Received: 31 December 2023 | Revised: 6 July 2024 | Accepted: 9 July 2024

DOI: 10.1111/1750-3841.17270

Journal of Food Science WILEY

REVIEW ARTICLE

Concise Reviews and Hypotheses in Food Science

The multifactorial phenomenon of enzymatic hydrolysis resistance in unripe banana flour and its starch: A concise review

Siphosethu R. Dibakoane[1,2] | **Laura Suzanne Da Silva**[3] | **Belinda Meiring**[3] | **Tonna A. Anyasi**[2,4] | **Victor Mlambo**[1] | **Obiro Cuthbert Wokadala**[1]

[1]School of Agricultural and Natural Sciences, University of Mpumalanga, Nelspruit, South Africa

[2]Agro-Processing and Postharvest Technologies Division, Agricultural Research Council – Tropical and Subtropical Crops, Nelspruit, South Africa

[3]Department of Biotechnology and Food Technology, Tshwane University of Technology, Pretoria, South Africa

[4]Food and Markets Department, Natural Resources Institute, University of Greenwich, Chatham Maritime, UK

Correspondence

Obiro Cuthbert Wokadala, School of Agricultural and Natural Sciences, University of Mpumalanga, Corner R40 and D725 Road, Nelspruit 1200, South Africa. Email: obiro.wokadala@ump.ac.za

Funding information

National Research Foundation (NRF) of South Africa, Grant/Award Number: Grant116308

Abstract

Unripe banana flour starch possesses a high degree of resistance to enzymatic hydrolysis, a unique and desirable property that could be exploited in the development of functional food products to regulate blood sugar levels and promote digestive health. However, due to a multifactorial phenomenon in the banana flour matrix—from the molecular to the micro level—there is no consensus regarding the complex mechanisms behind the slow enzymatic hydrolysis of unripe banana flour starch. This work therefore explores factors that influence the enzymatic hydrolysis resistance of raw and modified banana flour and its starch including the proportion and distribution of the amorphous and crystalline phases of the starch granules; granule morphology; amylose–amylopectin ratio; as well as the presence of nonstarch components such as proteins, lipids, and phenolic compounds. Our findings revealed that the relative contributions of these factors to banana starch hydrolytic resistance are apparently dependent on the native or processed state of the starch as well as the cultivar type. The interrelatability of these factors in ensuring amylolytic resistance of unripe banana flour starch was further highlighted as another reason for the multifactorial phenomenon. Knowledge of these factors and their contributions to enzymatic hydrolysis resistance individually and interconnectedly will provide insights into enhanced ways of extraction, processing, and utilization of unripe banana flour and its starch.

KEYWORDS

amylolytic resistance, banana starch, enzymatic hydrolysis, granule morphology, unripe banana flour

1 | INTRODUCTION

Resistant starch (RS) can be described as the food starch component that cannot be digested in the mouth and small intestine of a healthy individual (Juarez-Garcia et al., 2006). Unripe banana flour is rich in RS (Thakorlal et al., 2010) ranging from about 17% to 80% (Bi et al., 2017; Cahyana et al., 2019; Falodun et al., 2019) depending on the cultivar type and its native or processed state. RS consists of physically inaccessible starches classified as resistant starch 1 (RS1); ungelatinized starches classified as resistant starch 2 (RS2); retrograded starches classified as resistant starch 3 (RS3); modified starches classified as resistant starch 4 (RS4); and starch nutrient complexes classified as resistant starch 5 (RS5) (Okumus et al., 2018; Tian & Sun, 2020). Recently, it was suggested that amylose–lipid complexes (Qin et al., 2019; Tang et al., 2022) and starch–polyphenol V-type complexes (Romero Hernández et al., 2022) should be classified as RS5 due to their ability to hinder the attachment of starch digestive enzymes to starch in the upper gastrointestinal tract. Unripe banana flour native RS is mainly categorized as RS2 (Li et al., 2020) and can be fermented by the microflora in the large intestine (Hernández-Nava et al., 2009).

Starch granules in different plant products, such as bananas, exist in a semicrystalline form and comprise both the amorphous regions, consisting of amylose and branched amylopectin, as well as the long-chain amylopectin crystalline regions that twist into H-bonded double helices (Butterworth et al., 2011). Classification of these starches according to their amylose/amylopectin results in normal starches with 25%–30% amylose and 70%–75% amylopectin; waxy starches with high amylopectin content of 98%–99%; and high-amylose starches with 50%–70% amylose content (Bello-Perez et al., 2020). This grouping ultimately influences the resistance of the starch to enzymatic hydrolysis. Wang et al. (2022) noted that factors including starch concentration, source, type, and concentration of enzymes, as well as amylose content, influence amylolysis. The RS fraction is therefore starch that resists alpha-amylase and pullulanase hydrolysis in vitro beyond 120 min compared to rapidly digestible and slowly digestible starch (Lockyer & Nugent, 2017). Thus, digestion of RS does not take place in the upper gastrointestinal tract, but RS is fermented by microbes in the colon, which produce short-chain fatty acids (SCFAs) such as acetate, butyrate, isobutyrate, isovalerate, and propionate (Koh et al., 2016; Meenu & Xu, 2019); gases such as methane, hydrogen, and CO_2; as well as alcohols including methanol and ethanol (Birt et al., 2013). Produced SCFAs are then utilized as a source of energy in the body and are of great importance to colonic health (Phothisoot et al., 2023). Butyrates formed through the fermentation of RS are the preferred matrix by colonocytes. The SCFA butyrates have also been implicated in increasing colonic blood circulation, reducing luminal pH, and preventing the formation of abnormal colonic cell populations (Lockyer & Nugent, 2017). Products of RS fermentation are absorbed from the gut into the blood flow where they are transported into the adipose and skeletal muscle tissue and undertake a unique role in improving insulin sensitivity (Lockyer & Nugent, 2017).

Currently, RS is classified as a prebiotic due to its fermentation by microbes in the colon and the corresponding production of SCFAs, which benefit the host (Moore et al., 2015). The major phyla that have been implicated in RS fermentation include *Actinobacterium*, *Bacteroidetes*, and *Firmicutes* (Birt et al., 2013). Other RS hydrolyzing strains identified in fecal samples of humans include *Bifidobacterium* spp., implicated for lactate and acetate production; *Bacteroides* spp., implicated for acetate and propionate production; as well as *Fusobacterium* and *Butyrivibrio*, implicated for butyrate production (Meenu & Xu, 2019). Intake of RS in diets has also been linked to an increase in the number and activities of microbes such as *Lactobacillus*, *Bifidobacterium*, *Lachnospiraceae*, *Ruminococcaceae*, and *Clostridium* spp. (Zeng et al., 2017; Meenu & Xu, 2019). RS thus functions as both a prebiotic and a symbiotic by promoting the growth and actions of probiotic bacteria as well as interacting with other prebiotic dietary fibers such as beta-glucan (Raigond et al., 2015). Upon absorption, RS undertakes some of its probiotic roles by protecting ingested organisms on their way to the colon, thus increasing the amount of these beneficial organisms in the colon (Raigond et al., 2015).

The RS present in unripe banana flour can modulate metabolic activity and hence provide protective effects against illnesses such as cardiovascular diseases, cancer, diabetes, and obesity (2019Khoozani et al., 2019; Li et al., 2020; Sidhu & Zafar, 2018; Singh et al., 2016). RS has been shown to reduce the incidence of noncommunicable diseases, lowering GI and cholesterol, as well as exerting positive effects against cardiovascular diseases, colorectal cancer, diabetes, and obesity (Phothisoot et al., 2023). Furthermore, RS, which is classified as a dietary fiber, contributes to fecal bulking and hinders the growth of cancerous lesions of the bowel in vivo (Moore et al., 2015). Rabbani et al. (2009) showed that a diet containing RS from unripe banana had a positive effect on controlling shigellosis in children after 5 days of feeding, through a decrease in stool volume while increasing the concentrations of acetate, butyrate, and propionate in the feces of the children. In the United States, the daily recommended intake of RS is 6 g RS/meal. However, Americans are reported to consume an estimated daily intake of ~5 g RS/day (Birt et al., 2013). Despite the beneficial health effects derived from unripe banana flour-resistant starch (UBFRS), there exists a paucity of literature attempting to integrate and elucidate the mechanisms by which unripe banana flour starch resists enzymatic hydrolysis. Therefore, this review aims to elaborate on the intricacies of the resistance mechanisms of unripe banana flour starch to enzymatic hydrolysis.

2 | AMYLOLYSIS RESISTANCE IN STARCH

The enzymatic hydrolysis of starch has been shown to occur at the interface between the solid phase, which is the starch, and the liquid phase, which is the conveyed enzyme acting on the starch (Wang et al., 2022). Naguleswaran et al. (2014) opined that the occurrence of amylolysis, which is the hydrolysis of starch granules by amylases, takes place via diffusion to a solid surface, adsorption, and catalysis. Furthermore, amylopectin molecules that possess greater levels of short chains and degree of branching but have more compact structure, high molecular density, high molar mass, and smaller molecular size are less susceptible to amylolytic attack (Naguleswaran et al., 2014). Enzymatic hydrolysis of amylose and amylopectin molecules is therefore determined by the structure of the granules or the arrangement of the crystallites within the granules (Dhital et al., 2015). In determining the mechanism of resistance to amylolytic attack, Ma et al. (2020) proposed that the presence of double helix order within or outside the crystal structure; the stabilization of the double helices into perfect crystalline structures; the presence of unevenly packed or isolated single helices; the entrapment of amorphous region within imperfect crystals; and the high molecular order formed by the attachment of short branch length amylopectin to the linear chain through covalent bonds are some mechanisms responsible for the resistance of starch to amylolytic attack.

Starch when heated in the presence of sufficient water undergoes gelatinization and increased digestibility. Upon cooling, high-amylose starches retrograde, forming crystalline regions that are inaccessible to enzymatic hydrolysis (Birt et al., 2013). The A-, B-, and C-type crystalline arrangement patterns of the different polymeric forms affect amylase hydrolysis2018. It has been observed that the shorter A-type double helices and interior crystallites are more vulnerable to enzymatic hydrolysis than the longer chains and stable helices, which are seen to be more resistant (Singh et al., 2010). The surface properties of starch granules are other contributory factors to the enzymatic hydrolysis of starches2018. Dhital et al. (2015) stated that cracks, pores, interior channels, and surface damage in starch granules further exacerbate enzymatic adsorption and binding to these molecules. Singh et al. (2010) showed that the presence of pores and pinholes in starches facilitates hydrolytic attack through penetration by amylases, thus resulting in endocorrosion, while starches (cooked or native) with smooth surfaces and without pores displayed very high resistance to enzymatic hydrolysis. However, starches from other food sources such as potatoes have been reported to be eroded through exocorrosion, which involves erosion of the granule surface during enzymatic hydrolysis.

Mass transfer limitations occurring due to the viscosity of the starch are known to influence the hydrolysis rate of starch (Singh et al., 2010). Available points for enzymatic attacks are increased due to a higher degree of branching. However, this higher degree of branching may lead to an increase in stearic hindrance (implicated in hindering enzymatic hydrolysis) and hence the mass transfer resistance. The molecular weight distribution of starches has also been reported as affecting enzymatic hydrolysis. Starches with higher molecular weight can experience sharp hindrances at the active centers of the enzyme (Singh et al., 2010). The large size of the starch granule compared to the alpha-amylase molecule provides avenues for attack by amylase by making available many possible sites for attachment of the enzyme (Butterworth et al., 2011). Similarly, the amylose–amylopectin ratio has been found to affect enzymatic hydrolysis as amylopectin is more easily digested than amylose in native starches. Das et al. (2022) showed that enzymatic treatment of banana flour led to increased amylose content in the modified flour due to amylopectin digestion by amylopullulanase. Amylopectin digestion has been attributed to its larger size with a greater surface area per molecule when compared to amylose (Singh et al., 2010). Furthermore, the glucose chains of amylose that are compactly held together by hydrogen bonds make amylose less susceptible to enzymatic hydrolysis than amylopectin with more branched glucose chains (Singh et al., 2010). Nonstarch ingredients including proteins and lipids reduce the surface accessibility of the granules by blocking the adsorption sites of the binding enzymes (Singh et al., 2010). Amylase hydrolysis can only occur by the binding of glucan chains through some glucose units to their subsites located at the active center, with the number of these subsites in an active center varying from four to nine. Wang et al. (2022) stated that different enzymes including alpha-amylase, beta-amylase, isoamylase, glycosyl-transferase, and glucosidase are involved in the enzymatic hydrolysis of starch. However, the hydrolysis of alpha-(1,4) glycosidic bonds in starch polymers is catalyzed by alpha-amylase through endocorrosion.

