UNIVERSIDADE FEDERAL DE VIÇOSA

Avaliação de polifenóis em cardápios escolares e formulação de farinhas de co-produtos de umbu-cajá e graviola

Valéria Silva de Lana
Magister Scientiae

VIÇOSA - MINAS GERAIS
2025

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Dissertação apresentada à Universidade Federal de Viçosa, como parte das exigências do Programa de Pós-Graduação em Ciência da Nutrição, para obtenção do título de *Magister Scientiae*.

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VIÇOSA - MINAS GERAIS
2025

Ficha catalográfica elaborada pela Biblioteca Central da Universidade Federal de Viçosa - Campus Viçosa

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L243a
2025

Lana, Valéria Silva de, 1999-
Avaliação de polifenóis em cardápios escolares e formulação de farinhas de coprodutos de umbu-cajá e graviola / Valéria Silva de Lana. – Viçosa, MG, 2025.
1 dissertação eletrônica (103 f.): il. (algumas color.).

Texto em português e inglês.
Orientador: Izabela Maria Montezano de Carvalho.
Dissertação (mestrado) - Universidade Federal de Viçosa, Departamento de Nutrição e Saúde, 2025.
Inclui bibliografia.
DOI: https://doi.org/10.47328/ufvbbt.2025.204
Modo de acesso: World Wide Web.

1. Umbu-cajá. 2. Graviola. 3. Polifenóis. 4. Cascas - Reaproveitamento. 5. Farinhas como alimento. 6. Merenda escolar. I. Carvalho, Izabela Maria Montezano de, 1983-. II. Universidade Federal de Viçosa. Departamento de Nutrição e Saúde. Programa de Pós-Graduação em Ciência da Nutrição. III. Título.

CDD 22. ed. 641.344

Bibliotecário(a) responsável: Euzébio Luiz Pinto CRB-6/3317

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APROVADA: 24 de fevereiro de 2025.

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AGRADECIMENTOS

A Deus pela presença em todos os momentos da minha vida e por me guiar e cuidar nas dificuldades;
Aos meus pais, por batalharem sem medir esforços para proporcionar apoio, conforto, segurança e amor;
Às minhas irmãs por confiarem no meu potencial e estarem sempre presentes mesmo à distância;
Aos meus amigos, pelo apoio, paciência, compreensão e convivência diária;
À minha orientadora Profa. Izabela Maria Montezano de Carvalho, pelos conselhos, amizade e confiança depositadas em mim. Por ser fonte de inspiração e coragem todos os dias. Agradeço pelos ensinamentos e por garantir que concluísse uma etapa tão importante sob sua orientação;
À minha coorientadora Profa. Hércia Stampini Duarte Martino, por ser fonte de inspiração e admiração todos os dias, pela orientação e ideias brilhantes e pela confiança no meu trabalho;
Ao Laboratório de Análise de Alimentos, onde passei boa parte do meu tempo, agradecimento especial à equipe do laboratório pela convivência, pelos cafés da tarde e pelos biscoitinhos. Vocês tornaram os desafios mais tranquilos de serem enfrentados;
A todos que contribuíram para o desenvolvimento deste estudo;
Aos membros da banca, pela disponibilidade para contribuir com a conclusão dessa etapa;
À Universidade Federal de Viçosa, ao Departamento de Nutrição e Saúde e ao PPGCN pelo ensino de excelência e por todo suporte, por me guiarem por um caminho novo e cheio de expectativas;
À Fundação Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES), pela concessão da bolsa de estudos;
E a todos que, durante algum momento desse caminho, se fizeram presentes para a concretização de um sonho, muito obrigada!

Este trabalho foi apoiado pelo “Conselho Nacional de Desenvolvimento Científico e Tecnológico - CNPq” (CNPq/MCTI/FNDCT N° 18/2021, bolsa 422847/2021-6).

O presente trabalho foi realizado com apoio da Coordenação de Aperfeiçoamento de Pessoal de Nível Superior – Brasil (CAPES) – Código de Financiamento 001.

"[...] *Não havíamos marcado hora, não havíamos marcado lugar. E, na infinita possibilidade de lugares, na infinita possibilidade de tempos, nossos tempos e nossos lugares coincidiram. E deu-se o encontro* [...]." (Rubens Alves)

RESUMO

LANA, Valéria Silva de, M.Sc., Universidade Federal de Viçosa, fevereiro de 2025. Avaliação de polifenóis em cardápios escolares e formulação de farinhas de co-produtos de umbu-cajá e graviola
. Orientadora: Izabela Maria Montezano de Carvalho. Coorientadora: Hercia Stampini Duarte Martino.

