potato 25% : Cassava 75%

Analysis of functional properties

Swelling power and solubility: Swelling power and solubility were determined according to the method of Awokoya (Awokoya *et al.*, 2011). One gram of starch was dispersed in 20 ml distilled water and heated at controlled temperatures (50–90 °C) for 30 min. The mixture was centrifuged at 3,000 × g for 10 min, and the supernatant was carefully decanted. Soluble solids in the supernatant were dried at 105 °C and weighed to calculate solubility as a percentage of the initial dry starch. Swelling power was expressed as the ratio of water retained in the sediment to the initial dry starch mass.

$$Solubility\ (\%) = \frac{Weight\ of\ dried\ supernatant}{Weight\ of\ dry\ starch\ sample} \times 100$$

$$Swelling\ pPower\ (g/g) = \frac{Weight\ of\ the\ sediment}{Weight\ of\ dry\ starch\ sample}$$

Water absorption ratio (WAR): WAR was measured following the protocol of Anderson, *et al.* (Anderson *et al.*, 1969). One gram of starch was mixed with 10 ml distilled water, heated to boiling and maintained for 30 min with intermittent stirring. After cooling to 25 °C, the suspension was centrifuged at 3,000 × g for 10 min. The hydrated sample was weighed, and WAR was calculated as the ratio of water absorbed to the initial dry starch weight. This method reflects the ability of starch to imbibe water during cooking.

$$WAR\ (g/g) = \frac{Weight\ of\ hydrated\ starch\ after\ centrifugation}{Weight\ of\ dry\ starch\ sample} \times 100$$

Water absorption capacity (WAC) and oil absorption capacity (OAC): WAC and OAC were measured as described by Whistler and BeMiller

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(Whistler and BeMiller, 2009). A 0.5 g sample was dispersed in 10 ml distilled water (for WAC) or refined vegetable oil (for OAC) in centrifuge tubes. The mixture was vortexed for 1 min, held at room temperature for 30 min with occasional inversion, and then centrifuged at 3,000 × g for 20 min. Unbound fluid was decanted, and the sediment was weighed. The capacities were calculated as grams of water or oil retained per gram of dry starch.

$$WAC\ (g/g) = \frac{Weight\ of\ hydrated\ sedimant - weight\ of\ the\ dry\ sample}{Weight\ of\ dry\ sample} \times 100$$

$$OAC\ (g/g) = \frac{Weight\ of\ oil\ retained\ sediment - weight\ of\ the\ dry\ sample}{Weight\ of\ dry\ sample}$$

Bulk density (BD) and tapped density (TD): BD and TD were assessed according to Navaf (Navaf *et al.*, 2020). Approximately 5–10 g of starch was gently filled into a 10 ml graduated cylinder without compacting, and the unsettled volume was recorded. BD was expressed as the sample mass divided by this volume. The cylinder was then tapped 100 times to obtain a reduced volume, and TD was calculated as mass divided by tapped volume.

$$BD\ (g/ml) = \frac{Mass\ of\ the\ sample\ (g)}{Unsettled\ volume\ (ml)}$$

$$TD\ (g/ml) = \frac{Mass\ of\ the\ sample\ (g)}{Tapped\ volume\ (ml)}$$

Data analysis

All experiments were performed in triplicate, and data were expressed as mean ± standard deviation (SD). Two-way analysis of variance (ANOVA) was conducted using R software (Version 2025.05.1+513) to determine the effects of starch type and modification on functional properties. Also, Tukey's post hoc test was applied to compare means at a significance level of P-value<0.05.

Results

Swelling power

The swelling power of native starches exhibited (**Table 2**) notable variability, ranging from 0.02±0.001 g/g for SP starch to 0.27±0.004 g/g for the 50:50 SP–CS composite, reflecting inherent differences in granule size, amylose-to-amylopectin ratio, and intermolecular interactions within the starch network. Upon modification with amylase and Gallic acid, swelling power was significantly altered (P<0.05), with values narrowing to a range of 0.13–0.18 g/g across all modified samples. Notably, the 50:50 modified starch composite exhibited the highest swelling power (0.18±0.002 g/g) among the treated samples.

Solubility

Native starches from SP and cassava exhibited the highest solubility, with values of 10.00±0.05% and 10.00±0.02%, respectively (**Table 2**). In contrast, native starch composites showed lower solubility (~2%), indicating inherent structural differences between single-source and composite starches. Upon modification with amylase and Gallic acid, solubility was significantly enhanced (P<0.05), particularly in the 75:25 and 25:75 SP–cassava composites, which both reached 8%.