3 | BANANA FLOUR RS CHARACTERISTICS AND AMYLOLYTIC RESISTANCE

3.1 | Starch molecular composition

Starch, which is made up of amylose and amylopectin, comprises a highly branched amylopectin molecule whose glucose bonds are joined by α-1,4 and α-1,6 linkages (Nasrin & Anal, 2014a). The α-1,6 branching points of amylopectin occur at every 24–30 glycosyl units and function as entry points for enzymes (Giacco et al., 2016; Nasrin & Anal, 2014a). The average number of UBFRS glucosyl units for amylopectin side chains was estimated to be 14.5 (Faisant et al., 1995; Langkilde et al., 2002), but more recent estimates place them at 5.0–20.3 glucosyl units (Yee et al., 2021). It has been reported that the average degree of polymerization (DPn) of banana RS was lower than that of jackfruit starch (Zhang et al., 2019), wheat amylopectin (Yoshio et al., 2011), and barley amylopectin (Tang et al., 2002). Furthermore, it was stated that UBFRS has less branching than other starches such as jackfruit and wheat starch (Bi et al., 2017; Das et al., 2022; Jiang et al., 2015; Yee et al., 2021). Longer chain branches of amylopectin produce more crystalline structures due to the presence of hydrogen bonds among such chains (Jane et al., 1997). However, Bi et al. (2017) and Chávez-Salazar et al. (2017) suggested that the amylopectin architecture can vary between cultivars. Hence, it could be imperative to consider the genome groupings during the selection of bananas for unripe banana flour production given their influence on the architecture of amylopectin chains.

The amylose content of native nonmodified banana starch ranges from 30% to 41.66% (Bi et al., 2017; Das et al., 2022). Pelissari et al. (2012) demonstrated that banana starch with 35.1% amylose content had higher RS content (50.3%) compared to flour with 23.1% amylose content (45.1% RS). Unripe banana flour starch amylose is a slightly branched molecule whose glucose bonds are joined mainly by α-1,4 linkages (Magallanes-Cruz et al., 2017). Amylose has very limited α-1,6 glucosyl bonds compared to amylopectin (Giacco et al., 2016); therefore, the presence of a limited number of α-1,6 glucosyl bonds could result in amylose undergoing slower hydrolysis compared to amylopectin. It has also been suggested that amylose is less digestible compared to amylopectin because it has a limited surface area exposed to digestion due to its ability to interact with other components such as lipids (Englyst & Cummings, 1985; Jenkins et al., 1988). Amylose–lipid complexes and amylopectin–lipid complexes (to a lesser extent) lead to higher resistance to hydrolysis due to the formation of more resistant V-type crystalline structures (RS5) (Wokadala et al., 2012). Hence, a higher amylose content would lead to a higher resistance to hydrolysis.

Nevertheless, the enzyme resistance capacity of banana starch has been attributed to the ratio of amylose and amylopectin present in unripe banana flour (Bi et al., 2017; Soares et al., 2011; Vatanasuchart et al., 2012). Conventionally, starch consists of 20%–30% amylose and 70%–80% amylopectin (Abdullah et al., 2018); however, the amylopectin content of unripe banana flour is generally higher than its amylose content (Kumar et al., 2019; Salazar et al., 2022). The amylose–amylopectin ratio influences the glycemic index (GI) of starchy foods (Björck et al., 1994); hence, a high amylose–amylopectin ratio is desirable as it improves the RS content of composite foods (Åkerberg et al., 1998). Ademosun et al. (2021) showed that the GI value of wheat noodles supplemented with unripe plantain flour and orange peels (35.03–53.09) was significantly lower than that of the control samples without unripe banana flour (58.23–66.21). A higher amylose content in unripe banana flour used in the study could have resulted in an increased RS content, thus leading to the lowering of the GI value.

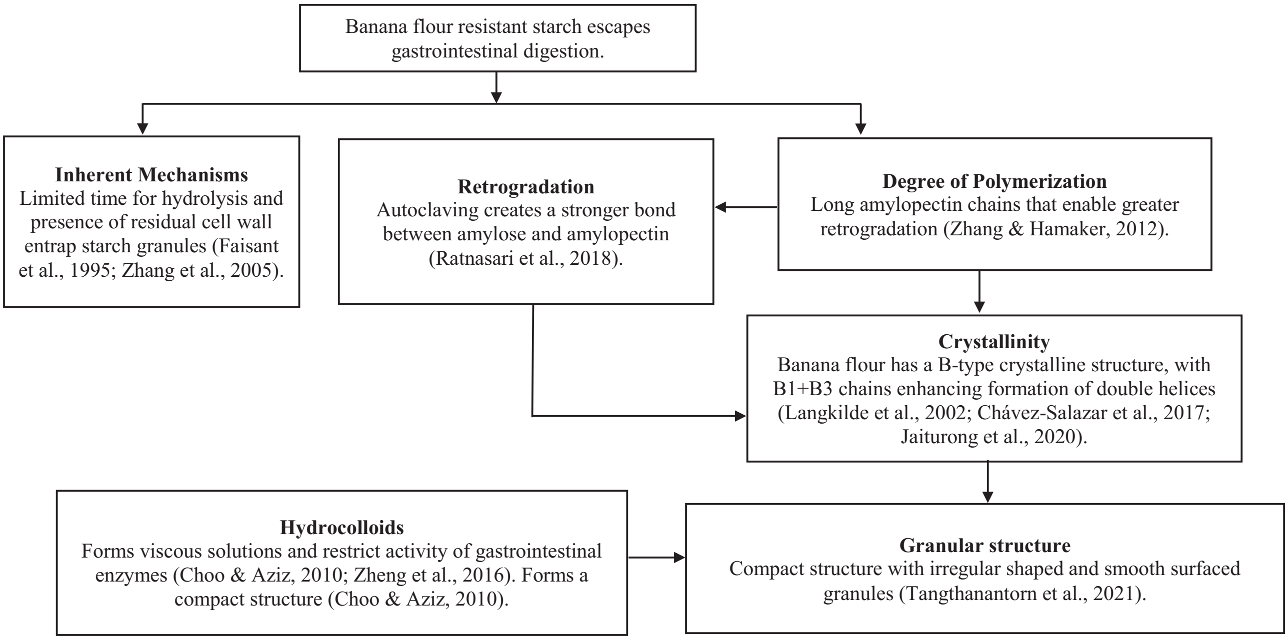
3.2 | Supramolecular structure

The structural properties of unripe banana flour and banana RS are summarized in Table 1. X-ray diffraction assessments showed that UBFRS (RS2) has a B-type crystalline structure (Faisant et al., 1995; Langkilde et al., 2002), which possesses a characteristically lower degree of branching, higher crystallinity, and longer amylopectin chains compared to the A-type crystalline structure (Chávez-Salazar et al., 2017; Kim et al., 2015; Martens et al., 2018). The longer amylopectin chains present in the B-crystalline structure produce a more crystalline structure and increased crystallinity compared to the A-type structure by enhancing the formation of double helical structures (Chávez-Salazar et al., 2017). Higher crystallinity results in the formation of a compact granular structure present in UBFRS, which hinders the starch granules (RS2) from being invaded by enzymes, resulting in reduced susceptibility to enzymatic hydrolysis (Ma & Boye, 2018; Tangthanantorn et al., 2021). Unlike the A-type crystalline structures, the B-type crystalline starch (Figure 1) tends to have a less porous structure because its branching sites are predominantly located in the amorphous zones (Kim et al., 2015). In addition, the longer B-type amylopectin chains are thought to further contribute to resistance through interactions with lipids and amylose, further preventing hydration and allowing for more extensive retrogradation (Zhang & Hamaker, 2012).

However, not all UBFSs consist of B-type crystalline structures (Table 1). Native RS2 prepared from Brazilian and Taiwanese banana cultivars have been reported to contain both B- and C-type crystalline structures (Cordoba et al., 2018; Wang et al., 2017). Cordoba et al. (2018) showed that Terra Pla´tan (AAB) displayed a B-type crystalline structure, while Catura cavendish (AAA) and Prata Ana (AAB) cultivars displayed a C-type crystalline structure. Likewise, *Musa* AAA Cavendish and *Musa* ABB Bluggoe RS (RS2) displayed a C-type crystalline structure, while *Musa* ABB Pisang Awak and *Musa* AA Pisang Mas RS (RS2) displayed a B-type crystalline structure (Wang et al., 2017). The relative resistance in C-type nonmodified UBFS compared to A-type crystallinity starches results from the fact that the C-type crystalline structures are structurally intermediate between the A-

TABLE 1 Degree of polymerization, crystallinity, and granular morphology of banana flour resistant starch.

Type	Description	Degree of crystallinity	Type of crystallinity	Size	Shape	Degree of polymerization	Reference
RS2	Native (uncooked)		B-type			14.5 glucosyl units	Langkilde et al., 2002
RS2	Native (uncooked)				Intact, irregularly shaped granules with a smooth surface.		Thakorlal et al., 2010
RS2	Native (uncooked and centrifuged)	21.76–27.75	B- and C-type	29.07–39.17 µm	Ellipsoidal and flattened shape		Cordoba et al., 2018
RS3	Enzymatically modified		C_A-type		Tough and rigid.		Das et al., 2022
RS3	Autoclaved and debranched		C-type		Cavities and channels in starch granules		González-Soto et al., 2007
RS3	Modified (autoclaved, debranched, and enzymatically modified)				Irregular shaped		Khawas & Deka, 2017
RS2	Native (centrifuged/uncooked)		B- and C-type	5–90 µm	Elliptic, triangular shaped with smooth surfaces		Wang et al., 2017
RS2	Raw (uncooked)					18.9–20.3 glucosyl units	Yee et al., 2021



Banana flour resistant starch escapes gastrointestinal digestion.

Inherent Mechanisms Limited time for hydrolysis and presence of residual cell wall entrap starch granules (Faisant et al., 1995; Zhang et al., 2005).

Retrogradation Autoclaving creates a stronger bond between amylose and amylopectin (Ratnasari et al., 2018).

Degree of Polymerization Long amylopectin chains that enable greater retrogradation (Zhang & Hamaker, 2012).

Crystallinity Banana flour has a B-type crystalline structure, with B1+B3 chains enhancing formation of double helices (Langkilde et al., 2002; Chávez-Salazar et al., 2017; Jaiturong et al., 2020).

Hydrocolloids Forms viscous solutions and restrict activity of gastrointestinal enzymes (Choo & Aziz, 2010; Zheng et al., 2016). Forms a compact structure (Choo & Aziz, 2010).

Granular structure Compact structure with irregular shaped and smooth surfaced granules (Tangthanantorn et al., 2021).

FIGURE 1 Mechanisms of resistance of unripe banana flour starch to enzymatic hydrolysis.

and B-type and thus would have relatively higher resistance compared to type A (Ma & Boye, 2018). Modified (autoclaved, debranched, and enzymatically modified) UBFRS (RS3) displays C- and C_A-type crystalline structure (Das et al., 2022; González-Soto et al., 2007). The C_A-type could therefore be more susceptible to enzymatic hydrolysis compared to the C-type crystalline structure due to the presence of short amylopectin chains, evidence of porous surfaces, and surface area effects, which result in high enzymatic hydrolysis.

4 | GRANULAR MORPHOLOGY AND ENZYMATIC HYDROLYSIS RESISTANCE

4.1 | Granule shape

There are diverse reports on the granular shape and surface structure of unripe banana flour starch (Table 1). The raw UBFRS-RS2 possesses intact, irregular, and ellipsoidal-shaped starch granules with smooth surfaces (Figure 1), whose structures are dependent on the banana cultivar (Cordoba et al., 2018; Thakorlal et al., 2010; Wang et al., 2017). Pelissari et al. (2012) demonstrated that RS2 present in unripe banana flour and starch granules displayed irregularly shaped and compact granules in both elongated and spheroid forms. Hoffmann Sardá et al. (2016) showed that raw unripe banana flour from Brazilian banana cultivars had spherical and elongated granules. Several reports provide evidence of the influence of cultivar type on the granular morphology of unripe banana flour starch. For example, Gao et al. (2016) reported that Cavendish had an irregular-shaped structure, while plantain had small and ellipsoidal granules. Anyasi et al. (2017) reported that *Luvhele* and *Mabonde* banana flour had polygonal- and oval-shaped granules, while *Muomva-red* and *William* banana flour had elongated, polygonal, and spherical-shaped granules. Soares et al. (2011) found that desert banana cultivars (Pacovan and Mysore) had small and leaf-like structures, while plantains (Terra and Figo) had round and elongated structures.

Irregular- and ellipsoidal-shaped starch granules tend to be more susceptible to enzymatic hydrolysis compared to other starch granules because they have a larger surface area and volume (Gao et al., 2016). Likewise, leaf-like starch granules found in unripe bananas tend to have higher susceptibility to enzymatic hydrolysis relative to rounded granules due to the arrangement of amylopectin molecules (Soares et al., 2011). The presence of smooth surfaces with no signs of degradation has been reported on the surface of Terra, Figo, and Pacovan banana cultivars (Soares et al., 2011). The presence of smooth surfaces is thought to restrict the invasion of starch granules by α-amylase. However, this explanation may not be generally applicable given that pits have been reported on the surface of the granules of Mysore banana cultivars, yet they have similar resistance to digestion as cultivars with smooth surfaces (Soares et al., 2011).