Os compostos fenólicos possuem reconhecida atividade antioxidante e benefícios à saúde, incluindo a prevenção de doenças crônicas. A análise quantitativa desses compostos é limitada por custos e falta de padronização. Ferramentas como o banco de dados Phenol-Explorer ajudam a integrar informações sobre sua presença em alimentos o que pode ser útil para o planejamento de cardápios saudáveis, como os cardápios escolares que preconizam não só a utilização de alimentos in natura e minimamente processados como também alimentos regionais. No Brasil, alimentos regionais como graviola e umbu-cajá são ricos em compostos bioativos e são muito utilizados na indústria para produção de polpas, mas seus coprodutos, como cascas e sementes, são subutilizados, apesar de conterem compostos bioativos. A reutilização desses materiais promove a sustentabilidade, alinhando-se aos Objetivos de Desenvolvimento Sustentável da ONU, e pode reduzir desperdícios, estimulando a economia local e a sustentabilidade. Este trabalho teve como objetivo analisar cardápios escolares de Sergipe, avaliando nutrientes e compostos fenólicos, além de caracterizar coprodutos de frutas regionais como forma de identificar potencial para o seu aproveitamento. Como resultados foram produzidos dois artigos: O artigo 1 examinou cardápios de 319 escolas de 75 municípios do estado de Sergipe, analisando o conteúdo calórico, nutrientes e polifenóis. Essas variáveis foram agrupadas por similaridade e submetidas à análise de agrupamento por meio da distância euclidiana e do método de Ward. A regressão linear destacou que a presença de alimentos e ingredientes culinários regionais influenciou positivamente a concentração de flavonoides e ácidos fenólicos nos cardápios. Alimentos in natura e minimamente processados foram positivamente associados aos flavonoides e ácidos fenólicos. Já os alimentos ultraprocessados apresentaram associações negativas aos flavonoides. O artigo 2 avaliou as características nutricionais e tecnológicas por meio da caracterização centesimal e perfil antioxidante das farinhas de coprodutos de umbu-cajá e graviola, além da citotoxicidade em células saudáveis. Os resultados demonstraram que as farinhas são ricas em fibra alimentar, pobres em proteínas e possuem teor mínimo de umidade recomendado para

classificação como farinhas (<15%). A composição mineral das farinhas apresentou notável presença de cobre, ferro, zinco, manganês e boro. A avaliação da capacidade antioxidante demonstrou a presença de antioxidantes resistentes ao processamento, indicados por alta capacidade antioxidante. O perfil fenólico constatou a presença de rutina e ácido p-cumárico nas farinhas elaboradas. O teste de citotoxicidade demonstrou que ambas as farinhas não exerceram efeitos prejudiciais em células saudáveis de fibroblastos de acordo com o ensaio metil-tiazolil-tetrazólio (MTT). Neste sentido, a inclusão de polifenóis na alimentação escolar requer estratégias sustentáveis, como o uso de alimentos regionais e o aproveitamento de coprodutos. A ausência de frutas regionais nas refeições destaca a necessidade de melhorias nos cardápios. Frutas como a graviola e o umbu-cajá podem ser usados na forma de polpa para sucos, enquanto casca e bagaço podem ser secos e moídos em farinha, um ingrediente minimamente processado. Essa abordagem enriquece os cardápios, promove a sustentabilidade e incentiva o aproveitamento integral dos alimentos. Estudos sobre biodisponibilidade de minerais e efeitos das fibras na microbiota intestinal são fundamentais para validar seus benefícios.

Palavras-chave: Compostos fenólicos; Phenol-Explorer; Frutas regionais; Subprodutos

ABSTRACT

LANA, Valéria Silva de, M.Sc., Universidade Federal de Viçosa, February, 2025. **Evaluation of polyphenols in school menus and formulation of flours from umbu-caja and soursop co-products**. Adviser: Izabela Maria Montezano de Carvalho. Co-adviser: Hercia Stampini Duarte Martino.

Phenolic compounds have recognized antioxidant activity and health benefits, including the prevention of chronic diseases. Quantitative analysis of these compounds is limited by cost and the absence of standardization. Tools such as the Phenol-Explorer database help to integrate information on their presence in foods, which can be useful for planning healthy menus, such as school menus that recommend not only the use of fresh and minimally processed foods, but also regional foods. In Brazil, regional foods such as soursop and umbu-caja are rich in bioactive compounds and are widely used in the industry for pulp production, but their co-products, such as peels and seeds, are underutilized, despite containing bioactive compounds. Reusing these materials promotes sustainability, in line with the UN's Sustainable Development Goals, and can reduce waste, stimulating the local economy and sustainability. The aim of this study was to analyze school menus in Sergipe, assessing nutrients and phenolic compounds, as well as characterizing regional fruit co-products in order to identify potential for their use. Two articles were produced as a result: Article 1 examined menus from 319 schools in 75 municipalities in the state of Sergipe, analyzing calorie content, nutrients and polyphenols. These variables were grouped by similarity and subjected to cluster analysis using the Euclidean distance and Ward's method. The linear regression showed that the presence of regional foods and culinary ingredients positively influenced the concentration of flavonoids and phenolic acids in the menus. Fresh and minimally processed foods were positively associated with flavonoids and phenolic acids. Ultra-processed foods, on the other hand, showed negative associations with flavonoids. Article 2 evaluated the nutritional and technological characteristics by means of the proximal characterization and antioxidant profile of umbu-cajá and soursop coproduct flours, as well as their cytotoxicity in healthy cells. The results showed that the flours are rich in dietary fiber, low in protein and have the minimum moisture content recommended for classification as flours (<15%). The mineral composition of the flours showed a notable presence of copper, iron, zinc, manganese and boron. The evaluation of the antioxidant capacity showed the presence of antioxidants resistant to processing, indicated by high antioxidant capacity. The phenolic profile showed the presence of rutin

and p-coumaric acid in the flours produced. The cytotoxicity test showed that both flours had no harmful effects on healthy fibroblast cells according to the methyl-thiazolyl-tetrazolium (MTT) assay. In this sense, the inclusion of polyphenols in school meals requires sustainable strategies, such as the use of regional foods and the utilization of co-products. The absence of regional fruits in meals highlights the need to improve menus. Fruits such as soursop and umbu-cajá can be used in the form of pulp for juices, while peel and pomace can be dried and ground into flour, a minimally processed ingredient. This approach enriches menus, promotes sustainability and encourages the full use of food. Studies on the bioavailability of minerals and the effects of fiber on the intestinal microbiota are essential to validate its benefits.

