



CAMILA MARIANE DA SILVA SOARES

**CHARACTERIZATION OF YELLOW PUÇÁ (*Mouriri
Grandiflora*) AND IMPACTS OF IN VITRO SIMULATED
GASTROINTESTINAL DIGESTION ON THE CHEMICAL,
BIOLOGICAL AND ANTIOXIDANT PROPERTIES OF
POLYPHENOLS**

**LAVRAS – MG
2026**

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Thesis presented to the Federal University of
Lavras, as part of the requirements of the
Graduate Program in Food Science, area of
concentration in Food Sciences, to obtain the title
of Doctor.

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Advisor

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*Dedico esta tese à minha família, cujo suporte
emocional, compreensão e paciência tornaram
possível a realização deste sonho.
Nada disso teria sentido sem vocês.*

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*“Sonhar é essencial, mas transformar sonhos exige
compromisso.”
Paulo Freire*

ABSTRACT

The yellow puçá (*Mouriri grandiflora*), a fruit native to the Amazon and Cerrado biomes, remains underexplored from a chemical, functional and technological perspective, despite its traditional use by local communities. In view of the growing demand for foods rich in bioactive compounds and the scientific interest in the bioaccessibility of polyphenols, this thesis aimed to perform the systemic characterization of the pulp, peels and seeds of the fruit, as well as to evaluate the impacts of simulated gastrointestinal digestion in vitro on its chemical, biological and antioxidant properties. The nutritional characterization showed distinct compositional profiles between the fractions, with variations in the distribution of macronutrients, organic acids, fatty acids and triglycerides. The phytochemical analyses showed that, although the pulp is the traditional edible fraction, peels and seeds showed higher levels of total phenolic compounds and high antioxidant activity, standing out as promising matrices for technological use. The identification of individual phenolics by liquid chromatography revealed a diversified profile of metabolites, reinforcing the functional potential of the fruit. The application of the INFOGEST protocol allowed the evaluation of the behavior of polyphenols throughout the digestive phases, indicating significant changes in the phenolic profile, with a reduction of sensitive compounds and an increase in the bioaccessible fraction due to the release of the food matrix. Modulation of antioxidant activity during digestion was also observed, showing that bioaccessibility also depends on structural transformations. In an integrated way, the results demonstrate that the yellow puçá has high functional potential, especially with the full use of its fractions, contributing to the development of natural ingredients and strategies aligned with the circular economy, in addition to expanding the knowledge about the interaction between plant matrix and gastrointestinal digestion.

Keyword: Native fruits; brazilian biodiversity; polyphenols; INFOGEST protocol, amazonian fruit.

RESUMO

O puçá-amarelo (*Mouriri grandiflora*), fruto nativo dos biomas Amazônia e Cerrado, permanece subexplorado sob a perspectiva química, funcional e tecnológica, apesar de seu uso tradicional por comunidades locais. Diante da crescente demanda por alimentos ricos em compostos bioativos e do interesse científico na bioacessibilidade de polifenóis, esta tese teve como objetivo realizar a caracterização sistêmica da polpa, cascas e sementes do fruto, bem como avaliar os impactos da digestão gastrointestinal simulada *in vitro* em suas propriedades químicas, biológicas e antioxidantes. A caracterização nutricional evidenciou perfis composicionais distintos entre as frações, com variações na distribuição de macronutrientes, ácidos orgânicos, ácidos graxos e triglicerídeos. As análises fitoquímicas mostraram que, embora a polpa seja a fração comestível tradicional, cascas e sementes apresentaram maiores teores de compostos fenólicos totais e elevada atividade antioxidante, destacando-se como matrizes promissoras para aproveitamento tecnológico. A identificação de fenólicos individuais por cromatografia líquida revelou um perfil diversificado de metabólitos, reforçando o potencial funcional do fruto. A aplicação do protocolo INFOGEST permitiu avaliar o comportamento dos polifenóis ao longo das fases digestivas, indicando alterações significativas no perfil fenólico, com redução de compostos sensíveis e aumento da fração bioacessível devido à liberação da matriz alimentar. Observou-se ainda modulação da atividade antioxidante durante a digestão, evidenciando que a bioacessibilidade depende também de transformações estruturais. De forma integrada, os resultados demonstram que o puçá-amarelo possui elevado potencial funcional, especialmente com o aproveitamento integral de suas frações, contribuindo para o desenvolvimento de ingredientes naturais e estratégias alinhadas à economia circular, além de ampliar o conhecimento sobre a interação entre matriz vegetal e digestão gastrointestinal.

Palavras-Chave: Frutos nativos; biodiversidade brasileira; polifenóis; protocolo INFOGEST, fruto amazônico.

IMPACTS INDICATORS

The present thesis presents relevant impacts in the scientific, technological, environmental, and socioeconomic dimensions, resulting from the unprecedented investigation on the yellow puçá (*Mouriri grandiflora*) and the application of a standardized methodology of simulated gastrointestinal digestion in vitro to evaluate the bioaccessibility of phenolic compounds. In the scientific field, the study contributes to the advancement of knowledge by providing original data on the nutritional composition, phytochemical profile and antioxidant activity of the different fractions of the fruit (pulp, peel and seeds), as well as on the structural and functional changes of these compounds throughout the digestive phases. The integrated approach adopted expands the understanding of the relationship between food matrix and bioaccessibility, providing consistent evidence that can support future investigations, including in vivo studies, metabolomic analyses and technological applications. From a technological perspective, the results demonstrate the potential for full appreciation of the fruit, especially of the fractions traditionally considered co-products. The identification of husks and seeds as expressive sources of phenolic compounds and antioxidant activity reinforces the possibility of their use as functional ingredients, natural extracts or inputs for innovative food formulations. In this way, the research offers technical subsidies for the development of products with greater added value, in line with the demands for natural and sustainable ingredients. In the environmental field, the thesis contributes to the promotion of strategies based on the circular economy, by highlighting the potential for reuse of plant fractions that are often discarded. The scientific appreciation of a fruit native to the Amazon and Cerrado biomes also reinforces the importance of biodiversity conservation and the sustainable use of Brazilian natural resources, integrating science, sustainability, and innovation. From a socioeconomic point of view, the results can stimulate the strengthening of regional production chains, adding value to native species that are still underexploited. The generation of applied knowledge about yellow puçá expands the possibilities of insertion of this fruit in specialized markets, potentially contributing to the development of the bioeconomy and to the generation of income in agroextractivist contexts. In this way, the impacts of this thesis go beyond the generation of analytical data, configuring itself as a strategic contribution to scientific advancement, technological innovation, environmental sustainability and the valorization of national biodiversity. The work has the potential for university extension by providing information that can be transferred to the productive sector, research institutions and technical assistance agencies, indirectly benefiting producers, consumers and society. Thus, its impacts are mainly concentrated in the thematic areas of Health, Environment and Technology and Production of the National Extension Policy and are aligned with the Sustainable Development Goals (SDGs) of the UN 2030 Agenda, especially SDG 2 (Zero Hunger and Sustainable Agriculture), SDG 3 (Health and Well-Being), SDG 9 (Industry, Innovation and Infrastructure), SDG 12 (Responsible Consumption and Production) and SDG 15 (Life on Land), reinforcing the potential of research to promote innovation, sustainability and appreciation of Brazilian natural resources.

INDICADORES DE IMPACTO

A presente tese apresenta impactos relevantes nas dimensões científica, tecnológica, ambiental, e socioeconômica, decorrentes da investigação inédita sobre o puçá-amarelo (*Mouriri grandiflora*) e da aplicação de metodologia padronizada de digestão gastrointestinal simulada *in vitro* para avaliação da bioacessibilidade de compostos fenólicos. No âmbito científico, o estudo contribui para o avanço do conhecimento ao disponibilizar dados originais sobre a composição nutricional, o perfil fitoquímico e a atividade antioxidante das diferentes frações do fruto (polpa, casca e sementes), bem como sobre as alterações estruturais e funcionais desses compostos ao longo das fases digestivas. A abordagem integrada adotada amplia a compreensão acerca da relação entre matriz alimentar e bioacessibilidade, fornecendo evidências consistentes que podem subsidiar investigações futuras, incluindo estudos *in vivo*, análises metabolômicas e aplicações tecnológicas. Sob a perspectiva tecnológica, os resultados demonstram o potencial de valorização integral do fruto, especialmente das frações tradicionalmente consideradas coprodutos. A identificação de cascas e sementes como fontes expressivas de compostos fenólicos e atividade antioxidante reforça a possibilidade de seu aproveitamento como ingredientes funcionais, extratos naturais ou insumos para formulações alimentícias inovadoras. Dessa forma, a pesquisa oferece subsídios técnicos para o desenvolvimento de produtos com maior valor agregado, alinhados às demandas por ingredientes naturais e sustentáveis. No campo ambiental, a tese contribui para a promoção de estratégias baseadas na economia circular, ao evidenciar o potencial de reaproveitamento de frações vegetais frequentemente descartadas. A valorização científica de um fruto nativo dos biomas Amazônia e Cerrado também reforça a importância da conservação da biodiversidade e do uso sustentável dos recursos naturais brasileiros, integrando ciência, sustentabilidade e inovação. Do ponto de vista socioeconômico, os resultados podem estimular o fortalecimento de cadeias produtivas regionais, agregando valor a espécies nativas ainda subexploradas. A geração de conhecimento aplicado sobre o puçá-amarelo amplia as possibilidades de inserção desse fruto em mercados especializados, contribuindo potencialmente para o desenvolvimento da bioeconomia e para a geração de renda em contextos agroextrativistas. Dessa maneira, os impactos desta tese ultrapassam a geração de dados analíticos, configurando-se como contribuição estratégica para o avanço científico, a inovação tecnológica, a sustentabilidade ambiental e a valorização da biodiversidade nacional. O trabalho possui potencial de extensão universitária ao disponibilizar informações que podem ser transferidas para o setor produtivo, instituições de pesquisa e órgãos de assistência técnica, beneficiando indiretamente produtores, consumidores e a sociedade. Dessa forma, seus impactos concentram-se principalmente nas áreas temáticas de Saúde, Meio Ambiente e Tecnologia e Produção da Política Nacional de Extensão e estão alinhados aos Objetivos de Desenvolvimento Sustentável (ODS) da Agenda 2030 da ONU, especialmente o ODS 2 (Fome Zero e Agricultura Sustentável), ODS 3 (Saúde e Bem-Estar), ODS 9 (Indústria, Inovação e Infraestrutura), ODS 12 (Consumo e Produção Responsáveis) e ODS 15 (Vida Terrestre), reforçando o potencial da pesquisa para promover inovação, sustentabilidade e valorização dos recursos naturais brasileiros.

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FIRST ONE

1 GENERAL INTRODUCTION

The growing interest in functional foods and natural ingredients has driven scientific research of native and underutilized fruits, especially those from regions with a wide variety of endemic species, such as Brazil. Brazilian biodiversity is home to a wide variety of plant species with high nutritional and functional potential, many of which remain little explored from a scientific and technological point of view (Morais *et al.*; 2025; Morais *et al.*, 2022. Soares *et al.*, 2021). Several studies have shown that exotic native fruits are important sources of bioactive compounds, including vitamins, carotenoids, dietary fibers, and phenolic compounds, which have antioxidant properties and may contribute to the prevention of chronic non-communicable diseases (Rufino *et al.*, 2010; Silva *et al.*, 2021; Soares *et al.*, 2022; Silva *et al.*, 2025; Tischer *et al.*, 2023;). In this context, valuing food biodiversity has been considered a promising strategy for the development of new functional foods and nutraceutical ingredients, in addition to contributing to environmental conservation and the strengthening of sustainable production chains.

Among the bioactive compounds present in foods of plant origin, polyphenols have received great attention from the scientific community due to their wide structural diversity and the multiple biological effects associated with their consumption (Morais *et al.* 2025; Honaiser *et al.*, 2025; Quero *et al.*, 2020). These compounds include different chemical classes, such as phenolic acids, flavonoids, tannins, and stilbenes, which have a high capacity to neutralize reactive oxygen species and modulate physiological processes related to oxidative stress and inflammation (Ribeiro *et al.*, 2019; Koch. 2019; Di Lorenzo *et al.*, 2021; Zaini *et al.*, 2022; Zecrumah *et al.*, 2023; Mérida *et al.*, 2023). Epidemiological and experimental studies have shown that diets rich in foods containing phenolic compounds are associated with reduced risk of several chronic diseases, including cardiovascular disease, type 2 diabetes, and some types of cancer (Schulz *et al.*, 2019; Shahidi and Pan, 2020; Giacco *et al.*, 2020; Morais *et al.*, 2022; El-Hadary, Sulieman, El-Shorbagy, 2023; Lippolis *et al.*, 2023; Wisnuwardani, 2025).

Despite the recognized importance of these compounds, the presence of polyphenols in food does not necessarily guarantee their biological effects on the human body. The efficacy of these compounds depends on their bioaccessibility and bioavailability, which are influenced by factors related to both the molecular structure of the compounds and the characteristics of the food matrix and the physiological conditions of the digestive process (Shahidi and Pan, 2020; García-Pérez *et al.*, 2023; Morais *et al.*, 2025).

During the digestive process, several factors can influence the release, stability, and transformation of phenolic compounds. Among these factors, pH variations along the gastrointestinal tract, the action of digestive enzymes, the presence of bile salts, and interactions between phenolic compounds and structural components of the food matrix, such as proteins and polysaccharides (Shahidi *et al.*, 2018; Deng *et al.*, 2018; Di Lorenzo *et al.*, 2021; Odriozola-Serrano *et al.*, 2023).

Given these limitations, *in vitro* simulated gastrointestinal digestion models have been widely used to investigate the behavior of bioactive compounds during the human digestive process. These models allow the reproduction, under controlled conditions, of the main stages of digestion, including the oral, gastric, and intestinal phases, making it possible to evaluate the release, transformation, and stability of bioactive compounds present in food (Fernández-Jalao *et al.*, 2020; Farias *et al.*, 2021). Among the available methods, the INFOGEST digestion protocol stands out, developed by an international consortium of researchers with the objective of standardizing *in vitro digestion methodologies* and improving comparability between scientific studies (Minekus *et al.*, 2014; Brodkorb *et al.*, 2019).

The application of the INFOGEST protocol has allowed significant advances in the understanding of the bioaccessibility of phenolic compounds in different food matrices. Recent studies demonstrate that gastrointestinal digestion can promote both the release and degradation of phenolic compounds, resulting in significant changes in their antioxidant activity and biological potential (Morais, *et al.*, 2025; Soares *et al.*, 2025; Rocchetti *et al.*, 2024; Morales and Iriondo-Dehond, 2025; Souza *et al.*, 2025). In addition, the composition of the food matrix and the presence of fiber and other macronutrients can significantly influence the stability and bioaccessibility of these compounds during the digestive process (Deng *et al.*, 2022; Latif *et al.*, 2025).

In parallel with methodological advances in the evaluation of the bioaccessibility of bioactive compounds, scientific interest in the investigation of native Brazilian fruits has grown, particularly those from biomes such as the Cerrado and the Amazon. These biomes have high biodiversity and are home to numerous fruit species with nutritional and functional potential (Rocha, De Carvalho, Conte-Junior, 2025; Morais *et al.*, 2025, Costa *et al.*, 2025; Carvalho, Barcia, Schmiele, 2025).

Among these native species, the yellow puçá (*Mouriri grandiflora*) stands out, a typical fruit of the Amazon and the Cerrado, widely used by local communities due to its food and medicinal properties (Soares *et al.*, 2025; Lima *et al.*, 2022). Ethnobotanical studies indicate that puçá-amarelo is traditionally consumed *in natura* or in the form of juices and artisanal

preparations, being recognized for its slightly acidic flavor and characteristic aroma (Oliveira *et al.*, 2016; Soares *et al.*, 2025). Morphologically, the fruit has intense yellow pulp, thin skin and high yield of edible part, in addition to having small and easily separable seeds. The plant, belonging to the Melastomataceae family, adapts well to environments with dense vegetation and poor soils, which reinforces its ecological importance in maintaining native flora and feeding local fauna (Ulloa Ulloa *et al.*, 2022; Soares *et al.*, 2025). Its cultural, nutritional and environmental relevance makes the yellow puçá a promising model for chemical and technological investigations in the field of food science.

In addition to the fruit pulp, different fractions of the puçá, including peels and seeds, can have significant concentrations of bioactive compounds. In this context, the full use of fruits and their co-products has received great attention in the context of sustainability and circular economy in the agri-food chain. Residues generated during fruit processing often have a high content of phenolic compounds and dietary fibers, and can be used as alternative sources of functional ingredients for industrial applications (Cano-Lou *et al.*, 2026; Soares *et al.*, 2022; Kirchherr *et al.*, 2017).

The investigation of the bioaccessibility of phenolic compounds present in native fruits and their co-products represents, therefore, an essential approach to understand the real functional potential of these foods. The application of standardized simulated digestion methodologies allows us to assess how these compounds are released, transformed, or degraded throughout the digestive process, providing fundamental information about their stability and potential bioavailability (Lippolis *et al.*, 2023; Dantas *et al.*, 2023; Morais *et al.*, 2025; Fernandes *et al.*, 2025). Thus, the investigation of the chemical composition, antioxidant properties and bioaccessibility of phenolic compounds present in puçá-amarelo through simulated gastrointestinal digestion represents a relevant scientific approach to expand knowledge about the functional potential of this native species. In addition to contributing to the appreciation of Brazilian food biodiversity, studies of this nature can provide fundamental information for the development of new functional foods and nutraceutical ingredients, promoting innovation, sustainability and food safety. Therefore, the present study evaluates the chemical composition, antioxidant properties and bioaccessibility of phenolic compounds present in puçá-amarelo (*Mouriri grandiflora*) and its co-products, using the INFOGEST simulated gastrointestinal digestion protocol, in order to understand the impact of the digestive process on its chemical and biological properties. The work was structured in the form of chapters, the first refers to the literature review "*Bioaccessibility of phenolic compounds in Brazilian native fruits: insights from INFOGEST's in vitro digestion models*", in the process of

submission to the journal Food Reviews International. In the second chapter, the article *"Systemic characterization of the pulp, peels and seeds of the yellow puçá (Mouriri grandiflora) and impacts of in vitro simulated gastrointestinal digestion on the chemical, biological and antioxidant properties of polyphenols"* published in the.