Table 2. Solubility (%) and Swelling power (g/g) of single and composite native and modified starches.

Starch combination	Native starch	Modified starch
Swelling power		
Sweet potato	2.0 ± 0.01^{a}	13.0 ± 0.02^{c}
Cassava	3.5 ± 0.02^{ab}	14.0 ± 0.01^{c}
SP (75): CS (25)	22.0 ± 0.05^{d}	14.0 ± 0.01^{c}
SP (50): CS (50)	27.0 ± 0.04^{e}	18.0 ± 0.02^{d}
SP (25): CS (75)	21.0 ± 0.02^{c}	16.0 ± 0.01^{d}
Solubility		
Sweet potato	10.0 ± 0.05^{a}	4 .0± 0.02^{d}
Cassava	10.0 ± 0.02^{a}	8.0 ± 0.04^{b}
SP (75): CS (25)	2.0 ± 0.01^{c}	8.0 ± 0.03^{b}
SP (50): CS (50)	2.0 ± 0.01^{c}	6 .0± 0.01^{c}
SP (25): CS (75)	2.0 ± 0.02^{c}	8.0 ± 0.02^{b}

Values are Mean±SD (n=3). Values with different letters (a, b, c, ...) within a column indicate significant differences at P<0.05, as determined by ANOVA followed by Tukey's post hoc test.

WAR

The WAR of native starches was comparatively low, with cassava starch exhibiting 1.72±0.01 g/g, reflecting the limited availability of hydrophilic sites and compact granule structure (**Table 3**). Upon modification with amylase and Gallic acid, WAR increased significantly (P<0.05) across all samples,

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indicating enhanced water-binding capacity. Modified cassava starch, for example, showed a WAR of 88.00±0.05 g/g, while among blends, the 50:50 modified starch composite achieved the highest WAR (10.40±0.05 g/g), representing a substantial increase from the native version (7.52±0.02 g/g).

WAC and OAC

All modified starches showed statistically significant increases in both WAC and OAC compared to their native counterparts (P<0.05) (**Table 4**). For example, the WAC of SP starch increased from 154.00±2.15 g/g in its native form to 190.00±1.58 g/g after modification, while OAC improved from 125.00±2.58 g/g to 238.00± 5.64 g/g. Composite samples, such as the 25:75 SP–CS (SP25:CS75) blend, also demonstrated remarkable improvements, reaching a WAC of 180.00±4.20 g/g and an OAC of 231.00±2.85 g/g.

Table 3. Water absorption ratio (g/g) of single and composite native and modified starches.

Starch combination	Native starch	Modified starch
Sweet potato	1.66 ± 0.02^{a}	3.78 ± 0.02^{b}
Cassava	1.72 ± 0.01^{a}	9.88 ± 0.05^{e}
SP (75): CS (25)	5.87 ± 0.05^{c}	6.21 ± 0.04^{e}
SP (50): CS (50)	7.52 ± 0.02^{d}	10.40 ± 0.05^{e}
SP (25): CS (75)	6.41 ± 0.03^{c}	7.85 ± 0.04^{d}

Values are Mean±SD (n=3). Values with different letters (a, b, c, …) within a column indicate significant differences at P<0.05, as determined by ANOVA followed by Tukey's post hoc test.

Table 4. Water and oil absorption capacities (g/g) of single and composite native and modified starches.

Starch combination	Native starch WAC	Native starch OAC	Modified starch- WAC	Modified starch -OAC
Sweet potato	154.00 ± 2.15^{d}	125.00 ± 2.58^{c}	190 .00± 1.58^{b}	238.00 ± 5.64^{a}
Cassava	105.00 ± 1.36^{e}	163.00 ± 4.56^{a}	120.00 ± 4.63^{e}	204.00± 2.58^{c}
SP (75): CS (25)	125.00 ± 1.58^{c}	132.00 ± 1.02^{b}	175.00 ± 3.65^{c}	218.00 ± 4.56^{b}
SP (50): CS (50)	98.00 ± 2.58^{f}	174.00 ± 2.36^{a}	123.00 ± 2.15^{d}	207.00 ± 1.89^{c}
SP (25): CS (75)	135.00 ± 3.85^{b}	199 .00± 2.98^{a}	180 .00± 4.20^{b}	231.00 ± 2.85^{a}

Values are Mean±SD (n=3). Values with different letters (a, b, c, …) within a column indicate significant differences at P<0.05, as determined by ANOVA followed by Tukey's post hoc test.