Keywords: Phenolic compounds; Phenol-Explorer; Regional fruits; By-products

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LISTA DE ABREVIATURAS E SIGLAS

AA	Antioxidant activity
ABTS	2,2′-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid)-diammonium salt
AOAC	Association of Official Analytical Chemists
BC	Bioactive compounds
CF	Compostos fenólicos
CI	Culinary Ingredients
DPPH	2,2-diphenyl-1-picrylhydrazyl
FAO	Food and Agriculture Organization
FLV	Flavonoids
FMPF	Fresh and minimally processed foods
FRAP	Ferric reducing antioxidant power
HPLC	High Performance Liquid Chromatography
ICP-OES	Inductively Coupled Plasma Atomic Emission Spectroscopy
IN	In natura and minimally processed foods
JECFA	Joint Expert Committee on Food Additives
MTT	Methyl-Thiazolyl-Tetrazolium
n.d.	Not detected
n.i	Non identified
OD	Optical density
OHC	Oil holding capacity
OP	Other polyphenols
PC	Phenolic compounds
PCI	Processed Culinary ingredients
PF	Processed foods
PNAE	Programa Nacional de Alimentação Escolar
PT	Total polyphenols
RDA	Recommended dietary allowance
Rt	Retention time
SCF	Soursop co-product flour
SEDUC	Secretaria de Estado da Educação e da Cultura de Sergipe
SS	Soluble solids
TEAC	Trolox equivalent antioxidant capacity

TF	Total flavonoids
TP	Total phenolic
TPS	Technical preparation sheet
TPTZ	2,4,6-Tris(2-piridil)-s-triazina
TRF	Total regional foods
UCF	Umbu-caja co-product flour
UP	Ultra-processed foods
WHC	Water holding capacity

SUMÁRIO

1 INTRODUÇÃO GERAL

Os compostos fenólicos (CF) são produtos do metabolismo secundário das plantas e amplamente encontrados em alimentos de origem vegetal. Tais compostos bioativos são reconhecidos por sua alta atividade antioxidante e seus potenciais benefícios à saúde. Além disso, os CF desempenham um papel importante na prevenção de doenças crônicas não transmissíveis (DCNT), como as cardiovasculares, o diabetes tipo 2 e alguns tipos de câncer (El-Saadony et al., 2024; Zhang et al., 2022). O padrão alimentar caracterizado pelo consumo de alimentos industrializados e pobre em alimentos *in natura* constitui um dos principais fatores que contribuem para o aumento da incidência de tais doenças. Diante disso, a associação entre uma alimentação baseada em alimentos *in natura* e minimamente processados com a redução do risco para as DCNT, tem impulsionado estudos que explorem a presença de agentes protetores da saúde nesse contexto, como é o caso dos CF (Ahlawat et al., 2024; Cheynier et al., 2012).

A análise laboratorial de CF em alimentos ainda é um processo restrito e de difícil acesso no meio acadêmico. Isso se deve a fatores como o custo de análises, a falta de padronização de metodologias e à grande variedade de compostos existentes. Para superar tal limitação, a análise de dados secundários tem sido empregada como uma ferramenta útil, sobretudo quando o objetivo é avaliar uma grande variedade de alimentos (Aguilera; Martín-Cabrejas; González de Mejía, 2016; El-Saadony et al., 2024; Liu, 2013). O Phenol-Explorer constitui uma dessas ferramentas, disponibilizando uma base de dados que abrange 452 alimentos, nos quais é possível obter informações acerca do conteúdo de fenólicos totais e do detalhamento de classes específicas, como antocianinas, lignanas, flavonoides e suas subclasses. Dessa maneira, bancos de dados podem contribuir para a realização de pesquisas que objetivam analisar polifenóis em um grande volume de alimentos, como nos estudos de consumo alimentar em populações e nas análises de cardápios para coletividades, como é o caso dos cardápios escolares (Neveu et al., 2010; Rothwell et al., 2016).

As atuais recomendações para os cardápios do Programa Nacional de Alimentação Escolar (PNAE) podem favorecer a oferta de CF à população atendida, uma vez que prioriza a aquisição de alimentos *in natura* e minimamente processados, que constituem as principais fontes de CF da alimentação. Considerando que o grau de processamento dos alimentos impacta diretamente no seu conteúdo de CF, é fundamental compreender em que medida isso ocorre para aprimorar a seleção dos alimentos que irão compor as preparações dos cardápios, incluindo os escolares no âmbito do PNAE. Outra recomendação do PNAE, que pode contribuir para o

aumento da oferta de CF na alimentação, é o estímulo à compra de alimentos regionais, especialmente frutas e hortaliças. A implementação de alimentos regionais no planejamento de cardápios escolares, além de valorizar a cultura alimentar local, promove a diversificação da dieta e o fortalecimento da agricultura familiar (Brasil, 2024; Brasil, 2020). Alimentos típicos de diferentes regiões brasileiras como a seriguela, o pequi e o umbu, são ricos em CF e apresentam elevado potencial funcional (Brasil, 2015). Entretanto, muitos desses alimentos ainda são subutilizados, especialmente no que tange ao aproveitamento integral, como cascas, sementes e bagaços, que muitas vezes são descartados durante o processamento (Banerjee et al., 2017; Munekata et al., 2023).