The morphology of starch granules and their susceptibility to enzymatic hydrolysis can further be influenced by food processing techniques such as heating, milling, and baking (Anyasi et al., 2017). Autoclaved and enzymatically (amylopullulanase) modified UBFRS (RS3) samples displayed irregularly shaped granules with porous, tough, and rigid surfaces (Das et al., 2022; González-Soto et al., 2007). Similar granular morphologies were reported for autoclaved, debranched, and enzymatically treated banana RS (Khawas & Deka, 2017). The presence of pores on the surface of processed starch granules provides a pathway for endocorrosion. Hence, autoclaved and enzymatically (amylopullulanase) modified banana RS (RS3) samples have increased susceptibility to enzymatic hydrolysis compared to native banana RS (RS2) due to differences in granule structure.

4.2 | Granule size

The diameter of unripe banana flour starch granules influences its susceptibility to enzymatic hydrolysis. Unripe banana flour starch granules have been reported to possess varied diameters ranging from 1.4 to 500 µm (Bezerra et al., 2013; Falade & Olugbuyi, 2010; Gao et al., 2016; Giraldo-Gómez et al., 2019; Khoozani et al., 2020; Padhi & Dwivedi, 2022; Salazar et al., 2022; Thakaeng et al., 2021). It has been suggested that small-sized starch granules are more susceptible to enzymatic hydrolysis compared to large-sized starch granules due to their large surface area to volume ratio (Gao et al., 2016; Jiang et al., 2015), while large rounded and elongated granules may show few signs of degradation (Soares et al., 2011).

Foam-mat, hot-air, oven, and sun drying tend to disrupt the molecular order of starch granules in banana flour due to high heat levels and longer drying times, resulting in relatively small-sized granules (Falade & Olugbuyi, 2010; Khoozani et al., 2020). Furthermore, it was also reported that the granule diameter (20–47.3 µm [Espinosa-Solis et al., 2009; Pelissari et al., 2012]; 5–90 µm [Wang et al., 2017]) of native (raw) unripe banana RS (RS2) was greater than that of enzymatically modified UBFRS (RS3) (20 µm) (Das et al., 2022).

5 | NONSTARCH COMPONENTS AND THE SUSCEPTIBILITY OF UNRIPE BANANA STARCH TO ENZYMATIC HYDROLYSIS

5.1 | Native hydrocolloids (proteins and nonstarch polysaccharides)

Hydrocolloids are long-chain protein or polysaccharide polymers with the ability to bind water and form gels or viscous dispersions (Woomer & Adedeji, 2021). Hydrocolloids interact with starch granules by forming physical barriers and viscous solutions around the surface of granules, resulting in the reduction of the activity of gastrointestinal enzymes (Faisant et al., 1995; Tester et al., 2006; Zheng et al., 2016). Interactions of starch granules and nonstarch polysaccharides have been shown to affect the rate and extent of digestion of starch by prolonging the time required for effective hydrolysis (Zhang & Hamaker, 2012; Zhang et al., 2005).

The protein content of unripe banana flour is generally reported to be low (<5%) (Kumar et al., 2019; Rachman et al., 2021; Thakaeng et al., 2021), with higher concentrations (>5%) reported in the peels (de Angelis-Pereira et al., 2016; do Prado Ferreira & Teixeira Tarley, 2020), as well as with increasing ripeness of the fruit (Campuzano et al., 2018). Miao et al. (2015) showed that the presence of proteins in unripe banana flour starch contributes to amylolytic resistance, as proteins interact with starch to reduce the rate of α-amylase hydrolysis. The interaction is thought to result in the formation of a three-way complex between fatty acids, amylose, and proteins (Shah et al., 2011; Zhang et al., 2010). This interaction occurs even with low protein concentrations (3.68%–8.02%), which could be up to 10 times less than starch (70.0%–84.4%) (Campuzano et al., 2018; Zhang et al., 2003), thus enabling, to a certain degree, resistance to enzymatic hydrolysis in unripe banana flour.

Fiber increases the viscosity of foods and limits enzymatic digestion by reducing access of gastrointestinal enzymes to the substrates and thereby reducing the conversion rate of starch into glucose (Abutair et al., 2016). The pectin content of unripe banana flour has been reported to range between 3.29% and 5.61% (Bi et al., 2017). Pectin is regarded as a hydrocolloid alongside gelatin, alginate, carrageenan, gellan, and agar (Goff & Guo, 2019; Milani & Maleki, 2012; 2010), as well as a soluble fiber (Martínez et al., 2015). Insoluble dietary fiber components including lignin, cellulose, and hemicellulose have been reported to be present in unripe banana flour (Silva et al., 2020; 2014). Ng et al. (2020) suggested that the barrier formed by soluble fiber protects starch granules against water penetration, which limits the swelling and gelatinization capacity of starch and reduces the ability of enzymes to access the starch due to limited entry or attachment points.

5.2 | Lipids

The rate and extent of digestion of starch are also influenced by its lipids composition and extent of phosphorylation (Qi & Tester, 2016; Qi et al., 2018). The presence of lipids in unripe banana flour has been reported by several authors and is said to be $\leq$2.0% (w/w) (Dan, 2011; Haslinda et al., 2009; Hoffmann Sardá et al., 2016; Kongolo et al., 2017; Menezes et al., 2011). Banana peel possesses higher lipid content when compared to the pulp; hence, the lipid content of unripe banana flour with peels is higher than without peels (Haslinda et al., 2009; Hoffmann Sardá et al., 2016). Interaction of amylose and lipids through amylose–lipid complex formation reduces susceptibility to hydrolysis by α-amylase (Wokadala et al., 2012). Lipids may also reduce α-amylase hydrolysis complexing with longer amylopectin chains (Tester et al., 2006). It has been suggested that the interaction of lipid and amylose induces a change in the torsion angles of glycosidic bonds and forms a helical structure, which reduces the binding activity of amylolytic enzymes and the formation of enzyme–substrate complexes (Chi et al., 2022; Li et al., 2021). However, the degree to which lipid complexation contributes to the hydrolysis resistance of banana flour starch is not clear and requires further investigation.

5.3 | Polyphenols

Numerous studies have reported the presence of polyphenols in unripe banana flour (Anyasi et al., 2018; Pico, Corbin, et al., 2019; Pico, Xu, et al., 2019). The polyphenols reported to be present in bananas include anthocyanins (cyanidin, delphinidin, and their derivatives in both fruit pulp and peel), chlorogenic acid, ferulic acid, flavan-3-ols (catechins, epicatechins, and gallic acid), flavonoids (myricetin and quercetin), and tannins (Anyasi et al., 2018; Bennett et al., 2010; 2006 Maseko et al., 2024; 2015Pico, Corbin, et al., 2019; Pico, Xu, et al., 2019). Phenolic compounds have been reported to be present in both banana peel and pulps (Castelo-Branco et al., 2017), although banana peels contain more polyphenols compared to the pulp (Agama-Acevedo et al., 2016; Castelo-Branco et al., 2017; Maseko et al., 2024).

Polyphenols facilitate the escape of starch from digestion in the small intestine by inhibiting the activity of α-amylase and glucosidase, resulting in intact starch being transported into the colon (Gu et al., 2020; Liu et al., 2017). Furthermore, polyphenols can interact with starch molecules via covalent or noncovalent bonds to form starch–polyphenol complexes (Chi et al., 2022). The presence of starch–polyphenol complexes has been suggested to occur in unripe banana flour (Choo & Aziz, 2010; Pico, Xu, et al., 2019; Sarawong et al., 2014); hence, unripe banana flour is considered a natural source of starch–polyphenol complexes (Hernández et al., 2022). Polyphenols contain hydroxyl groups that enable them to interact with starch via noncovalent bonds such as hydrogen bonds and electrostatic and ionic interactions (Chi et al., 2017; Giuberti et al., 2020; Ngo et al., 2022).

Polyphenols can reduce the activity of enzymes and starch digestibility by binding to starch-digesting enzymes and forming V-type inclusion complexes through hydrophobic interaction and hydrogen bonding with amylose and amylopectin side chains (Han et al., 2020; Hernández et al., 2022; Kan et al., 2022; Ngo et al., 2022). The extent to which the polyphenol inhibition mechanism contributes to the overall resistance to enzymatic hydrolysis is, however, difficult to ascertain (Hernández et al., 2022). Providing more information on the mechanisms responsible for starch–polyphenol complexes will enhance the exploitation of polyphenols in increasing the resistance of unripe banana flour starch. Furthermore, optimizing processing conditions to maintain the structure of the starch–polyphenol complexes present in unripe banana flour and increasing the production and application of banana peel flour as a standalone functional ingredient or composited with banana pulp flour will enhance the application of polyphenols for increasing the resistance of unripe banana flour starch.

6 | GENOME GROUPINGS AND THE RS CONTENT OF UNRIPE BANANA FLOUR

The effects of the genomic and subgenomic group and cultivar type on the RS content of unripe banana flour are summarized in Table 2. Kumar et al. (2019) found that the RS content of unripe banana flour prepared from cultivars belonging to the ABB genomic group (44.4%–46.5%) was greater than those in AAA (32.7%) and AAB genomic group (31.3%–35.6%). Vatanasuchart et al. (2012) showed that the RS content of unripe banana flour from the BBB (68.1%) genomic group was greater than the AA (52.2%–57.2%), AAA (57.0%), and ABB (56.6%–61.8%) genomic groups. It has been suggested that the B genomic group tends to have more RS content compared to the A genomic group due to its high starch and protein content, larger granule size, and higher amylose content (Annor et al., 2016; Gibert et al., 2014; Narayana et al., 2017)

However, recent studies have suggested that the genomic group has no significant effect on the RS content of unripe banana flour. Chang et al. (2022) found that there was no significant difference in the RS content of the total starch composition of unripe banana flour from cultivars belonging to the AA (90.38% dry basis of total starch), AAA (89.20%–90.86% dry basis of total starch), and ABB (90.58% dry basis of total starch) genomic groups. This conforms with the findings of Reis et al. (2019), who suggested that the genomic group had no effect on the RS content of unripe banana flour. Kongolo et al. (2017) found that for cultivars grown under the same agronomic conditions, the RS content varied. The RS content of the AA genomic group (39.6%–47.3%) was similar to that of the AAA (38.6%–47.1%) and AAB (39.2%) genomic groups but slightly higher than the AAAB genomic group (19.9%–47.4%). Cordoba et al. (2018) also found that there was an overlap in the RS content of AAB (24.79%–34.85%) and AAA (31.92%) genomic groups. Other studies on unripe banana flour suggested that cultivars from the AAA genomic group with RS content of 4.83%–57.49% (Campuzano et al., 2018; Khoozani et al., 2019; Menezes et al., 2011; Rayo et al., 2015; Rosado et al., 2020; Tian et al., 2020) had RS content that overlapped with that of the ABB cultivar types, which ranged from 8.60% to 68.8% (Haslinda et al., 2009; Nasrin et al., 2015; Rodríguez-Ambriz et al., 2008; Vatanasuchart et al., 2012). The effect of the genomic group could therefore be attributed to differences in agronomic conditions, maturity at harvest, as well as postharvest processing.

7 | EFFECT OF MODIFICATIONS ON THE RS CONTENT OF UNRIPE BANANA FLOUR

The effect of physical and chemical modifications on the RS content of unripe banana flour is summarized in Table 3 and Figure 2.

7.1 | Physical modifications

Heat–moisture treatment (HMT) generally reduces the RS content of unripe banana flour. Heat disrupts the molecular order of starch granules resulting in irreversible changes such as granule swelling, melting of native crystalline structure, starch solubilization, and loss of birefringence (Anyasi et al., 2017). Zhang and Hamaker (2012) demonstrated that cooking at 100°C for 10–20 min reduced the RS content of banana flour. De la Rosa-Millan et al. (2014) showed that the RS content of flour prepared from bananas that had been cooked for 5 min was 61.5% compared to 66.5% in raw banana flour. Similarly, Rodríguez-Damian et al. (2013) found that the RS content of banana flour cooked for 5 min (58.5%) was greater than that of banana flour samples cooked for 15–25 min (23.3%–23.7%). Given that the RS content of unripe banana flour tends to decrease with cooking time (De la Rosa-Millan et al., 2014; Rodríguez-Damian et al., 2013), shorter cooking times could be applied to limit the destruction of the structural integrity of starch granules and thus minimize the reduction in RS content of banana flour.

Annealing and HMT followed by storage have been shown to improve the RS content of unripe banana flour2010. The RS content of unripe banana flour subjected to annealing and storage was greater than that of cooked banana flour samples (De la Rosa-Millan et al., 2014). Cahyana et al. (2019) demonstrated that annealing produced higher RS content in unripe banana flour

TABLE 2 Effect of genomic and subgenomic group classification on unripe banana flour (UBF)-resistant starch content.

