

RESISTANT STARCH IN BANANAS AND OTHER FRUITS: STRUCTURAL TO MOLECULAR CHARACTERISTICS FOR NUTRITIONAL SIGNIFICANCE AND HEALTH BENEFITS OF CONSUMING

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ABSTRACT

Resistant starch (RS) is the fraction of starch that escapes digestion in the small intestine and is instead fermented by microbes in the colon. Bananas, especially when unripe, are among the richest fruit sources of RS, accounting for 40-60% of their total starch content. As bananas ripen, their starch converts into soluble sugars, resulting in a decrease in RS content. Jack Fruit and Bread Fruit also contain significant levels of RS, ranging from 15-35%, which vary based on the cultivar, maturity, and processing methods. The structural properties of RS in fruits are influenced by the ratio of amylose to amylopectin, granule shape, and crystalline structure. Unripe banana starch exhibits B-type crystallinity, making it resistant to enzymatic digestion. As the fruit matures, crystallinity diminishes, affecting its digestibility. Processing methods like heat treatment, retrogradation, enzymatic modification, and dehydration can enhance RS formation by 20-50%. RS derived from fruits serves as dietary fiber and a texture enhancer in foods. The fermentation of RS produces short-chain fatty acids, notably butyrate, which supports gut health. RS content varies with genotype, maturity, and processing, necessitating optimized processing and cultivar selection for functional foods.

KEYWORDS: Resistant Starch, Granule morphology, Molecular characteristics, Digestibility assessment.

1 INTRODUCTION

Starch is the primary polysaccharide stored in plants and plays a crucial role in human nutrition. It consists of α -D-glucose units and is composed of two major fractions, amylose and amylopectin (1). Amylose is predominantly linear chain of α -D-glucose units linked through α -(1,4) glycosidic bonds. In contrast, amylopectin possesses a highly branched structure in which glucose units are linked by α -(1,4) glycosidic bonds with branching points formed by α -(1,6) linkages (2). Amylose forms helical and compact structures that are less accessible to enzymatic digestion and contribute to gel formation, retrogradation, and resistant starch formation. Conversely, amylopectin, due to its branched architecture, is more susceptible to enzymatic hydrolysis and is mainly responsible for starch viscosity, swelling, gelatinization, and rapid digestibility (3). Based on digestion kinetics, starch is classified into three major categories: rapidly digestible starch (RDS), which is hydrolysed within 20 minutes during in vitro digestion; slowly digestible starch (SDS), which is digested between 20 and 120 minutes; and resistant starch (RS), which escapes digestion in the small intestine and reaches the colon, where it undergoes microbial fermentation (4). Fruits, especially unripe tropical varieties like bananas, plantains, breadfruit, jackfruit, papaya, and exotic fruits such as durian and cempedak, are a significant source of resistant starch. This resistant starch, acting as a natural dietary fiber, offers numerous health benefits. The rise in unhealthy lifestyles has contributed to an increase in diseases like diabetes and obesity across different age groups. Fruits high in resistant starch improve glycemic control, insulin sensitivity, lipid profiles, and gut barrier integrity, thus aiding in the management of type II diabetes, colon cancer, and cardiovascular diseases (5). As a prebiotic, resistant starch fosters the growth of beneficial microbes and the production of short-chain fatty acids (6). Unripe bananas and plantains have the highest resistant starch content, with raw unripe banana flour containing 68–81% resistant starch by dry weight, depending on the cultivar and ripeness (7). Breadfruit and jackfruit seeds are also notable sources, with modified unripe breadfruit containing about 29% resistant starch (8) and jackfruit seed starch containing over 60% resistant starch, which increases with heat or acid treatments (9, 10). To understand this resistant starch more clearly, we should have a clear knowledge of the components and types of resistant starch.

2 TYPES OF RESISTANT STARCH:

There are five categories of Resistant Starch as shown in Figure 1. Resistant Starch 1 is characterized by a cell wall barrier that renders starch physically inaccessible and unable to be hydrolyzed, and is typically found in partially milled grains and cereals. RS 2 is present in plantains, high-amylose corn, ginkgo biloba, and raw potatoes, known as crude starch, which remains undigested owing to its crystalline structure (11). Type RS3 refers to retrograded starch, which is difficult for amylase to break down because of the crystallization that occurs during the cooling and storage phases following gelatinization. This type of starch is found in foods like potatoes, rice, oatmeal, and stale bread that have been cooked and then allowed to cool (12). RS4 is a starch that undergoes chemical modifications, such as substitution, conversion, and cross-linking, to become resistant to enzymatic breakdown. In this altered form, it serves as a functional food additive (13). The final type, RS5, is created by combining long, straight starch chains with free fatty acids, resulting in a helical structure that is difficult to digest. Additionally, it includes resistant maltodextrin, a novel, non-viscous dietary fiber produced by deliberately rearranging the starch molecules (14-16).