O Brasil é um país muito extenso e possui uma grande variedade de árvores frutíferas consideradas regionais, muitas das quais ainda são pouco exploradas (Camacam; Messias, 2022), como o umbu-cajá (*Spondias bahiensis*) e a graviola (*Annona muricata*), que são espécies tropicais típicas do semiárido brasileiro (Carvalho; Conte-Junior, 2021). Em seus ciclos naturais, os frutos são colhidos por meio de sistemas extrativistas, sem a aplicação de quaisquer insumos ou práticas agrícolas. Ambas são árvores consideradas resistentes às condições climáticas e os frutos possuem uma composição nutricional rica e elevado conteúdo de compostos bioativos (Freitas et al., 2013; Souza et al., 2020).

Os frutos do umbu-cajá são possuem quantidades notáveis de vitamina C, retinol, carotenoides e vitamina B6, além de outros compostos bioativos como os flavonoides. A graviola é rica em carboidratos, vitaminas do complexo B e minerais como potássio, magnésio, fósforo e cálcio (Santos et al., 2010; Brasil, 2011). Durante a despolpa dessas frutas, o material fibroso (cascas e sementes) é separado do restante da fruta. Esse processo gera produtos secundários com concentrada quantidade de fibras alimentares, proteínas, minerais e compostos bioativos presentes nas cascas e sementes, com potencial para reaproveitamento, tornando-se um coproduto de valor agregado (Lira Junior et al., 2005; Ribeiro et al., 2022). Pesquisas recentes demonstraram que os coprodutos têm uma quantidade semelhante ou até maior de compostos bioativos em comparação com as partes de frutas consumidas convencionalmente (Lebaka et al., 2021; Marcillo-Parra et al., 2021; Ramos et al., 2023).

O processamento de frutas resulta em um material com potencial nutricional, mas que normalmente não tem um destino específico (Reguengo et al., 2022). Nesse sentido, o aproveitamento de produtos derivados do processamento agroindustrial é uma prática justificável por uma série de razões fundamentais, que vão desde a redução de desperdício até o estímulo à economia e à sustentabilidade ambiental. Essas estratégias de aproveitamento possuem respaldo pela Organização das Nações Unidas (ONU), em que, na Agenda 2030,

contempla no seu Objetivo de Desenvolvimento Sustentável (ODS) número 12, a proposta de garantir padrões sustentáveis de produção e consumo, com o objetivo de reduzir as perdas e desperdícios de alimentos ao longo das cadeias de produção e abastecimento até 2030, visando também o aproveitamento de recursos que originalmente seriam descartados (Organização das Nações Unidas; 2015; Leão et al., 2017; Tiwari et al., 2022). Muitos desses produtos não utilizados têm se mostrado viáveis para inclusão na dieta humana, especialmente como matérias-primas para formulações de alimentos ou recuperação de compostos bioativos. Embora muitas espécies de frutas nativas façam parte da diversidade alimentar das comunidades locais, há poucos estudos disponíveis sobre a importância nutricional dos produtos gerados a partir da despolpa de frutas (Silva, 2020; Batista et al., 2023).

Diante do exposto, este trabalho contempla dois estudos: o primeiro, se propõe a analisar a oferta de nutrientes e CF em cardápios da alimentação escolar da rede pública estadual de ensino em Sergipe. Trata-se de uma abordagem *in silico*, realizando a modelagem estatística e ajuste dos modelos entre o conteúdo de nutrientes e compostos fenólicos em cardápios escolares e o grau de processamento de alimentos, assim como a ocorrência de alimentos regionais dos escolares na rede pública estadual de Sergipe. Para o segundo estudo, dois materiais foram escolhidos para conduzir uma análise de caracterização. Originalmente, era proposto realizar uma análise de compostos fenólicos em frutos regionais presentes no cardápio. Entretanto, ao longo do estudo, constatou-se que não havia presença destes alimentos nos cardápios e, assim, optou-se por escolher duas frutas da safra à época (umbu-cajá e graviola) e analisar os seus coprodutos.

2 OBJETIVO GERAL

Analisar a oferta de polifenóis em cardápios planejados para a alimentação escolar e avaliar produtos do processamento de umbu-cajá (*Spondias bahiensis*) e graviola (*Annona muricata*) como potencial estratégia para aumento do consumo de polifenóis.

2.1 OBJETIVOS ESPECÍFICOS

- Avaliar a previsão da oferta de nutrientes e o perfil de polifenóis dos cardápios escolares da rede estadual de ensino de Sergipe.
- Caracterizar a composição nutricional, perfil fenólico, atividade antioxidante e propriedades tecnológicas, bem como avaliar a toxicidade de produtos secundários ao processamento dos frutos regionais umbu-cajá e graviola.