SECOND PART

ARTICLE 1: BIOACCESSIBILITY OF PHENOLIC COMPOUNDS IN BRAZILIAN NATIVE FRUITS: INSIGHTS FROM INFOGEST IN VITRO DIGESTION MODELS

Article submitted in Food Reviews International

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Abstract

Brazilian native fruits are recognized as rich sources of bioactive compounds, particularly polyphenols and carotenoids, associated with antioxidant activity and health benefits. In addition to their nutritional value, these fruits support biodiversity valorization and sustainable development, especially through the use of agro-industrial coproducts. However, most studies emphasize phytochemical composition, with limited focus on the fate of these compounds during digestion. Standardized in vitro models, such as the INFOGEST protocol, enable the evaluation of bioaccessibility and stability. This review highlights recent advances and proposes an integrated perspective linking composition, coproducts, and digestion to better assess the functional potential of native fruits.

Keywords: brazilian native fruits; phenolic compounds; bioaccessibility; in vitro digestion, INFOGEST protocol, circular economy.

Introduction

Brazilian biomes, globally recognized for their fruit diversity, constitute important reservoirs of native fruits with high nutritional, functional and socioeconomic potential^[1] The Amazon, Cerrado, Caatinga, and Atlantic Forest, among other biomes, are home to fruit species that have historically played fundamental roles in feeding traditional communities and that, in

recent decades, have attracted increasing scientific attention due to their bioactive properties and technological applicability ^[2]. These fruits have historically played a fundamental role in the diet and livelihood of traditional communities, while in recent decades they have attracted increasing scientific attention due to their nutritional value, phytochemical richness, and technological potential for the development of functional foods and nutraceutical products ^[3, 4].

Native Brazilian fruits are widely recognized as important sources of bioactive compounds, such as total phenols, flavonoids, anthocyanins, condensed tannins, and carotenoids. These molecules are associated with several biological activities, including antioxidant, anti-inflammatory, and photoprotective effects, which may contribute to the prevention of chronic non-communicable diseases ^[2]. In addition to their health-related properties, these compounds also play a relevant role in determining sensory attributes such as color, flavor, and aroma, reinforcing the technological and commercial value of these plant matrices ^[1, 5].

Despite the growing body of literature describing the chemical composition and functional properties of native fruits, an important challenge in food science is understanding how these compounds behave during digestion and how much of them becomes available for absorption in the human gastrointestinal tract. The biological effects attributed to phytochemicals are strongly influenced by their bioaccessibility and bioavailability, which depend on complex interactions between the food matrix, digestive conditions, and the physicochemical stability of the compounds. In this context, *in vitro* gastrointestinal digestion models have emerged as valuable tools for evaluating the stability, transformation, and potential bioaccessibility of food bioactive compounds ^[6, 7]. Developed by an international consortium of researchers, the INFOGEST protocol simulates the oral, gastric, and intestinal phases of human digestion under controlled laboratory conditions^[8].

Recent studies applying the INFOGEST protocol to yellow puçá and other Brazilian species have demonstrated that gastrointestinal digestion can significantly influence the chemical stability and bioaccessibility of phenolic compounds, carotenoids, and other phytochemicals^[9, 10]. However, although several investigations have examined individual fruit species or isolated bioactive compounds, the current body of knowledge remains fragmented. In particular, there is limited integration between studies addressing phytochemical composition, the valorization of agro-industrial coproducts, and the digestive behavior of these compounds. Therefore, a comprehensive synthesis of the available literature is needed to clarify the relationships among Brazilian native fruits, their phenolic profiles, the potential use of agro-industrial coproducts, and the bioaccessibility of these bioactive compounds when evaluated using standardized in vitro digestion models.

Thus, the present review analyzed the current scientific literature on Brazilian native fruits as sources of bioactive compounds, with emphasis on phenolic compounds, the valorization of fruit coproducts, and the application of the INFOGEST in vitro digestion protocol to assess the stability and bioaccessibility of these compounds. By integrating these perspectives, this review seeks to contribute to the advancement of research on functional foods and the development of innovative food systems based on native plant resources, aligned with current global challenges in nutrition, health, and sustainability.

Methodology and objectives of this review

This review was based on a structured survey of peer-reviewed scientific literature, retrieved from the ScienceDirect, Scopus, Web of Science and Google Scholar databases, complemented by consultations to the CAPES Catalogue of Theses and Dissertations. The search mainly covered publications from the last five years, with an emphasis on the most recent studies to contemplate scientific advances related to Brazilian native fruits and their bioactive compounds. The search strategies used keywords in Portuguese, English and Spanish, including

terms such as native fruits, bioactive compounds, phenolic compounds, flavonoids, carotenoids, antioxidant activity, agro-industrial co-products, in vitro gastrointestinal digestion, simulated digestion and INFOGEST protocol. Original articles, review articles, book chapters, dissertations, and theses that addressed the chemical composition, bioactive potential, co-product valorization, and bioaccessibility of bioactive compounds were included. Publications without peer review, studies with insufficient methodological information, and studies not directly related to the objectives of this review were excluded.

The objectives of this review are to provide a comprehensive and integrated analysis of Brazilian native fruits as promising sources of bioactive compounds, establishing a connection between fundamental scientific knowledge and their potential applications in the areas of food, nutrition, health and sustainability. The review critically examines the physicochemical characteristics, nutritional composition, phytochemical profiles, and bioactive properties of these fruits, with special emphasis on phenolic compounds and their contribution to antioxidant activity. Additionally, this review addresses the application of the INFOGEST protocol as a standardized method for the simulation of gastrointestinal digestion in vitro, highlighting its relevance in the evaluation of the stability, transformation and bioaccessibility of the bioactive compounds present in native fruits and their co-products. The main factors that influence the release and availability of these compounds throughout the digestive process are discussed, as well as the prospects for their use.

The relevance of this work lies in its critical and multidisciplinary approach, which integrates information on chemical composition, bioactivity, in vitro digestion and sustainability. By gathering and analyzing recent scientific evidence, this review contributes to the understanding of the potential of Brazilian native fruits and their co-products as sources of bioactive compounds, in addition to identifying knowledge gaps and directing future research aimed at the development of innovative, sustainable and health-promoting products.

Native fruits Brazilian as a source of phenolic compound

Brazil is home to one of the greatest biodiversities on the planet, with a wide diversity of fruit species distributed in different biomes^[1]. Although these fruits are traditionally valued for their nutritional and socioeconomic relevance, recent evidence has highlighted their role as expressive sources of phenolic compounds, considered key determinants of their functional potential^[2].

Species such as açaí, bacaba, buriti, jabuticaba, and pequi have a high concentration of phenolic acids, flavonoids, and anthocyanins, often at levels comparable to or higher than those of widely traded fruits ^[2, 11]. This compositional variability is associated with both taxonomic diversity (e.g., Arecaceae, Myrtaceae, Caryocaraceae) and environmental factors, which results in highly heterogeneous and still insufficiently standardized phenolic profiles^[12, 13]. In particular, Amazonian palm Arecaceae fruits have been associated with high polyphenol contents and strong antioxidant activity, often demonstrated by assays such as ORAC and DPPH^[14].

Although multiple biological activities have been attributed to phenolic compounds, including antioxidant, anti-inflammatory, and cardioprotective effects, these results are often based on isolated experimental models, with limited extrapolation to actual physiological conditions ^[15–17]. Additionally, possible synergistic effects between phenolic subclasses remain poorly elucidated, especially in complex food matrices.

Table 1 shows the diversity of phenolic compounds present in these species, as well as the predominance of specific classes in certain fruits and fractions (pulp, peel, and seeds). However, methodological differences in extraction and quantification limit comparability between studies and hinder the consolidation of compositional patterns.

Table 1. Major phenolic compounds in Brazilian native fruits and associated biological activities

Fruit (Scientific name)	Major phenolic compounds	Phenolic class	Fruit fraction	Content (range)*	Reported bioactivities	References
Açaí (<i>Euterpe oleracea</i>)	Cyanidin-3-glucoside, cyanidin-3-rutinoside	Anthocyanins	Pulp	200–600 mg/100 g	Antioxidant, cardioprotective	Rufino et al. ^[18] ; Silva et al. ^[19]
Jabuticaba (<i>Plinia cauliflora</i>)	Delphinidin, cyanidin, ellagic acid, quercetin derivatives	Anthocyanins / Phenolic acids / Flavonoids	Peel	300–800 mg/100 g	Antioxidant, anti-inflammatory	Inada et al. ^[20] ; Oliveira et al. ^[21]
Camu-camu (<i>Myrciaria dubia</i>)	Ellagic acid, quercetin, catechin	Phenolic acids / Flavonoids	Pulp/ Seed	150–400 mg/100 g	Antioxidant, immunomodulatory	Fidelis et al. ^[22] , Fidelis et al. ^[23] Neves et al. ^[24]
Cagaita (<i>Eugenia dysenterica</i>)	Gallic acid, quercetin	Phenolic acids	Pulp	80–200 mg/100 g	Antioxidant, antimicrobial	Santana et al. ^[25] , Silva et al. ^[26]
Murici (<i>Byrsonima crassifolia</i>)	Gallic acid, catechin	Phenolic acids / Flavanols	Pulp	70–180 mg/100 g	Antioxidant	Fernandes et al., 2024

Fruit (Scientific name)	Major phenolic compounds	Phenolic class	Fruit fraction	Content (range)*	Reported bioactivities	References
Buriti (<i>Mauritia flexuosa</i>)	Ferulic acid, p-coumaric acid	Phenolic acids	Pulp/ Peel	60–150 mg/100 g	Antioxidant, photoprotective	Silva et al. ^[27] , Sousa et al. ^[28]
Buritirana (<i>Mauritiella armata</i>)	Gallic acid, caffeic acid, quercetin derivatives	Phenolic acids / Flavonoids	Pulp	90–210 mg/100 g	Antioxidant, anti-inflammatory	Morais et al. ^[10]
Puçá amarelo (<i>Mouriri grandiflora</i>)	Ascorbic acid, Malic acid	Phenolic acids	Pulp/ Peel and Seed	50–310 mg/100 g	Antioxidant	Soares et al. ^[9]
Umbu (<i>Spondias tuberosa</i>)	Quinic acid	Phenolic acids	Pulp	40–120 mg/100 g	Antioxidant	Gualberto et al. ^[29]
Araçá-boi (<i>Eugenia stipitata</i>)	Catechin, fertaric acid, vanillic acid, cinnamic acid, gallic acid hexoside	Phenolic acids / Flavonoids	Pulp/ Seed	80–200 mg/100 g	Antioxidant	Araujo et al., ^[30]
Pequi (<i>Caryocar brasiliense</i>)	Gallic acid	Phenolic acids	Pulp/ Peel and Seed	60–140 mg/100 g	Antioxidant	Silva et al. ^[31] , Martins et al. ^[32]

Another critical aspect refers to the bioaccessibility and metabolism of phenolic compounds. A significant fraction is linked to the food matrix, reducing its immediate bioavailability^[33, 34]. These compounds can be metabolized by the gut microbiota, generating bioactive metabolites, but there is still a lack of studies that integrate chemical characterization, simulated digestion, and biological evaluation^[8, 35]. In addition, factors such as processing and storage can compromise the stability of these compounds, directly impacting their functionality^[36].

Despite the high potential, many species remain underexploited, with fragmented data and the absence of structured production chains^[1, 36]. This gap limits not only industrial application, but also the advancement of scientific knowledge about the real contribution of the phenolic compounds of these fruits to human health.

Thus, advances in the area depend on more integrated and standardized approaches, which consider not only the phenolic composition, but also its bioaccessibility, stability, and biological relevance under physiologically realistic conditions. This perspective is essential to overcome current limitations and enable the rational use of Brazilian native fruits as sources of phenolic compounds in food systems and functional applications.

Coproducts of native fruits as sources of phenolics

Despite the growing number of studies dedicated to the characterization of the phenolic profile of Brazilian native fruits, most investigations remain focused on the edible fraction traditionally consumed, while other parts of the fruit remain relatively little explored. Evidence indicates that the distribution of bioactive compounds in the plant matrix is highly heterogeneous, with expressive concentrations often associated with co-products generated during agro-industrial processing. Peels, seeds, bagasse, and residual cakes represent a substantial portion of plant biomass that is often discarded or underutilized, despite having a high concentration of bioactive compounds and nutrients of functional and technological

interest[22]. It is estimated that between 30% and 60% of the total mass of many tropical fruits is discarded during processing, depending on the species and the technology employed. In native species such as jabuticaba, buriti, pequi, and bacaba, these residual fractions concentrate significant amounts of dietary fiber, phenolic compounds, and minerals, often at levels higher than those observed in the pulp ^[1, 2, 12].

This distribution shows that the conventional edible fraction represents only part of the functional potential of these plant matrices. Peels and seeds act as relevant reservoirs of polyphenols, often showing higher concentration and chemical diversity than the pulp, suggesting that the current model of fruit use still significantly underexploits this potential^[36]. Despite these advances, the literature remains fragmented and marked by the absence of standardized methodological approaches, especially with regard to the integration between chemical composition, bioaccessibility, and stability of these compounds when incorporated into different food matrices.

In this context, the valorization of food co-products has been driven by strategies aimed at the full use of raw materials and the transition to more sustainable food systems. The incorporation of flours and extracts obtained from peels and bagasse in products such as breads, snacks, beverages, and food bars has shown potential to increase the fiber content and improve the antioxidant profile of formulations ^[19, 38, 39]. At the same time, these materials have been explored as alternative sources for the extraction of bioactive compounds, including polyphenols, carotenoids, and vegetable oils. Emerging techniques such as ultrasound-assisted extraction, microwaves, and pressurized fluids have been applied to increase extraction yields and reduce the environmental impact of processes^[8, 40].

The valorization of these co-products is also part of the circular economy, which proposes the reintegration of residual flows into new production cycles, reducing losses along food chains^[22]. In this model, biomass flows are directed to biological or technical cycles,

prioritizing reuse, recycling, and material recovery rather than disposal ^[41, 42]. Despite broad conceptual recognition of this paradigm, its effective implementation in the tropical fruit sector still faces structural challenges, including logistical limitations, lack of technological standardization, and uncertainties regarding economic viability at an industrial scale^[43, 44].

From a nutritional point of view, fruit co-products have a high content of dietary fibers, particularly insoluble fractions associated with the plant structural matrix. These components may exert relevant physiological effects, including modulation of the gut microbiota and the production of short-chain fatty acids, metabolites associated with metabolic and immune health^[35]. However, much of the available evidence derives from experimental studies or in vitro models, which limits direct extrapolation to human physiological conditions and highlights the need for more robust clinical investigations.

Another relevant challenge refers to the stability of bioactive compounds during processing and storage. Technological processes such as drying, grinding, and extrusion can promote the degradation or structural transformation of polyphenols, thereby compromising their bioactivity and bioavailability^[45, 46]. Thus, the optimization of processing parameters and the development of strategies for the protection of these compounds become essential to enable their industrial application ^[36]. At the same time, sensory factors and consumer acceptance remain determinant for the viability of products formulated with food co-products, requiring a balance between nutritional functionality and sensory quality^[47].

In the case of native Brazilian fruits, the valorization of co-products also has relevant socioeconomic implications, and can contribute to the strengthening of local production chains and sustainable agroextractive systems. The use of these materials as food ingredients can generate greater added value when compared to traditional use as fertilizers or animal feed^[48–50]. In addition, residues from tropical and native fruits have been recognized as promising sources of bioactive compounds and minerals with potential application in different industrial

sectors^[51, 52].

The consolidation of the use of food co-products still depends on the generation of more robust scientific evidence related to the chemical composition, safety and stability of these materials. Global food safety guidelines indicate that the use of ingredients derived from plant biomass should be underpinned by detailed chemical characterization, toxicological evaluation, and strict control of processing conditions and microbiological quality ^[53]. In this context, integrated approaches involving phytochemical characterization, bioaccessibility assessment, and technological development are essential to position co-products from Brazilian native fruits as strategic raw materials for food innovation and bioeconomy advancement.

Simulation of gastrointestinal digestion in vitro

The simulation of gastrointestinal digestion in vitro has been consolidated as an essential tool for evaluating the bioaccessibility of nutrients and bioactive compounds present in complex food matrices. These experimental models seek to reproduce, in a controlled and reproducible manner, the main physicochemical and enzymatic conditions found throughout the human gastrointestinal tract, allowing the investigation of the phenomena of release, transformation and solubilization of compounds during digestion^[6]. Unlike conventional chemical composition analyses, in vitro digestion assays provide more realistic information on the fraction of compounds potentially available for intestinal absorption. This is particularly relevant in the case of phenolic compounds and carotenoids, whose bioactivity depends not only on the total concentration in the food, but also on its ability to be released from the food matrix during digestion^[1].

In vitro digestion models can be classified into static, semi-dynamic, and dynamic. Static models, widely used due to their simplicity and low cost, simulate oral, gastric, and intestinal phases through the sequential addition of enzyme solutions and pH adjustment. Although they do not completely reproduce the physiological dynamics of the digestive tract,

these models have good reproducibility and are suitable for comparative studies^[6, 54]. In native Brazilian fruits, studies using *in vitro* digestion have shown that a significant fraction of phenolic compounds undergo degradation or transformation during digestion, while another portion remains bioaccessible, and may exert biological effects on the small intestine or colon^[8]. These results reinforce the importance of considering digestion as a critical step in the evaluation of the functional potential of foods.

During the gastric phase, factors such as acidic pH and pepsin activity can promote the release of compounds bound to the protein matrix, while favoring the degradation of more sensitive compounds^[55]. In the intestinal phase, the action of pancreatic enzymes and bile salts directly influences the solubilization of lipophilic compounds, such as carotenoids and tocopherols^[2]. The interaction between bioactive compounds and other components of the food matrix, such as fibers and lipids, plays a decisive role in bioaccessibility. Since, the presence of lipids can favor the mycellization and absorption of lipophilic compounds, while fibers can limit and modulate the release of phenolics, depending on their structure and degree of polymerization^[35].

In addition to the evaluation of bioaccessibility, *in vitro* digestion models have been used to investigate changes in antioxidant activity throughout the digestive process. Several studies report that, although there is a reduction in total antioxidant activity after digestion, the metabolites formed may have high bioactivity, especially in the intestinal context^[1]. The applicability of these models also extends to the evaluation of food co-products, allowing us to understand the digestive fate of compounds present in husk, seed, and pomace meals. These studies are fundamental to support the use of these ingredients as functional and prebiotic sources^[36].