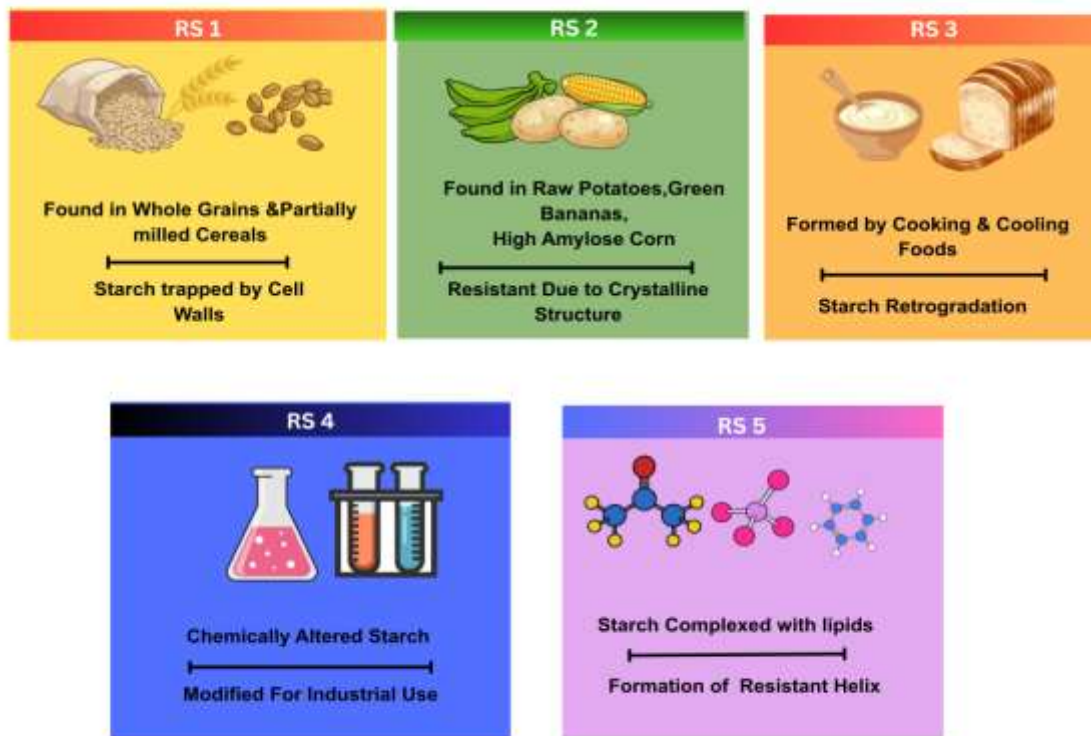


Figure 1 Types of Resistant Starch

3 OCCURRENCE OF RS IN FRUITS:

Unripe green bananas contain a higher concentration of resistant starch compared to dessert banana varieties as shown in Figure 2. Cooking bananas such as Saba and Monthan exhibited a higher resistant starch (RS) content of 65.56% compared to plantain and dessert banana varieties. The resistant starch levels in various green banana starches ranged from 49.8% to 65.56% (17). Numerous studies have been conducted on this subject across various countries, including Nigeria, Indonesia. The chemical composition of starches from various banana types reveals notable differences in moisture, ash, lipid, protein, total starch, fiber, and amylose content. Amylose content, which is pivotal in determining resistant starch (RS) levels, varies significantly among these types, ranging from approximately 13.36% to 42.07%. Generally, higher amylose content is associated with increased RS levels because amylose tends to form tightly packed, crystalline structures that resist enzymatic breakdown. For example, (Hom Tong var.) (AAA) obtained from local market Talad Thai of Thailand, which has an amylose content of 13.36% (18). Similarly, Banana (Kapas var.) (AAB) and Banana (Kepok var.) (ABB) from Jati Gede, Indonesia have high amylose contents of 38.63% and 40.88%, respectively, indicating higher RS levels (19). Total starch content, which ranges from about 63.90% to over 99%, also influences RS formation, as a higher starch concentration provides more material for RS development. Varieties with moderate to high amylose and total

starch content, such as Banana (Mysore var.)(AAB) from Sousa, state of Paraiba, Brazil with 37.88% amylose (20) and Banana (Terra var.) green bananas (*Musa paradisiaca*) of Monthan from local markets of Salem, Tamil Nadu with 35.00% amylose and 94.80% total starch, are expected to exhibit elevated RS levels (21). Conversely, varieties with lower amylose and starch contents, like Banana (Hom Tong var.) (18) and Banana (Valery var.) with 19.32% amylose (22), may have reduced RS amounts. Although minor components like lipids and proteins can affect RS formation through interactions with starch molecules, amylose remains the primary determinant. Therefore, the observed differences in amylose and total starch contents among banana starches suggest corresponding variations in RS content, with varieties higher in amylose generally leading to greater resistant starch levels. (23). African breadfruit(*Treculia Africana* L.)starch contains 69% total starch and 30% amylose. Although the resistant starch content of African breadfruit kernel starch has not been directly quantified in modification studies, it undergoes modifications such as acid-thinning, which reduce pasting and gelatinization. These traits may enhance resistance to digestion, similar to resistant starch, in tropical fruit sources (24). Guava (*Psidium guajava* L.) polysaccharides reduced obesity markers in high-fat diet mice, (where they fed mice guava extract to mice for 11 weeks) via microbiota modulation and butyric acid production, effects attributable to their resistant starch fraction (25). Jack Fruit (*Artocarpus heterophyllus* L.) contains 60-80% of seeds, after consuming the fruits, the seeds which is the major source of carbohydrate that is thrown away and in some places it is cooked and eaten. The seeds are composed of roughly 16–25% starch and possess an amylose content that varies between 22.10% and 38.34% indicating their potential as a source of resistant and slowly digestible starch (26). Jackfruit seed flour (JFS) from "Thong Prasert" cultivar exhibits significantly higher native resistant starch type 2 (RS2) content than mung bean, cassava, and rice starches, though lower than raw banana starch and commercial Hi-maize 260, positioning it as a promising unconventional tropical fruit seed source for RS applications (27). Breadfruit (*Artocarpus atilis*) flour has been shown to contain over 70% starch, which has been modified to give about 50% resistant starch type 3 (28, 29).

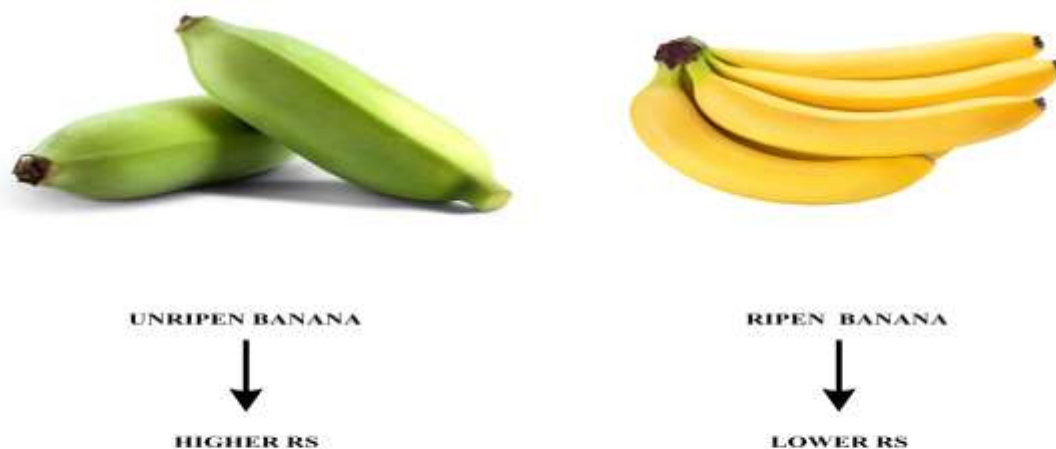


Figure 2 RS Concentration Ripe Vs Unripe.

4 PHYSIOCHEMICAL AND FUNCTIONAL PROPERTIES OF BANANA RESISTANT STARCH:

4.1 Pasting, Gelatinization and Retrogradation:

Banana starch is characterized by a high peak viscosity, minimal breakdown, and moderate setback, making it an effective thickening agent that remains stable under heat and shear conditions, thus serving as a substitute for cross-linked starches (30, 31). Gelatinization typically occurs between 60–71 °C for various banana types, and this range can be extended through modifications such as annealing and heat-moisture treatment (31). Annealing and certain chemical modifications can reduce retrogradation and enhance paste stability, while treatments like HMT and specific enzyme applications can increase crystallinity and raise RS levels upon cooling (32-34).