3 REVISÃO BIBLIOGRÁFICA

3.1 Os compostos fenólicos e sua importância

Compostos fenólicos são metabólitos secundários encontrados em uma variedade de plantas, como frutas e vegetais. Os metabólitos secundários são sintetizados a partir dos primários (carboidratos, lipídios e aminoácidos) pela via do chiquimato/fenilpropanóide onde ocorre transporte do carbono da glicólise e da via pentose fosfato para sintetizar uma ampla gama de compostos monoméricos e poliméricos. Com base nas suas vias de síntese, os compostos secundários são classificados em três grupos principais: compostos fenólicos (exemplo: ácidos fenólicos, flavonoides, ligninas), terpenos (exemplo: carotenoides e esteróis) e compostos contendo nitrogênio (como alcaloides) (Ahlawat et al., 2024; Cheynier et al., 2012).

Os compostos fenólicos possuem uma estrutura química comum que compreende um anel aromático com um ou mais substitutos hidroxila que podem ser divididos em várias classes. Os principais compostos fenólicos incluem: flavonoides, ácidos fenólicos, taninos, estilbenos e lignanas. Até o momento, mais de 8.000 estruturas fenólicas foram identificadas, que incluem mais de 4.000 flavonoides (Ahlawat et al., 2024; El-Saadony et al., 2024; Zhang et al., 2022). Nas plantas, esses compostos fornecem funções essenciais na reprodução e crescimento, como mecanismo de defesa contra estresses bióticos e abióticos e também contribuem para cor, sabor e aroma (Liu, 2013).

Além de suas funções nas plantas, os compostos fenólicos em nossa dieta podem fornecer benefícios adicionais à saúde, como a redução do risco de desenvolvimento de doenças crônicas. Os polifenóis não apenas protegem a célula e os componentes celulares de danos oxidativos, mas também reduzem o risco de estresse oxidativo associado a diferentes doenças como as degenerativas (Mutha; Tatiya; Surana, 2021), hipercolesterolemia, hiperglicemia, hiperlipidemia e insurgência de câncer (Abbas et al., 2017). Uma dieta equilibrada associando o consumo de frutas, hortaliças, chá e café é naturalmente rica em polifenóis além de fibras, vitaminas e minerais, componentes importantes em todas as fases da vida (Aguilera; Martín-Cabrejas; González de Mejía, 2016; El-Saadony et al., 2024; Liu, 2013).

Embora ainda não exista uma recomendação para consumo de polifenóis, é essencial ter informações detalhadas e abrangentes sobre a natureza e as quantidades de polifenóis encontrados nos principais alimentos consumidos em nossa dieta. Entretanto, essas informações são ainda pouco acessíveis. Um dos motivos é a grande diversidade de estruturas e de métodos analíticos usados para a quantificação de compostos fenólicos, isso resulta na dispersão dos dados em incontáveis fontes na literatura. Além disso, existe uma falta de padronização dos

métodos analíticos. Por fim, o conteúdo de polifenóis em um determinado alimento pode variar muito de acordo com a região de cultivo, variedade, condições agrícolas e climáticas, além de condições de armazenamento e processamento (Neveu et al., 2010; Rothwell et al., 2016).

Para contornar um pouco dessa falta de acessibilidade de informações, estimar a quantificação de compostos fenólicos por meio de banco de dados é uma alternativa interessante. Nesse sentido, tem-se explorado o banco de dados Phenol-Explorer, que contém dados para todos os polifenóis conhecidos em alimentos. Dados de composição foram coletados de publicações científicas, avaliados e usados para calcular valores de conteúdo médio representativos para 502 polifenóis em 452 alimentos (Neveu et al., 2010; Rothwell et al., 2016). Embora haja a limitação que esse não seja um banco brasileiro, e, portanto, muitos alimentos regionais permanecem de fora dessa análise, há muitos alimentos que compõem a base de alimentação servindo como um apoio para melhorar a oferta de compostos bioativos para coletividades. Esse banco de dados é um instrumento fundamental para, por exemplo, melhorar a construção de cardápios escolares, como os elaborados através do PNAE, por meio da estimação indireta do conteúdo de compostos fenólicos nos principais alimentos consumidos por uma comunidade.

3.2 O que é o Programa Nacional de Alimentação Escolar (PNAE)

Promover o consumo de alimentos vegetais ricos em compostos fenólicos desde a infância implica a necessidade de melhorar os sistemas de fornecimento e distribuição de frutas e vegetais *in natura* ou minimamente processados, garantir sua segurança, qualidade e acesso a populações mais vulneráveis. Nesse contexto, insere-se o Programa Nacional de Alimentação Escolar (PNAE), instituído pela Lei Federal nº 11.947 que regulamenta o fornecimento de alimentação escolar. É uma das políticas públicas mais longevas no Brasil na área de segurança alimentar e nutricional e é considerado um dos poucos programas do mundo a ser universal e gratuito, considerado pela Food and Agriculture Organization (FAO) um dos melhores do mundo. Surgiu na década de 40 com o intuito de combater no país os altos índices de desnutrição e doenças associadas a comorbidades alimentares pelos estudantes (Bicalho et al., 2021; Nero et al., 2022) e hoje tem como objetivo garantir ao menos uma refeição completa e saudável por dia para crianças em idade escolar (Silva et al., 2024) garantindo, dessa forma, a Segurança Alimentar e Nutricional (SAN).