Genomic group	Subgenomic group	Cultivar	Resistant starch content (%)	References
AA	Inarnibal	Pisang Lemark	47.30	Kongolo et al., 2017
AA	Sucrier	Lady Finger	39.60	
AAA	Cavendish	Gross Michel	37.90	
AAA	Cavendish	IBP. 5.61	47.10	
AAA	Sucrier	Khai Thong	42.10	
AAA	Red	Green red	38.90	
AAB	Pome	Pome	39.20	
AAAB	Pome synthetic hybrid	FHIA 18	19.90	
AAAB	Goldfinger selection	Goldfinger	47.20	
AAAB	Goldfinger selection	PKZ	47.40	
AAA	Cavendish	Grand Naine	32.70	Kumar et al., 2019
ABB		Monthan	44.35	
ABB		Saba	46.50	
AAB		Nendran	35.55	
AAB		Populu	31.28	
ABB	Plantain	Mzuzu	90.58	Chang et al., 2022
AAA	Cavendish	Malindi	90.86	
AA	Pisane litin	Mshale	90.38	
AAA	Cavendish	Bukoba	90.69	
	Dwarf red banana	*Guihongjiao No. 1*	86.15	Bi et al., 2017
ABB	Pisang Awak	*Jinfen No. 1*	85.88	
AAA	Cavendish	*Williams* B_6	90.46	
AAA	Cavendish		57.48 (standard UBF)	Hoffmann Sardá et al., 2016
AAA	Cavendish		43.25 (standard peel UBF)	
AAA		Kluai Hom	57.20	Vatanasuchart et al., 2012
AA		*Kluai Khai*	52.20	
AA		*Kluai Lembuenang*	57	
ABB		*Kluai Namwa*	56.60	
ABB		*Kluai Hakmak*	61.80	
BBB		*Kluai Hin*	68.80	
AAB		*Terra Pla´tano*	34.85	Cordoba et al., 2018
AAA		*Caturra Cavendish*	31.92	
AAB		*Prata Ana*	24.79	
AAA	Cavendish	*Nanicão*	40.90–58.50	Tribess et al., 2009
AAA	Cavendish	*Nanica*	32.20–46.72	Khoozani et al., 2019

(Continues)

TABLE 2 (Continued)

Genomic group	Subgenomic group	Cultivar	Resistant starch content (%)	References
AAA	Cavendish	*Nanicão*	48.99	Menezes et al., 2011
AAA	Cavendish	*Nanicão*	53.95–57.49 (unmodified)	Rayo et al., 2015
AAA	Cavendish		33.86	Bezerra et al., 2013
AAA	Cavendish	*Pei-Chiao*	30.30	Liao & Hung, 2015
	Cavendish	*Nanicão*	38.92–44.27	La Fuente & Tadini, 2017
AAA	Cavendish		22.80	Rosado et al., 2020
AAA			11.12–38.28	Campuzano et al., 2018
AAA	*Musa acuminata* Colla		4.33	Tian et al., 2020
AAA		*Nanicão*	8.20	Menezes et al., 2010
AAA	Gros Michel	*Kluai Hom Thong*	41.74–46.35	Virulchatapan & Luangsakul, 2020
AAA		*Nanicão*	8.20	Menezes et al., 2010
AAB			30.40	Rodríguez-Ambriz et al., 2008
AAB	Harton		31	Gutiérrez, 2018
ABB	*Sapientum*	*Kluai Nam Wa*	32.26–48.88	Moongngarm et al., 2014
	Musa paradisiaca L.		17.50	Juarez-Garcia et al., 2006
	Musa paradisiaca	*Terra*	49.50	Pelissari et al., 2012
ABB	Awak		39.50	Haslinda et al., 2009
			39.50	
ABB	*Musa paradisiaca*		67	
		Pei Chiao	42.23	Li et al., 2020
		Fomosana	34.00	
		Tai-Chiao	30.03	
		Nam	40.25	Ramos et al., 2009
		Fhia 01	10.01	
		Chifre de Vaca	61.05	Reis et al., 2019
		Comprida	60.79	
		Curare Enano	68.50	
		Mongolo	65.92	
		Pinha	53.12	

(Continues)

TABLE 2 (Continued)

Genomic group	Subgenomic group	Cultivar	Resistant starch content (%)	References
		Red Yade	43.44	
		Samura B	52.33	
		Terra Ana Branca	46.26	
		Terra Ponta Aparada	56.13	
		Terra Sem Nome	21.15	
		Tipo Velhaca	55.91	
		Trouis Vert	60.59	
		Terra Maranhão	62.10	
		D'Angola	46.35	
		Terrinha	40.14	
		BRS SCS Belluna	59.14	
		BRS Platina	67.43	
	Grand Naine		51.37	
		Pacovan	70.06	
		Prata Anã	58.30	
	Plantain	*Red Essong* (French bunch)	33.30–50.23	Udomkun et al., 2021
	Plantain	*Mbouroukou 3 - False Horn bunch*	35.36–53.11	
	Plantain hybrids	*PITA 14*	32.72–39.67	
	Plantain hybrids	*PITA 27*	30.91–49.89	
	Musa sapientum, triploid hybrid banana		7.90–8.90	Chaipai et al., 2018
	Musa paradisiaca		24.60	Hernández-Nava et al., 2011
		Kluai Namwa plantain	8.60	Nasrin et al., 2015

(80%–85%) compared to HMT and dual retrogradation (45%–60%). Annealing has been shown to increase the RS content of unripe banana flour by preserving the compact granule surface and increasing the crystallinity of the starch (Cahyana et al., 2019). Rodríguez-Damian et al. (2013) showed that the RS content of banana flour (24.0%–29.0%) subjected to HMT and storage for 25 min was greater than that of banana flour (23.3%) cooked for 25 min. Bello-Perez et al. (2015) further demonstrated that the RS content of banana flour spaghetti stored for 5 days increased from 3.2% to 7.0% (w/w). HMT minimizes starch granule damage, while storage facilitates the retrogradation of the starch granules by enabling the reassociation of amylose and the formation of a more crystalline structure, which results in improved RS yields. The increase in crystallinity can also be attributed to the strong bond formed by amylose and amylopectin due to the autoclaving process (González-Soto et al., 2007).

7.2 | Chemical and enzymatic modifications

Gutiérrez (2018) reported that phosphated plantain flour recorded the highest RS content (80%) compared to the acetylated, methylated, and oxidized plantain flour

TABLE 3 Effect of modification type on unripe banana flour-resistant starch content.

Modification type	Modifying agent	Modifying conditions	Resistant starch content (%)	References
Physical modifications				
Cooking	H_2O	100°C	10.50–61.50	Zhang & Hamaker, 2012
Cooking	H_2O	95°C	12.10–61.50	De la Rosa-Millan et al., 2014
Cooking and annealing	H_2O	64°C for 24 h	18.70–64.10	
Cooking, annealing, and storage	H_2O	64°C for 24 h; 4°C for 7 days (storage: refrigeration)	23.20–64.30	
Cooking	H_2O	95°C for 15–25 min	23.34–58.50	Rodríguez-Damian et al., 2013
Cooking and heat–moisture treatment	H_2O	30% distilled water; 120°C for 24 h (heating/drying)	22.90–55.83	
Cooking, heat–moisture treatment, and storage (refrigeration)	H_2O	30% distilled water; 120°C for 24 h (heating/drying); −20°C for 7 days (storage: refrigeration)	21.70–59.40	
Heat–moisture treatment	H_2O	30% distilled water, 100°C for 8 h (heating/drying), and 4–5°C for 24 h (refrigeration)	45–50	Cahyana et al., 2019
Annealing		70% distilled water (w/w) and water bath at 55°C for 12 h	80–85	
Dual retrogradation		Water suspension (1:5.5 [w/v]); 100°C for 30 min (heating); and 4°C for 48 h (refrigeration)	50–60	
Autoclaving		121°C for 30 min	25.25–30.30	Liao & Hung, 2015
Blanching		100°C for 1 min	38.40–52.21	
Blanching	H_2O	100°C for 1 min	37.33–55.11	
Blanching	H_2O	100°C for 1 min	34.32–52.35	
Blanching	H_2O		32.53–52.67	
Cooked			12.40	
Heat treated (uncooked)			81.19 g	
Cooked			10.90	
Cooked			93.68	
Chemical modifications				
Native			31	Gutiérrez, 2018
Acetylation	Acetic anhydride ($(CH_3CO)_2O$)	7.5 mL = 0.079 mol	39.10	
Carboxymethylation	Monochloroacetic acid ($ClCH_2CO_2H$)	4.7 mL = 0.079 mol	39	
Methylation	Dimethyl sulfate ($(CH_3)_2SO_4$)	7.5 mL = 0.079 mol	78	
Oxidation	Hydrogen peroxide (H_2O_2)	6.2 mol—30% (0.079 mol)	23	
Phosphation	Sodium trimetaphosphate (STMP—$Na_3P_3O_9$)		82.10	
Native			40.69	Almanza-Benitez et al., 2015

(Continues)

TABLE 3 (Continued)

Modification type	Modifying agent	Modifying conditions	Resistant starch content (%)	References
Chemical	Hydrochloric acid		44.87	
Native	Thermoplastic plantain flour films		1.99	Gutiérrez & Álvarez, 2016
Crosslinking	Aloe vera gel		2.12–3.05	
Native (Red Essong French bunch)			33.30–50.23	Udomkun et al., 2021
Chemical	Citric acid	10 g/L citric acid for 10 min	37.65–54.32	
Native (Mbouroukou 3 - False Horn bunch)			35.36–53.11	
Chemical	Citric acid	10 g/L citric acid for 10 min	39.44–56.37	
Native (PITA 14)			32.72–39.67	
Chemical	Citric acid	0 g/L citric acid for 10 min	36.28–53.63	
Native (PITA 27)			30.91–49.89	
Chemical	Citric acid	0 g/L citric acid for 10 min	34.40–53.97	
Native (uncooked)			85.67 g	Sánchez-Rivera et al., 2017
Chemical (uncooked)	Citric acid		93.87 g	

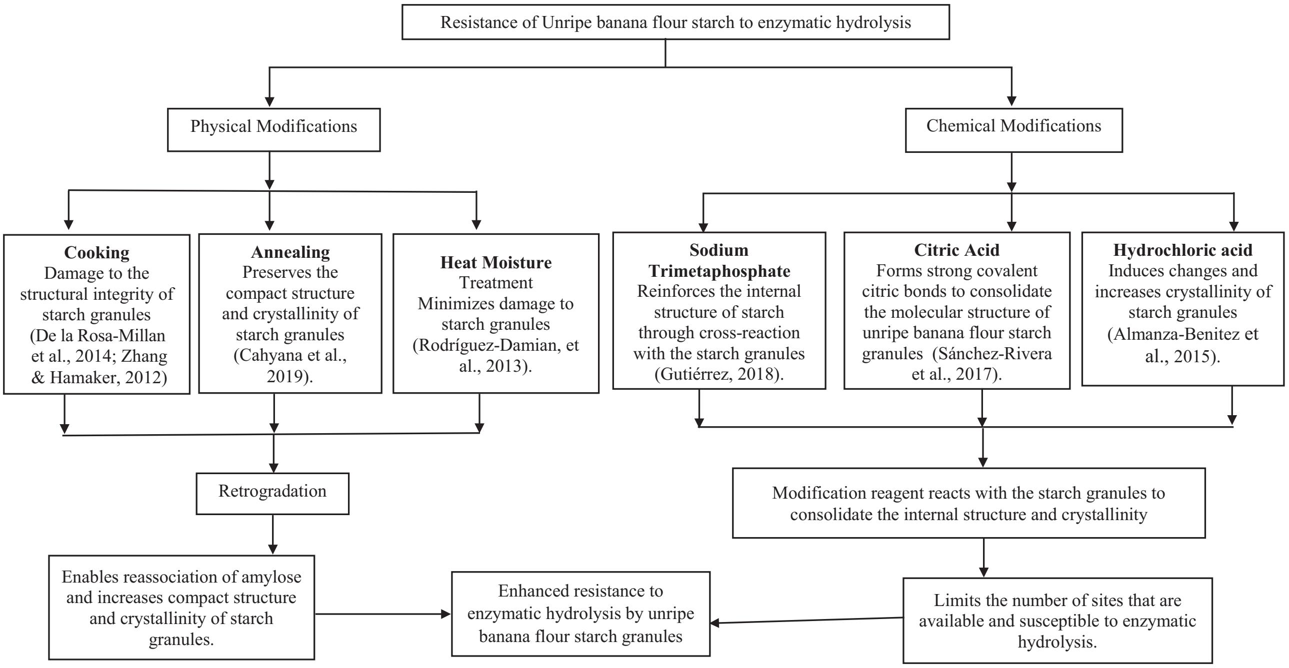


FIGURE 2 Effects of physical and chemical modifications and their associated mechanisms on the resistance of banana flour starch granules to enzymatic hydrolysis.