From a methodological point of view, the standardization of *in vitro* digestion assays represents a significant advance in the area. The adoption of international and recognized

protocols allows the comparison of results between different studies and food matrices, strengthening the robustness of the scientific evidence generated^[54, 56]. Despite their advantages, it is important to emphasize that in vitro models have limitations and do not completely replace in vivo studies. However, they constitute a fundamental intermediate step, reducing costs, experimental time, and the use of animal models, in addition to providing subsidies for the design of more targeted clinical studies. Thus, the simulation of gastrointestinal digestion in vitro represents a strategic and indispensable approach to understanding the real functional potential of native Brazilian fruits, contributing to the generation of scientific knowledge for concrete technological and nutritional applications.

***In vitro* gastrointestinal digestion models applied to fruits**

Static models

Static in vitro gastrointestinal digestion models operate as discontinuous batch systems designed to sequentially reproduce the oral, gastric, and intestinal stages of digestion. In these approaches, the food sample is combined with simulated digestive fluids and enzymes under controlled conditions of pH, temperature, and incubation time specific to each phase. However, these systems do not replicate the continuous physiological dynamics of the human digestive tract, such as gradual gastric emptying, progressive secretion of digestive fluids, intestinal flow, or nutrient absorption processes^[54, 57]. Despite these simplifications, static models are extensively used to evaluate bioaccessibility, the fraction of compounds released from the matrix and potentially available for absorption, as well as the stability of bioactive molecules such as phenolics, carotenoids, and peptides, the release of nutrients, and changes in the physicochemical characteristics of the digesta. Their widespread adoption is largely attributed to methodological simplicity, relatively low operational cost, high experimental reproducibility, and compatibility with conventional laboratory infrastructure^[54, 58, 59].

The methodological premise of static digestion models is the maintenance of constant

experimental conditions within each phase, including fixed pH values for gastric and intestinal stages, defined food-to-fluid ratios, and standardized enzyme compositions. This standardization facilitates comparisons across different studies and food matrices but limits the model's capacity to represent temporal variations and physiological heterogeneity typical of in vivo digestion.

These models are widely applied in research on plant-derived foods, particularly fruits, and have increasingly been employed in studies of Brazilian native fruits due to their remarkable phytochemical diversity and structural complexity^[20, 60–62]. The use of advanced analytical techniques, including HPLC-DAD, UHPLC-ESI-MS/MS, and LC-QTOF-MS, allows detailed monitoring of structural modifications in phenolic compounds throughout simulated digestion. These transformations may involve the breakdown of polymeric or conjugated molecules into simpler phenolic units, alkaline degradation during the intestinal stage, or the formation of non-covalent associations with dietary fibers, polysaccharides, and plant proteins present in the food matrix^[63, 64]. Furthermore, static digestion systems are commonly combined with functional analyses, such as measurements of antioxidant capacity based on electron- or hydrogen-transfer mechanisms (e.g., ABTS, ORAC, FRAP) and enzyme inhibition assays related to carbohydrate and lipid metabolism, enabling correlations between chemical transformations and potential biological activity.

Dynamic models

Dynamic in vitro digestion systems were developed to more closely emulate the physiological processes occurring in the human gastrointestinal tract by incorporating continuous, time-dependent variations in parameters such as pH, secretion of digestive fluids, mechanical mixing, gastric emptying, and intestinal transit. These systems frequently rely on automated control mechanisms, pumps, and interconnected compartments to reproduce sequential digestive events under controlled kinetic conditions^[65–67]. In contrast to static

approaches, dynamic models allow the gradual addition of enzymes and bile, automated pH regulation via sensors and controllers, and mechanical simulation of gastrointestinal motility. These features enable sampling at multiple time points and, in some configurations, the removal of digested fractions through membrane systems such as dialysis to mimic nutrient absorption, thereby maintaining conditions closer to those observed in vivo^[66]. Representative dynamic digestion systems currently used in food research, together with their main characteristics, limitations, and typical applications, are summarized in Table 2.

Table2. Comparison of representative dynamic *in vitro* gastrointestinal digestion models used in food research.

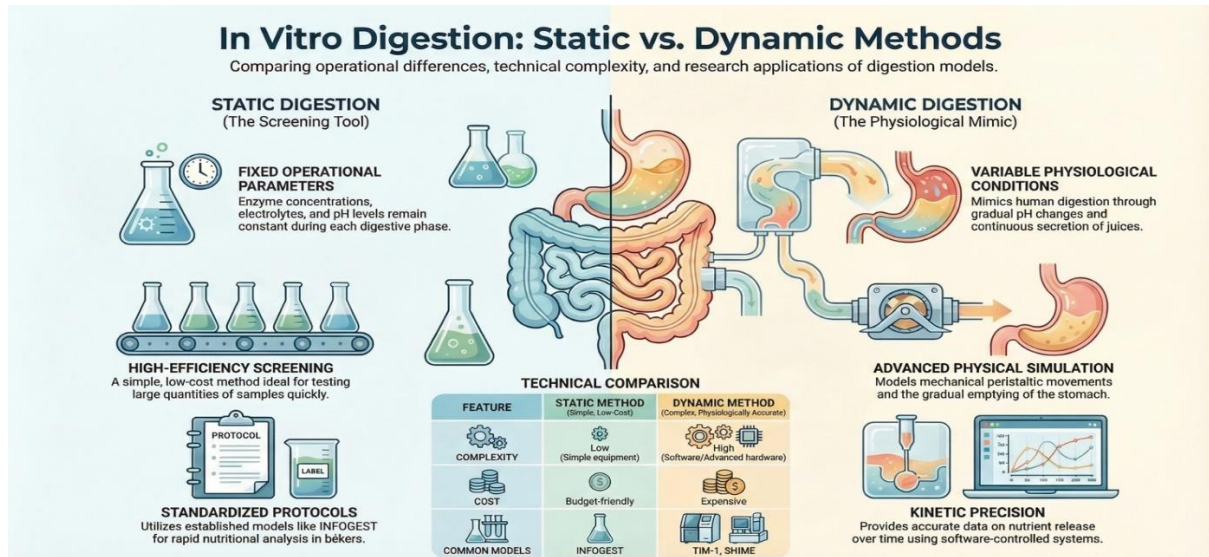
Model	Key features	Cost / complexity	Main limitations	Typical applications	References
TIM-1 (TNO Intestinal Model)	Multicompartment system simulating stomach, duodenum, jejunum, and ileum; automated control of pH, enzyme secretion, gastric emptying, and peristaltic mixing; dialysis membranes simulate nutrient absorption	Very high	Expensive infrastructure; specialized operation; limited accessibility across laboratories	Bioaccessibility of nutrients and phytochemicals; digestion kinetics; evaluation of complex food matrices	Dupont et al. ^[68]
Dynamic Gastric Model (DGM)	Mechanical simulation of gastric peristalsis, secretion, and gradual gastric emptying	High	Restricted to gastric phase; requires coupling with intestinal models	Gastric digestion kinetics; disintegration of solid foods; release of bioactive compounds	Dupont et al. ^[68] ; Singh ^[69]
DIDGI® system	Two-compartment model simulating stomach and small intestine with dynamic pH regulation, enzyme secretion, and gastric emptying	Moderate–high	Limited simulation of intestinal absorption; fewer compartments than TIM	Bioaccessibility studies of nutrients and polyphenols; digestion of functional foods	Li et al. ^[70] ; Ji et al. ^[71]
SHIME® (Simulator of the Human Intestinal Microbial)	Multicompartment system including stomach, small intestine, and colon fermentation; simulates gut microbiota metabolism	Very high	High operational complexity; microbiota variability; long experimental duration	Interaction between digestion and gut microbiota; metabolism of polyphenols and dietary	Dupont et al. ^[68] ; Ji et al. ^[71]

Model	Key features	Cost / complexity	Main limitations	Typical applications	References
Ecosystem)				fibers	
Human Gastric Simulator (HGS)	Mechanical simulation of gastric mixing and peristalsis with controlled acidification and enzyme addition	Moderate–high	Focus mainly on gastric digestion; no simulation of intestinal absorption	Physical breakdown of foods; digestion of structured matrices (gels, emulsions)	Singh ^[69]

Dynamic digestion models are particularly valuable when the objective is to examine the kinetics of compound release rather than relying solely on endpoint measurements. They are also advantageous for assessing the time-dependent stability of sensitive compounds such as anthocyanins, curcuminoids, and alkaloids, investigating the digestion of structurally complex food systems, including emulsions, oleogels, and protein gels, and establishing correlations between in vitro observations and physiological responses. In practice, these systems tend to provide improved predictive capability for processes occurring in the upper gastrointestinal tract, especially in the stomach and small intestine. However, their implementation requires specialized equipment, rigorous methodological standardization, longer experimental durations, and significantly higher operational costs. The mechanical complexity of these systems and variability in parameter settings, such as gastric emptying profiles and pH curves, may also complicate direct comparisons between laboratories^[72, 73].

Dynamic digestion models offer a more physiologically relevant simulation of gastrointestinal processes than static approaches, especially by reproducing the dynamic changes in pH, enzyme secretion, gastric emptying, and gastrointestinal transit over time^[57, 65, 74, 75]. However, static models remain the most widely employed in food and nutrition research due to their simplicity, reproducibility, and cost-effectiveness^[6, 76]. Therefore, a direct comparison between these two approaches is essential to highlight their characteristics, advantages, limitations e aplicações adequadas. Figure 1 summarizes the main differences between static and dynamic models of in vitro digestion, providing an overview of their operating principles and applicability in gastrointestinal digestion studies.

Figure 1. Comparison between static and dynamic in vitro digestion methods.



INFOGEST Protocol

The INFOGEST protocol emerged as an international initiative with the objective of standardizing in vitro gastrointestinal digestion methods used in bioaccessibility and bioavailability studies of nutrients and bioactive compounds. Developed by a consortium of researchers from different fields, the protocol was originally published by Minekus et al.^[6] and has since been widely adopted and refined by the scientific community. The main contribution of the INFOGEST protocol lies in the precise definition of the experimental conditions for each phase of digestion, including composition of digestive solutions, enzyme concentrations, incubation times, temperature, and pH adjustments. This standardization reduces the methodological variability observed in previous studies and allows for a more reliable comparison of results between different laboratories and food matrices^[4, 77, 78].

The protocol includes three main phases: oral, gastric, and intestinal. In the oral phase, mastication and the action of salivary α -amylase are simulated, with a short contact time and a pH close to neutrality. This step, although often neglected in older protocols, can significantly influence the physical structure of the food matrix and the initial release of water-soluble compounds^[6]. The gastric phase is characterized by the reduction of pH and the action of pepsin, simulating the acidic environment of the human stomach. During this stage, protein denaturation, release of protein-associated compounds, and possible structural degradation of

compounds sensitive to acidic pH occur. Studies with native fruits have shown that this phase has a significant impact on the stability of anthocyanins and other polyphenols^[1]. In the intestinal phase, the INFOGEST protocol includes the addition of pancreatin and bile salts, simulating digestion in the small intestine. This step is crucial for the mycellization of lipophilic compounds and for the evaluation of the bioaccessible fractions of carotenoids, tocopherols, and fatty acids present in fruits and co-products^[2].

One of the advantages of the INFOGEST protocol is its flexibility, allowing controlled adaptations according to the objective of the study, such as the inclusion of a simulated colonic phase or the evaluation of specific digestion for different population groups. However, these adaptations must be clearly described and justified to maintain comparability of results^[79]. Studies applying the INFOGEST protocol to native Brazilian fruits have revealed that the bioaccessibility of phenolic compounds varies widely between species and strongly depends on the food matrix. In some cases, there is an increase in the bioaccessible fraction after digestion, while in others, there is a significant reduction, reinforcing the complexity of digestive interactions^[8]. Examples of these variations in individual phenolic compounds identified in different native fruits and their bioaccessibility after static digestion are summarized in Table 3.

Table 3. Phenolic compounds identified in different fruits and their contents before and after in vitro digestion, as well as the bioaccessibility (%) determined by the static model INFOGEST.

Fruit (species)	<i>In vitro</i> method	Compounds	Before digestion	After digestion	Bioaccessibility (%)	References
Yellow Puçá (<i>Mouriri grandiflora</i>)	Static digestion model INFOGEST, and the result expressed in $\mu\text{g g}^{-1}$	Gallic acid	319.03	80.30	25.17	Soares et al. ^[9]
		Theobromine	4.65	1.58	33.97	
		Catechin	4.15	4.02	96.86	
		Chlorogenic acid	1.24	0.76	61.30	
		Caffeic acid	0.62	0.46	74.20	
		<i>p</i> -coumaric acid	0.61	0.06	22.95	
		<i>m</i> -coumaric acid	0.43	0.23	13.95	
Guapeva (<i>Pouteria gardneriana</i>)	Static digestion model INFOGEST, and the result expressed in $\mu\text{g g}^{-1}$	<i>trans</i> -cinnamic acid	0.08	0.06	75.00	Morais et al. ^[62]
		Trigonelline	65.67	7.25	86.98	
		Gallic acid	452.63	180.42	55.67	
		Theobromine	19.64	1.17	17.71	
		Catechin	23.98	11.23	46.83	
		Chlorogenic acid	77.05	10.61	40.03	
		Caffeic acid	87.78	10.94	12.46	
Jaboticaba (<i>Plinia jaboticaba</i>)	Static digestion, and the result expressed	Vanillin	11.87	5.10	42.97	Inada et al. ^[20]
		Gallic acid	237.00	161.70	68.22	
		Ellagic acid	151.40	337.50	222.91	

Yellow Puçá (<i>Mouriri grandiflora</i>)	Static digestion model INFOGEST, and the result expressed in $\mu\text{g g}^{-1}$	in mg 100 g^{-1}	Myricetin	5.60	4.90	87.50	Soares et al. ^[9]
			Myricetin-3- <i>O</i> -rhamnoside	9.50	8.80	92.63	
			Quercetin	1.10	4.50	409.09	
			Quercetin-3- <i>O</i> -rutinoside	115.60	193.70	167.56	
			Cyanidin-3- <i>O</i> -glucoside	1112.00	213.20	18.17	
			Delphinidin-3- <i>O</i> -glucoside	159.40	35.01	21.96	
			Trigonelline	1.42	0.25	17.60	
			Gallic acid	652.01	200.64	30.77	
			Theobromine	0.34	0.24	70.58	
			Catechin	0.80	0.73	91.25	
Baru (<i>Dipteryx alata</i>)	Static digestion model INFOGEST, and the result is expressed in mg 100 g^{-1}		Chlorogenic acid	0.62	0.30	48.38	Fernandes et al. ^[80]
			Caffeic acid	1.19	0.76	63.86	
			<i>p</i> -coumaric acid	1.52	0.10	6.57	
			<i>trans</i> -cinnamic acid	3.32	0.77	23.19	
			Caffeic acid	1.61	1.74	47.22	
			Vanillic acid *	4.82	3.95	28.86	
			Trans-caftaric acid *	8.43	7.17	79.38	
			Ferulic acid *	1.29	1.18	45.30	
			Epigallocatechin *	1.73	1.83	49.44	
			Epigallocatechin gallate *	0.75	0.32	16.02	
			Naringin *	4.82	6.32	47.48	

Quercetin-3-glucoside	0.57	8.85	19.85
Procyanidin B1	10.98	4.89	11.88
Procyanidin B2	49.82	10.55	5.85

Note: Before digestion refers to the analysis of the matrix in its original state, performed without subjecting it to any stage of the simulated *in vitro* gastrointestinal digestion process. After digestion, the final phase of the simulated *in vitro* gastrointestinal digestion is the intestinal phase.

The protocol has also been widely used in the evaluation of food co-products, allowing to demonstrate that traditionally discarded fractions can present high bioaccessibility of bioactive compounds, especially after intestinal digestion. These findings strengthen the scientific argument in favor of the full use of native fruits^[36]. From an analytical point of view, the application of INFOGEST is often combined with chromatographic and spectrophotometric techniques for the quantification of bioactive compounds before and after digestion. This integrated approach makes it possible to assess not only total bioaccessibility, but also qualitative changes in the phytochemical profile^[14].

Despite its numerous methodological advantages, the INFOGEST protocol has limitations inherent to static models of in vitro digestion. The system predominantly reproduces the luminal stages of digestion and, therefore, does not contemplate subsequent physiological processes, such as intestinal absorption, hepatic metabolism, systemic circulation and excretion, which are determinant steps for the effective bioavailability of nutrients and bioactive compounds. Thus, the results obtained mainly reflect the potentially bioaccessible fraction, and not necessarily the fraction biologically available in the human body. In addition, the model simplifies several aspects of gastrointestinal physiology, including dynamic pH variations, continuous secretion of digestive enzymes, gastrointestinal motility, and interactions with the gut microbiota, factors that can influence the stability and structural transformation of phenolic compounds during digestion^[6, 76]. In summary, the INFOGEST protocol is currently the most widely adopted methodological standard for in vitro gastrointestinal digestion studies, especially for comparative evaluation of the bioaccessibility of bioactive compounds. When interpreted within its experimental scope and, where possible, combined with complementary approaches such as cellular models of intestinal absorption, simulated colonic fermentation systems, or dynamic digestion models, the results obtained provide robust and comparable scientific evidence. Its application in research involving native Brazilian fruits has contributed

significantly to understanding the digestive fate of phenolic compounds and to support the valorization of these matrices in the context of functional nutrition, food technology and health promotion.

Conclusions

Brazilian native fruits are promising sources of bioactive compounds with significant potential to promote human health; however, their biological effects depend on the stability and bioaccessibility of these compounds during gastrointestinal digestion. This review highlights the importance of standardized in vitro digestion methods, especially the INFOGEST protocol, as reliable tools to assess the digestive fate of phytochemicals and generate comparable results across studies. Current evidence demonstrates that the bioaccessibility of phenolic compounds is strongly influenced by the food matrix and digestive conditions, reinforcing the need to evaluate these compounds beyond their initial concentrations. While static digestion models remain the most widely used due to their simplicity, reproducibility, and cost-effectiveness, dynamic models provide a more physiologically relevant simulation of gastrointestinal processes and represent an important direction for future research. The integration of standardized digestion protocols with advanced analytical techniques and complementary approaches will contribute to a better understanding of the health potential of Brazilian native fruits, supporting their appreciation as sustainable sources of functional ingredients and promoting their application in food, nutritional, and nutraceutical development.