O PNAE abrange todas as escolas públicas e comunitárias do sistema de educação básica, desde creche, jardim de infância, ensino fundamental, ensino médio até a educação para jovens adultos, seja o ensino em turno parcial ou integral (Sidaner et al., 2013). Além disso,

desde 2009, é obrigatório que 30 % do orçamento de alimentação do PNAE seja destinado à compra de alimentos diretamente da agricultura familiar, proporcionando uma alimentação mais diversificada e regionalizada aos estudantes, fortalecendo a renda familiar e contribuindo para a economia local. Além de incentivar a produção de alimentos culturalmente apropriados pelas populações locais, por meio da agricultura familiar de caráter agroecológico (Brasil, 2024).

Antes da instituição de uma legislação mais robusta, a merenda escolar era composta em sua grande parte por alimentos enlatados, embutidos, óleo, leite em pó, arroz, macarrão, biscoitos, açúcar, sal e outros alimentos industrializados (Affonso et al., 2024). Em 2020, foi publicada a resolução FNDE n° 06 que apresentou alterações significativas quanto aos aspectos nutricionais dos cardápios incorporando recomendações alinhadas ao Guia Alimentar para População Brasileira (Brasil, 2014) que adota o sistema de classificação NOVA (Monteiro et al., 2019). Esse sistema categoriza os alimentos de acordo com a extensão e a finalidade do processamento industrial, recomendando o uso de uma ampla variedade de alimentos *in natura* ou minimamente processados em detrimento de alimentos ultraprocessados (Azevedo et al., 2023).

O consumo de diferentes tipos e grandes variedades de alimentos não processados ou minimamente processados é aconselhável para melhorar a ingestão de nutrientes e a qualidade da dieta tornando-as mais sustentáveis (Azevedo et al., 2023). Nesse sentido, a adequada oferta nutricional nos cardápios da alimentação institucional é essencial não só para a promoção da saúde e combate à fome, mas também como a oferta de compostos não nutrientes que apresentam benefícios adicionais, a exemplo dos compostos fenólicos. Sabe-se que a grande maioria dos compostos fenólicos existentes são de origem vegetal e alinhado a isso, a nova resolução do PNAE recomenda que a maioria dos alimentos adquiridos para a alimentação escolar sejam *in natura* ou minimamente processados, majoritariamente de origem vegetal, ao passo que limita a oferta de alimentos processados, ultraprocessados e ingredientes culinários. A resolução também estabelece parâmetros mínimos para a oferta de alimentos que são fonte de ferro e vitamina A (Brasil, 2020), elementos essenciais para o crescimento e aprendizagem adequados.

Entretanto, ainda há muito para se avançar. Embora haja a recomendação de que alimentação escolar seja baseada em alimentos *in natura* ou minimamente processados, estudos relatam um grande número de alimentos ultraprocessados em cardápios escolares. Azevedo et al. (2023) analisaram a participação média dos grupos de alimentos, segundo a classificação NOVA, do total de energia adquirida para o Brasil, e embora o maior percentual seja de

alimentos i*n natura* ou minimamente processados (44,07%; IC95%: 43,79 - 44,35) os ultraprocessados ocupam uma fatia grande com cerca de 29,88% (IC95%: 29,62 - 30,15). Os menores percentuais foram encontrados para ingredientes culinários processados e alimentos processados, com 20,09% (IC95%: 19,89 - 20,29) e 5,96% (IC95%: 5,85 - 6,06), respectivamente. Teo (2018) também encontrou resultados parecidos em que a oferta média de ultraprocessados em alimentos escolares foi de 21,8% da energia total das aquisições do PNAE para o período de pesquisa 2008-2010 em Santa Catarina (Brasil). Entretanto, apesar da alta porcentagem de ultraprocessados que os cardápios escolares ofertaram nessas pesquisas, a legislação brasileira vem se atualizando e se tornando cada vez mais rígida em relação a oferta de alimentos processados e ultraprocessados. Até 2024, a Resolução n° 06 de 2020 permitia uma oferta de até 20% de alimentos ultraprocessados nos cardápios escolares, em 2025 o percentual reduziu para 15% e a expectativa que para 2026 esse percentual seja reduzido para 10% (Brasil, 2025).

Por fim, é importante que se planeje os cardápios pensando na inserção cada vez maior de alimentos *in natura* e minimamente processados, além de alimentos regionais que priorizam a cultura e a economia local. Entre esses alimentos, destacam-se as frutas por serem uma importante fonte de vitaminas, minerais, fibras e compostos fenólicos.

3.3 Frutas regionais do semiárido nordestino

O Brasil é um dos países com maior biodiversidade existente, sendo responsável por cerca de 10–20% das espécies vivas conhecidas no mundo. Quanto à variedade da flora, existem mais de 30 mil espécies diferentes de angiospermas (plantas com flores e frutos) coabitando os diferentes biomas do país (Floresta Amazônica, Caatinga, Cerrado, Mata Atlântica, Pantanal e Pampa). Embora grande parte da biodiversidade biológica e química do Brasil permaneça inexplorada, ela apresenta importância econômica com potencial para contribuir nos setores farmacêutico, de cosméticos, químico e de alimentos (Pilon et al., 2017; Valli; Russo; Bolzani, 2018; Neri-Numa et al., 2018; Schulz et al., 2020; Brasil, 2022).