4.2 Water/Oil absorption, Gel Strength, Rheology:

Native banana starch has a moderate capacity to absorb water (~1.3–1.5 g/g) and slightly less oil, with moderate swelling and solubility (30, 35). Techniques such as pre-gelatinization or acetylation can enhance water and oil

absorption and emulsion capacity, although they may reduce gel firmness or peak viscosity depending on the intensity of the treatment (36, 37). Changes observed in fruits during different treatment/modification compared native starch is mentioned in

Table 1

Table 1 Changes observed in fruits during different treatments/modification compared to native starch

Modification/Treatment	Property Affected	Changes observed(Compared to native starch)	References
Native banana starch	Pasting, gelatinization, water absorption	High peak viscosity, minimal breakdown, moderate setback; moderate water(1.3-1.5g/g) and oil absorption	(30, 31, 38)
Annealing	Gelatinization, retrogradation	Extended gelatinization temperature range; reduced retrogradation; improved paste stability	(31, 32)
Heat-moisture treatment(HMT)	Gelatinization, crystallinity, RS content	Modified gelatinization behavior; increased crystallinity and resistant starch formation upon cooling	(31, 33)
Chemical modification	Retrogradation	Lower retrogradation tendency; enhanced paste stability	(34)
Enzyme treatment	Crystallinity, RS content	Increased crystallinity and higher resistant starch levels after cooling	(33)
Pre-gelatinization	Water/oil absorption gel strength	Increased water and oil absorption; possible reduction in gel firmness or peak viscosity depending on treatment intensity	(37)
Acetylation	Water/oil absorption emulsifying ability	Enhanced water and oil absorption and emulsion capacity; may reduce gel strength at higher substitution	(39)

5 STRUCTURAL CHARACTERISTICS OF BANANA AND OTHER FRUIT RESISTANT STARCH:

5.1 Structural Characteristics:Crystalline polymorph:

“Crystalline arrangement refers to the highly ordered molecular organization of starch polymers, primarily formed by the double-helical alignment of amylopectin chains, which imparts structural rigidity and enzymatic resistance to starch granules.”The crystalline arrangement is related to resisted starch because of its enzyme resistance, the digestive enzymes cannot penetrate the ordered crystals. Starch is characterized by three primary crystalline polymorphs A, B, and C each resulting from distinct arrangements of amylopectin double helices. These polymorphs exhibit unique structural features, water content, and digestibility, which are influenced by the lengths of amylopectin chains and the distribution of branch points (40). A-type polymorphs are distinguished by a dense orthogonal packing of 6-fold helices with minimal water content (8 molecules per unit cell), short amylopectin chains (degree of polymerization, DP < 20), and branch points distributed across both crystalline and amorphous regions (40-42). These features create "weak points" that facilitate rapid enzymatic digestion. In contrast, B-type polymorphs, commonly found in high-amylose banana starches, form open hexagonal arrays of 12-fold helices with water-filled channels (36 molecules per unit cell), longer amylopectin chains (DP(Degree of Polymerization) > 22), and branches concentrated solely in amorphous regions, thereby enhancing resistance to enzymatic digestion and promoting the formation of resistant starch (41). C-type polymorphs represent hybrids that combine features of both A- and B-type structures, as observed in certain fruits, and exhibit mixed diffraction patterns along with intermediate properties (40, 43). Saba (cooking banana) exhibits the highest crystallinity at 52.02%, followed by Popoulu (cooking banana) at 48.10%. Monthan (cooking banana) has a crystallinity of 35.11%, while Nendran (plantain) shows 25.03%. Grand Naine (dessert) has the lowest crystallinity level at 21.19% (44). Monthan, Saba, and Popoulu varieties demonstrated a B-type polymorph (45). This B-type polymorph is characterized by double D-glucose molecules, resulting in an open structure that connects macromolecules and water molecules within the double helices of amylopectin (19). It has been noted in previous studies that cooking bananas typically exhibit a B-type pattern (45). Grand Naine and Nendran varieties showed a C-type polymorph, which is a blend of A and B types. Notably, Nendran, a plantain banana, also displayed a C-type pattern (45). The kernels of jackfruit, litchi, longan, loquat, and mango fruits were found to contain over 50% starch. The starches derived from these five kernels exhibited significantly different morphologies and sizes, yet all contained approximately 25% amylose (46). The starches from jackfruit, longan, and mango demonstrated A-type crystallinity, whereas those from loquat and litchi exhibited C-type crystallinity. Notable differences were also observed in terms of relative crystallinity, short-range ordered structure, and crystalline lamellar intensity among the five kernel starches (46). Jackfruit starch (JS) is characterized by an A-type crystalline structure. This determination is derived from X-ray diffraction patterns, which reveal peaks at 15 degree, 17 degree, 18 degree,

and 23 degree. These peaks correspond to d-spacings of 5.8, 5.2, 4.8, and 3.8 Å, respectively (26). It is noteworthy that while cereal starches typically display an A-type crystalline pattern and fruit starches, such as those from bananas, frequently exhibit a C-type pattern, jackfruit starch, despite being classified as a fruit starch, demonstrates an A-type crystal structure (26, 47). The crystallinity of Chinese jackfruit starch has been documented to range from 24.21% to 36.75%, while that of Mexican jackfruit starch ranges from 26.3% to 28.0% (48, 49). The starch nanoparticles derived from green bananas (GBSNPs) showed a B-type crystalline structure. Prior to digestion, these nanoparticles exhibited prominent X-ray diffraction peaks at 17° and moderate peaks at 5.5°, 15°, 22°, and 24° (2θ), with a crystallinity level of 23.6%.(50). The in vitro digestion process of Cv.Giant Cavendish which lasted for 20, 60, 120, and 180 minutes, the GBSNPs retained their B-type crystalline polymorph. Nevertheless, as the digestion time increased, both the intensity of the B-type signature reflections and the overall degree of crystallinity diminished (50). The crystalline structure of peach palm fruit starch is a C-type polymorph combining A-type and B-type starches (51). The C-type polymorph contains short amylopectin branch chains of A-type and long chains of B-type. The starch's relative crystallinity is considered low for starch with less than 20% apparent amylose (51). According to the X-ray diffraction analysis, the starches derived from avocado seed (ASS), chempedak seed (CPSS), and Pouteria campechiana seed (PCSS) all showed an A-type crystal structure. Conversely, the starch from sweetsop pulp (STS) displayed a B-type crystal structure (52). All these are mentioned in

Table 2

Table 2 Crystalline Polymorphs of Banana and Other Fruits

Fruit/Variety	Fruit Type	Crystalline/Polymorphic Type	Structural Remarks	References
Saba	Cooking Banana	B-type	Highest crystallinity (52.02%) typical of cooking bananas	(44, 45)
Popoulu	Cooking Banana	B-type	High crystallinity (48.10%)	(44, 45)
Monthan	Cooking Banana	B-type	Crystallinity 35.11%	(44, 45)
Nendran	Plantain banana	C-type	Blend of A and B type polymorphs: Crystallinity 25.03%	(44, 45)
Grand Naine	Dessert banana	C-type	Lowest crystallinity (21.19%)	(44, 45)
Green Banana Starch nanoparticles (GBSNPs)	Giant Cavendish banana	B-type	Retained B-type during in-vitro digestion; crystallinity decreased with digestion time	(53)
Jackfruit (kernel starch)	Fruit kernel	A-type	Peaks at 15°, 17°, 18°, 23°; crystallinity 24.21-36.75)% (China), 26.3-28.0% (Mexico)	(46, 49)
Litchi (kernel Starch)	Fruit Kernel	C-type	Approx 25% of Amylose	(46)
Longan(kernel starch)	Fruit Kernel	A-type	Approx 25% of Amylose	(46)
Loquat (kernel starch)	Fruit Kernel	C-type	Approx 25% of Amylose	(46)
Mango (kernel starch)	Fruit kernel	A-type	Approx 25% of Amylose	(46)
Peach Palm Fruit Starch	Fruit	C-type	Combination of A-type and B-type; low relative crystallinity	(51)
Avocado seed starch (ASS)	Seed Starch	A-type	Identified by X-ray diffraction	(52)
Chempedak seed starch (CPSS)	Seed starch	A-type	Identified by X-ray diffraction	(52)
Pouteria campechiana seed starch (PCSS)	Seed Starch	A-type	Identified by X-ray diffraction	(52)
Sweetsop pulp starch(STS)	Fruit Pulp	B-type	Identified by X-ray	(52)