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No potential conflict of interest was reported by the author(s).

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ARTICLE 2: SYSTEMIC CHARACTERIZATION OF THE PULP, PEELS, AND SEEDS OF THE YELLOW PUÇÁ (MOURIRI GRANDIFLORA) AND IMPACTS OF SIMULATED IN VITRO GASTROINTESTINAL DIGESTION ON THE CHEMICAL, BIOLOGICAL, AND ANTIOXIDANT PROPERTIES OF POLYPHENOLS

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Systemic Characterization of the Pulp, Peels, and Seeds of the Yellow Puçá (*Mouriri grandiflora*) and Impacts of Simulated In Vitro Gastrointestinal Digestion on the Chemical, Biological, and Antioxidant Properties of Polyphenols

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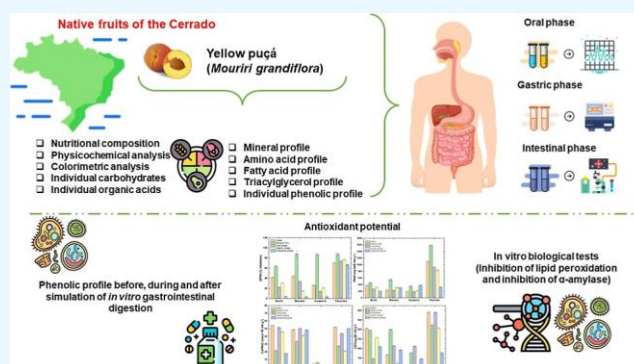
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ABSTRACT: The yellow puçá (*Mouriri grandiflora*), a typical plant of the Cerrado, has nutritional and bioactive properties that are still little explored. The objective of this study was to evaluate the nutritional composition, physicochemical, and phytochemical properties, and to verify the effects of the in vitro gastrointestinal digestion simulation process on the phenolic profile, antioxidant potential, and in vitro biological properties of the pulp, peels, and seeds of the yellow puçá. The seeds presented high lipids, proteins, and fibers (17.35, 21.56, and 35.00 g 100 g⁻¹, respectively). On the other hand, the pulp presented high contents of fructose (17.11 mg g⁻¹) and malic acid (315.67 mg g⁻¹). The peels presented the highest results for phytochemical analyses, amino acid profile, and mineral analysis. Regarding the phenolic profile, gallic acid was the compound marjoritary for pulp, peels, and seeds (652.01, 173.94, and 319.03 μg g⁻¹, respectively). However, all phenolic compounds analyzed showed significant reductions when compared to the undigested sample and the intestinal phase ($p < 0.05$), indicating degradation processes or the formation of new compounds. Considerable decreases in the antioxidant potential and in vitro biological properties were observed. This is the first study to detail the complete characterization of all yellow puçá fractions.



1, INTRODUCTION

The Brazilian Cerrado is widely recognized as a biodiversity hotspot due to its vast variety of species, many of which remain underexploited and underutilized. Among these, the fruits of the Cerrado stand out, presenting not only pleasant and unique sensory characteristics but also significant nutritional, functional, and therapeutic potential due to the presence of bioactive and antioxidant compounds, such as phenolics, vitamins, and carotenoids, among other compounds.^{1,2} Although some of these species are already used, primarily in

a limited manner by local communities, being consumed in their natural state or processed into juices, liqueurs, ice creams, jams, and artisanal sweets, there is considerable exploration potential that has not yet been fully exploited.^{3,4} A relevant example of this wealth is the yellow puçá (*Mouriri grandiflora*), a species that has characteristics that are particularly favorable for a healthy and nutritious diet.

M. grandiflora, which belongs to the Melastomataceae family, is a fruit plant that produces edible fruits, popularly known as yellow puçá or camutim. Its fruits have a globose berry that contains a single seed surrounded by a fleshy orange pulp with a sweet flavor, and the remains of a calyx crown its yellow peel. These characteristics give the puçá a highly appreciated flavor.⁵ The medium-sized tree has elongated branches with small flowers ranging from white to pink along its woody branches.⁶ Rufino et al.⁷ highlighted the potential of puçá fruit as a valuable resource, owing to its significant levels of bioactive compounds, including vitamin C, carotenoids, and phenolic

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compounds. The synergistic effects of these components contribute to the fruit's exceptional antioxidant capacity, positioning it as a promising candidate for sustainable agricultural practices and innovative product development. In addition to its considerable nutritional value, puçá has amazing potential for producing food and nutraceutical products, with potential benefits for preventing several diseases.⁵

Polyphenols present in native fruits have attracted increasing interest due to their therapeutic potential and health benefits, especially in preventing heart and neurological diseases.⁸ However, polyphenol absorption is inefficient, as most of these compounds are in their conjugated form and need to be hydrolyzed by intestinal enzymes or the microbiota of the digestive system to be absorbed. It is estimated that only 5–10% of ingested polyphenols are absorbed in the small intestine, while the majority are metabolized in the large intestine.^{9,10} Understanding the physicochemical transformations that foods undergo during digestion and the factors that impact nutrient bioaccessibility, bioavailability, and digestibility is crucial for developing functional foods. Bioaccessibility specifically refers to the proportion of a compound that is liberated from the food matrix and dissolved in the aqueous phase of the chyme, thereby becoming available for absorption across the intestinal membrane into the bloodstream.^{11,12} Bioavailability, on the other hand, describes the total amount of a compound released and absorbed, reaching the bloodstream, and distributed to different tissues throughout the body.¹³ Digestibility concerns explicitly the fraction of food components that transform potentially accessible material (present in soluble and insoluble digest fractions) through the physical and chemical processes that occur in the lumen.^{11,14} A more comprehensive understanding of these processes is essential for determining optimal daily nutrient intake recommendations and identifying processing conditions that enhance the health-promoting properties of bioactive compounds.¹⁵ Despite the growing scientific interest in native fruits of the Brazilian Cerrado, such as the yellow puçá (*M. grandiflora*), there are still important gaps in the literature regarding the complete characterization of its nutritional, functional, and bioactive properties, especially related to the digestibility, bioaccessibility and bioactivity of its compounds after the gastrointestinal digestion process. Most studies are limited to identifying the basic composition or consumption by local communities, neglecting crucial steps such as simulating in vitro digestion, which is essential to assess the real availability of bioactive compounds in the human body. Furthermore, to date, there are no studies in the literature that address the different parts of the fruit (pulp, peel, and seeds) in an integrated manner, disregarding its full potential for the development of sustainable functional foods and nutraceuticals. Therefore, this study stands out for using the standardized INFOGEST protocol to simulate gastrointestinal digestion in vitro, enabling a comprehensive and comparative evaluation of the physicochemical transformations, the release of bioactive compounds, and their antioxidant and biological properties in different fractions of the yellow puçá. This approach is unprecedented for this species and represents a significant advance in the understanding of the bioavailability and full use of underexploited native fruits, contributing to the valorization of the Cerrado biodiversity and the development of new products with functional and therapeutic appeal.

2. MATERIALS AND METHODS

2.1. Chemical Reagents. The reagents neocuproine, thiobarbituric acid, potassium sulfate, sodium carbonate,

sodium molybdate, ethanol, aluminum chloride, hydrochloric acid, 2,2-diphenyl-1-picrylhydrazyl (DPPH[•]), tripyridyl triazine (TPTZ), ferric chloride, ascorbic acid, potassium persulfate, potassium permanganate and α -amylase (30 U mg⁻¹), pepsin (535 U mg⁻¹), pancreatin (activity equivalent to 8× USP), and bile salts, such as sodium glycodeoxycholate (0.8 mM), sodium taurodeoxycholate hydra (0.45 mM), and sodium taurocholate hydrate (0.75 mM). Standards glucose, xylose, fructose, cellobiose, arabinose, citric acid, ascorbic acid, malic acid, and tartaric acid were used to analyze individual carbohydrates and organic acids. All other standards and reagents were purchased from Sigma-Aldrich (São Paulo, Brazil). All chemical reagents were of analytical grade.

2.2. Samples. The fruits of the yellow puçá (*M. grandiflora*) were collected in the Cerrado of the municipality of Lajeado, Tocantins, Brazil (Latitude—9°61'84" S, Longitude—48°35'51" W) between October and December 2022. The fruits were harvested manually and then subjected to cleaning, selection, and classification processes, with only ripe fruits in good condition being selected. The fruits were sanitized in chlorinated water at 100 mg L for 15 min. After sanitization, the fruit fractions (pulp, peels, and seeds) were separated manually. After separation, the fruit fractions were dried in a circulation oven at 50 °C for 48 h. After drying, the fractions were ground in a commercial blender until they reached a standard particle size of 1.00 mm. They were stored in low-density polyethylene plastic bags at −20 °C until the analysis.

2.3. Nutricional and Physicochemical Composition. All analyses were determined according to the Association of Official Analytical Chemists.¹⁶ Methods 923.03 and 925.09, respectively, were used to determine ash and moisture content. The fat content was evaluated by the Soxhlet method, with *n*-hexane as the extracting solvent, during 6 h of reflux (method 920.85). The protein content was determined by the Kjeldahl method, using a conversion factor of 6.25 (method 920.87). The fibers were determined in a muffle furnace at 550 °C by gravimetric test (method 941.43). Carbohydrates were obtained by calculating the difference between the other constituents. The titratable acidity (947.05), pH (981.12), and soluble solids (932.12) were also analyzed. The total energy value was estimated considering the conversion factors of 4 kcal g⁻¹ for proteins and carbohydrates and 9 kcal g⁻¹ for lipids, and expressed in kcal 100 g⁻¹ of the sample. Color analysis was performed at 25 °C using a digital colorimeter (Minolta CR4000, D₆₅ light source in *L**, *a**, *b**, chroma, and hue color space in the CIELAB system). All results were expressed as mean ± standard deviation (g 100 g⁻¹). The extraction of starch and pectin from the pulp, peels, and seeds of the yellow puçá followed the protocols proposed by de Castro et al.¹⁷ with modifications made by Alves Morais et al.¹⁸ and Torralbo et al.¹⁹ respectively. The starch and pectin yields will be calculated using eq 1. The yields will be expressed in grams per hundred grams (g 100 g⁻¹).

$$y = \frac{p}{Bi} \times 100 \quad (1)$$

where y is the percentage yield of extracted starch or pectin, $p(g)$ is the mass of extracted starch or pectin in grams, and $B_i(g)$ is the initial mass of the sample.

2.4. Individual Carbohydrates and Organic Acids. For the quantification of individual carbohydrates and organic acids, the extracts were prepared as described by Alves Morais et al.,²⁰ and the injection was made with 10 μ L of each extract. Carbohydrates and organic acids were determined by high-performance liquid chromatography (HPLC) using an Agilent model 1260 Infinity II equipment equipped with a refractive index detector (RID) at 40 °C. The analysis was performed with a Supelcogel C_{610H} chromatographic column (30 cm $\times$ 7.8 mm) and a Supelguard C_{610H} precolumn (5 cm $\times$ 4.6 mm) from Sigma-Aldrich. The mobile phase consisted of a 0.1% phosphoric acid (H₃PO₄) solution in deionized water, with a flow rate of 0.5 mL min⁻¹, total run time of 30 min, and oven temperature of 40 °C. Glucose, xylose, fructose, cellobiose, and arabinose were used as standards for the identification of carbohydrates, and citric, ascorbic, malic, and tartaric acids were used as standards for the identification of organic acids at a wavelength of 215 nm. The results were expressed in mg g⁻¹.

2.5. Bioactive Compounds. **2.5.1. Recovery of the Extracts from Yellow Puçá Parts and Total Phenolic Content.** To obtain extracts from the pulp, peels, and seeds of yellow puçá, 25 g of the samples were diluted in 100 mL of 80% (v/v) ethanol. Subsequently, the samples were subjected to sonication using an ultrasonic bath (EGS 5HD, 40 kHz, 300 W, Enge Solutions, São Paulo, Brazil) at 40 °C for 60 min. Afterward, the extracts were filtered (Whatman no. 541, 125 mm) and stored in amber vials at -20 °C for later analysis. These extracts were used to determine the phytochemical content of the respective fractions. Total phenolic contents were determined using the Folin–Ciocalteu spectrophotometric method according to Singleton and Rossi.²¹ For this purpose, 1 mL of the fruit fraction extracts was added to a solution composed of 0.2 mL of Folin–Ciocalteu reagent (10%), 2 mL of distilled water, and 1 mL of sodium carbonate (4%). The mixture was homogenized and kept in the dark for 2 h at room temperature. The absorbance was recorded at a wavelength of 750 nm, and the results were reported as milligrams of gallic acid equivalents per 100 g (mg GAE 100 g⁻¹).

2.5.2. Total Flavonoids and Anthocyanins. Flavonoid and anthocyanin content were determined using the method outlined by Francis and Markakis.²² In brief, 1 g of each dried sample was dissolved in 10 mL of an extraction solution consisting of 1.5 N HCl in 85% ethanol (v/v). The samples were homogenized, transferred to a 50 mL volumetric flask, and extracted for 13 h under refrigeration in an inert light environment. After extraction, the extracts were filtered using Whatman no. 1 filter paper. Absorbance was measured at 535 nm for anthocyanins and at 374 nm for flavonoids using a spectrophotometer. The results were expressed in mg per 100 g (mg 100 g⁻¹).

2.5.3. Total Carotenoids. Carotenoid extraction was conducted following the protocol described by Kimura and Rodriguez-Amaya.²³ In brief, 1.0 g of each part of the yellow puçá fruit (pulp, peel, and seeds) was weighed and ground with 30 mL of acetone. This mixture was then transferred to a separatory funnel containing 50 mL of petroleum ether and filtered 3 times until the residue became colorless. The solution obtained was transferred to a separatory funnel containing 50 mL of petroleum ether and washed 3 times in sequence until the residue was colorless. Subsequently, 50 mL of acetone was added to the extract, followed by the controlled

addition of 300 mL of distilled water. After separation of the phases, the lower fraction—composed predominantly of water and traces of solvents—was discarded. The remaining ethereal phase was then washed repeatedly (5 times) with distilled water to eliminate residues of water-soluble solvents. Then, the absorbance of the ethereal fraction was determined by spectrophotometry in the ultraviolet–visible (UV–vis) region. To quantify the carotenoids, the absorbance recorded at 450 nm was used, together with the molar extinction coefficient of β -carotene in petroleum ether (ϵ = 2592), as described in eq 2. The results were expressed as mean $\pm$ standard deviation, in micrograms of total carotenoids per gram of sample (μ g β -carotene g⁻¹).

$$\text{Total carotenoids} = \frac{A \times V \times 10^4}{E^{1\%}_{1\text{cm}} \times m} \quad (2)$$

where, A = extract absorbance at 450 nm; V = volume of the volumetric (mL); m = sample mass (g); $E^{1\%}_{1\text{cm}}$ = extinction coefficient.

1.1. Mineral Profile. Minerals were determined using two digestion methods: nitro-perchloric, at a controlled temperature of 210 °C, and sulfuric, at a controlled temperature of 350 °C, according to the methodology described by Silva.²⁴ First, 0.5 g of the sample was placed in a digestion tube, where 4 mL of concentrated solvent was added for both methods. Subsequently, the sample was taken to the digestion block for 2 h. After cooling, another 2 mL of nitric acid and 2 mL of concentrated hydrogen peroxide were added, and the sample was returned to the digestion block for another 2 h. Then, the volume of the samples was completed with ultrapure water to 25 mL in a volumetric flask, and then the samples were filtered through filter paper. Nitro-perchloric digestion was used to analyze the contents of magnesium, potassium, phosphorus, boron, sulfur, copper, manganese, zinc, and iron. The sulfuric digestion product was used to determine total nitrogen by micro-Kjeldahl distillation. Phosphorus was quantified with a molecular absorption spectrophotometer (model 700 Plus, FEMTO, São Paulo, Brazil), operating in the 660 nm range, while boron and sulfur were analyzed in the 420 nm range. Potassium was detected by flame photometry according to model 400 from Corning (New York, USA). Magnesium, zinc, copper, and iron contents were obtained with a flame atomic absorption spectrophotometer, model AAnalyst-400, from PerkinElmer (Waltham, USA). The results were expressed as mean $\pm$ standard deviation (g 100 g⁻¹).

1.2. Amino Acids. Determining amino acids present in the pulp, peel, and seeds of yellow puçá was carried out following the protocol proposed by Hagen, Frost, and Augustin²⁵ and White, Hart, and Fry.²⁶ When weighing the sample and adding it to a hydrolysis tube, 9 mL of 6 N HCl with 3% w/v phenol was added, and the tube was aspirated, sealed, and placed in a thermal reaction block, leaving it for 24 h at 110 °C. Then, an aliquot of the internal standard AAAB was added and diluted to the tabulated volume. Following the dilution table, an aliquot was filtered and dried at 70 mTorr in a trap system with cryogenic nitrogen. It was neutralized with a solution of 0.2 N sodium acetate trihydrate, HPLC grade methanol, and 99% triethylamine (4:4:2 w/w/v) and dried again. At this step, PITC is added to derivatize the amino acids released by hydrolysis, forming PTC-amino acid. To the tube containing the derivatized amino acid crystals, 500 μ L of diluent was added. Detection was performed at 254 nm by reversed-phase chromatography (30 μ L injection loop, pH 6.40, in a binary linear gradient with a flow rate of 1 mL min⁻¹ and column temperature of 58 °C), with eluent A being a buffer of 0.14 N

sodium acetate, acetonitrile (240 mL 2000 mL⁻¹ of 0.14 N sodium acetate), and triethylamine (1 mL 2000 mL⁻¹ of 0.14 N sodium acetate). Eluent B was a 6:4 solution of acetonitrile (HPLC grade) and Milli-Q water (v/v). Quantification was performed using AAAB, aminobutyric acid, as an internal standard. Results are expressed as mean $\pm$ standard deviation (g 100 g⁻¹).