(23%–78%). The highest RS value of phosphated flour was attributed to the crosslinking reaction of granules with sodium trimetaphosphate (STMP), which was thought to reinforce the internal structure of the starch (Gutiérrez, 2018). Furthermore, Sánchez-Rivera et al. (2017) demonstrated that the RS content of raw and cooked plantain flour treated with citric acid was greater than native and heat-treated raw and cooked plantain flour. Similarly, Udomkun et al. (2021) found that the RS content of citric acid-treated plantain flours was higher than blanched and native plantain flours. The highest RS content of citric acid-treated plantain flours was attributed to its citric acid crosslinking effect, which facilitated the formation of strong covalent citric bonds to consolidate the molecular structures of unripe banana flour and limit the number of sites susceptible to enzymatic attacks (Sánchez-Rivera et al., 2017). Moreover, unripe plantain films substituted with *Aloe vera* gel possessed higher RS content compared to native plantain films, thus suggesting that plantain flour films are susceptible to crosslinking with the citric acid present within *Aloe vera* gel (Gutiérrez & Álvarez, 2016). It has been suggested that unripe plantain flour modified with citric acid and phosphate can be used as an ingredient to develop functional gluten-free products due to the high RS content (Gutiérrez, 2018; Udomkun et al., 2021). The RS of plantain flour (44.9%) modified with hydrochloric acid was found to be greater than nonmodified plantain flour (40.69%) (Almanza-Benitez et al., 2015). The increase in the RS level of unripe plantain flour treated with hydrochloric acid was attributed to the hydrolysis of the amorphous zones of starch and an increase in the crystallinity and change in the structure of the starch granules (Almanza-Benitez et al., 2015).

The susceptibility of unmodified unripe banana flour starch to enzymatic hydrolysis is related to changes in thermal properties induced by processing2010. Several studies have shown that the gelatinization temperature of debranched and enzymatically modified banana flour RS (103–106°C) was greater than the unmodified one (69.1–72.5°C) (Cordoba et al., 2018; Das et al., 2022; Yee et al., 2021). Similarly, the enthalpy of gelatinization (4.4–188.2 J/g) and gelatinization temperature (68.5–147°C) of the debranched, enzymatically modified, and lintnerized banana RS were greater compared to the unmodified UBFRS (6.1–13.9 J/g and 37.6–72.5°C) (Aparicio-Saguilán et al., 2005, 2008; Cordoba et al., 2018; Das et al., 2022; Lehmann et al., 2002; Nasrin & Anal, 2014b; Yee et al., 2021). The RS content of enzymatically modified banana flour using amylopullulanase treatment was observed to be higher at 68.99% when compared to the unmodified banana flour with an RS content of 38.5% (Das et al., 2022). The enzymatic treatment was seen to lead to an increase in amylopectin degradation, thus increasing the amylose content and resulting in a more ordered crystalline state. These findings suggest that chemical and enzymatic modification methods can be employed in developing unripe banana flour and starch-functional foods with high RS content.

8 | CONCLUSION

The resistance to enzymatic hydrolysis of UBFRS can be attributed to a multi-phenomenon mechanism that is primarily dependent on the status of the banana flour: native or processed. Factors such as the proportion of crystalline and amorphous phases, type, and their distribution; starch granule shape, size, and structure; amylose–amylopectin ratios; and the presence of other native nonstarch components such as proteins, lipids, and polyphenols have all been implicated as other mechanisms known to influence the resistance of both native and modified UBFRS. The DPn, degree of amylopectin branching, and amylose–amylopectin ratio were observed to have more influence on the enzymatic resistance of processed UBFRS compared to the native one. Furthermore, unmodified UBFRS was seen to be less susceptible to enzymatic hydrolysis due to its B-type crystalline structure, irregularly shaped granules, and compact granular structure. However, recent findings demonstrated that modification of unripe banana starch through autoclaving, debranching, and enzymatic treatment results in greater susceptibility to enzymatic hydrolysis due to the formation of a C_A- and C-type crystalline structure as well as porous structures on the granule surface. With differences in banana genome groupings contributing to the RS content in unripe banana flour, further research is needed on the contributions of these factors to enzymatic hydrolysis resistance in the different genome groups, both individually and interconnectedly.

AUTHOR CONTRIBUTIONS

Siphosethu R. Dibakoane: Writing—original draft; writing—review and editing. **Laura Suzanne Da Silva**: Writing—review and editing. **Belinda Meiring**: Writing—review and editing. **Tonna A. Anyasi**: Supervision; writing—review and editing. **Victor Mlambo**: Supervision; writing—review and editing. **Obiro Cuthbert Wokadala**: Conceptualization; supervision; funding acquisition; writing—review and editing.

ACKNOWLEDGMENTS

This work is supported by the National Research Foundation (NRF) of South Africa (grant number 116308). We acknowledge Professor Naushad Emmambux of the University of Pretoria for his advice on starch chemistry.

CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

ORCID

Obiro Cuthbert Wokadala https://orcid.org/0000-0002-0292-400X

REFERENCES

Abdullah, A. H. D., Chalimah, S., Primadona, I., & Hanantyo, M. H. G. (2018). Physical and chemical properties of corn, cassava, and potato starchs. *IOP Conference Series: Earth and Environmental*

Science, *160*, Article 012003. https://doi.org/10.1088/1755-1315/160/1/012003

Abutair, A. S., Naser, I. A., & Hamed, A. T. (2016). Soluble fibers from psyllium improve glycemic response and body weight among diabetes type 2 patients (randomized control trial). *Nutrition Journal*, *15*, Article 86. https://doi.org/10.1186/s12937-016-0207-4

Ademosun, A. O., Odanye, O. S., & Oboh, G. (2021). Orange peel flavored unripe plantain noodles with low glycemic index improved antioxidant status and reduced blood glucose levels in diabetic rats. *Journal of Food Measurement and Characterization*, *15*(4), 3742–3751. https://doi.org/10.1007/s11694-021-00953-3

Agama-Acevedo, E., Sañudo-Barajas, J. A., Vélez De La Rocha, R., González-Aguilar, G. A., & Bello-Peréz, L. A. (2016). Potential of plantain peels flour (*Musa paradisiaca* L.) as a source of dietary fiber and antioxidant compound. *CyTA - Journal of Food*, *14*(1), 117–123. https://doi.org/10.1080/19476337.2015.1055306

Åkerberg, A., Liljeberg, H., & Björck, I. (1998). Effects of amylose/amylopectin ratio and baking conditions on resistant starch formation and glycaemic indices. *Journal of Cereal Science*, *28*(1), 71–80. https://doi.org/10.1006/jcrs.1997.0173

Almanza-Benitez, S., Osorio-Díaz, P., Méndez-Montealvo, G., Islas-Hernández, J. J., & Bello-Perez, L. A. (2015). Addition of acid-treated unripe plantain flour modified the starch digestibility, indigestible carbohydrate content and antioxidant capacity of semolina spaghetti. *LWT - Food Science and Technology*, *62*(2), 1127–1133. https://doi.org/10.1016/j.lwt.2015.02.031

Annor, G. A., Asamoah-Bonti, P., & Sakyi-Dawson, E. (2016). Fruit physical characteristics, proximate, mineral and starch characterization of FHIA 19 and FHIA 20 plantain and FHIA 03 cooking banana hybrids. *SpringerPlus*, *5*(1), Article 796. https://doi.org/10.1186/s40064-016-2465-1

Anyasi, T. A., Jideani, A. I., & Mchau, G. R. (2017). Effects of organic acid pretreatment on microstructure, functional and thermal properties of unripe banana flour. *Journal of Food Measurement and Characterization*, *11*(1), 99–110.

Anyasi, T. A., Jideani, A. I. O., & McHau, G. R. A. (2018). Phenolics and essential mineral profile of organic acid pretreated unripe banana flour. *Food Research International*, *104*, 100–109. https://doi.org/10.1016/j.foodres.2017.09.063

Aparicio-Saguilán, A., Flores-Huicochea, E., Tovar, J., García-Suárez, F., Gutiérrez-Meraz, F., & Bello-Pérez, L. A. (2005). Resistant starch-rich powders prepared by autoclaving of native and lintnerized banana starch: Partial characterization. *Starch - Stärke*, *57*(9), 405–412. https://doi.org/10.1002/star.200400386

Aparicio-Saguilán, A., Gutiérrez-Meraz, F., García-Suárez, F. J., Tovar, J., & Bello-Pérez, L. A. (2008). Physicochemical and functional properties of cross-linked banana resistant starch. Effect of pressure cooking. *Starch - Stärke*, *60*(6), 286–291. https://doi.org/10.1002/star.200600584

Bennett, R. N., Shiga, T. M., Hassimotto, N. M., Rosa, E. A., Lajolo, F. M., & Cordenunsi, B. R. (2010). Phenolics and antioxidant properties of fruit pulp and cell wall fractions of postharvest banana (*Musa acuminata* Juss.) cultivars. *Journal of Agricultural and Food Chemistry*, *58*(13), 7991–8003.

Bello-Perez, L. A., Flores-Silva, P. C., Agama-Acevedo, E., & Tovar, J. (2020). Starch digestibility: past, present, and future. *Journal of the Science of Food and Agriculture*, *100*(14), 5009–5016. https://doi.org/10.1002/jsfa.8955

Bello-Perez, L. A., Flores-Silva, P. C., Utrilla-Coello, R. G., Agama-Acevedo, E., & Hamaker, B. R. (2015). In Vitro Starch Digestibility of gluten-free spaghetti based on maize, chickpea, and unripe plantain flours. *Cereal Chemistry*, *92*(2), 171–176. https://doi.org/10.1094/CCHEM-06-14-0124-R

Bezerra, C. V., Amante, E. R., de Oliveira, D. C., Rodrigues, A. M. C., & da Silva, L. H. M. (2013). Green banana (*Musa cavendishii*) flour obtained in spouted bed—Effect of drying on physico-chemical, functional and morphological characteristics of the starch. *Industrial Crops and Products*, *41*, 241–249. https://doi.org/10.1016/j.indcrop.2012.04.035

Bi, Y., Zhang, Y., Jiang, H., Hong, Y., Gu, Z., Cheng, L., Li, Z., & Li, C. (2017). Molecular structure and digestibility of banana flour and starch. *Food Hydrocolloids*, *72*, 219–227. https://doi.org/10.1016/j.foodhyd.2017.06.003

Birt, D. F., Boylston, T., Hendrich, S., Jane, J.-L., Hollis, J., Li, L., McClelland, J., Moore, S., Phillips, G. J., Rowling, M., Schalinske, K., Scott, M. P., & Whitley, E. M. (2013). Resistant starch: Promise for improving human health. *Advances in Nutrition*, *4*, 587–601. https://doi.org/10.3945/an.113.004325

Björck, I., Granfeldt, Y., Liljeberg, H., Tovar, J., & Asp, N. G. (1994). Food properties affecting the digestion and absorption of carbohydrates. *The American Journal of Clinical Nutrition*, *59*(3), 699S–705S. https://doi.org/10.1093/ajcn/59.3.699S

Butterworth, P. J., Warren, F. J., & Ellis, P. R. (2011). Human α-amylase and starch digestion: An interesting marriage. *Starch/Starke*, *63*, 395–405.

Cahyana, Y., Wijaya, E., Halimah, T. S., Marta, H., Suryadi, E., & Kurniati, D. (2019). The effect of different thermal modifications on slowly digestible starch and physicochemical properties of green banana flour (*Musa acuminata colla*). *Food Chemistry*, *274*, 274–280. https://doi.org/10.1016/j.foodchem.2018.09.004

Campuzano, A., Rosell, C. M., & Cornejo, F. (2018). Physicochemical and nutritional characteristics of banana flour during ripening. *Food Chemistry*, *256*, 11–17. https://doi.org/10.1016/j.foodchem.2018.02.113

Castelo-Branco, V. N., Guimarães, J. N., Souza, L., Guedes, M. R., Silva, P. M., Ferrão, L. L., Miyahira, R. F., Guimarães, R. R., Freitas, S. M. L., Citelli, M., Zago, L., & Reis, M. C. D. (2017). The use of green banana (*Musa balbisiana*) pulp and peel flour as an ingredient for tagliatelle pasta. *Brazilian Journal of Food Technology*, *20*, Article e2016119.

Chaipai, S., Kriangsinyot, W., & Srichamnong, W. (2018). Effects of ripening stage and cooking methods on available glucose, resistant starch and estimated glycemic index of bananas (*Musa sapientum*; Nam-wa variety). *Malaysian Journal of Nutrition*, *24*(2), 269–279.