BD and TD

Densities, including BD and TD, are important indicators of the flow properties, packing ability, and compressibility of powdered starches (**Table 5**). Both BD and TD increased significantly following modification with amylase and Gallic acid. For instance, the 50:50 modified SP–CS composite displayed a BD of 1.01±0.004 g/ml and a TD of 1.03±0.003 g/ml, compared to native values of 0.83±0.001 g/ml and 0.87±0.002 g/ml, respectively.

Table 5. BD and TD (g/ml) of single and composite native and modified starches.

Starch combination	Native starch -BD	Native starch-TD	Modified starch BD	Modified starch TD
Sweet potato	0.86 ± 0.005^{b}	0.90 ± 0.004^{c}	0.89 ± 0.005^{b}	0.97 ± 0.005^{a}
Cassava	0.87 ± 0.002^{b}	0.90 ± 0.002^{c}	0.89 ± 0.003^{b}	0.96 ± 0.002^{a}
SP (75): CS (25)	0.84 ± 0.001^{c}	0.87 ± 0.001^{d}	0.84 ± 0.004^{c}	0.90 ± 0.003^{b}
SP (50): CS (50)	0.83 ± 0.001^{c}	0.87 ± 0.002^{d}	1.01 ± 0.004^{a}	1.03 ± 0.003^{a}
SP (25): CS (75)	0.90 ± 0.002^{a}	0.91 ± 0.002^{b}	0.93 ± 0.003^{a}	0.94 ± 0.004^{a}

Values are Mean±SD (n=3). Values with different letters (a, b, c, …) within a column indicate significant differences at P<0.05, as determined by ANOVA followed by Tukey's post hoc test.

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Discussion

The functional properties of native and modified starches showed notable variations following enzymatic and Gallic acid modification. These treatments significantly influenced the functional parameters, such as swelling power, solubility, WAR, WAC, OAC, BD and TD.

The observed reduction in swelling power after modification aligns with previous studies, which reported that enzymatic hydrolysis combined with polyphenolic interactions can reinforce the internal starch structure, thereby restricting excessive granule swelling (Deng *et al.*, 2023, Wang *et al.*, 2015). Partial hydrolysis of starch granules by amylase cleaves α-1,4 glycosidic bonds, decreasing granule integrity and limiting the capacity to entrap water (Wang *et al.*, 2024). Concurrently, Gallic acid may interact with starch chains through hydrogen bonding or weak cross-linking, creating a denser network that physically limits granule expansion (Zhang and Hamaker, 2009). This combined effect leads to more uniform swelling behavior among modified samples, as evidenced by the lower variability in measured values.

Furthermore, the formation of starch-Gallic acid complexes may alter the amorphous and crystalline regions of the granules, reducing their susceptibility to water penetration and swelling (Che *et al.*, 2024). Such modifications are particularly relevant for many applications, where controlled swelling is desirable for consistent textural properties, viscosity, and film-forming ability. In comparison, native starches, particularly single- source starches like SP, show highly variable swelling due to differences in granule morphology and amylose-rich regions, which restrict water uptake in some granules while allowing excessive swelling in others.

Therefore, the enzymatic-polyphenolic modification strategy not only standardizes swelling behavior, but also improves the functional predictability of composite starch food formulations. Additionally, the highest swelling power observed in the 50:50 starch composite suggests a compositional synergy between SP and CS starches, which enhances water retention capacity to some extent.

The increase in the solubility of starch composites suggests that enzymatic hydrolysis by amylase and the formation of starch–polyphenol complexes contribute to structural loosening and thereby enhancing solubility. This observation aligns with findings by Che (Che *et al.*, 2024), who reported improved starch solubility following polyphenol binding and enzymatic action. Similarly, studies have shown that enzymatic hydrolysis of starches, such as those derived from cassava, can lead to increased solubility due to partial degradation of starch granules and the release of soluble sugars (Jorge *et al.*, 2023, Mohamed *et al.*, 2021). Moreover, the interaction between starch and polyphenols can modify both crystalline and amorphous regions of starch granules, facilitating water penetration and further improving solubility (Echave *et al.*, 2024). These structural changes are particularly relevant in food processing applications, where enhanced solubility contributes to better dispersion, and texture in formulated products.