1.3. Fatty Acid Profile and Triacylglycerol Composition. Lipids were extracted from the samples using the method (920.85) described by the Association of Official Analytical Chemists,¹⁶ followed by fatty acid esterification as described by Hartman and Lago.²⁷ The fatty acid methyl esters (FAMES) obtained were analyzed using gas chromatography coupled with flame ionization detection (GC-FID) on a Shimadzu GC 2010 chromatograph (Agilent Technologies Inc., Palo Alto, CA, USA), which was fitted with a flame ionization detector. Injection was performed in split mode (ratio 1:50) using a capillary column (SPTM-2560 Supelco, 100 m $\times$ 0.25 mm $\times$ 0.20 μ m; Supelco Inc., Bellefonte, PA, USA). The temperature ramp involved heating the samples from 140 °C (for 5 min) to 240 °C (for 30 min) at a rate of 4 °C min⁻¹, with the injector and detector set at 260 °C, and helium was used as the carrier gas. Fatty acids were identified and quantified by comparing the retention times of the sample peaks with those of the fatty acid standard mixture (Supelco₃₇ Component FAME; Supelco, Bellefonte, PA, USA). The quantification was performed by normalization of the area and expressed as a percentage. The triacylglycerol profile of oils extracted from the pulp, peels, and seeds of yellow puçá was determined using the ProOleos software, as designed by Filho et al.²⁸ This approach employs a random distribution model for fatty acids, assuming no positional preference between the *sn*-1,3 and *sn*-2 sites on the glycerol backbone, and relies on the fatty acid composition of the sample as the primary input for analysis.

1.4. Antioxidant Potential. **2.9.1. DPPH Scavenging Assay.** The DPPH assay will follow the protocol established by Brand-Williams, Cuvelier, and Berset.²⁹ Absorbance measurements will be recorded at a wavelength of 517 nm. The outcomes will be determined based on the radical scavenging activity and the extent of radical consumption in the reaction mixture. The results will be expressed as a percentage of inhibition, calculated using eq 3.

$$\% \text{ inhibition} = \left[1 - \left(\frac{A_A}{A_B} \right) \right] \times 100 \quad (3)$$

where A_A is the absorbance value of the sample, while A_B denotes the absorbance value of the control.

2.9.2. Ferric Reducing Assay (FRAP). The ferric reduction assay (FRAP) was determined according to Benzie and Strain.³⁰ The FRAP solution was prepared by adding 2.5 mL of TPTZ solution (10 mmol L⁻¹) diluted with HCl (40 mmol L⁻¹), 2.5 mL of ferric chloride hexahydrate (20 mmol L⁻¹), and 25 mL of sodium acetate buffer solution (pH 3.6). In a microplate, 20 μ L of the obtained extracts and 280 μ L of the FRAP reagent were added. The microplate was kept protected from light at 37 °C for 5 min. The absorbance of the solutions was measured at 593 nm. The reducing power of iron was determined from an ascorbic acid standard curve, and the results were expressed in mg of ascorbic acid equivalent per 100 g of sample (mg AAE 100 g⁻¹).

2.9.3. Total Reducing Capacity (TRC). The total reducing capacity of hydrophilic and lipophilic compounds was evaluated according to the protocol proposed by Berker et al.³¹ In a centrifuge tube, 75 μ L of the Folin–Ciocalteu reagent diluted in isobutanol 1:2 (v/v) was added to 50 μ L of the sample. After 2 min, 875 μ L of a 0.1 mol L⁻¹ NaOH solution and 1.50 mL of ultrapure water were added to the mixture, which was then shaken for 10 s. The tubes remained at rest, protected from light, at room temperature, for 20 min. After the reaction, 250 μ L of the solution was transferred to a microplate. The absorbance of the solutions was measured at 665 nm. The total reducing capacity was determined from a quercetin standard curve, and the results were expressed as mg of quercetin equivalent per 100 g of sample (mg QE 100 g⁻¹).

2.9.4. Copper Reducing Assay (CUPRAC). The antioxidant activity evaluation based on the CUPRAC method was performed according to Apak et al.³² First, 50 μ L of the obtained extracts were added to a test tube, followed by 1 mL of copper chloride (CuCl₂), 1 mL of neocuproine (C₁₄H₁₂N₂), and 1 mL of ammonium acetate (pH 7.0). Then, 250 μ L of the solution was transferred to a microplate and kept at rest, protected from light, at room temperature, for 30 min. The absorbance of the solutions was measured at 450 nm. The reducing capacity of cupric ions was determined from a Trolox standard curve, and the results were expressed in mmol of Trolox equivalent per 100 g of sample (mmol TE 100 g⁻¹).

1.5. In Vitro Biological Properties. **2.10.1. Inhibition of Lipid Peroxidation (ILP).** The inhibition of lipid peroxidation induced by Fe²⁺ in phospholipids, triacylglycerols, and proteins present in egg yolk will be assessed following the methodology outlined by Daker et al.³³ and Margraf et al.,³⁴ with slight modifications. The formation of peroxidation products will be monitored by measuring absorbance at 517 nm. A control assay (AC) will be simultaneously performed using ultrapure water instead of the extracts. The percentage of lipid peroxidation inhibition will be calculated using eq 4.

$$ILP = \frac{A_c - A_a}{A_c} \times 100 \quad (4)$$

where ILP is the inhibition of lipid peroxidation (%); A_c is the absorbance of the control sample; A_a is the absorbance of the sample.

2.10.2. α -Amylase Inhibition. The α -amylase inhibitory activity of the extracts was assessed following the method proposed by Ademiluyi and Oboh. In this procedure, 50 μ L of the extract was combined with 500 μ L of an α -amylase solution, prepared in a 20 mM sodium phosphate buffer (pH 6.9), and incubated at 25 °C for 10 min. Subsequently, 500 μ L of a 1% starch solution was introduced into the mixture, and the incubation proceeded at 25 °C for an additional 10 min. The reaction was then triggered by adding 1 mL of a 60 nM 3,5-dinitrosalicylic acid (DNS) solution and then heating the mixture at 95 °C for 5 min. After the heating step, the samples were rapidly cooled in an ice bath, maintained at approximately 4 °C. To halt the reaction, 10 mL of distilled water was added, and the absorbance of the final solution was measured at 540 nm using a UV–vis spectrophotometer. A control sample was prepared by replacing the extract with 50 μ L of 20 mM sodium

Table 1. Nutritional Composition, Carbohydrate and Organic Acid Profile, and Phytochemicals of the Pulp, Peels, and Seeds of the Yellow Puçá (Mouriri grandiflora)^a

Nutritional composition (g 100 g ⁻¹)	Pulp	Peel	Seeds
Moisture	68.26 ± 0.07	66.61 ± 0.02	53.96 ± 0.03
Ash	0.86 ± 0.01	0.98 ± 0.01	1.81 ± 0.01
Lipids	1.75 ± 0.11	2.56 ± 0.01	17.35 ± 0.04
Proteins	7.33 ± 0.20	7.45 ± 0.01	21.56 ± 0.01
Fibers	5.55 ± 0.09	9.03 ± 0.18	35.00 ± 2.90
Total carbohydrates*	21.8 ± 0.27	22.4 ± 0.18	5.33 ± 0.31
Energetic value (kcal 100 g ⁻¹)	132.27 ± 0.40	142.44 ± 0.19	263.71 ± 0.79
Starch	1.18 ± 0.10	1.17 ± 0.30	1.23 ± 0.50
Pectin	28.84 ± 3.00	28.26 ± 0.70	
Soluble solids	16.8 ± 0.17	16.7 ± 0.00	
pH	6.13 ± 0.25	5.81 ± 0.12	5.93 ± 0.01
Titrate acidity**	2.60 ± 0.20	3.80 ± 0.07	3.70 ± 0.61
L*	34.02 ± 1.27	26.10 ± 3.21	
*Hue (h)	71.80 ± 1.10	82.61 ± 0.83	
Chroma (C*)	43.58 ± 3.66	17.02 ± 0.80	
Carbohydrates (mg g ⁻¹)			
Sucrose	nd	nd	nd
Fructose	17.11 ± 0.23	3.47 ± 0.17	0.97 ± 0.02
Glucose	nd	nd	nd
Xylose	4.27 ± 0.10	2.91 ± 0.04	1.32 ± 0.01
Cellobiose	nd	nd	nd
Organic Acids (mg g ⁻¹)			
Ascorbic acid	55.96 ± 1.14	51.03 ± 0.24	68.41 ± 0.92
Tartaric acid	21.09 ± 0.33	111.48 ± 0.37	117.55 ± 0.40
Malic acid	315.67 ± 0.71	160.12 ± 0.08	nd
Citric acid	24.76 ± 1.48	88.11 ± 1.31	nd
Phytochemicals			
Total phenolics (mg GAE 100 g ⁻¹)	340.30 ± 1.20	840.96 ± 2.74	1010.57 ± 0.95
Carotenoids (μg β-carotene 100 g ⁻¹)	12.44 ± 0.35	17.40 ± 0.19	2.65 ± 0.10
Total anthocyanins (mg 100 g ⁻¹)	2.97 ± 0.12	6.43 ± 0.39	6.53 ± 0.92
Total flavonoids (mg 100 g ⁻¹)	35.76 ± 0.05	128.50 ± 0.071	160.00 ± 0.41

^aValues expressed as mean ± standard deviation (*n* = 3); *carbohydrates calculated by difference; **values expressed in g citric acid 100 g⁻¹; nd: not detected.; GAE: gallic acid equivalent.

phosphate buffer (pH 6.9), and α-amylase inhibition was calculated based on the method described in eq 5.

$$\alpha\text{-Amylase inhibition} = \frac{A_c - A_a}{A_c} \times 100 \quad (5)$$

where *A_c* is the absorbance of the control sample, and *A_a* is the absorbance of the sample.

1.6. Simulation of In Vitro Gastrointestinal Digestion.

The pulp, peel, and seed fractions of the yellow puçá were subjected to in vitro digestion simulation to verify the stability of the polyphenols during digestion. This step will be carried out based on the simulated digestion protocol proposed by INFOGEST.^{11,12} The method is divided into three phases: oral, gastric, and intestinal. For the oral phase of digestion, 0.25 g of dried samples were combined with 3.5 mL of simulated salivary fluid, 25 μL of CaCl₂ solution, and 0.975 μL of distilled water. The pH of the resulting mixture was adjusted to 7.0 using a 0.1 M sodium hydroxide (NaOH) solution, then 0.5 mL of α-amylase (75 U mL⁻¹) was added. The samples were then incubated at 37 °C under constant stirring at 100 rpm for 2 min. For the gastric phase, 7.5 mL of simulated gastric fluid, 20 μL of CaCl₂ solution, and 0.700 μL of distilled water were added to the same reaction vessel used in the oral phase. The pH of the mixture was then adjusted to 3.0 with hydrochloric acid (HCl, 6 mol L⁻¹), and 1.6 mL of

pepsin (2000 U mL⁻¹) was added. The samples were subsequently incubated in a thermostatically controlled water bath at 37 °C for 2 h. For the intestinal phase, 11 mL of bile salts, and 1.46 mL of NaOH (1 mol L⁻¹) were introduced into the gastric phase reaction tubes. The pH was then neutralized to 7.0, adding 5 mL of pancreatin (800 U mL⁻¹). The digestion process continued for 2 h at 37 °C under continuous stirring to mimic physiological conditions. After each digestion phase, enzymatic activity was stopped by placing the samples in an ice bath maintained at approximately 4 °C. Control experiments were conducted using identical proportions of samples and reagents, excluding digestive enzymes and bile salts. After each digestion phase, the resulting liquid was analyzed for antioxidant activity and phenolic compound profile, with comparisons made with samples before in vitro digestion. The method aims to evaluate digestion products and micronutrient release. As responses to the different phases of simulated digestion in vitro, the phenolic profile, antioxidant analyses (DPPH, FRAP, CU-PRAC, CRT), and biological properties (inhibition of lipid peroxidation and inhibition of α-amylase) will be evaluated, as well as the calculation of bioaccessibility for the phenolic profile of the extracts.

1.7. Individual Phenolic Compounds. The phenolic composition of the pulp, peel, and seed extracts of yellow puçá was evaluated by high-performance liquid chromatography (HPLC) before and after each step of in vitro digestion (oral, gastric, and intestinal phases). Before injection, the extracts were filtered through a 0.22 μm membrane filter with a diameter of 13 mm. The identification and quantification of phenolic compounds were performed using an HPLC-DAD-UV-VIS system (AB Sciex LLC, Framingham, USA), equipped with a quaternary pump, degasser, injector, system controller, column oven, diode array detector, refractive index detector, and collector. The analysis was performed under the following conditions: column temperature maintained at 35°C, Shim-pack VP-ODS analytical column (250 mm $\times$ 4.6 mm $\times$ 5 μm), and Shim-pack GVP-ODS guard column (10 mm $\times$ 4.6 mm $\times$ 5 μm). The mobile phase comprised two components: phase A, a 2% (v/v) aqueous solution of glacial acetic acid, and phase B, a mixture of methanol, water, and acetic acid in a 70:28:2% (v/v) ratio. The system functioned with a flow rate of 1.0 mL min⁻¹, utilizing a gradient elution technique over a total runtime of 65 min. An injection volume of 20 μL was employed, and the phenolic compounds were detected at 280 nm. Calibration curves were obtained in duplicate, using ten different concentrations of each standard. The calibration curves were: trigonelline ($y = 49881x - 553.26$, $r^2 = 0.9986$), theobromine ($y = 29533x - 47.97$, $r^2 = 0.9989$), catechin ($y = 11371x - 567.84$, $r^2 = 0.9993$), vanillin ($y = 69247x + 254.33$, $r^2 = 0.9986$), and gallic acid ($y = 3994x + 8047$, $r^2 = 0.9962$), syringic acid ($y = 49401x + 706.65$, $r^2 = 0.9990$), chlorogenic acid ($y = 29451x - 2224.70$, $r^2 = 0.9992$), caffeic acid ($y = 67928x - 2499.20$, $r^2 = 0.9991$), *p*-coumaric ($y = 100602x - 4097.70$, $r^2 = 0.9989$), *o*-coumaric ($y = 116320x - 6657$, $r^2 = 0.9990$), *m*-coumaric acid ($y = 130558x - 2496.20$, $r^2 = 0.9996$), ferulic acid ($y = 52609x - 5197.10$, $r^2 = 0.9997$), rosmarinic acid ($y = 30075x - 5006.10$, $r^2 = 0.9984$), *trans*-cinnamic acid ($y = 177527x - 8753$, $r^2 = 0.9989$) and resveratrol ($y = 79962x - 11,037$, $r^2 = 0.9983$). The phenolic compounds were identified by comparing the sample peaks' retention times with the standards' retention times. The results were expressed in micrograms per gram ($\mu\text{g g}^{-1}$).

1.8. Bioaccessibility Index. The bioaccessibility of phenolic compounds released from the pulp, peels, and seeds of yellow puçá during simulated gastrointestinal digestion and made available for absorption into the systemic circulation was assessed using the bioaccessibility index (eq 6).³⁶

$$\text{Bioaccessibility index (\%)} = \frac{A}{B} \times 100 \quad (6)$$

where A = total phenolic content after in vitro digestion and B = total phenolic content before in vitro digestion.

1.9. Statistical Analyses. Results are presented as mean $\pm$ standard deviation. Data were organized and processed in Excel spreadsheets. Significant differences were detected for the simulation of in vitro gastrointestinal digestion using the Tukey test or *t*-test to compare means at a significance level of 5% ($p < 0.05$) using Statistica 10.0 software (StatSoft Inc., Tulsa, OK, USA) when necessary. OriginPro 2022® software (OriginLab Corporation, Northampton, MA, USA) was used to analyze and plot the graphs.

3. RESULTS AND DISCUSSION

3.1. Nutritional, Physicochemical, and Phytochemical Composition of the Pulp, Peels, and Seeds of the Yellow Puçá.

Table 1 shows the results of the nutritional and physicochemical characterization of the pulp, peels, and seeds of the yellow puçá. As expected, the pulp and peels presented the highest moisture contents, with 68.26 and 66.61 g 100 g⁻¹, respectively. High moisture content is directly related to the stability, quality, composition, and yield of the fruit. In this context, special care during harvesting, transportation, and processing of the yellow puçá is essential, as high moisture levels favor the development of deteriorating microorganisms, which can compromise the shelf life of the fruit.¹⁸ da Silva et al.³⁷ observed higher moisture values in the black puçá (*Mouriri pusa*) pulp and peels, with 75.95 and 78.45 g 100 g⁻¹, respectively. The observed differences can be attributed to factors such as harvest time, soil type, geographic variability, solar incidence, and variation among the analyzed species.

Regarding lipids, the results indicate low levels in the pulp and peel of the yellow puçá, with 1.75 and 2.56 g 100 g⁻¹, respectively, while the seeds presented a high content of 17.35 g 100 g⁻¹ (Table 1). This lipid distribution pattern is typical in fruits since, in most plants, lipids are predominantly stored in the endosperm of the seeds due to genetic and metabolic factors that regulate the storage of energy reserves. da Silva et al.³⁷ observed similar behavior for the black puçá, with low lipid levels in the pulp and peel and a high seed content. Regarding the protein content, it is essential to emphasize that, as defined by da Silva Sousa et al.,³⁸ for a food to be considered a good source of protein, it must contain at least 12 g 100 g⁻¹. Based on this criterion, the consumption of 100 g of pulp, peels, and seeds would represent 87.96%, 89.04%, and 100%, respectively, of the recommended daily protein intake for a healthy adult. These data highlight the nutritional potential of yellow puçá seeds, which, in addition to their high lipid content, also contribute significantly to the protein intake in the diet.

The results obtained for carbohydrates and energy values indicate differences in the content between the analyzed samples. The pulp and peels presented relatively similar carbohydrate contents, with 21.80 and 22.41 g 100 g⁻¹, respectively, while the seeds had relatively lower levels (5.33 g 100 g⁻¹). These carbohydrates are largely responsible for the energy value of the different fractions of the fruit. The pulp and peel have energy values of 132.27 and 142.44 kcal 100 g⁻¹, respectively, while the seeds have a higher energy value (263.71 kcal 100 g⁻¹), which can be attributed to the higher lipid content (17.35 g 100 g⁻¹) in the seeds, compared to the pulp and peel (1.75 and 2.56 g 100 g⁻¹, respectively). This higher lipid content in the seeds justifies their higher energy value since lipids provide a more concentrated amount of energy per gram compared to carbohydrates.³⁹ Therefore, the distribution of lipids and carbohydrates among the different components directly reflects the caloric and nutritional value of each part of the fruit.