Chang, L., Yang, M., Zhao, N., Xie, F., Zheng, P., Simbo, J., Yu, X., & Du, S.-k. (2022). Structural, physicochemical, antioxidant and in vitro digestibility properties of banana flours from different banana varieties (*Musa* spp.). *Food Bioscience*, *47*, Article 101624. https://doi.org/10.1016/j.fbio.2022.101624

Chávez-Salazar, A., Bello-Pérez, L. A., Agama-Acevedo, E., Castellanos-Galeano, F. J., Álvarez-Barreto, C. I., & Pacheco-Vargas, G. (2017). Isolation and partial characterization of starch from banana cultivars grown in Colombia. *International Journal of Biological Macromolecules*, *98*, 240–246. https://doi.org/10.1016/j.ijbiomac.2017.01.024

Chi, C., Li, X., Zhang, Y., Chen, L., Li, L., & Wang, Z. (2017). Digestibility and supramolecular structural changes of maize starch by non-covalent interactions with gallic acid. *Food & Function*, *8*(2), 720–730. https://doi.org/10.1039/C6FO01468B

Chi, C., Shi, M., Zhao, Y., Chen, B., He, Y., & Wang, M. (2022). Dietary compounds slow starch enzymatic digestion: A review. *Frontiers in Nutrition*, *9*, Article 1004966.

Choo, C. L., & Aziz, N. A. A. (2010). Effects of banana flour and β-glucan on the nutritional and sensory evaluation of noodles. *Food Chemistry*, *119*(1), 34–40. https://doi.org/10.1016/j.foodchem.2009.05.004

Cordoba, L. D. P., da Silva, R. G., Gomes, D. D. S., Schnitzler, E., & Waszczynskyj, N. (2018). Brazilian green banana. *Journal of Thermal Analysis & Calorimetry*, *134*(3), 2065–2073. https://doi.org/10.1007/s10973-018-7374-9

Cornejo-Ramírez, Y. I., Martínez-Cruz, O., Del Toro-Sánchez, C. L., Wong-Corral, F. J., Borboa-Flores, J., & Cinco-Moroyoqui, F. J. (2018). The structural characteristics of starches and their functional properties. *CyTA - Journal of Food*, *16*(1), 1003–1017. https://doi.org/10.1080/19476337.2018.1518343

Dan, M. C. T. (2011). *Avaliação da potencialidade da farinha de banana verde como ingrediente funcional: Estudo in vivo e in vitro*. Universidade de São Paulo.

Das, M., Rajan, N., Biswas, P., & Banerjee, R. (2022). A novel approach for resistant starch production from green banana flour using amylopullulanase. *LWT - Food Science and Technology*, *153*, Article 112391. https://doi.org/10.1016/j.lwt.2021.112391

de Angelis-Pereira, M. C., Barcelos, M. D. F. P., Pereira, R. C., Pereira, J. D. A. R., & de Sousa, R. V. (2016). Chemical composition of unripe banana peels and pulps flours and its effects on blood glucose of rats. *Nutrition & Food Science*, *46*(4), 504–516.

De la Rosa-Millan, J., Agama-Acevedo, E., Osorio-Díaz, P., & Bello-Pérez, L. (2014). Effect of cooking, annealing and storage on starch digestibility and physicochemical characteristics of unripe banana flour. *Revista Mexicana de Ingeniería Química*, *13*(1), 151–163.

Dhital, S., Butardo, V. M., Jr., Jobling, S. A., & Gidley, M. J. (2015). Rice starch granule amylolysis—Differentiating effects of particle size, morphology, thermal properties and crystalline polymorph. *Carbohydrate Polymers*, *115*, 305–316. https://doi.org/10.1016/j.carbpol.2014.08.091

do Prado Ferreira, M., & Teixeira Tarley, C. R. (2020). Assessment of in vitro bioacessibility of macrominerals and trace elements in green banana flour. *Journal of Food Composition and Analysis*, *92*, Article 103586. https://doi.org/10.1016/j.jfca.2020.103586

Englyst, H. N., & Cummings, J. H. (1985). Digestion of the polysaccharides of some cereal foods in the human small intestine. *American Journal of Clinical Nutrition*, *42*(5), 778–787. https://doi.org/10.1093/ajcn/42.5.778

Espinosa-Solis, V., Jane, J.-l., & Bello-Perez, L. A. (2009). Physicochemical characteristics of starches from unripe fruits of mango and banana. *Starch - Stärke*, *61*(5), 291–299. https://doi.org/10.1002/star.200800103

Faisant, N., Buléon, A., Colonna, P., Molis, C., Lartigue, S., Galmiche, J. P., & Champ, M. (1995). Digestion of raw banana starch in the small intestine of healthy humans: Structural features of resistant starch. *British Journal of Nutrition*, *73*(1), 111–123. https://doi.org/10.1079/BJN19950013

Falade, K. O., & Olugbuyi, A. O. (2010). Effects of maturity and drying method on the physico-chemical and reconstitution properties of plantain flour. *International Journal of Food Science & Technology*, *45*(1), 170–178. https://doi.org/10.1111/j.1365-2621.2009.02118.x

Falcomer, A. L., Riquette, R. F. R., de Lima, B. R., Ginani, V. C., & Zandonadi, R. P. (2019). Health benefits of green banana consumption: A systematic review. *Nutrients*, *11*(6), Article 1222. https://doi.org/10.3390/nu11061222

Falodun, A. I., Ayo-Omogie, H. N., & Awolu, O. O. (2019). Effect of different drying techniques on the resistant starch, bioactive components, physicochemical and pasting properties of Cardaba banana flour. *Acta Universitatis Cinbinesis, Series E: Food Technology*, *23*(1), 35–42.

Gao, H., Huang, S., Dong, T., Yang, Q., & Yi, G. (2016). Analysis of resistant starch degradation in postharvest ripening of two banana cultivars: Focus on starch structure and amylases. *Postharvest Biology and Technology*, *119*, 1–8.

Giacco, R., Vitale, M., & Riccardi, G. (2016). Pasta: Role in diet. In B. Caballero, P. M. Finglas, & F. Toldrá (Eds.), *Encyclopedia of food and health* (pp. 242–245). Academic Press.

Gibert, O., Alemán, S., Guzman, R., Raymundez, M., Laurentin, A., Manzanilla, E., Ricci, J., & Perez, E. (2014). Morphometric variations of banana starches issued from various genomic groups and in vitro starch digestibility. *Enliven: Journal of Dietetics Research and Nutrition*, *1*(1), Article 003.

Giraldo-Gómez, G. I., Rodríguez-Barona, S., & Sanabria-González, N. R. (2019). Preparation of instant green banana flour powders by an extrusion process. *Powder Technology*, *353*, 437–443. https://doi.org/10.1016/j.powtec.2019.05.050

Giuberti, G., Rocchetti, G., & Lucini, L. (2020). Interactions between phenolic compounds, amylolytic enzymes and starch: An updated overview. *Current Opinion in Food Science*, *31*, 102–113. https://doi.org/10.1016/j.cofs.2020.04.003

Goff, H. D., & Guo, Q. (2019). The role of hydrocolloids in the development of food structure. In F. Spyropoulos, A. Lazidis, & I. Norton (Eds.), *Handbook of food structure development* (pp. 1–28). The Royal Society of Chemistry.

González-Soto, R. A., Mora-Escobedo, R., Hernández-Sánchez, H., Sánchez-Rivera, M., & Bello-Pérez, L. A. (2007). The influence of time and storage temperature on resistant starch formation from autoclaved debranched banana starch. *Food Research International*, *40*(2), 304–310. https://doi.org/10.1016/j.foodres.2006.04.001

Gu, C., Suleria, H. A. R., Dunshea, F. R., & Howell, K. (2020). Dietary lipids influence bioaccessibility of polyphenols from black carrots and affect microbial diversity under simulated gastrointestinal digestion. *Antioxidants*, *9*(8), Article 762. https://www.mdpi.com/2076-3921/9/8/762

Gutiérrez, T. J. (2018). Plantain flours as potential raw materials for the development of gluten-free functional foods. *Carbohydrate Polymers*, *202*, 265–279. https://doi.org/10.1016/j.carbpol.2018.08.121

Gutiérrez, T. J., & Álvarez, K. (2016). Physico-chemical properties and in vitro digestibility of edible films made from plantain flour with added Aloe vera gel. *Journal of Functional Foods*, *26*, 750–762. https://doi.org/10.1016/j.jff.2016.08.054

Han, M., Bao, W., Wu, Y., & Ouyang, J. (2020). Insights into the effects of caffeic acid and amylose on in vitro digestibility of maize starch-caffeic acid complex. *International Journal of Biological Macromolecules*, *162*, 922–930. https://doi.org/10.1016/j.ijbiomac.2020.06.200

Harnly, J. M., Doherty, R. F., Beecher, G. R., Holden, J. M., Haytowitz, D. B., Bhagwat, S., & Gebhardt, S. (2006). Flavonoid content of

U.S. fruits, vegetables, and nuts. *Journal of Agricultural and Food Chemistry*, *54*(26), 9966–9977. https://doi.org/10.1021/jf061478aSC

Haslinda, W. H., Cheng, L. H., Chong, L. C., & Aziah, A. A. N. (2009). Chemical composition and physicochemical properties of green banana (*Musa acuminata* × *balbisiana Colla* cv. *Awak*) flour. *International Journal of Food Sciences and Nutrition*, *60*(sup4), 232–239. https://doi.org/10.1080/09637480902915525

Hernández-Nava, R., Bello-Pérez, L., Martín-Martínez, S., Hernández-Sánchez, H., & Mora-Escobedo, R. (2011). Effect of extrusion cooking on the functional properties and starch components of lentil/banana blends: Response surface analysis. *Revista Mexicana de Ingenieria Quimica*, *10*(3), 409–419.

Hernández-Nava, R., Berrios, J. D. J., Pan, J., Osorio-Diaz, P., & Bello-Perez, L. A. (2009). Development and characterization of spaghetti with high resistant starch content supplemented with banana starch. *Food Science and Technology International*, *15*(1), 73–78.

Hoffmann Sardá, F. A., de Lima, F. N. R., Lopes, N. T. T., Santos, A. d. O., Tobaruela, E. d. C., Kato, E. T. M., & Menezes, E. W. (2016). Identification of carbohydrate parameters in commercial unripe banana flour. *Food Research International*, *81*, 203–209. https://doi.org/10.1016/j.foodres.2015.11.016

Jane, J.-L., Wong, K.-S., & McPherson, A. E. (1997). Branch-structure difference in starches of A- and B-type X-ray patterns revealed by their Naegeli dextrins. *Carbohydrate Research*, *300*(3), 219–227. https://doi.org/10.1016/S0008-6215(97)00056-6

Jenkins, D. J. A., Wolever, T. M. S., & Jenkins, A. L. (1988). Starchy foods and glycemic index. *Diabetes Care*, *11*(2), 149–159. https://doi.org/10.2337/diacare.11.2.149

Jiang, H., Zhang, Y., Hong, Y., Bi, Y., Gu, Z., Cheng, L., & Li, Z., & Li, C. (2015). Digestibility and changes to structural characteristics of green banana starch during in vitro digestion. *Food Hydrocolloids*, *49*, 192–199.

Juarez-Garcia, E., Agama-Acevedo, E., SÁYago-Ayerdi, S. G., RodrÍGuez-Ambriz, S. L., & Bello-PÉRez, L. A. (2006). Composition, digestibility and application in breadmaking of banana flour. *Plant Foods for Human Nutrition*, *61*(3), 131–137. https://doi.org/10.1007/s11130-006-0020-x

Kan, L., Capuano, E., Oliviero, T., & Renzetti, S. (2022). Wheat starch-tannic acid complexes modulate physicochemical and rheological properties of wheat starch and its digestibility. *Food Hydrocolloids*, *126*, Article 107459. https://doi.org/10.1016/j.foodhyd.2021.107459

Khawas, P., & Deka, S. C. (2017). Effect of modified resistant starch of culinary banana on physicochemical, functional, morphological, diffraction, and thermal properties. *International Journal of Food Properties*, *20*(1), 133–150. https://doi.org/10.1080/10942912.2016.1147459

Khoozani, A. A., Bekhit, A. E.-D. A., & Birch, J. (2019). Effects of different drying conditions on the starch content, thermal properties and some of the physicochemical parameters of whole green banana flour. *International Journal of Biological Macromolecules*, *130*, 938–946. https://doi.org/10.1016/j.ijbiomac.2019.03.010

Khoozani, A. A., Birch, J., & Bekhit, A. E.-D. A. (2020). Textural properties and characteristics of whole green banana flour produced by air-oven and freeze-drying processing. *Journal of Food Measurement and Characterization*, *14*, 1533–1542.

Kim, M. A., Choi, S. J., & Moon, T. W. (2015). Digestibility of retrograded starches with A- and B-type crystalline structures. *Journal of the Korean Society for Applied Biological Chemistry*, *58*(4), 487–490. https://doi.org/10.1007/s13765-015-0069-z

Koh, A., Vadder, F. D., Kovatcheva-Datchary, P., & Backhed, F. (2016). From dietary fiber to host physiology: Short-chain fatty acids as key bacterial metabolites. *Cell*, *165*, 1332–1345.