Entre as regiões fontes dessa biodiversidade está o Semiárido brasileiro, que se estende por oito estados da Região Nordeste (Alagoas, Bahia, Ceará, Paraíba, Pernambuco, Piauí, Rio Grande do Norte e Sergipe) além do Norte de Minas Gerais (Medeiros, 2012). Caracteriza-se por apresentar irregularidade na distribuição das chuvas, solos rasos e pedregosos, conferindo baixa capacidade de reter água além de alta taxa de evapotranspiração (Marengo; Alves; Alvala, 2018). A vegetação é composta principalmente por plantas xerófitas, tais como cactos, adaptadas ao clima seco (Camacam, Messias; 2022).

Devido a essa heterogeneidade do relevo, clima e solo, na região Nordeste do Brasil é encontrada intensa biodiversidade (Coradin; Camillo; Pareyn, 2018) com um grande número de espécies frutíferas nativas, exóticas e endêmicas. Embora pouco exploradas, possuem grande potencial na agroindústria, como é o caso da *Spondias tuberosa, Spondias mombin e Spondias purpurea* (umbu, cajá e seriguela, respectivamente) além de outras como o *Psidium cattleianum, Talisia esculenta, Manilkara zapota* e *Annona Muricata* (araçá, pitomba, sapoti e graviola, respectivamente) (Almeida et al., 2011). Essas frutas são normalmente comercializadas *in natura* ou processadas na forma de polpas, sucos e outros produtos alimentícios. Devido à utilização comercial desses frutos, estudos vêm abordando as suas características de cultivo, assim como as características físico-químicas, maturação e estabilidade, os constituintes químicos e os coprodutos após o processamento (Silva et al., 2014).

Essas frutas, muitas vezes produzidas em sistemas agroecológicos, além de preservarem o meio ambiente e representarem fonte de renda para os agricultores familiares, são particularmente nutritivas, versáteis e saborosas (Brack et al., 2020) apresentando níveis consideráveis de minerais e vitaminas, capacidade antioxidante e compostos bioativos diversificados (Almeida et al., 2011; Assis et al., 2022). Assim, explorar frutas regionais como o umbu-cajá (*Spondias bahiensis*) e a graviola (*Anonna muricata*) é uma estratégia essencial para valorizar a biodiversidade, fortalecer a economia local e promover a sustentabilidade.

3.4 A fruta umbu-cajá (*Spondias bahiensis*)

O umbu-cajá é uma espécie endêmica do Brasil e nativa da região Nordeste (Alagoas, Bahia, Pernambuco, Sergipe). Ocorre em regiões semiáridas e também em zonas úmidas das áreas litorâneas e Mata Atlântica. Presume-se ser resultante do cruzamento natural entre cajá (*Spondias mombin* L.) e umbu (*Spondias tuberosa* Arr. Cam.) (Araújo et al., 2018; Santos; Araújo; Lemos, 2018; Souza, 1998). Botanicamente, essa fruta é caracterizada como um fruto carnoso do tipo drupa, contendo um endocarpo inserido em uma matriz fibrosa e a polpa é espessa com sabor doce acidulado. O formato é arredondado ou oblongo, a casca lisa e fina e seu peso pode variar de 5 a 100g a depender do genótipo. No estágio amadurecido, a sua coloração é amarela clara bem viva e na fase de maturação é verde escura (Araújo et al., 2018). As plantas de umbu e umbu-cajá apresentam mecanismos de sobrevivência à escassez hídrica, pois dispõem de raízes que contém xilopódios, armazenando água e outros elementos necessários à sua nutrição (Lima Filho, 2011).

Figura 1 - Frutos maduros de *Spondias bahiensis.*

Fonte: Araújo et al., 2018

Dentro da própria espécie são encontradas intensas diferenças morfológicas entre folhas e frutos, devido à variabilidade genética (Lira Júnior, 2005). O rendimento médio da fruta é de 55% a 65% em polpa (Brandão et al., 2018) devido ao mesocarpo espesso e suculento semelhante àquela encontrada nos frutos do umbu (*Spondias tuberosa*) (Sousa, 2020). Por isso, essa fruta é uma das principais matérias-primas de muitas agroindústrias de polpa do Nordeste (Gondim et al, 2013). Os frutos de umbu-cajá têm aroma e sabor agradáveis, além de serem rico em vitamina C e compostos bioativos, como beta caroteno e rutina, tornando a fruta desejável para o consumo *in natura* ou na forma processada, como polpa, sucos, picolés e sorvetes; ou na culinária, como componente de bebidas e doces (Assis et al, 2020; Assis et al., 2022).

O teor de vitamina C presente naturalmente nas frutas é um parâmetro nutricional de grande importância devido ao seu elevado poder antioxidante na prevenção e redução do risco de diversas doenças. Santos e colaboradores (2010), determinaram a composição centesimal de polpas de umbu-cajá (Tabela 1) e perceberam que em 100g de polpa continha em média 14,8 mg de vitamina C, ou seja, um copo de 300 ml de polpa representa aproximadamente 26% da Ingestão Diária Recomendada (IDR) para adultos dessa vitamina.