The results obtained for the pectin content in the pulp and peel of yellow puçá (28.84 and 28.26 g 100 g, respectively) indicate that these fractions present concentrations compatible with those of fruits rich in pectin, suggesting that they can be used as substitutes for commercial pectin. However, no positive results were found for the seeds of yellow puçá. Similar results were described by da Silva et al.³⁷ when

investigating black puçá, where the authors also observed high pectin contents in the pulp and peel but not in the seed. These values are consistent with those found in other tropical fruits, such as mango,⁴⁰ and indicate that yellow puçá can be a promising source of pectin for food applications. The pH and titratable acidity results of the different fractions of yellow puçá (Table 1) indicate an expected correlation between these two variables. The pulp, with a pH of 6.13, presents the lowest titratable acidity (2.60 g citric acid 100 g⁻¹), consistent with its greater alkalinity. On the other hand, the peels and seeds, with lower pH (5.81 and 5.93, respectively), have higher titratable acidity values (3.80 and 3.70 g citric acid 100 g⁻¹, respectively). This pattern is commonly found in peels and seeds since there is an increase in the concentration of organic acids, such as citric and malic acid, which contribute to a more acidic pH and higher titratable acidity.⁴¹ These results on the distribution of acids in the different fractions of the fruit indicate that the pulp of the yellow puçá is more neutral, while the peels and seeds are more acidic, which may influence its use in food applications.

The results obtained for carbohydrates reveal a distinct distribution among the fruit fractions (Table 1). The pulp presented the highest concentrations of fructose and xylose (17.11 and 4.27 mg g⁻¹, respectively), which is in line with the carbohydrate profile observed in many native fruits such as bacupari, tucuma, pupunha and yellow puçá, where reducing sugars, such as fructose, predominate due to their rapid conversion into energy sources.^{37,38} The absence of sucrose in the three fractions may suggest a distinct metabolic pathway for forming sugars in the yellow puçá, emphasizing fructose. Since, in more acidic environments, sucrose undergoes a hydrolysis process, where its molecule breaks down and is partially divided into the monosaccharide fructose and glucose.⁴² Compared to other traditional fruits, such as apple⁴³ and mango,⁴⁴ which have more balanced carbohydrate profiles between fructose and glucose, the yellow puçá stands out for its predominance of fructose in the pulp and a moderate xylose content in the skins and seeds, which may reflect differences in the specific metabolic characteristics of the plant.

Regarding organic acids, the acidity of yellow puçá is distributed differently among the fruit fractions, with emphasis on the pulp, which presented the highest levels of malic, ascorbic, and citric acid (315.67, 55.96 and 24.76 mg g⁻¹, respectively) (Table 1). These compounds synergistically act as significant protectors on the myocardium act in ischemic lesions and, together with their salts, contribute to iron absorption.^{45,46} On the other hand, tartaric acid, found in higher concentrations in the peels and seeds (111.48 and 117.55 mg g⁻¹, respectively), is a compound commonly associated with fruits with an acidic flavor and a potent antioxidant. The higher concentration of tartaric acid in the peels and seeds, compared to the pulp (21.09 mg g⁻¹), may reflect a defense mechanism or a specific functional adaptation of these fractions about abiotic stresses.⁴⁷ These findings on the composition of organic acids have direct implications for the methods and processes to be applied to the different fruit fractions, influencing their sensory and functional properties.

The results for phenolic and carotenoid compounds in yellow puçá indicate a significant distribution of these phytochemicals in the different fractions of the fruit (Table 1). The peel and seeds showed high concentrations of phenolic compounds (840.96 and 1010.57 mg GAE 100 g⁻¹,

respectively). The peels and seeds of many native tropical fruits, such as buriti and açaí, also demonstrate high levels of phenolics, exerting protective and antioxidant action.^{48,49} The pulp, on the other hand, showed a more moderate concentration of phenolics (340.30 mg GAE 100 g⁻¹). da Silva et al.,³⁷ when studying black puçá, reported a relatively lower number of phenolic compounds in the pulp (288.08 mg GAE 100 g⁻¹) compared to the peels and seeds. Despite this, all fractions of the yellow puçá can be classified as good sources of phenolic compounds since, according to criteria established by Vasco et al.,⁵⁰ fruits with levels higher than 500 mg GAE 100 g⁻¹ are considered rich in this compound. Therefore, the presence of phenolic compounds is relevant, as they contribute significantly to foods' antioxidant activity and exert potential therapeutic effects.⁵¹

The distribution of carotenoids and anthocyanins in the different fractions of yellow puçá suggests a phytochemical profile with nutritional and functional relevance (Table 1). Carotenoids are more concentrated in the peel (17.40 µg 100 g⁻¹) compared to the pulp (12.44 µg 100 g⁻¹), while the seeds present very low levels (2.65 µg 100 g⁻¹). Carotenoids play crucial roles as precursors of vitamin A and antioxidants, contributing to eye health and physiological processes such as reducing blood glucose and reducing neurodegenerative diseases, in addition to favoring the body's immune action.^{52,53} In contrast, anthocyanins, responsible for the color of several fruits, are more abundant in the peels (6.43 mg 100 g⁻¹) and seeds (6.53 mg 100 g⁻¹). Anthocyanins, with their antioxidant, anti-inflammatory, and potentially anticancer properties, add functional value to the yellow puçá, making its external fractions (peels) an important source of bioactive pigments.⁵⁴

3.2. Mineral Profile. The mineral profile of the pulp, peels, and seeds of the yellow puçá presents significant differences in mineral concentrations (Table 2). The pulp presents

Table 2. Mineral Profile of the Pulp, Peel, and Seeds of Yellow Puçá (*Mouriri grandiflora*)^a

Minerals (g 100 g ⁻¹)	Pulp	Peel	Seeds
Nitrogen (g kg ⁻¹)	8.06 ± 0.20	9.63 ± 0.11	22.57 ± 0.28
Phosphorus (g kg ⁻¹)	0.54 ± 0.01	0.80 ± 0.05	1.44 ± 0.43
Potassium (g kg ⁻¹)	7.00 ± 0.33	9.00 ± 0.30	6.00 ± 0.71
Calcium (g kg ⁻¹)	0.80 ± 0.02	1.60 ± 0.48	0.70 ± 0.09
Magnesium (g kg ⁻¹)	0.90 ± 0.06	1.10 ± 0.05	1.00 ± 0.01
Sulfur (g kg ⁻¹)	0.15 ± 0.01	0.36 ± 0.01	0.96 ± 0.01
Boron (mg kg ⁻¹)	3.80 ± 0.19	5.03 ± 0.20	7.13 ± 0.29
Copper (mg kg ⁻¹)	0.28 ± 0.05	2.03 ± 0.74	1.41 ± 0.91
Iron (mg kg ⁻¹)	28.96 ± 0.20	34.31 ± 0.71	36.58 ± 0.57
Manganese (mg kg ⁻¹)	0.75 ± 0.05	1.37 ± 0.06	1.11 ± 0.09
Zinc (mg kg ⁻¹)	3.22 ± 0.03	4.76 ± 0.29	8.13 ± 0.58

^aValues expressed as mean ± standard deviation (n = 2).

satisfactory levels of nitrogen and potassium (8.06 and 7.00 g kg⁻¹, respectively), demonstrating that it is rich in minerals essential for cell growth and muscle function. The recommended dietary intake of potassium for a healthy individual of both sexes is 4.70 g day⁻¹.⁵⁵ Therefore, the consumption of 100 g of both fractions of the yellow puçá would be enough to meet the entire daily demand. Therefore, potassium is crucial for electrolyte balance and blood pressure control, contributing to the prevention of cardiovascular diseases. However, a diet deficient in potassium can cause leg cramps, slow reflexes, muscle weakness, and irregular heartbeat, among other

pathologies.¹ On the other hand, it was observed that boron (7.13–13.80 mg kg⁻¹) presented satisfactory concentrations among the different compounds. This mineral is fundamental to bone health and hormonal regulation and is a potent antioxidant.⁵⁶

On the other hand, the peels have higher concentrations of copper (2.03 mg kg⁻¹) and zinc (4.76 mg kg⁻¹) (Table 2), which make them a good option for bone health and strengthening the immune system.⁵⁷ In addition, they stand out for the significant amount of iron (34.31 mg kg⁻¹), an essential mineral for the formation of hemoglobin and the transport of oxygen in the blood, which may be particularly beneficial in preventing anemia.⁵⁸ The seeds of the yellow puçá, with high levels of iron (36.58 mg kg⁻¹) and zinc (8.13 mg kg⁻¹), have a highly nutritious mineral profile with notable benefits for immune function, wound healing, and maintenance of cellular integrity.⁵⁷ Zinc, in particular, is an essential cofactor for several antioxidant enzymes and plays a crucial role in cellular metabolism and the immune response.⁵⁹ Thus, each fraction of the yellow puçá contributes differently to promoting health, with concentrations of minerals that benefit everything from muscle function to strengthening the immune, bone, and neurological systems, and can be applied in various industrial sectors, such as cosmetics and pharmaceuticals.

3.3. Amino Acids. Analyzing essential and nonessential amino acids in different yellow puçá parts reveals significant nutritional value information (Table 3). Essential amino acids, which are indispensable for a series of metabolic functions and cannot be synthesized by the human body, are present in higher concentrations in the seeds, followed by the skin and pulp (3.09, 1.55, and 0.99 g 100 g⁻¹, respectively). Among

Table 3. Amino acids from the pulp, peel, and seeds of the yellow puçá (*Mouriri grandiflora*).

Amino acids (g 100 g ⁻¹)	Pulp	Peel	Seeds
Essentials			
Histidine	0.07 ± 0.01	0.11 ± 0.01	0.24 ± 0.03
Threonine	0.11 ± 0.02	0.17 ± 0.02	0.22 ± 0.05
Tyrosine	0.05 ± 0.01	0.10 ± 0.05	0.32 ± 0.02
Valina	0.16 ± 0.02	0.25 ± 0.00	0.40 ± 0.01
Methionine	0.01 ± 0.00	0.01 ± 0.00	0.06 ± 0.00
Isoleucine	0.12 ± 0.01	0.18 ± 0.07	0.30 ± 0.05
Leucine	0.21 ± 0.05	0.32 ± 0.06	0.73 ± 0.01
Phenylalanine	0.11 ± 0.02	0.18 ± 0.01	0.53 ± 0.05
Lysine	0.15 ± 0.02	0.23 ± 0.02	0.29 ± 0.06
Tryptophane	nd	<0.01	<0.01
Σ essentials	0.99 ± 0.05	1.55 ± 0.09	3.09 ± 0.28
Nonessential			
Glutamic acid	0.45 ± 0.05	0.53 ± 0.03	1.67 ± 0.01
Serina	0.26 ± 0.01	0.31 ± 0.01	0.32 ± 0.09
Glycine	0.16 ± 0.07	0.26 ± 0.01	0.72 ± 0.09
Taurine	<0.01	<0.01	<0.01
Arginine	0.18 ± 0.03	0.25 ± 0.02	1.84 ± 0.01
Alanine	0.24 ± 0.06	0.29 ± 0.10	0.36 ± 0.03
Proline	0.23 ± 0.01	0.30 ± 0.01	0.39 ± 0.05
Cystine	0.06 ± 0.00	0.05 ± 0.01	0.20 ± 0.02
Hydroxyproline	0.02 ± 0.00	0.03 ± 0.00	0.04 ± 0.01
Σ No essentials	1.60 ± 0.09	2.02 ± 0.11	5.54 ± 0.52
Σ Total amino acids	2.59 ± 0.08	3.57 ± 0.20	8.63 ± 0.74

Values expressed as mean ± standard deviation ($n = 2$); nd.: not detected; < 0.01 = Below the limit of quantification (LOQ < 0.01).

these amino acids, leucine and valine stand out in the seeds, with 0.73 and 0.40 g 100 g⁻¹, respectively. This same behavior can be observed by da Silva et al.³⁷ when studying the seed of the yellow puçá (*M. pusa*), where the authors reported higher concentrations of essential amino acids in the seed (3.45 g 100 g⁻¹), with leucine and valine as the major amino acids (0.82 and 0.44 g 100 g⁻¹, respectively). Leucine, in addition to being crucial for synthesizing muscle proteins, plays a central role in the process of muscle recovery and growth, contributes to controlling blood glucose levels, and helps regulate energy metabolism.⁶⁰ Valine, which also participates in protein synthesis, is essential for maintaining the body's nitrogen balance and immune system function.^{61,62} Both substances are indispensable, as they cannot be produced by the body and are required through food, especially in high-quality protein sources, such as yellow puçá seeds.

Regarding nonessential amino acids, which are synthesized by the body, the data indicate a similar pattern. The concentrations of nonessential amino acids in the peel and pulp follow a pattern similar to that observed for essential amino acids, with the seeds presenting the highest results (5.54 g 100 g⁻¹), followed by the peel and pulp (2.02 and 1.60 g 100 g⁻¹, respectively) (Table 3). The differentiation between the parts of the plant reinforces the importance of the seeds as a primary source of amino acids, both essential and nonessential, for the human diet. The main nonessential amino acids found in the seeds of the yellow puçá were glutamic acid, arginine, and glycine (1.67, 1.84, and 0.72 g 100 g⁻¹, respectively). Thus, raw materials that present satisfactory concentrations of these amino acids act in metabolism and in maintaining health, in addition to being essential in the transmission of nerve signals to the brain, being one of the main excitatory neurotransmitters, in addition to acting in the cellular energy production cycle and the metabolism of ammonium, aiding in the detoxification of the organism, and acting in the production of nitric oxide, a compound that plays an essential role in the dilation of blood vessels and increasing circulation, contributing to cardiovascular health.^{60,63,64} Together, these amino acids not only perform essential metabolic functions but are also closely related to the proper functioning of the nervous system, cardiovascular health, and the energy balance of the organism. Thus, the amino acid profile of the different fractions of yellow puçá indicates its potential for nutritional and food applications, highlighting its importance in a balanced diet.

3.4. Fatty Acids. Table 4 shows that all oils analyzed presented higher concentrations of saturated fatty acids (93.34–98.35%) than monounsaturated (1.25–6.38%) and polyunsaturated (0.12–0.40%) fatty acids. These results indicate that the oils obtained from yellow puçá are rich in saturated lipids, comparable to other oilseeds native to Brazil, such as coconut oil (37.01–63.60%).⁶⁵ Saturated fatty acids are widely used due to their stability and oxidation resistance, making these lipids ideal for formulating food products with greater shelf life, such as margarine and frying oils.⁶⁶ The main saturated fatty acids present in the yellow puçá oils obtained from the pulp, peels, and seeds were palmitic (54.22–76.40%) and myristic (8.06–13.71%) acids (Table 4). The majority presence of these fatty acids gives the oil stability, increasing its resistance to oxidation and degradation at high temperatures, in addition to having antimicrobial and anti-inflammatory properties.⁶⁷ However, the inclusion of oils rich in saturated fats should be evaluated with caution since uncontrolled consumption can negatively impact health, increase LDL

Table 4. Fatty Acid Profile of the Pulp, Peels, and Seeds of the Yellow Puçá (*Mouriri grandiflora*)

Fatty acids (%)	Pulp	Peel	seeds
Butyric (C4:0)	13.24 ± 0.85	nd	nd
Caproic (C6:0)	0.43 ± 0.03	0.63 ± 0.05	0.04 ± 0.00
Caprylic (C8:0)	nd	4.31 ± 0.32	nd
Capric (C10:0)	nd	0.57 ± 0.01	nd
Lauric (C12:0)	1.03 ± 0.02	1.92 ± 0.21	0.05 ± 0.01
Myristic (C14:0)	13.13 ± 1.87	13.71 ± 0.84	8.06 ± 0.21
Pentadecanoic (C15:0)	nd	9.18 ± 0.57	12.58 ± 1.32
Palmitic (C16:0)	55.10 ± 0.83	54.22 ± 1.58	76.40 ± 1.41
Heptadecanoic (C17:0)	10.24 ± 0.14	12.24 ± 0.92	nd
cis-10-heptadecanoic (C17:1)	nd	0.95 ± 0.06	nd
Stearic (C18:0)	0.17 ± 0.03	1.19 ± 0.12	0.25 ± 0.12
Oleic (C18:1)	6.38 ± 0.20	0.30 ± 0.06	2.48 ± 0.20
Linoleic (C18:2)	0.08 ± 0.01	0.37 ± 0.02	0.12 ± 0.05
Linolenic (C18:3)	0.20 ± 0.05	0.03 ± 0.01	nd
Arachidic (C20:0)	nd	0.22 ± 0.05	0.02 ± 0.00
Heneicosanoic (C21:0)	nd	0.16 ± 0.07	nd
Σ Saturated (SFA)	93.34 ± 0.47	98.35 ± 0.28	97.40 ± 0.81
Σ Monounsaturated (MUFA)	6.38 ± 0.20	1.25 ± 0.12	2.48 ± 0.33
Σ Polyunsaturated (PUFA)	0.28 ± 0.07	0.40 ± 0.05	0.12 ± 0.02

^aValues expressed as mean ± standard deviation (*n* = 2); nd: not detected.

cholesterol levels in the blood, and trigger inflammatory processes in the body, contributing to the development of chronic conditions, such as hypertension and type 2 diabetes.⁶⁸ In addition to the nutritional implications, the fatty acids identified in the different oils from the parts of the yellow puçá can be applied in technological and industrial processes. Butyric acid, present in the pulp (13.24%), is particularly relevant for the cosmetics and pharmaceutical industries due to its antimicrobial and anti-inflammatory properties.⁶⁹ Medium-chain fatty acids, such as caprylic (4.31%) and pentadecanoic (9.18%), present in the peel, have applications in the production of biodiesel and the food industry due to their efficiency in emulsification and potential antibacterial effect.⁷⁰ Additionally, the high levels of oleic acid present in the pulp and seeds (6.38 and 2.48%, respectively) and stearic acid in the peels (1.19%) can be used for the formulation of healthy food products, such as vegetable oils and margarine, which benefit from the stability of these fatty acids at high temperatures.⁶⁸ The lipid profile observed in yellow puçá, with a combination of saturated and unsaturated fatty acids, presents great potential for developing new ingredients in several industries, taking advantage of these compounds' nutritional and technological characteristics.