6 MOLECULAR CHARACTERISTICS:

6.1 Genomic control of starch structure;RS in banana:

Starch is composed of linear amylose and branched amylopectin, with the branching pattern and chain length playing a crucial role in determining digestibility and the formation of RS (54, 55). Amylopectin is primarily synthesized by soluble starch synthase, debranching enzyme, and starch branching enzyme (SBE) (56). SBE dictates the number and location of α -1,6 branches, which significantly affect the yield and nutritional value of amylopectin (57-59). Generally, a lower number of branches and longer chains (high "apparent amylose") promote RS, whereas dense short-chain branching leads to starch that is rapidly digestible.

6.2 Banana SBE gene family Domain Structure:

Bananas and other starchy fruits have a type of starch called resistant starch (RS). This is controlled by many genes that help make and manage starch. One important part of RS is amylose, which is made by a gene called granule-bound starch synthase I (GBSSI). In bananas, a gene named MaGBSSI-3 is important for making amylose in the fruit (60). Another group of enzymes, called starch branching enzymes (SBE II), helps shape another starch part called amylopectin. For example, MaSBE2.3 is one of these enzymes. If SBE activity is low, it makes longer starch chains that are harder to break down, which helps form RS. A study found three important mediators (60). A detailed study found seven SBE genes in one banana type (*Musa acuminata*) and six in another (*Musa balbisiana*) (61, 62). These genes are on chromosomes 4, 5, and 6 and are in three groups (SBE I, SBE II, SBE III-like). Most SBE proteins have three main parts that help them work, but some proteins (like MaSBE2.2, 2.5, MbSBE2.2) are missing parts and might work differently. The key enzyme for fruit starch and RS is SBE2.3. Studies of banana roots, leaves, and fruits showed that MaSBE2.3 and MbSBE2.3 are most active in fruits (RPKM > 11–33) as they grow (60). SBE2.3 activity rises from 0 to 80 days after flowering, then drops as the fruit ripens, matching starch changes. Reducing MaSBE2.3 lowers starch and amylopectin, while increasing it raises these levels. SBE2.3 is crucial for making branched amylopectin in bananas. Less branching can increase RS type 2 in bananas by reducing SBE2.3 activity (60). Also, a factor called MaMYB3 slows down starch breakdown in ripening bananas, keeping RS longer.

7 HEALTH IMPLICATIONS AND PHYSIOLOGICAL RELEVANCE OF BANANA AND FRUIT DERIVED RESISTANT STARCH:

7.1 Physiological Relevance:

In a crossover study involving 39 adults with insulin resistance, participants alternated between a diet rich in resistant starch (48–66 g/day sourced from Hi-Maize cornstarch) and a diet low in resistant starch (3–4 g/day from Melojel) over two-week periods, following an initial baseline diet. Researchers collected fecal samples to monitor shifts in the gut microbiome and performed blood tests to assess the glucose and insulin responses. This study underscores the significance of resistant starch in nurturing beneficial gut bacteria and enhancing blood sugar management in individuals at risk. The results showed that a high intake of resistant starch (HRS) modified the gut microbiome, increasing the abundance of Firmicutes, such as *Faecalibacterium prausnitzii* and *Roseburia*, which led to higher butyrate production and proteins related to lipid metabolism. Furthermore, it improved post-meal blood glucose and insulin levels, contributing to better glycemic control, but also raised Trimethylamine N-oxide (TMAO) levels, a marker linked to heart disease risk, highlighting the importance of tailoring RS benefits to individual needs (63).

Recent meta-analyses conducted between 2018 and 2023, which synthesized data from numerous randomized trials, demonstrated that resistant starch (RS) consistently reduces blood glucose and insulin responses by decelerating digestion, akin to the effects of dietary fiber (64). A comprehensive review encompassing over 40 studies revealed that a single dose of RS significantly attenuated the postprandial blood glucose surge in individuals with pre diabetes and diabetes, while regular consumption (ranging from 6 to 40 grams per day) resulted in a more modest reduction in both postprandial and fasting blood glucose levels, particularly with RS2 (raw starches such as green bananas), but not with RS3 (65). These effects vary by health status (66-68).

Resistant starch (RS) sourced from bananas and other fruits, especially from green or unripe bananas, positively influences blood sugar levels, lipids, body weight, and gut health. This is primarily achieved through its fermentation in the colon into short-chain fatty acids (SCFAs). All the health benefits are illustrated in Figure 3 and precisely represented in **Error! Reference source not found.**

7.2 Digestive fate and Colonic fermentation:

Banana Flour Resistant Starch (BFRS) derived from unripe banana flour enhances SCFAs through colonic fermentation in a dose-dependent manner, with varying proportions of acetate (average 32.3 mmol L⁻¹, 50–70%), propionate (12.1 mmol L⁻¹), and butyrate (10.4 mmol L⁻¹) (69-71). Acetate facilitates systemic energy and glucose absorption (71-74); propionate helps reduce cholesterol, insulin, and triglyceride levels; while butyrate promotes colon health and aids in preventing colorectal cancer by influencing metabolism, cell proliferation, and apoptosis, outperforming other SCFAs. These variations are influenced by the type of RS (RS2 leads to acetate; RS3 leads to propionate) and individual factors (6, 75-78) prebiotic, BFRS selectively alters the microbiota to benefit the host, though the mechanisms require further exploration for specific applications, such as using butyrate for cancer prevention. Green banana starch and flour hold promise in this regard (70, 79-81). In studies involving human intubation, freeze-dried green banana flour demonstrated its role as a significant source of resistant starch. About 84% of the starch consumed reached the terminal ileum, primarily in the form of intact RS 2-type granules and soluble oligosaccharides, before being almost entirely fermented in the colon. Importantly, conventional in vitro

RS assays only measured the insoluble granular portion, thus underestimating the actual amount of fermentable starch that enters the large intestine (82).