Kongolo, J. I., Da Silva, L. S., Wokadala, O. C., Du Plessis, B., Husselman, J., Ngcobo, M. E. K., Emmambux, N. M., & Daneel, M. (2017). Pasting, thermal, gel texture, resistant starch and colour properties of unripe banana flour from 10 desert banana varieties cultivated in South Africa. *Journal of Food Measurement and Characterization*, *11*(3), 1056–1064. https://doi.org/10.1007/s11694-017-9481-x

Kumar, P. S., Saravanan, A., Sheeba, N., & Uma, S. (2019). Structural, functional characterization and physicochemical properties of green banana flour from dessert and plantain bananas (*Musa* spp.). *LWT - Food Science and Technology*, *116*, Article 108524. https://doi.org/10.1016/j.lwt.2019.108524

La Fuente, C. I., & Tadini, C. C. (2017). Unripe banana flour produced by air-drying assisted with ultrasound–description of the mechanisms involved to enhance the mass transfer in two approaches. *International Journal of Food Engineering*, *13*(11), Article 20170178.

Langkilde, A. M., Champ, M., & Andersson, H. (2002). Effects of high-resistant-starch banana flour (RS2) on in vitro fermentation and the small-bowel excretion of energy, nutrients, and sterols: An ileostomy study. *The American Journal of Clinical Nutrition*, *75*(1), 104–111.

Lehmann, U., Jacobasch, G., & Schmiedl, D. (2002). Characterization of resistant starch type III from banana (*Musa acuminata*). *Journal of Agricultural and Food Chemistry*, *50*(18), 5236–5240.

Li, L., Liu, Z., Zhang, W., Xue, B., & Luo, Z. (2021). Production and applications of amylose-lipid complexes as resistant starch: Recent approaches. *Starch - Stärke*, *73*(5-6), Article 2000249. https://doi.org/10.1002/star.202000249

Li, M.-C., Chou, C.-F., Hsu, S.-C., & Lin, J.-S. (2020). Physicochemical characteristics and resistant starch of different varieties of banana from Taiwan. *International Journal of Food Properties*, *23*(1), 1168–1175. https://doi.org/10.1080/10942912.2020.1788077

Liao, H.-J., & Hung, C.-C. (2015). Chemical composition and in vitro starch digestibility of green banana (cv. Giant Cavendish) flour and its derived autoclaved/debranched powder. *LWT - Food Science and Technology*, *64*(2), 639–644.

Liu, Q. F., Yan, Z., & Feng, W. J. (2017). Fracture behaviors for a functionally graded superconducting cylinder with a cylindrical crack. *Mechanics of Advanced Materials and Structures*, *24*(2), 176–182. https://doi.org/10.1080/15376494.2015.1124954

Lockyer, S., & Nugent, A. P. (2017). Health effects of resistant starch. *Nutrition Bulletin*, *42*, 10–41. https://doi.org/10.1111/nbu.12244

Ma, Z., & Boye, J. I. (2018). Research advances on structural characterization of resistant starch and its structure-physiological function relationship: A review. *Critical Reviews in Food Science and Nutrition*, *58*(7), 1059–1083. https://doi.org/10.1080/10408398.2016.1230537

Ma, Z., Hu, X., & Boye, J. I. (2020). Research advances on the formation mechanism of resistant starch type III: A review. *Critical Reviews in Food Science and Nutrition*, *60*(2), 276–297. https://doi.org/10.1080/10408398.2018.1523785

Magallanes-Cruz, P. A., Flores-Silva, P. C., & Bello-Perez, L. A. (2017). Starch structure influences its digestibility: A review. *Journal of Food Science*, *82*(9), 2016–2023. https://doi.org/10.1111/1750-3841.13809

Martens, B. M. J., Gerrits, W. J. J., Bruininx, E. M. A. M., & Schols, H. A. (2018). Amylopectin structure and crystallinity explains variation in digestion kinetics of starches across botanic sources in an in vitro pig model. *Journal of Animal Science and Biotechnology*, *9*(1), Article 91. https://doi.org/10.1186/s40104-018-0303-8

Martínez, M. E. S., Aguilar Méndez, M. A., Sánchez Solís, A., & Vieyra, H. (2015). Thermoplastic biodegradable material elaborated from unripe banana flour reinforced with metallocene catalyzed polyethylene. *Polymer Engineering & Science*, *55*(4), 866–876. https://doi.org/10.1002/pen.23954

Maseko, K. H., Regnier, T., Meiring, B., Wokadala, O. C., & Anyasi, T. A. (2024). *Musa* species variation, production, and the application of its processed flour: A review. *Scientia Horticulturae*, *325*, Article 112688. https://doi.org/10.1016/j.scienta.2023.112688

Meenu, M., & Xu, B. (2019). A critical review on anti-diabetic and anti-obesity effects of dietary resistant starch. *Critical Reviews in Food Science and Nutrition*, *59*(18), 3019–3031. https://doi.org/10.1080/10408398.2018.1481360

Menezes, E. W., Dan, M. C., Cardenette, G. H., Goñi, I., Bello-Pérez, L. A., & Lajolo, F. M. (2010). In vitro colonic fermentation and glycemic response of different kinds of unripe banana flour. *Plant Foods for Human Nutrition*, *65*(4), 379–385.

Menezes, E. W., Tadini, C. C., Tribess, T. B., Zuleta, A., Binaghi, J., Pak, N., Vera, G., Dan, M. C. T., Bertolini, A. C., Cordenunsi, B. R., & Lajolo, F. M. (2011). Chemical composition and nutritional value of unripe banana flour (*Musa acuminata*, var. Nanicão). *Plant Foods for Human Nutrition*, *66*(3), 231–237. https://doi.org/10.1007/s11130-011-0238-0

Miao, M., Jiang, B., Cui, S. W., Zhang, T., & Jin, Z. (2015). Slowly digestible starch—A review. *Critical Reviews in Food Science and Nutrition*, *55*(12), 1642–1657. https://doi.org/10.1080/10408398.2012.704434

Milani, J., & Maleki, G. (2012). Hydrocolloids in food industry. *Food Industrial Processes - Methods and Equipment*, *2*, 2–37.

Moongngarm, A., Tiboonbun, W., Sanpong, M., Sriwong, P., Phiewtong, L., Prakitrum, R., & Huychan, N. (2014). Resistant starch and bioactive contents of unripe banana flour as influenced by harvesting periods and its application. *American Journal of Agricultural and Biological Sciences*, *9*(3), 457–465.

Moore, S. A., Ai, Y., Chang, F., & Jane, J.-L. (2015). Effects of alpha-amylase reaction mechanisms on analysis of resistant-starch contents. *Carbohydrate Polymers*, *115*, 465–471.

Naguleswaran, S., Vasanthan, T., Hoover, R., & Bressler, D. (2014). Amylolysis of amylopectin and amylose isolated from wheat, triticale, corn and barley starches. *Food Hydrocolloids*, *35*, 686–693. https://doi.org/10.1016/j.foodhyd.2013.08.018

Narayana, C., Jeyabaskaran, K., & Mustaffa, M. (2017). Chemical and mineral composition of four cultivars of banana (*Musa* sp.) belonging to different genomic groups grown in India. *International Journal of Current Microbiology and Applied Science*, *6*(6), 2862–2867.

Nasrin, T. A. A., & Anal, A. K. (2014a). Resistant starch: Properties, preparations and applications in functional foods. In A. Noomhorm, I. Ahmad, & A. K. Anal (Eds.), *Functional foods and dietary supplements: Processing effects and health benefits* (pp. 228–253). Wiley.

Nasrin, T. A. A., & Anal, A. K. (2014b). Resistant starch III from culled banana and its functional properties in fish oil emulsion. *Food Hydrocolloids*, *35*, 403–409.

Nasrin, T. A. A., Noomhorm, A., & Anal, A. K. (2015). Physico-chemical characterization of culled plantain pulp starch, peel starch, and flour. *International Journal of Food Properties*, *18*(1), 165–177. https://doi.org/10.1080/10942912.2013.828747

Ng, Y. V., Tengku Ismail, T. A., & Wan Ishak, W. R. (2020). Effect of overripe banana in developing high dietary fibre and low glycaemic index cookie. *British Food Journal*, *122*(10), 3165–3177. https://doi.org/10.1108/BFJ-12-2019-0934

Ngo, T. V., Kusumawardani, S., Kunyanee, K., & Luangsakul, N. (2022). Polyphenol-modified starches and their applications in the food industry: Recent updates and future directions. *Foods*, *11*(21), Article 3384. https://www.mdpi.com/2304-8158/11/21/3384

Okumus, B. N., Tacer-Caba, Z., Kahraman, K., & Nilufer-Erdil, D. (2018). Resistant starch type V formation in brown lentil (*Lens culinaris* Medikus) starch with different lipids/fatty acids. *Food Chemistry*, *240*, 550–558. https://doi.org/10.1016/j.foodchem.2017.07.157

Padhi, S., & Dwivedi, M. (2022). Physico-chemical, structural, functional and powder flow properties of unripe green banana flour after the application of Refractance window drying. *Future Foods*, *5*, Article 100101. https://doi.org/10.1016/j.fufo.2021.100101

Pelissari, F. M., Andrade-Mahecha, M. M., do Amaral Sobral, P. J., & Menegalli, F. C. (2012). Isolation and characterization of the flour and starch of plantain bananas (*Musa paradisiaca*). *Starch - Stärke*, *64*(5), 382–391. https://doi.org/10.1002/star.201100133

Pereira, A., & Maraschin, M. (2015). Banana (*Musa* spp) from peel to pulp: Ethnopharmacology, source of bioactive compounds and its relevance for human health. *Journal of Ethnopharmacology*, *160*, 149–163. https://doi.org/10.1016/j.jep.2014.11.008

Perera, A., Meda, V., & Tyler, R. T. (2010). Resistant starch: A review of analytical protocols for determining resistant starch and of factors affecting the resistant starch content of foods. *Food Research International*, *43*, 1959–1974.

Phothisoot, T., Gavahian, M., & Lin, J. (2023). Sustainable superheated steam drying of unripe banana (*Musa acuminata* cv. Hom Thong): Effects on resistant starch and physicochemical–functional properties. *Starch - Stärke*, *00*, 00–00. https://doi.org/10.1002/star.202300101

Pico, J., Corbin, S., Ferruzzi, M. G., & Martinez, M. M. (2019). Banana flour phenolics inhibit trans-epithelial glucose transport from wheat cakes in a coupled in vitro digestion/Caco-2 cell intestinal model. *Food & Function*, *10*(10), 6300–6311. https://doi.org/10.1039/C9FO01679A

Pico, J., Xu, K., Guo, M., Mohamedshah, Z., Ferruzzi, M. G., & Martinez, M. M. (2019). Manufacturing the ultimate green banana flour: Impact of drying and extrusion on phenolic profile and starch bioaccessibility. *Food Chemistry*, *297*, Article 124990. https://doi.org/10.1016/j.foodchem.2019.124990

Qi, X., Al-Ghazzewi, F. H., & Tester, R. F. (2018). Dietary fiber, gastric emptying, and carbohydrate digestion: A mini-review. *Starch - Stärke*, *70*(9-10), Article 1700346. https://doi.org/10.1002/star.201700346

Qi, X., & Tester, R. F. (2016). Effect of composition and structure of native starch granules on their susceptibility to hydrolysis by amylase enzymes. *Starch - Stärke*, *68*(9-10), 811–815. https://doi.org/10.1002/star.201600063

Qin, R., Yu, J., Li, Y., Copeland, L., Wang, S., & Wang, S. (2019). Structural changes of starch–lipid complexes during postprocessing and their effect on in vitro enzymatic digestibility. *Journal of Agricultural and Food Chemistry*, *67*(5), 1530–1536.

Rabbani, G. H., Ahmed, S., Hossain, I., Islam, R., Marni, F., Akhtar, M., & Majid, N. (2009). Green banana reduces clinical severity of childhood shigellosis: A double-blind, randomized, controlled clinical trial. *The Pediatric Infectious Disease Journal*, *28*(5), 420–425.

Rachman, A., Brennan, M. A., Morton, J., & Brennan, C. S. (2021). Starch pasting properties, and the effects of banana flour and cassava flour addition to semolina flour on starch and amino acid digestion. *Starch - Stärke*, *73*(1-2), Article 2000137. https://doi.org/10.1002/star.202000137

Raigond, P., Ezekiel, R., & Raigond, B. (2015). Resistant starch in food: A review. *Journal of the Science of Food and Agriculture*, *95*, 1968–1978.

Ramos, D. P., Leonel, M., & Leonel, S. (2009). Resistant starch in green banana flour/Amido resistente em farinhas de banana verde. *Alimentos e Nutricao (Brazilian Journal of Food and Nutrition)*, *20*(3), 479–484.