3.5. Triacylglycerol Composition. The triacylglycerol (TAG) compositions of the oil samples are presented in Table 5, considering only fatty acids >0.5% of the total peak area. The main triacylglycerols in the pulp oil were BuPP, PMP, PPP, and HPP, representing approximately 54% of the triacylglycerol composition. On the other hand, the main TAGs found in the oil obtained from the peels were PMP, PPp, PPP, and PHP, representing approximately 55% of the total triacylglycerols. The seeds, however, presented the TAGs MPp, MPP, PePP, and PPP as major compounds, the sum of these four composing approximately 87% of the TAGS of the seed oil of the yellow puçá. The composition of the oils of the

different fractions of the yellow puçá was predominantly composed of diunsaturated TAGs (PMP and HPP), followed by triunsaturated (PPP). These results corroborate the fatty acid composition shown in Table 4, demonstrating that it is rich in saturated fatty acids such as palmitic, myristic, and heptadecanoic acid. This same behavior can be observed by Alves Morais et al.¹⁸ when studying the TAGS profile in the different fractions of the buritirana fruit (*Mauritiella armata*), and found that the oils were composed of diunsaturated TAGs (POO and PLO), followed by monounsaturated (POP). Consequently, the predominant triacylglycerol (TAG) identified across all oil samples was PPP, with concentrations of 18.12%, 18.58%, and 45.53% in oils derived from the pulp, peels, and seeds, respectively. These findings highlight a clear correlation between the variations in the TAG composition of the oils and the distinct fatty acid profiles of each sample, underscoring how the lipid composition in different plant fractions influences the overall lipid structure and properties of the oils.

3.6. In Vitro Gastrointestinal Digestion and Bioaccessibility of Individual Phenolic Compounds

The phenolic profile of the pulp, peels, and seeds of yellow puçá before and after the in vitro digestion process is presented in Tables 6, 7 and 8, respectively. It is crucial to understand the behavior of the compounds throughout the digestive process, as their bioaccessibility is essential for potential health benefits, requiring concentrations available for intestinal absorption after digestion.⁷¹ Statistical analysis indicated significant differences between all individual phenolics, evidencing considerable variations throughout the stages of the digestive process (*p* < 0.05). Among the compounds identified in the extracts before in vitro digestion, gallic acid was the major compound for the pulp, peels, and seeds (652.01, 173.94, and 319.03 $\mu\text{g g}^{-1}$, respectively). These results are highly satisfactory since fruits rich in gallic acids, such as yellow puçá, offer several health benefits, including antioxidant, anti-inflammatory, and antimicrobial properties, in addition to contributing to cellular protection against oxidative damage, strengthening the immune system and preventing cardiovascular diseases.^{72–74} The analysis of the phenolic profile of the pulp revealed the presence of 12 individual phenolic compounds, of which only eight were bioaccessible at the end of the digestion process. The peels presented 12 compounds before digestion; only six remained bioaccessible in the intestinal phase. On the other hand, the seeds presented a more balanced behavior, with eight phenolic compounds quantified before digestion and 7 maintaining their bioaccessibility after the completion of the reactions (Tables 6–8). In all samples, a significant trend (*p* < 0.05) of reduction in phenolic compounds was observed throughout the in vitro gastrointestinal simulation. This pattern is consistent with previous studies that highlight the influence of enzymes and pH during digestion, resulting in phenolic compounds' degradation, modification, and transformation.⁷⁵ It is worth noting that the concentrations of phenolic compounds presented in Tables 6–8 correspond to values normalized to the volume of the initial sample, considering the dilutions caused by the addition of oral, gastric, and intestinal fluids during the simulation. This normalization ensures an accurate bioaccessibility comparison between the different digestive process stages.⁷¹ When evaluating the bioaccessibility of the compounds, it is noted that catechin was the phenolic that presented the

Table 5. Triacylglycerol Composition of Oils from Pulp, Peel, and Seed Oils of Puca (*Mouriri grandiflora*)^a

Triacylglycerol (%)			Triacylglycerol (%)		
ECN	Acronym	Pulp	Peel	Seeds	
22:0	BuMBu	0.74 ± 0.02			
24:0	BuPBu	3.12 ± 0.24			
25:0	BuHBu	0.57 ± 0.06			
32:0	BuMM	0.74 ± 0.09			
34:0	BuMP	6.20 ± 0.52			
35:0	BuMH	1.15 ± 0.02			
36:0	BuPP	13.02 ± 1.30			
36:1	BuOM	0.72 ± 0.02			
37:0	BuPH	4.85 ± 0.09			
38:0	CapPP		0.62 ± 0.01		
38:0	CMP		2.24 ± 0.72		
38:1	BuOP	3.02 ± 0.28			
39:0	CPeP		1.50 ± 0.10		
39:1	BuOH	0.56 ± 0.07			
40:0	CPP		4.42 ± 0.86		
41:0	CHP		2.00 ± 0.33		
42:0	CaPP		0.61 ± 0.01		
42:0	PLaM		0.99 ± 0.04		
43:0	PLaPe		0.66 ± 0.03		
43:0	PeMM		0.60 ± 0.01		
44:0	LaPP	0.98 ± 0.09			
44:0	PMM	3.07 ± 0.32	3.56 ± 0.44		
44:0	PLaP		1.95 ± 0.46		
44:0	MPM			1.54 ± 0.01	
45:0	HMM	0.57 ± 0.08			
45:0	PMPe		4.78 ± 0.05		
45:0	PHLa		0.88 ± 0.02		
45:0	MHM		0.80 ± 0.06		
45:0	MPPe			4.78 ± 0.11	
46:0	PMP	12.93 ± 1.02	14.09 ± 0.63		
46:0	PPePe		1.60 ± 0.05		
46:0	PeHM		1.08 ± 0.05		
46:0	MPP			14.49 ± 0.59	
46:0	PePPe			3.71 ± 0.20	
47:0	HMP	4.78 ± 0.62			
47:0	PPeP		9.46 ± 0.96		
47:0	PHM		6.34 ± 0.57		
47:0	PePP			22.52 ± 1.51	
48:0	PPP	18.12 ± 0.94	18.58 ± 1.63	45.53 ± 1.20	
48:0	SMP		0.62 ± 0.06		
48:0	PHPe		4.25 ± 0.22		
48:0	MHH		0.71 ± 0.05		
48:1	POM	3.00 ± 0.54			
48:1	MOP				
49:0	HPP	10.07 ± 0.36			
49:0	PHP		12.55 ± 0.92		
49:1	HOM	0.55 ± 0.07			
49:1	PHepP		1.02 ± 0.09		
49:1	PeOP			1.47 ± 0.11	
50:0	HPH	1.85 ± 0.04			
50:0	SPP		1.23 ± 0.02	0.53 ± 0.02	
50:0	PHH		2.82 ± 0.09		
50:1	POP	6.30 ± 0.55		4.47 ± 0.33	
51:1	HOP	2.33 ± 0.41			
52:2	POO	0.73 ± 0.03			

^aResults are presented as mean ± standard deviation ($n = 2$); ECN, equivalent carbon number and double bonds; Bu, butyric acid; M, myristic acid; P, palmitic acid; H, heptadecenoic acid; O, oleic acid; La, lauric acid; S, stearic acid; Pe, pentadecanoic acid; Hep, *cis*-10-heptadecanoic acid; Ca, capric acid; C, caprylic acid; Bu, butyric acid; Cap, caproic acid.

Table 6. Bioaccessibility and Simulation of In Vitro Gastrointestinal Digestion of Yellow Puca Pulp^a

Individual phenolics ($\mu\text{g g}^{-1}$)	RT	Nondigested ethanol 70%	Oral phase	Gastric phase	Intestinal phase	Bioaccessibility (%)
Trigonelline	3.49	1.42 ± 0.02 ^b	0.93 ± 0.01 ^c	1.96 ± 0.09 ^a	0.25 ± 0.02 ^d	17.60
Gallic acid	6.65	652.01 ± 0.96 ^a	60.65 ± 0.20 ^d	280.71 ± 0.87 ^b	200.64 ± 0.02 ^c	30.77
Theobromine	9.30	0.34 ± 0.10 ^b	0.32 ± 0.08 ^b	0.68 ± 0.05 ^a	0.24 ± 0.01 ^b	70.58
Catechin	10.49	0.80 ± 0.14 ^b	0.33 ± 0.05 ^c	1.92 ± 0.10 ^a	0.73 ± 0.06 ^b	91.25
Chlorogenic acid	12.19	0.62 ± 0.09 ^a	nd	nd	0.30 ± 0.01 ^b	48.38
Caffeic acid	14.38	1.19 ± 0.05 ^b	0.89 ± 0.05 ^c	4.89 ± 0.09 ^a	0.76 ± 0.07 ^c	63.86
Syringic acid	15.39	nd	nd	nd	nd	
Vanillin	16.80	nd	nd	nd	nd	
<i>p</i> -coumaric acid	20.65	1.52 ± 0.03 ^b	0.37 ± 0.09 ^c	4.24 ± 0.67 ^a	0.10 ± 0.01 ^d	6.57
Ferulic acid	23.57	0.24 ± 0.05	nd	nd	nd	
<i>m</i> -coumaric acid	25.95	0.31 ± 0.05 ^b	0.17 ± 0.07 ^c	2.17 ± 0.11 ^a	nd	
<i>o</i> -coumaric acid	32.35	1.14 ± 0.01	nd	nd	nd	
Resveratrol	36.73	nd	nd	nd	nd	
Rosmarinic acid	42.83	13.30 ± 0.28	nd	nd	nd	
<i>trans</i> -cinnamic acid	50.43	3.32 ± 0.14 ^b	1.04 ± 0.04 ^c	11.16 ± 0.29 ^a	0.77 ± 0.05 ^d	23.19

^aResults are presented as mean ± standard deviation ($n = 2$); means followed by the same lowercase letter in the same row do not differ statistically using the Tukey or *t*-test at 5% probability ($p \leq 0.05$); nd: not detected; RT: retention time in minutes.

greatest bioaccessibility for pulp and seed (91.25 and 96.86%, respectively). On the other hand, gallic acid was the most bioaccessible compound in the yellow puca peel (93.11%). These values reflect high bioaccessibility, suggesting that these compounds' absorption and utilization in the gastrointestinal tract are maximized. This behavior can be attributed to the

interaction of phenolic compounds with food components, which can facilitate their release and absorption in the intestine. Recent studies indicate that gallic acid and catechin are widely found in various foods, and their availability for absorption by the body can be facilitated due to the formation of complexes with other substances present in the food.^{36,76–78}

Table 7. Bioaccessibility and Simulation of In Vitro Gastrointestinal Digestion of Yellow Puçá Peel^a

Individual phenolics ($\mu\text{g g}^{-1}$)	RT	Nondigested ethanol 70%	Oral phase	Gastric phase	Intestinal phase	Bioaccessibility (%)
Trigonelline	3.49	3.34 $\pm$ 0.20 ^a	1.33 $\pm$ 0.11 ^b	0.22 $\pm$ 0.09 ^d	0.99 $\pm$ 0.05 ^c	29.64
Gallic acid	6.65	173.94 $\pm$ 0.58 ^b	107.25 $\pm$ 0.85 ^d	228.12 $\pm$ 0.63 ^a	161.97 $\pm$ 0.74 ^c	93.11
Theobromine	9.30	nd	0.70 $\pm$ 0.01 ^b	9.23 $\pm$ 0.22 ^a	0.55 $\pm$ 0.06 ^c	
Catechin	10.49	3.42 $\pm$ 0.37 ^a	0.87 $\pm$ 0.20 ^b	nd	nd	
Chlorogenic acid	12.19	0.46 $\pm$ 0.06 ^b	0.44 $\pm$ 0.01 ^b	1.68 $\pm$ 0.10 ^a	nd	
Caffeic acid	14.38	8.66 $\pm$ 0.39 ^a	0.93 $\pm$ 0.08 ^b	0.32 $\pm$ 0.01 ^c	0.18 $\pm$ 0.02 ^c	2.07
Syringic acid	15.39	1.61 $\pm$ 0.05 ^a	0.07 $\pm$ 0.02 ^b	nd	nd	
Vanillin	16.80	nd	0.09 $\pm$ 0.01 ^b	0.34 $\pm$ 0.02 ^c	nd	
<i>p</i> -coumaric acid	20.65	0.14 $\pm$ 0.07 ^b	1.74 $\pm$ 0.01 ^a	0.05 $\pm$ 0.03 ^c	0.09 $\pm$ 0.01 ^c	64.28
Ferulic acid	23.57	nd	0.35 $\pm$ 0.02 ^a	0.25 $\pm$ 0.01 ^b	nd	
<i>m</i> -coumaric acid	25.95	3.69 $\pm$ 0.06 ^a	0.16 $\pm$ 0.04 ^b	0.11 $\pm$ 0.04 ^b	nd	
<i>o</i> -coumaric acid	32.35	0.12 $\pm$ 0.00 ^b	0.28 $\pm$ 0.05 ^a	0.24 $\pm$ 0.06 ^a	nd	
Resveratrol	36.73	1.45 $\pm$ 0.05 ^a	0.43 $\pm$ 0.02 ^c	1.08 $\pm$ 0.10 ^b	0.19 $\pm$ 0.01 ^d	13.10
Rosmarinic acid	42.83	2.30 $\pm$ 0.58	nd	nd	nd	
<i>trans</i> -cinnamic acid	50.43	0.34 $\pm$ 0.01 ^b	0.66 $\pm$ 0.06 ^a	0.15 $\pm$ 0.02 ^c	0.10 $\pm$ 0.01 ^c	29.41

^aResults are presented as mean $\pm$ standard deviation ($n = 2$); means followed by the same lowercase letter in the same row do not differ statistically using the Tukey or *t*-test at 5% probability ($p \leq 0.05$); nd: not detected; RT: retention time in minutes.

Table 8. Bioaccessibility and Simulation of In Vitro Gastrointestinal Digestion of Yellow Puçá Seeds^a

Individual phenolics ($\mu\text{g g}^{-1}$)	RT	Nondigested ethanol 70%	Oral phase	Gastric phase	Intestinal phase	Bioaccessibility (%)
Trigonelline	3.49	nd	0.05 $\pm$ 0.01 ^c	0.27 $\pm$ 0.02 ^b	0.38 $\pm$ 0.02 ^a	
Gallic acid	6.65	319.03 $\pm$ 0.40 ^b	nd	84.58 $\pm$ 0.56 ^c	80.30 $\pm$ 0.48 ^b	25.17
Theobromine	9.30	4.65 $\pm$ 0.09 ^b	0.10 $\pm$ 0.01 ^d	17.11 $\pm$ 0.28 ^a	1.58 $\pm$ 0.63 ^c	33.97
Catechin	10.49	4.15 $\pm$ 0.20 ^a	nd	0.74 $\pm$ 0.06 ^b	4.02 $\pm$ 0.85 ^a	96.86
Chlorogenic acid	12.19	1.24 $\pm$ 0.05 ^b	0.35 $\pm$ 0.01 ^d	1.77 $\pm$ 0.02 ^a	0.76 $\pm$ 0.02 ^c	61.30
Caffeic acid	14.38	0.62 $\pm$ 0.01 ^a	0.10 $\pm$ 0.01 ^c	0.10 $\pm$ 0.01 ^c	0.46 $\pm$ 0.00 ^b	74.20
Syringic acid	15.39	nd	nd	0.01 $\pm$ 0.00 ^a	0.03 $\pm$ 0.00 ^a	
Vanillin	16.80	nd	nd	nd	0.06 $\pm$ 0.00	
<i>p</i> -coumaric acid	20.65	0.61 $\pm$ 0.02 ^a	0.03 $\pm$ 0.02 ^c	nd	0.14 $\pm$ 0.01 ^b	22.95
Ferulic acid	23.57	nd	0.14 $\pm$ 0.02 ^a	0.17 $\pm$ 0.03 ^a	0.19 $\pm$ 0.02 ^a	
<i>m</i> -coumaric acid	25.95	0.43 $\pm$ 0.04 ^a	nd	0.05 $\pm$ 0.00 ^b	0.06 $\pm$ 0.00 ^b	13.95
<i>o</i> -coumaric acid	32.35	nd	nd	0.19 $\pm$ 0.03 ^a	0.23 $\pm$ 0.02 ^a	
Resveratrol	36.73	nd	nd	nd	nd	
Rosmarinic acid	42.83	nd	nd	nd	nd	
<i>trans</i> -cinnamic acid	50.43	0.08 $\pm$ 0.01 ^b	nd	0.15 $\pm$ 0.01 ^a	nd	

^aResults are presented as mean $\pm$ standard deviation ($n = 2$); means followed by the same lowercase letter in the same row do not differ statistically using the Tukey or *t*-test at 5% probability ($p \leq 0.05$); nd: not detected; RT: retention time in minutes.