7.3 Gut Microbiota:

Resistant starch (RS) is essential for preserving the gut barrier's integrity by aiding in the renewal of epithelial cells, (83) boosting mucin production, and fortifying tight junction proteins (84). Through microbial fermentation, RS generates short-chain fatty acids, notably butyrate, which enhance barrier function and decrease gut permeability as shown in Figure 3. Additionally, RS affects gut microbiota and mucosal immune responses, fostering immune tolerance and preventing pathogen invasion. RS also influences the gut–brain axis and the vascular elements of the gut barrier, underscoring its diverse role in both gastrointestinal and systemic health (85–88). The interaction between the gut and the immune system is a highly active process, with approximately 70% of immune cells located in the gut-associated lymphoid tissue (GALT). Resistant starch (RS) bypasses digestion in the upper gastrointestinal tract and reaches the colon, where it undergoes fermentation by gut microbiota, resulting in the production of short-chain fatty acids (SCFAs) such as acetate, propionate, and butyrate (89).

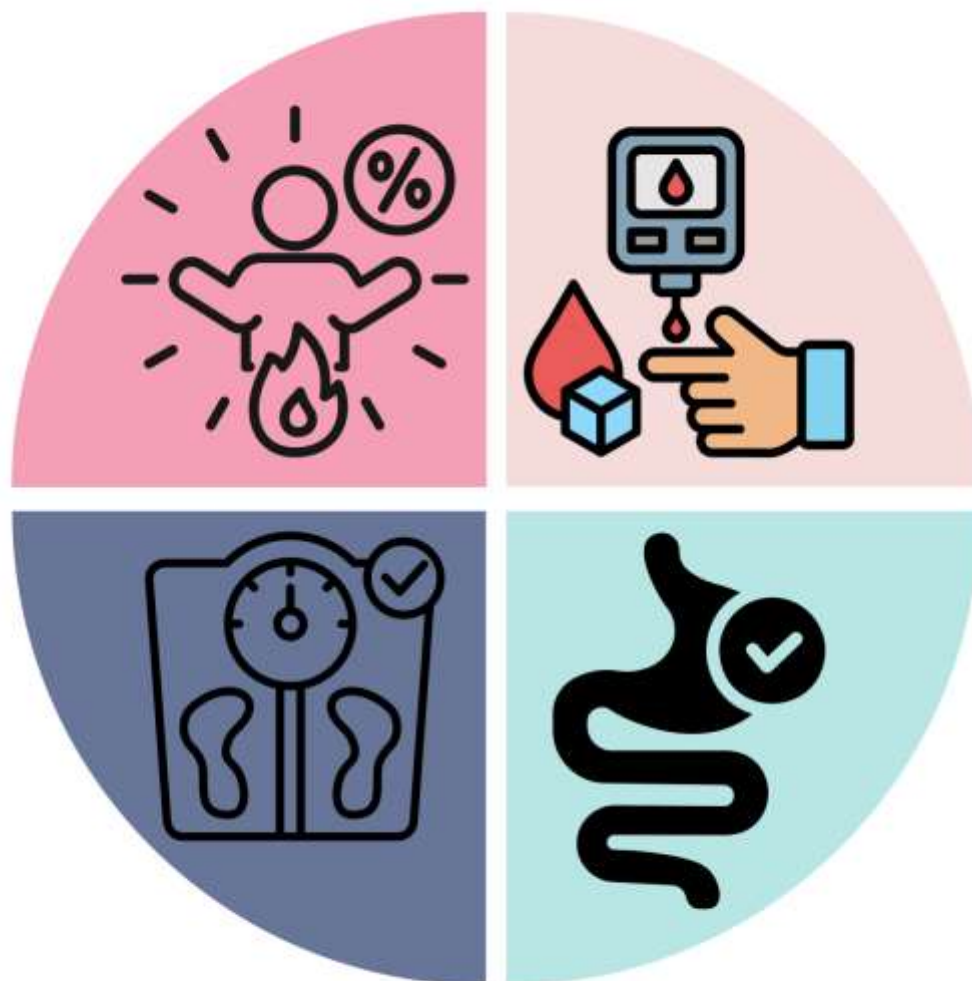


Figure 3 Health Benefits of RS

8 CONCLUSION

Green bananas and similar fruits are abundant in resistant starch (RS), which confers significant health benefits due to its distinctive molecular structure and granule formation. This structure inhibits digestion in the small intestine, resulting in advantageous fermentation in the colon. These characteristics contribute to the regulation of blood sugar levels, improvement of lipid metabolism, and enhancement of gut health, rendering RS a valuable functional ingredient for managing chronic metabolic conditions such as type 2 diabetes. The levels and physiological effects of RS are modulated by variations in amylose content, starch crystallinity, and granule morphology across different fruit varieties. Advances in molecular genetics and food processing techniques have the potential to further augment RS production and efficacy. The incorporation of RS derived from fruits into

dietary plans and functional foods presents promising potential for enhancing metabolic health while promoting sustainability through the utilization of fruit by-products. Continued research into the molecular mechanisms, fermentation characteristics, and clinical effects of fruit-derived RS will facilitate the development of personalized nutritional strategies and innovative health-promoting applications.

9 FUTURE PERSPECTIVES:

Future research should aim to clarify the effects of processing conditions and ingredient interactions on the resulting microbiome. Furthermore, it should assess the impact of the molecular, supra-molecular, and granular properties of unripe banana flour on the synthesis of short-chain fatty acids (90). Research should focus on determining how differences in resistant starch levels and physicochemical characteristics among various cultivars result in varying short-chain fatty acid profiles and Firmicutes/Bacteroidetes ratios in obese individuals, thus facilitating more precise prebiotic applications (90). Moreover, to gain insights into gut health and metabolic disorders, it is essential to employ advanced omics techniques to explore the fundamental mechanisms of resistant starch at the genetic, protein, and metabolic levels (91). Future research should aim to pinpoint and measure the exact bioactive compounds in green bananas that contribute to these health advantages. Additionally, it should investigate their potential use as functional ingredients in nutraceuticals and gluten-free products for managing diabetes (92).

Statements and Declaration:

The authors declare no competing interests.

Author contribution:

1. **H.J.S.** drafted the manuscript and performed the data collection.
2. **M.K, J.A., C.K, and G.G.K.S.** provided supplementary information, corrected the manuscript and supervised the work.
3. All authors have read and agreed to the published version of the manuscript.