The modified cassava starch exhibited a WAR of 9.88 g/g, while the CS–SP composite showed a slightly higher value of 10.4 g/g. These values are within the reported range (3.3–18 g/g) for root starches, depending on the botanical source and treatment (Adebowale *et al.*, 2002, Hoover, 2001). The increase in WAR after modification can be ascribed to the partial disruption of the granular crystalline regions and the exposure of hydrophilic hydroxyl groups, which promote water binding. Similarly, blending cassava starch with SP starch likely enhanced water uptake due to the higher amylopectin content and amorphous nature of SP starch, which facilitate hydration and swelling. Comparable increases in hydration capacity after starch modification or blending have been observed in previous studies (Gunaratne A, 2002, Nwokocha LM, 2011). The observed enhancements in the WAC of modified composite starches is primarily attributed to enzymatic depolymerization, which exposes additional hydroxyl groups and increases the hydrophilic surface area, facilitating greater water binding. Concurrently, the incorporation of

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Gallic acid and other polyphenolic groups enhances the hydrophobic and amphiphilic interactions, contributing to improved oil-binding capacity. Such structural modifications increase surface activity and improve the ability of starch granules to interact with both aqueous and lipid phases (Che *et al.*, 2024). These results are consistent with previous reports by Che, (Che *et al.*, 2024), who emphasized that starch microstructure, particularly porosity and surface roughness, is critical in determining both water and oil absorption properties. The improvement in WAC and OAC are particularly relevant in food processing applications, as it contributes to improved flavor retention, emulsification, and texture stabilization in various starch-based products (Rashwan *et al.*, 2024). Therefore, enzymatic-polyphenolic modification represents a practical approach to tailor the functional properties of starch composites for diverse industrial applications.

Higher BD and TD values generally reflect greater particle cohesion and reduced interstitial spaces, which improve powder handling, storage efficiency, and processability (Adebowale *et al.*, 2002, Thanyapanich *et al.*, 2021). Polyphenolic modification with Gallic acid may further enhance particle cohesion through hydrogen bonding, contributing to higher TD and improved compressibility. These changes imply enhanced flowability and compaction-properties that are particularly valuable in encapsulation, tablet formulation, and other powder-based industrial applications (Siriwachirachai and Pongjanyakul, 2022). In food industry, starches with favorable BD and TD support efficient blending in bakery premixes, instant foods, and powdered beverages, while also facilitating encapsulation and spray-drying processes (Marinopoulou *et al.*, 2025). Improved compressibility further benefits nutraceuticals and functional food formulations, enabling the use of starches as carriers or fillers in tablets, capsules, and other compaction-based delivery systems (Lawal, 2019). Thus, optimization of starch density characteristics contributes directly to enhanced product uniformity, stability, and overall processing efficiency.

Conclusion

The functional characterization of native and amylase–Gallic acid modified SP, CS, and their composite starches revealed significant provements n key physicochemical properties following modification. Modified starches showed more uniform swelling power and solubility, indicating controlled granule hydration and partial structural loosening due to enzymatic hydrolysis and polyphenolic interactions. WAR, WAC, and OAC were markedly enhanced, reflecting increased hydrophilic and hydrophobic binding sites that improve hydration, oil retention, and surface functionality. BD and TD also increased, suggesting better powder flowability and compaction, which are essential for industrial processing and formulation. These functional enhancements make the modified starches and their composites highly suitable for the development of value-added food products, such as instant foods, bakery items, sauces, and nutraceuticals, while also supporting nutritional security by improving digestibility, nutrient delivery, and product versatility.

Although the present study mainly focused on evaluating the functional and physicochemical changes in native and modified starches, additional structural characterization using Scanning Electron Microscopy (SEM), X-Ray Diffraction (XRD), or Fourier Transform Infrared Spectroscopy (FTIR) would further support the observed modifications and provide a better understanding of the structural changes involved. Overall, amylase–Gallic acid modification provides an effective approach to tailor starch functionality, creating starch- based ingredients that contribute to food innovation, value addition, and improved nutrition outcomes.

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Conflict of interest

The authors declared no conflicts of interest.

Authors' contributions

Sarasamma J designed the research; Binikumari B, Sanal A, and Sara Jose A conducted the study;

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Sarasamma J, Sobitha winston S, Appukuttan Nair S, and Govindakurup Retnamma D analysed data and wrote the paper; Sarasamma J had primary responsibility for final content. All authors read and approved the final manuscript.

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