This same behavior was observed by Morais et al.³⁶ when investigating the profile of phenolic compounds present in the pulp, peels and seeds of buritirana (*M. armata*). They identified significant variations between the respective phases of the in vitro gastrointestinal digestion simulation process and identified significant variations in the bioavailability and stability of phenolic compounds throughout the different digestive phases (oral, gastric and intestinal). These results show that each phase of the digestive process affects the different food matrices in a distinct and specific manner, reflecting both the release and the transformation of the bioactive compounds present. Such variations can be attributed to the physicochemical characteristics of the matrices—such as the composition of dietary fiber, the lipid and protein content, and the interaction with other constituents of the plant matrix—which modulate the solubilization and release of phenolic compounds.¹¹ In addition, the food matrix plays a crucial role in the protection and/or degradation of phenolic compounds during digestion. For example, flavonoids and phenolic acids can undergo structural transformations, such as

hydrolysis or conjugation, which affect their antioxidant activity and intestinal absorption.^{73–76}

It can be concluded that the bioaccessibility of phenolic compounds in the pulp, peels, and seeds of yellow puçá varies significantly during the simulated in vitro gastrointestinal digestion process. While the pulp showed remarkable bioaccessibility for catechin (91.25%) and a considerable presence of gallic acid (30.77%) in the intestinal phase, the peels demonstrated the highest bioaccessibility for gallic acid (93.11%) among all compounds. The seeds exhibited remarkable bioaccessibility of catechin (96.86%), suggesting that this compound is highly available for absorption, while gallic acid had a more limited bioaccessibility (25.17%). The results obtained indicate that the bioavailability of phenolic compounds present in yellow puçá is significantly modulated by intrinsic characteristics of its chemical structure, as well as by intermolecular interactions with other constituents of the food matrix during the different phases of the digestive process.^{11,12,36} The substantial variation observed in the bioaccessibility of these compounds throughout simulated in vitro gastrointestinal digestion can be attributed to the

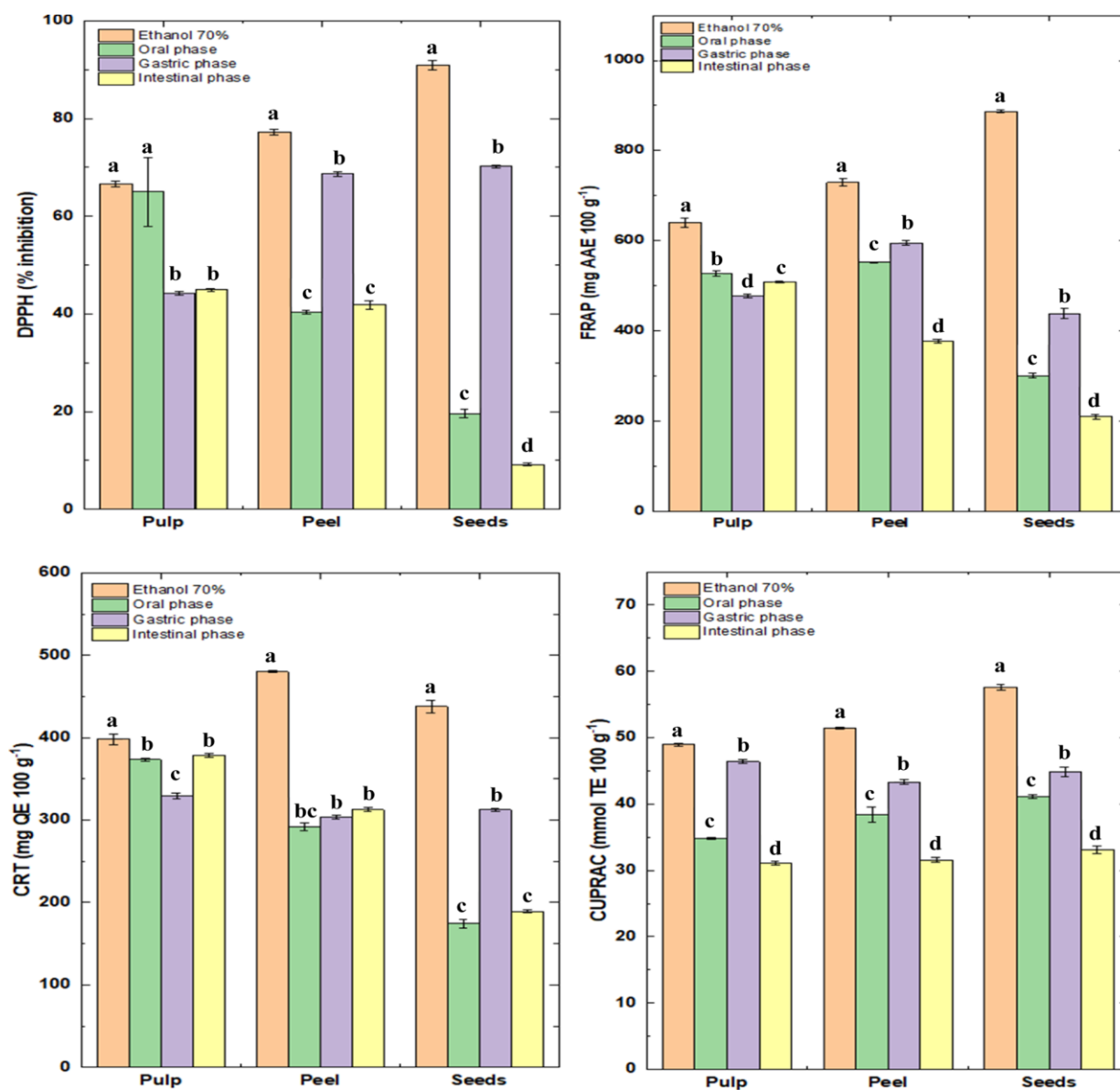


Figure 1. Antioxidant potential of the pulp, peels, and seeds of yellow puçá before, during, and after simulation of in vitro gastrointestinal digestion.

chemical stability of phenolics under different physiological conditions (pH, enzymatic activity, presence of bile salts), as well as to their ability to form complexes with macronutrients. Phenolic compounds can undergo degradation, isomerization, or enzymatic conjugation reactions during gastric and intestinal digestion, directly affecting their solubility and subsequent absorption.^{10–12}

Another relevant factor is the potential for enzymatic metabolism by digestive enzymes, such as esterases and glycosidases, which can modify the structure of phenolics, transforming them into metabolites of lower or higher polarity, which, in turn, influences their passive diffusion or active transport through intestinal cells.⁷⁵ Therefore, understanding the mechanisms that regulate the release and transformation of phenolic compounds during digestion requires an integrated

approach that considers both structural aspects of phenolics and the dynamic parameters of the food matrix. Overall, the presented data provide valuable insights into the digestive fate and bioaccessibility of phenolic compounds in yellow puçá, supporting its potential as a functional food ingredient.

3.7. Antioxidant Potential. The antioxidant potential of the pulp, peels, and seeds of yellow puçá was evaluated before and after the simulation of in vitro gastrointestinal digestion using the DPPH, FRAP, CUPRAC, and TRC assays, as shown in Figure 1. In general, a substantial reduction in the antioxidant activity of the different components of yellow puçá was observed during the digestive process, compared to the undigested samples. For the DPPH assay, the peels and seeds showed a significant decrease in antioxidant capacity ($p < 0.05$), with reductions of 39% (from 68.70 to 41.83%) and

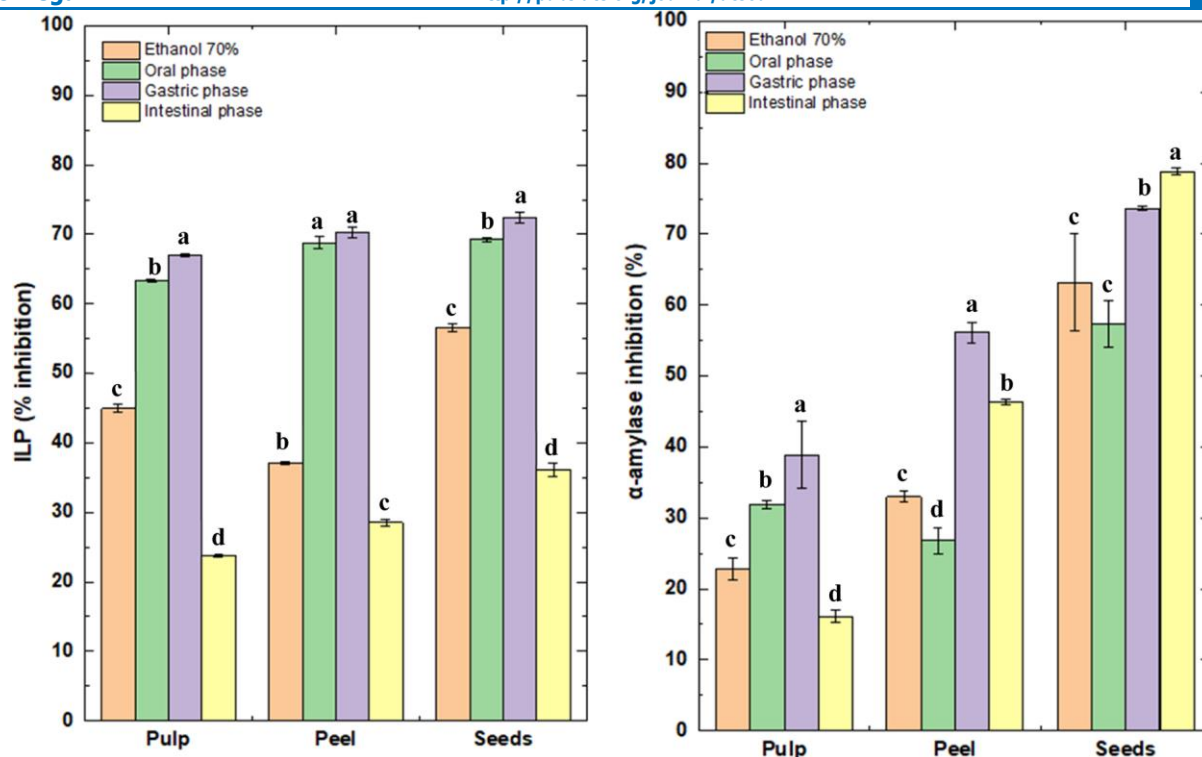


Figure 2. Biological properties of yellow puca pulp, peels, and seeds before, during, and after simulation of in vitro gastrointestinal digestion.

87% (from 70.21 to 9.13%), respectively, between the gastric and intestinal phases. The pulp, in turn, did not present a statistically significant difference between the respective phases. This phenomenon is related to the fact that free radicals, unstable molecules, can induce damage to various cellular structures, including lipids, proteins, and DNA, facilitating the process of premature aging and the development of multiple pathologies.⁷⁵ Therefore, antioxidant compounds that neutralize these reactive species play a crucial role in maintaining redox balance, preventing the emergence of chronic oxidative stress. However, for these compounds to exert their beneficial effects, it is essential to ensure their stability and activity during the in vitro digestive process to ensure their adequate intestinal absorption and subsequent transport to the target tissues.⁷⁹

The CRT and FRAP analyses presented in Figure 2 show a significant reduction in antioxidant activity throughout the simulated gastrointestinal digestion phases, particularly in the intestinal phase, for all yellow puca fractions (pulp, peel, and seed). While the ethanolic extracts presented the highest values for both CRT (397.90–479.90 mg QE 100 g⁻¹) and FRAP (640.96–887.12 mg AAE 100 g⁻¹), reflecting the high extraction of phenolic compounds under ideal solvent conditions. However, a significant decrease in these values was observed in the oral and gastric phases, and even more pronounced in the intestinal phase, suggesting degradation or structural transformation of the bioactive compounds due to changes in pH and enzymatic action. Notably, the husk showed greater relative stability in CRT between the gastric and intestinal phases (from 303.65 to 312.53 mg QE 100 g⁻¹), while the seed showed greater loss of antioxidant potential by the FRAP protocol (from 887.12 undigested extract to 209.87 mg AAE 100 g⁻¹ intestinal phase), indicating specific sensitivity of its phenolic matrix. These results reinforce the

importance of considering the real bioaccessibility of antioxidants, and not only their total levels, when evaluating the functional potential of foods.

The CUPRAC method showed the best results among the other antioxidant assays evaluated and showed the smallest reductions between the undigested sample and the intestinal phase ($p < 0.05$), with reductions of 36.50% (from 48.92 to 31.06 mmol TE 100 g⁻¹), 38.61% (from 51.43 to 31.57 mmol TE 100 g⁻¹) and 42.56% (from 57.60 to 33.08 mmol TE 100 g⁻¹), for pulp, peels and seeds, respectively. These reductions may be related to pH changes in the in vitro digestion simulation phases.⁸⁰ This suggests that even after the entire gastrointestinal digestion process, the antioxidant action responsible for the beneficial health effects of consuming these fruits, which are rich in antioxidants, may still be maintained.⁸¹ The data also indicate that most of the digested fractions presented favorable results, corroborating the results presented in the phenolic profile.

The decrease in antioxidant potential at the end of the simulated digestion process may suggest that the compounds are unstable against changes in pH and enzymatic activity. Antioxidant compounds demonstrate greater reactivity in acidic pH conditions, typical of gastric digestion, and less reactivity at pH close to neutrality, as occurs in intestinal digestion.⁸² In this context, during the digestive process, the biotransformation of the original compounds into others with lower antioxidant capacity may occur, or even the formation of new metabolites with preserved or potentially more effective antioxidant properties.^{83,84} However, it is possible to observe that most of the fractions, after digestion, presented satisfactory results in terms of antioxidant capacity, indicating that the antioxidant metabolites maintained their protective potential. This protective potential thus contributes to reducing

inflammation and chronic diseases, such as cardiovascular diseases, diabetes, and neurodegenerative diseases.

3.8. In Vitro Biological Properties. The in vitro biological properties of the pulp, peels, and seeds of yellow puçá are shown in Figure 2. The highest inhibition percentages were recorded in the samples from the gastric phase, both for lipid peroxidation inhibition (ILP) and for α -amylase inhibition, in all fruit fractions. In the ILP analysis, a significant increase was observed between the undigested samples and those from the gastric phase ($p < 0.05$). The pulp showed an increase of 45.74% (from 44.95% to 66.97% inhibition), the peels showed an increase of 89.37% (from 37.09% to 70.24%), and the seeds showed an increase of 15.78% (from 56.56% to 72.34%). Despite the promising results observed in the gastric phase, the last stage of the process, corresponding to the intestinal phase, showed significant reductions in the lipid peroxidation inhibition values ($p < 0.05$), with levels lower than those of the undigested samples, indicating substantial variations between the different matrices and the phases of the in vitro digestive process. This decrease in the inhibition potential throughout the digestive phases is corroborated by studies by Chen et al.,⁸⁵ which suggest that digestion can negatively impact the antioxidant potential, mainly due to the reduction in the content of phenolic compounds, which can be transformed into altered structural forms with distinct chemical properties. Additional factors, such as pH variations and particle size during the digestive process, can also contribute to the decrease in the inhibition potential observed in the different phases of digestion.

Figure 2 shows the results of α -amylase enzyme inhibition before, during, and after the simulated in vitro gastrointestinal digestion process. Notably, the gastric phase showed the highest inhibition levels (38.83 and 56.40% inhibition) for pulp and peels and decreased significantly in the intestinal phase with reductions of 58.48% (from 38.83 to 16.12%) and peels with reductions of 17.47% (from 56.10 to 46.30%) ($p < 0.05$). These results are by Burgos-Edwards et al.,⁸⁶ where the authors reported that the α -amylase inhibitory activities of two wild Chilean gooseberries (*Ribes magellanicum* and *Ribes punctatum*) decreased after gastric digestion. These differences may be due to the different compositions of the phytochemical components and their affinity with the respective phases. On the other hand, the highest percentage of seed inhibition is related to the intestinal phase (78.84%), with an increase of 7.08% between the gastric and intestinal phases, corroborating the findings of He et al.⁸⁷ when analyzing 22 juices from native and traditional fruits, noticed that after the gastric phase, some juices presented a higher percentage of inhibition compared to the other stages of the in vitro digestion process. These results suggest that the digestive process modulates the inhibitory activity of α -amylase differently, depending on the digestion phase and the phytochemical composition of the fractions analyzed.

4. CONCLUSIONS

The findings of this study highlight the nutritional and functional potential of the different fractions of yellow puçá. Peels and seeds were rich in lipids, proteins, and dietary fibers, while the pulp exhibited high levels of pectin, fructose, and malic acid—compounds of interest for food and health-related applications. The oils extracted showed a predominance of palmitic acid, indicating a saturated profile with potential thermal stability, making them suitable for industrial uses,

particularly in frying processes. All fractions demonstrated satisfactory levels of essential minerals, amino acids, and triacylglycerols. Gallic acid was the main phenolic compound identified in both fractions, reinforcing their value for pharmaceutical and cosmetic applications due to its well-documented antioxidant and bioactive properties. However, in vitro gastrointestinal digestion led to a marked reduction in phenolic content, antioxidant activity, and related biofunctions, especially during the intestinal phase. These results suggest that digestive conditions can significantly affect the stability and bioaccessibility of phenolic compounds, pointing to the occurrence of degradation or transformation mechanisms that warrant further investigation. Overall, this study underscores the complexity of the interactions between food bioactives and the human digestive system and emphasizes the importance of deepening our understanding of these processes to better harness the health benefits and industrial applications of underutilized native fruits such as yellow puçá.

ASSOCIATED CONTENT

Data Availability Statement

The datasets generated during and analyzed during the current study are available from the corresponding author upon reasonable request.

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THIRD PART

2 FINAL CONSIDERATIONS

This thesis contributes in an unprecedented way to the expansion of scientific knowledge about the yellow puçá (*Mouriri grandiflora*), a native fruit of the Amazon and Cerrado biomes, by performing its systemic characterization and investigating, in an integrated way, the impacts of in vitro simulated gastrointestinal digestion on the chemical, biological and antioxidant properties of its polyphenols.

The results obtained show that the puçá-amarelo has a relevant nutritional and phytochemical composition, with emphasis on the expressive presence of phenolic compounds distributed among pulp, peel and seeds. The detailed characterization of these fractions demonstrated that, in addition to the portion traditionally consumed, the fruit co-products are promising sources of bioactive compounds, reinforcing their potential for full use and technological valorization.

The application of the standardized protocol of simulated gastrointestinal digestion in vitro allowed a more realistic understanding of the behavior of polyphenols in relation to the physiological conditions of the digestive tract. It was observed that the digestive process promotes important structural transformations, resulting both in the partial degradation of certain compounds and in the release and increase of the bioaccessibility of other metabolites. These findings show that the simple quantification of phenolic compounds in the in natura matrix is not enough to predict its real functional potential, and it is essential to consider the digestive stage as a modulator of bioactivity.

Overall, the results indicate that puçá-amarelo has significant potential as a source of bioaccessible antioxidant compounds, which broadens its application perspectives in the development of functional foods, nutraceutical ingredients and innovative formulations. In addition, the use of husks and seeds reinforces the principles of the circular economy and the bioeconomy, adding value to fractions that are often neglected and contributing to the reduction of agro-industrial waste.

From a scientific perspective, this thesis fills relevant gaps in the literature by providing unprecedented data on the chemical composition, the profile of individual phenolics and the bioaccessibility of these compounds after simulated digestion. Such information can support future research involving in vivo assays, metabolomics studies, evaluation of specific biological effects and development of technological products based on native Brazilian fruits.

From a socio-environmental point of view, the appreciation of the yellow puçá reinforces the importance of Brazilian biodiversity as a strategic source of scientific innovation and

sustainable development. The integration between chemical characterization, bioaccessibility assessment and technological potential demonstrates that the rational and sustainable exploitation of native fruits can generate positive impacts on health promotion, the strengthening of local production chains and the conservation of biomes.

Finally, it is concluded that the yellow pudge stands out as a promising food matrix, whose functional potential is significantly influenced by the transformations that occur during digestion. The approach adopted in this research highlights the importance of integrated studies that consider not only the initial composition of foods, but also their behavior throughout the digestive process, consolidating advances in the field of Food Science and contributing to the appreciation of the Brazilian fruit richness

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