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Data sharing is not applicable to this article as no new datasets were created or analyzed during the current study. All data analyzed during this study are included in the cited published articles

REFERENCES:

1. Tester RF, Karkalas J, Qi X. Starch—composition, fine structure and architecture. *Journal of cereal science*. 2004;39(2):151-65.
2. Poláková M, Beláňová M, Petruš L, Mikušová K. Synthesis of alkyl and cycloalkyl α -D-mannopyranosides and derivatives thereof and their evaluation in the mycobacterial mannosyltransferase assay. *Carbohydrate research*. 2010;345(10):1339-47.
3. Tangsuphoom N, Coupland JN. Effect of surface-active stabilizers on the surface properties of coconut milk emulsions. *Food Hydrocolloids*. 2009;23(7):1801-9.
4. Zhang G, Hamaker BR. Slowly digestible starch: concept, mechanism, and proposed extended glycemic index. *Critical reviews in food science and nutrition*. 2009;49(10):852-67.
5. Bojarczuk A, Skąpska S, Khaneghah AM, Marszałek K. Health benefits of resistant starch: A review of the literature. *Journal of functional foods*. 2022;93:105094.
6. Bindels LB, Segura Munoz RR, Gomes-Neto JC, Mutemberezi V, Martínez I, Salazar N, et al. Resistant starch can improve insulin sensitivity independently of the gut microbiota. *Microbiome*. 2017;5(1):12.
7. Moongngarm A, Tiboobun W, Sanpong M, Sriwong P, Phiewtong L, Prakitrum R, et al. RESISTANT STARCH AND BIOACTIVE CONTENTS OF UNRIPE BANANA FLOUR AS INFLUENCED BY HARVESTING PERIODS AND ITS APPLICATION. *American Journal of Agricultural and Biological Sciences*. 2014;9(3).
8. Tran T, Nhan N, Hoang P. Study on the preparation of high-resistant starch content from unripe *Artocarpus altilis* fruits. *Vietnam Journal of Chemistry*. 2025.
9. Hao H, Minh Nhật Đ, Thanh M. Effects of Processing Conditions on Type 4 Resistant Starch Content from Jackfruit Seed Starch. *International Journal of Latest Engineering Research and Applications (IJLERA)*. 2024;9:06-14.
10. Ho HT, Dang NM, Mac TTH, Nguyen HTB. Effect of various heat processing methods on resistant starch level in jackfruit (*Artocarpus heterophyllus* Lam.) seed starch. *International Food Research Journal*. 2024.
11. Birt DF, Boylston T, Hendrich S, Jane J-L, Hollis J, Li L, et al. Resistant starch: promise for improving human health. *Advances in nutrition*. 2013;4(6):587-601.
12. Ma Z, Hu X, Boye JI. Research advances on the formation mechanism of resistant starch type III: A review. *Critical reviews in food science and nutrition*. 2020;60(2):276-97.

13. Goñi I, Garcia-Diz L, Mañas E, Saura-Calixto F. Analysis of resistant starch: a method for foods and food products. *Food chemistry*. 1996;56(4):445-9.
14. Astina J, Sapwarobol S. Resistant maltodextrin and metabolic syndrome: a review. *Journal of the American College of Nutrition*. 2019;38(4):380-5.
15. Han J-A, BeMiller JN. Preparation and physical characteristics of slowly digesting modified food starches. *Carbohydrate polymers*. 2007;67(3):366-74.
16. Hasjim J, Lee SO, Hendrich S, Setiawan S, Ai Y, Jane JL. Characterization of a novel resistant-starch and its effects on postprandial plasma-glucose and insulin responses. *Cereal chemistry*. 2010;87(4):257-62.
17. Kumar S, Arumugam S, Narayanan S, Shiva K, Ravi I, Muthu M, et al. Exploring differences in the physicochemical, functional, structural, and pasting properties of banana starches from dessert, cooking, and plantain cultivars (*Musa* spp.). *International Journal of Biological Macromolecules*. 2021;191.
18. Nimsung P, Thongngam M, Naivikul O, editors. *Compositions, Morphological and Thermal Properties of Green Banana Flour and Starch* 2007.
19. Marta H, Cahyana Y, Djali M, Arcot J, Tensiska T. A comparative study on the physicochemical and pasting properties of starch and flour from different banana (*Musa* spp.) cultivars grown in Indonesia. *International Journal of Food Properties*. 2019;22(1):1562-75.
20. Fontes S, Cavalcanti M, Candeia R, Almeida E. Characterization and study of functional properties of banana starch green variety of Mysore (*Musa* AAB - Mysore). *Food Science and Technology (Campinas)*. 2017;37.
21. Parimalavalli R, Babu D. Comparative Study on Properties of Banana Flour, Starch and Autoclaved Starch. *Trends in Carbohydrate Research*. 2014;6:38-44.
22. Utrilla-Coello RG, Rodríguez-Huezo ME, Carrillo-Navas H, Hernández-Jaimes C, Vernon-Carter EJ, Alvarez-Ramirez J. In vitro digestibility, physicochemical, thermal and rheological properties of banana starches. *Carbohydr Polym*. 2014;101:154-62.
23. Marta H, Cahyana Y, Djali M, Pramafisi G. The Properties, Modification, and Application of Banana Starch. *Polymers*. 2022;14(15):3092.
24. Oderinde AA, Ibikunle AA, Bakre LG, Babarinde NAA. Modification of African breadfruit (*Treculia africana*, Decne) kernel starch: Physicochemical, morphological, pasting, and thermal properties. *Int J Biol Macromol*. 2020;153:79-87.
25. Li Y, Bai D, Lu Y, Chen J, Yang H, Mu Y, et al. The crude guava polysaccharides ameliorate high-fat diet-induced obesity in mice via reshaping gut microbiota. *Int J Biol Macromol*. 2022;213:234-46.
26. Zhang Y, Li B, Xu F, He S, Zhang Y, Sun L, et al. Jackfruit starch: Composition, structure, functional properties, modifications and applications. *Trends in Food Science & Technology*. 2020;107.
27. Kittipongpatana OS, Kittipongpatana N. Resistant Starch Contents of Native and Heat-Moisture Treated Jackfruit Seed Starch. *The Scientific World Journal*. 2015;2015(1):519854.
28. Nuriah S, Zakaria Z, Hussin N, Ahmad F. The Effect of Processing Conditions on Production of Resistant Starch Type III (RS3) from Breadfruit Starch. *Journal Of Agrobiotechnology*. 2020;11:48-58.
29. Turi C, Liu Y, Ragone D, Murch S. Breadfruit (*Artocarpus* spp.): A Traditional Crop with the Potential to Prevent Hunger and Mitigate Diabetes in the Tropics. *Trends in Food Science & Technology*. 2015;45.
30. Yang M, Chang L, Jiang F, Zhao N, Zheng P, Simbo J, et al. Structural, physicochemical and rheological properties of starches isolated from banana varieties (*Musa* spp.). *Food Chemistry: X*. 2022;16.
31. De Barros Mesquita C, Leonel M, Franco C, Leonel S, Garcia É, Santos TD. Characterization of banana starches obtained from cultivars grown in Brazil. *International journal of biological macromolecules*. 2016;89:632-9.
32. Cahyana Y, Wijaya E, Halimah TS, Marta H, Suryadi E, Kurniati D. The effect of different thermal modifications on slowly digestible starch and physicochemical properties of green banana flour (*Musa acuminata* colla). *Food chemistry*. 2019;274:274-80.
33. Paramasivam S, Subramaniyan P, Thayumanavan S, Shiva K, Narayanan S, Raman P, et al. Influence of chemical modifications on dynamic rheological behaviour, thermal techno-functionalities, morpho-structural characteristics and prebiotic activity of banana starches. *International journal of biological macromolecules*. 2023:126125.
34. Carlos-Amaya F, Osorio-Díaz P, Agama-Acevedo E, Yee-Madeira H, Bello-Pérez L. Physicochemical and digestibility properties of double-modified banana (*Musa paradisiaca* L.) starches. *Journal of agricultural and food chemistry*. 2011;59 4:1376-82.
35. Thanyapanich N, Jimtaisong A, Rawdkuen S. Functional Properties of Banana Starch (*Musa* spp.) and Its Utilization in Cosmetics. *Molecules*. 2021;26.
36. Olatunde G, Arogundade L, Orija O. Chemical, functional and pasting properties of banana and plantain starches modified by pre-gelatinization, oxidation and acetylation. *Cogent Food & Agriculture*. 2017;3.
37. Nwakego A-OH, Opeyemi JO, Olufemi AO, David OT. Physicochemical, Functional, Pasting Properties and Fourier Transform Infrared Spectroscopy of Native and Modified Cardaba banana (*Musa* ABB) Starches. *Food Chemistry Advances*. 2022.
38. Thanyapanich N, Jimtaisong A, Rawdkuen S. Functional properties of banana starch (*Musa* spp.) and its utilization in cosmetics. *Molecules*. 2021;26(12):3637.