Rayo, L. M., Chaguri e Carvalho, L., Sardá, F. A. H., Dacanal, G. C., Menezes, E. W., & Tadini, C. C. (2015). Production of instant green banana flour (*Musa cavendischii*, var. Nanicão) by a pulsed-fluidized bed agglomeration. *LWT - Food Science and Technology*, *63*(1), 461–469. https://doi.org/10.1016/j.lwt.2015.03.059

Rebello, L. P. G., Ramos, A. M., Pertuzatti, P. B., Barcia, M. T., Castillo-Muñoz, N., & Hermosín-Gutiérrez, I. (2014). Flour of banana (*Musa* AAA) peel as a source of antioxidant phenolic compounds. *Food Research International*, *55*, 397–403. https://doi.org/10.1016/j.foodres.2013.11.039

Reis, R. C., de Souza Viana, E., de Assis, S. L. F., de Oliveira Sena, L., dos Santos de Souza, A., & Amorim, E. P. (2019). Promising green banana and plantain genotypes for making flour. *Pesquisa Agropecuária Brasileira*, *54*, Article e01303.

Rodríguez-Ambriz, S., Islas-Hernández, J., Agama-Acevedo, E., Tovar, J., & Bello-Pérez, L. (2008). Characterization of a fibre-rich powder prepared by liquefaction of unripe banana flour. *Food Chemistry*, *107*(4), 1515–1521.

Rodríguez-Damian, A. R., De, L., Rosa-Millán, J., Agama-Acevedo, E., Osorio-Díaz, P., & Bello-Pérez, L. A. (2013). Effect of different thermal treatments and storage on starch digestibility and physicochemical characteristics of unripe banana flour. *Journal of Food Processing and Preservation*, *37*(5), 987–998. https://doi.org/10.1111/j.1745-4549.2012.00737.x

Romero Hernández, H. A., Gutiérrez, T. J., & Bello-Pérez, L. A. (2022). Can starch-polyphenol V-type complexes be considered as resistant starch? *Food Hydrocolloids*, *124*, Article 107226. https://doi.org/10.1016/j.foodhyd.2021.107226

Rosado, C. P., Rosa, V. H. C., Martins, B. C., Soares, A. C., Santos, I. B., Monteiro, E. B., Moura-Nunes, N., da Costa, C. A., Mulder, A. D. R. P., & Daleprane, J. B. (2020). Resistant starch from green banana (*Musa* sp.) attenuates non-alcoholic fat liver accumulation and increases short-chain fatty acids production in high-fat diet-induced obesity in mice. *International Journal of Biological Macromolecules*, *145*, 1066–1072.

Saha, D., & Bhattacharya, S. (2010). Hydrocolloids as thickening and gelling agents in food: A critical review. *Journal of Food Science and Technology*, *47*(6), 587–597.

Salazar, D., Arancibia, M., Lalaleo, D., Rodríguez-Maecker, R., López-Caballero, M. E., & Montero, M. P. (2022). Physicochemical properties and filmogenic aptitude for edible packaging of Ecuadorian discard green banana flours (*Musa acuminanta* AAA). *Food Hydrocolloids*, *122*, Article 107048. https://doi.org/10.1016/j.foodhyd.2021.107048

Sánchez-Rivera, M. M., del Carmen Núñez-Santiago, M., Bello-Pérez, L. A., Agama-Acevedo, E., & Alvarez-Ramirez, J. (2017). Citric acid esterification of unripe plantain flour: Physicochemical properties and starch digestibility. *Starch - Stärke*, *69*(9-10), Article 1700019. https://doi.org/10.1002/star.201700019

Sarawong, C., Schoenlechner, R., Sekiguchi, K., Berghofer, E., & Ng, P. K. W. (2014). Effect of extrusion cooking on the physicochemical properties, resistant starch, phenolic content and antioxidant capacities of green banana flour. *Food Chemistry*, *143*, 33–39. https://doi.org/10.1016/j.foodchem.2013.07.081

Shah, A., Zhang, G., Hamaker, B. R., & Campanella, O. H. (2011). Rheological properties of a soluble self-assembled complex from starch, protein and free fatty acids. *Journal of Food Engineering*, *105*(3), 444–452. https://doi.org/10.1016/j.jfoodeng.2011.02.048

Sidhu, J. S., & Zafar, T. A. (2018). Bioactive compounds in banana fruits and their health benefits. *Food Quality and Safety*, *2*(4), 183–188. https://doi.org/10.1093/fqsafe/fyy019

Silva, V. D. M., Arquelau, P. B. F., Silva, M. R., Augusti, R., Melo, J. O. F., & Fante, C. A. (2020). Use of paper spray-mass spectrometry to determine the chemical profile of ripe banana peel flour and evaluation of its physicochemical and antioxidant properties. *Química Nova*, *43*, 579–585.

Singh, B., Singh, J. P., Kaur, A., & Singh, N. (2016). Bioactive compounds in banana and their associated health benefits—A review. *Food Chemistry*, *206*, 1–11. https://doi.org/10.1016/j.foodchem.2016.03.033

Singh, J., Dartois, A., & Kaur, L. (2010). Starch digestibility in food matrix: A review. *Trends in Food Science & Technology*, *21*, 168–180.

Soares, C. A., Peroni-Okita, F. H. G., Cardoso, M. B., Shitakubo, R., Lajolo, F. M., & Cordenunsi, B. R. (2011). Plantain and banana starches: Granule structural characteristics explain the differences in their starch degradation patterns. *Journal of Agricultural and Food Chemistry*, *59*(12), 6672–6681. https://doi.org/10.1021/jf201590h

Tang, H., Watanabe, K., & Mitsunaga, T. (2002). Structure and functionality of large, medium and small granule starches in normal and waxy barley endosperms. *Carbohydrate Polymers*, *49*(2), 217–224. https://doi.org/10.1016/S0144-8617(01)00329-0

Tang, J., Liang, Q., Ren, X., Raza, H., & Ma, H. (2022). Insights into ultrasound-induced starch-lipid complexes to understand physicochemical and nutritional interventions. *International Journal of Biological Macromolecules*, *222*, 950–960. https://doi.org/10.1016/j.ijbiomac.2022.09.242

Tangthanantorn, J., Wichienchot, S., & Sirivongpaisal, P. (2021). Development of fresh and dried noodle products with high resistant starch content from banana flour. *Food Science and Technology*, *00*, 00–00.

Tester, R. F., Qi, X., & Karkalas, J. (2006). Hydrolysis of native starches with amylases. *Animal Feed Science and Technology*, *130*(1), 39–54. https://doi.org/10.1016/j.anifeedsci.2006.01.016

Thakaeng, P., Boonloom, T., & Rawdkuen, S. (2021). Physicochemical properties of bread partially substituted with unripe green banana (*Cavendish* spp.) flour. *Molecules*, *26*(7), Article 2070. https://www.mdpi.com/1420-3049/26/7/2070

Thakorlal, J., Perera, C., Smith, B., Englberger, L., & Lorens, A. (2010). Resistant starch in Micronesian banana cultivars offers health benefits. *Pacific Health Dialog*, *16*(1), 49–60.

Tian, D.-D., Xu, X.-Q., Peng, Q., Zhang, Y.-W., Zhang, P.-B., Qiao, Y., & Shi, B. (2020). Effects of banana powder (*Musa acuminata* Colla) on the composition of human fecal microbiota and metabolic output using in vitro fermentation. *Journal of Food Science*, *85*(8), 2554–2564. https://doi.org/10.1111/1750-3841.15324

Tian, S., & Sun, Y. (2020). Influencing factor of resistant starch formation and application in cereal products: A review. *International Journal of Biological Macromolecules*, *149*, 424–431. https://doi.org/10.1016/j.ijbiomac.2020.01.264

Tribess, T. B., Hernández-Uribe, J. P., Méndez-Montealvo, M. G. C., Menezes, E. W., Bello-Perez, L. A., & Tadini, C. C. (2009). Thermal properties and resistant starch content of green banana flour (*Musa cavendishii*) produced at different drying conditions. *LWT - Food Science and Technology*, *42*(5), 1022–1025. https://doi.org/10.1016/j.lwt.2008.12.017

Udomkun, P., Masso, C., Swennen, R., Innawong, B., Alakonya, A., Fotso Kuate, A., & Vanlauwe, B. (2021). How does cultivar, maturation, and pre-treatment affect nutritional, physicochemical, and pasting properties of plantain flours? *Foods*, *10*(8), Article 1749. https://www.mdpi.com/2304-8158/10/8/1749

Vatanasuchart, N., Niyomwit, B., & Wongkrajang, K. (2012). Resistant starch content, in vitro starch digestibility and physico-chemical properties of flour and starch from Thai bananas. *Maejo International Journal of Science and Technology*, *6*(2), 259–271.

Virulchatapan, P., & Luangsakul, N. (2020). Effect of harvesting period on physicochemical properties and in vitro digestibility of banana flour. *International Journal of Agricultural Technology*, *16*, 517–528.

Wang, J., Huang, H. H., & Chen, P. S. (2017). Structural and physicochemical properties of banana resistant starch from four cultivars. *International Journal of Food Properties*, *20*(6), 1338–1347.

Wang, Y., Ral, J.-P., Saulnier, L., & Kansou, K. (2022). How does starch structure impact amylolysis? Review of current strategies for starch digestibility study. *Foods*, *11*, Article 1223. https://doi.org/10.3390/foods11091223

Wokadala, O. C., Ray, S. S., & Emmambux, M. N. (2012). Occurrence of amylose–lipid complexes in teff and maize starch biphasic pastes. *Carbohydrate Polymers*, *90*(1), 616–622. https://doi.org/10.1016/j.carbpol.2012.05.086

Woomer, J. S., & Adedeji, A. A. (2021). Current applications of gluten-free grains—A review. *Critical Reviews in Food Science and Nutrition*, *61*(1), 14–24. https://doi.org/10.1080/10408398.2020.1713724

Yee, J., Roman, L., Pico, J., Aguirre-Cruz, A., Bello-Perez, L. A., Bertoft, E., & Martinez, M. M. (2021). The molecular structure of starch from different Musa genotypes: Higher branching density of amylose chains seems to promote enzyme-resistant structures. *Food Hydrocolloids*, *112*, Article 106351. https://doi.org/10.1016/j.foodhyd.2020.106351

Yoshio, N., Maeda, I., & Hisamatsu, M. (2011). Relationship between fine structure and properties of gelatinization and retrogradation of starches from wheat harvested in Japan and Canada. *Journal of Applied Glycoscience*, *59*(1), 1–9. https://doi.org/10.5458/jag.jag.JAG-2011_009

Zeng, H., Huang, C., Lin, S., Zheng, M., Chen, C., Zheng, B., & Zhang, Y. (2017). Lotus seed resistant starch regulates gut microbiota and increases short-chain fatty acids production and mineral absorption in mice. *Journal of Agricultural and Food Chemistry*, *65*(42), 9217–9225.

Zhang, G., Maladen, M., Campanella, O. H., & Hamaker, B. R. (2010). Free fatty acids electronically bridge the self-assembly of a three-component nanocomplex consisting of amylose, protein, and free fatty acids. *Journal of Agricultural and Food Chemistry*, *58*(16), 9164–9170. https://doi.org/10.1021/jf1010319

Zhang, G., Maladen, M. D., & Hamaker, B. R. (2003). Detection of a novel three component complex consisting of starch, protein, and free fatty acids. *Journal of Agricultural and Food Chemistry*, *51*(9), 2801–2805. https://doi.org/10.1021/jf030035t

Zhang, P., & Hamaker, B. R. (2012). Banana starch structure and digestibility. *Carbohydrate Polymers*, *87*(2), 1552–1558.

Zhang, P., Whistler, R. L., BeMiller, J. N., & Hamaker, B. R. (2005). Banana starch: Production, physicochemical properties, and digestibility—A review. *Carbohydrate Polymers*, *59*(4), 443–458. https://doi.org/10.1016/j.carbpol.2004.10.014

Zhang, Y., Li, B., Zhang, Y., Xu, F., Zhu, K., Li, S., Tan, L., Wu, G., & Dong, W. (2019). Effect of degree of polymerization of amylopectin on the gelatinization properties of jackfruit seed starch. *Food Chemistry*, *289*, 152–159. https://doi.org/10.1016/j.foodchem.2019.03.033

Zheng, Z., Stanley, R., Gidley, M. J., & Dhital, S. (2016). Structural properties and digestion of green banana flour as a functional ingredient in pasta. *Food & Function*, *7*(2), 771–780. https://doi.org/10.1039/C5FO01156F

How to cite this article: Dibakoane, S. R., Da Silva, L. S., Meiring, B., Anyasi, T. A., Mlambo, V., & Wokadala, O. C. (2024). The multifactorial phenomenon of enzymatic hydrolysis resistance in unripe banana flour and its starch: A concise review. *Journal of Food Science*, *89*, 5185–5204. https://doi.org/10.1111/1750-3841.17270