39. Olatunde GO, Arogundade LK, Orija OI. Chemical, functional and pasting properties of banana and plantain starches modified by pre-gelatinization, oxidation and acetylation. *Cogent Food & Agriculture*. 2017;3(1):1283079.
40. Zhang L, Zhao Y, Hu W, Qian J-Y, Ding X-L, Guan C-R, et al. Multi-scale structures of cassava and potato starch fractions varying in granule size. *Carbohydrate Polymers*. 2018;200:400-7.
41. Jane J-I, Wong K-s, McPherson AE. Branch-structure difference in starches of A-and B-type X-ray patterns revealed by their Naegeli dextrans. *Carbohydrate Research*. 1997;300(3):219-27.
42. Bertoft E. Understanding starch structure: Recent progress. *Agronomy*. 2017;7(3):56.
43. Chi C, Li X, Huang S, Chen L, Zhang Y, Li L, et al. Basic principles in starch multi-scale structuration to mitigate digestibility: A review. *Trends in Food Science & Technology*. 2021;109:154-68.
44. Nathalie F, Buléon A, Colona P, Molis C, Lartigue S, Galmiche J, et al. Digestion of raw banana starch in the small intestine of healthy humans: Structural features of resistant starch. *The British journal of nutrition*. 1995;73:111-23.
45. Paramasivam SK, Arumugam S, Narayanan S, Shiva KN, Iyyakutty R, Muthu M, et al. Exploring differences in the physicochemical, functional, structural, and pasting properties of banana starches from dessert, cooking, and plantain cultivars (*Musa spp.*). *International journal of biological macromolecules*. 2021.
46. Guo K, Lin L, Fan X, Zhang L, Wei C. Comparison of structural and functional properties of starches from five fruit kernels. *Food Chemistry*. 2018;257.
47. Chávez-Salazar A, Bello-Pérez LA, Agama-Acevedo E, Castellanos-Galeano FJ, Álvarez-Barreto CI, Pacheco-Vargas G. Isolation and partial characterization of starch from banana cultivars grown in Colombia. *Int J Biol Macromol*. 2017;98:240-6.
48. Zhang Y, Zhang Y, Xu F, Li S, Tan L. Structural characterization of starches from Chinese jackfruit seeds (*Artocarpus heterophyllus Lam*). *Food Hydrocolloids*. 2018;80.
49. Madruga M, Albuquerque F, Silva I, Amaral D, Magnani M, Neto V. Chemical, morphological and functional properties of Brazilian jackfruit (*Artocarpus heterophyllus L.*) seeds starch. *Food chemistry*. 2014;143:440-5.
50. Jiang H, Zhang Y, Hong Y, Bi Y, Gu Z-b, Cheng L, et al. Digestibility and changes to structural characteristics of green banana starch during in vitro digestion. *Food Hydrocolloids*. 2015;49:192-9.
51. Ferrari Felisberto MH, Souza Costa M, Villas Boas F, Lopes Leivas C, Maria Landi Franco C, Michielon de Souza S, et al. Characterization and technological properties of peach palm (*Bactris gasipaes var. gasipaes*) fruit starch. *Food Res Int*. 2020;136:109569.
52. Li B, Zhang Y, Zhao Y, Luo W, Huang C, Khan M. Relationship between in vitro digestibility and multi-structures of four unconventional starches from Chinese tropical fruits (sweetsop, avocado, chempedak, and *Pouteria campechiana*) extracted using an ultrasound method. *Industrial Crops and Products*. 2023;192:116011.
53. Jiang H, Zhang Y, Hong Y, Bi Y, Gu Z, Cheng L, et al. Digestibility and changes to structural characteristics of green banana starch during in vitro digestion. *Food Hydrocolloids*. 2015;49:192-9.
54. Jobling SA, Schwall GP, Westcott RJ, Sidebottom CM, Debet M, Gidley MJ, et al. A minor form of starch branching enzyme in potato (*Solanum tuberosum L.*) tubers has a major effect on starch structure: cloning and characterisation of multiple forms of SBE A. *The Plant Journal*. 1999;18(2):163-71.
55. Wang J, Hu P, Lin L, Chen Z, Liu Q, Wei C. Gradually decreasing starch branching enzyme expression is responsible for the formation of heterogeneous starch granules. *Plant Physiology*. 2018;176(1):582-95.
56. AM M. Recent progress toward understanding biosynthesis of the amylopectin crystal. *Plant Physiol*. 2000;122:9898-997.
57. Pan T, Lin L, Wang J, Liu Q, Wei C. Long branch-chains of amylopectin with B-type crystallinity in rice seed with inhibition of starch branching enzyme I and IIb resist in situ degradation and inhibit plant growth during seedling development: Degradation of rice starch with inhibition of SBEI/IIb during seedling development. *BMC Plant Biology*. 2018;18(1):9.
58. Li C, Gilbert RG. Progress in controlling starch structure by modifying starch-branching enzymes. *Planta*. 2016;243(1):13-22.
59. Kim K-N, Guiltinan MJ. Identification of cis-acting elements important for expression of the starch-branching enzyme I gene in maize endosperm. *Plant physiology*. 1999;121(1):225-36.
60. Miao H, Sun P, Liu Q, Liu J, Jia C, Zhao D, et al. Molecular identification of the key starch branching enzyme-encoding gene SBE2. 3 and its interacting transcription factors in banana fruits. *Horticulture research*. 2020;7.
61. D'hont A, Denoeud F, Aury J-M, Baurens F-C, Carreel F, Garsmeur O, et al. The banana (*Musa acuminata*) genome and the evolution of monocotyledonous plants. *Nature*. 2012;488(7410):213-7.
62. Wang Z, Miao H, Liu J, Xu B, Yao X, Xu C, et al. *Musa balbisiana* genome reveals subgenome evolution and functional divergence. *Nature plants*. 2019;5(8):810-21.
63. Maier TV, Lucio M, Lee LH, VerBerkmoes NC, Brislawn CJ, Bernhardt J, et al. Impact of dietary resistant starch on the human gut microbiome, metaproteome, and metabolome. *MBio*. 2017;8(5):10.1128/mbio. 01343-17.
64. Baptista NT, Dessalles R, Illner AK, Ville P, Ribet L, Anton PM, et al. Harnessing the power of resistant starch: a narrative review of its health impact and processing challenges. *Front Nutr*. 2024;11:1369950.

65. Pugh JE, Cai M, Altieri N, Frost G. A comparison of the effects of resistant starch types on glycemic response in individuals with type 2 diabetes or prediabetes: A systematic review and meta-analysis. *Front Nutr.* 2023;10:1118229.
66. Vahdat M, Hosseini SA, Khalatbari Mohseni G, Heshmati J, Rahimlou M. Effects of resistant starch interventions on circulating inflammatory biomarkers: a systematic review and meta-analysis of randomized controlled trials. *Nutr J.* 2020;19(1):33.
67. Halajzadeh J, Milajerdi A, Reiner Ž, Amirani E, Kolahdooz F, Barekat M, et al. Effects of resistant starch on glycemic control, serum lipoproteins and systemic inflammation in patients with metabolic syndrome and related disorders: A systematic review and meta-analysis of randomized controlled clinical trials. *Crit Rev Food Sci Nutr.* 2020;60(18):3172-84.
68. Lu J, Ma B, Qiu X, Sun Z, Xiong K. Effects of resistant starch supplementation on oxidative stress and inflammation biomarkers: A systematic review and meta-analysis of randomized controlled trials. *Asia Pac J Clin Nutr.* 2021;30(4):614-23.
69. Muir JG, Lu ZX, Young GP, Cameron-Smith D, Collier GR, O'Dea K. Resistant starch in the diet increases breath hydrogen and serum acetate in human subjects. *The American journal of clinical nutrition.* 1995;61(4):792-9.
70. Silvester KR, Englyst HN, Cummings JH. Ileal recovery of starch from whole diets containing resistant starch measured in vitro and fermentation of ileal effluent. *The American journal of clinical nutrition.* 1995;62(2):403-11.
71. Phillips J, Muir JG, Birkett A, Lu ZX, Jones GP, O'Dea K, et al. Effect of resistant starch on fecal bulk and fermentation-dependent events in humans. *The American journal of clinical nutrition.* 1995;62(1):121-30.
72. Cummings JH, Beatty ER, Kingman SM, Bingham SA, Englyst HN. Digestion and physiological properties of resistant starch in the human large bowel. *British Journal of Nutrition.* 1996;75(5):733-47.
73. Langkilde AM, Champ M, Andersson H. Effects of high-resistant-starch banana flour (RS2) on in vitro fermentation and the small-bowel excretion of energy, nutrients, and sterols: an ileostomy study¹²³. *The American journal of clinical nutrition.* 2002;75(1):104-11.
74. Caballero B, Trugo L, Finglas P. *Encyclopedia of food sciences and nutrition: Volumes 1-10*2003.
75. Shinde T, Perera AP, Vemuri R, Gondalia SV, Beale DJ, Karpe AV, et al. Synbiotic supplementation with prebiotic green banana resistant starch and probiotic *Bacillus coagulans* spores ameliorates gut inflammation in mouse model of inflammatory bowel diseases. *European journal of nutrition.* 2020;59(8):3669-89.
76. Yuan H, Meng Y, Bai H, Shen D, Wan B, Chen L. Meta-analysis indicates that resistant starch lowers serum total cholesterol and low-density cholesterol. *Nutrition research.* 2018;54:1-11.
77. Robertson M, Currie J, Morgan L, Jewell D, Frayn K. Prior short-term consumption of resistant starch enhances postprandial insulin sensitivity in healthy subjects. *Diabetologia.* 2003;46(5):659-65.
78. Ribeiro Vieira C, Laurides Ribeiro de Oliveira Lomeu F, de Castro Moreira ME, Stampini Duarte Martino H, Ribeiro Silva R. Clinical application of a cocoa and unripe banana flour beverage for overweight women with abdominal obesity: Prospective, double-blinded and randomized clinical trial. *Journal of Food Biochemistry.* 2017;41(3):e12372.
79. Almeida-Junior L, Curimbaba T, Chagas A, Quaglio A, Di Stasi L. Dietary intervention with green dwarf banana flour (*Musa sp.* AAA) modulates oxidative stress and colonic SCFAs production in the TNBS model of intestinal inflammation. *Journal of Functional Foods.* 2017;38:497-504.
80. Roediger W. Utilization of nutrients by isolated epithelial cells of the rat colon. *Gastroenterology.* 1982;83(2):424-9.
81. Zeng H, Hamlin SK, Safratowich BD, Cheng W-H, Johnson LK. Superior inhibitory efficacy of butyrate over propionate and acetate against human colon cancer cell proliferation via cell cycle arrest and apoptosis: Linking dietary fiber to cancer prevention. *Nutrition Research.* 2020;83:63-72.
82. Faisant N, Buléon A, Colonna P, Molis C, Lartigue S, Galmiche J, et al. Digestion of raw banana starch in the small intestine of healthy humans: structural features of resistant starch. *British Journal of Nutrition.* 1995;73:111-23.
83. Turner JR. Intestinal mucosal barrier function in health and disease. *Nature reviews immunology.* 2009;9(11):799-809.
84. Suzuki T. Regulation of intestinal epithelial permeability by tight junctions. *Cellular and molecular life sciences.* 2013;70(4):631-59.
85. González-Bosch C, Boorman E, Zunszain PA, Mann GE. Short-chain fatty acids as modulators of redox signaling in health and disease. *Redox biology.* 2021;47:102165.
86. Nogal A, Valdes AM, Menni C. The role of short-chain fatty acids in the interplay between gut microbiota and diet in cardio-metabolic health. *Gut microbes.* 2021;13(1):1897212.
87. Baert F, Matthys C, Mellaerts R, Lemaître D, Vlaemynck G, Foulon V. Dietary intake of Parkinson's disease patients. *Frontiers in nutrition.* 2020;7:105.
88. Braniste V, Al-Asmakh M, Kowal C, Anuar F, Abbaspour A, Tóth M, et al. The gut microbiota influences blood-brain barrier permeability in mice. *Science translational medicine.* 2014;6(263):263ra158-263ra158.
89. Bindels LB, Walter J, Ramer-Tait AE. Resistant starches for the management of metabolic diseases. *Current Opinion in Clinical Nutrition & Metabolic Care.* 2015;18(6):559-65.

90. Dibakoane SR, Du Plessis B, Da Silva LS, Anyasi TA, Emmambux MN, Mlambo V, et al. Nutraceutical properties of unripe banana flour resistant starch: a review. *Starch-Stärke*. 2023;75(9-10):2200041.
91. Han J, Wu J, Liu X, Shi J, Xu J. Physiological effects of resistant starch and its applications in food: a review. *Food Production, Processing and Nutrition*. 2023;5(1):48.
92. Shini V, Billu A, Suvachan A, Nisha P. Exploring the nutritional, physicochemical and hypoglycemic properties of green banana flours from unexploited banana cultivars of southern India. *Sustainable Food Technology*. 2024;2(4):1113-27.