Tabela 1 - Composição centesimal da polpa do umbu-cajá *in natura*.

Componente	**Quantidade (100g)**
Energia (Kcal)	43,99
Umidade (%)	91,30
Proteína (g)	0,63
Carboidratos (g)	10,12
Lipídios totais (g)	0,11

Cinzas (g)	0,79
Fibra bruta (g)	1,36
Vitamina C (mg)	14,80
Cálcio (mg)	27,00
Ferro (mg)	0,41
Magnésio (mg)	15,00
Fósforo (mg)	166,00
Potássio (mg)	231,00
Sódio (mg)	1,00
B-caroteno (mcg)	24,00

Fonte: Santos et al., 2010; Biodiversity for Food and Nutrition, 2018

O fruto do umbu-cajazeiro é amplamente consumido nas regiões onde é cultivado e tem sido explorado devido ao seu alto potencial antioxidante, teor de polifenóis, valor nutricional, textura e aspectos sensoriais únicos. A *Spondias* spp. faz parte do bioma nativo brasileiro e é considerada um alimento fonte de compostos fenólicos bioacessíveis como: ácidos transcinâmicos, ácido gálico e catequinas (Carvalho; Conte-Júnior, 2021). Mesmo sendo pouco explorada, tem sido aplicada na indústria para desenvolvimento de novos produtos, como o Kombucha, processamento de sucos, polpa de fruta e substituição de farinhas com menores valores nutricionais pela farinha de umbu-cajá (Silva Júnior et al., 2021).

A utilização das partes não comestíveis do umbu-cajá que são diretamente descartadas (cascas e sementes), podem promover o desenvolvimento socioeconômico e contribuir para o desenvolvimento sustentável. As cascas, sementes e partes da polpa geralmente não consumidas, apresentam um alto potencial agroindustrial pelas características organolépticas presentes (Souza et al., 2017). Além disso, os coprodutos dessa fruta podem apresentar em sua composição nutricional compostos com atividades bioativas e características benéficas à saúde humanas tais como fibras alimentares e polifenóis (Lira-Junior et al., 2005; Correia et al., 2012).

O consumo adequado de fibras alimentares está associado à redução do risco de desenvolver doenças crônicas não transmissíveis, bem como à redução dos níveis séricos de lipídios, ao controle do peso e da glicemia pós-prandial, ao aumento da saciedade e à melhoria da função intestinal (Ötles; Ozgoz, 2014; He et al., 2022; Snauwaert et al., 2023). Um estudo com farinha de subprodutos de frutas e vegetais demonstrou a capacidade dessa matriz de ser fermentada pela microbiota intestinal, aumentando a produção de ácidos graxos de cadeia curta, como o butirato, e estimulando o desenvolvimento de *Bifidobacterium* e *Lactobacillus* (Andrade et al., 2020).

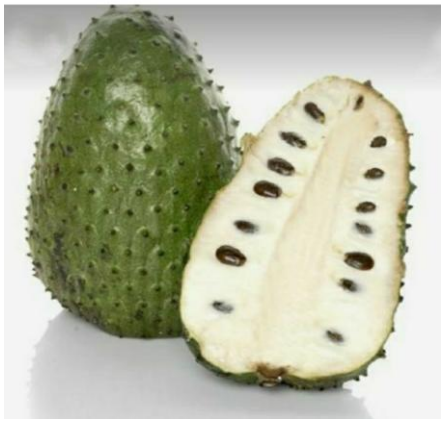
No umbu-cajá um dos polifenóis mais abundantes é a rutina, com cerca de 134,60 mg/100g (Dutra et al., 2017). A rutina possui um alto número de grupos hidroxila em sua estrutura, responsáveis pela neutralização dos radicais livres e, consequentemente, alta atividade antioxidante (Shen et al., 2022).

3.5 A fruta graviola (*Annona muricata)*

A graviola é de origem da região Neotropical e hoje é cultivada em todos os trópicos (Strijk et al., 2022). A espécie é nativa, mas não endêmica do Brasil, ocorrendo também em outras zonas tropicais da América do Sul. No Brasil, ocorre nas regiões Norte (Acre, Amazonas, Pará), Nordeste (Bahia), Centro-Oeste (Mato Grosso), Sudeste (Minas Gerais, Rio de Janeiro) e Sul (Rio Grande do Sul) (Coradin; Camillo; Pareyn, 2018).

A gravioleira é uma árvore tropical, amplamente difundida nas regiões litorâneas e no semiárido do Nordeste brasileiro, onde encontra condições ideais de clima e solo para o desenvolvimento. A árvore mede de 4 a 8 metros de altura, possui tronco reto, com copa pequena, estreita e pouco ramificada. Suas folhas são largas, verde-brilhosas. As flores são isoladas, grandes, amareladas e nascem no tronco ou nos ramos. Sua maior ocorrência é verificada nas regiões de clima quente e úmido. Quanto à frutificação das espécies cultivadas, esta ocorre durante o ano inteiro, sendo mais intensa nos meses de julho a outubro (Brasil, 2015).

Figura 2 - *Annona muricata* Linn



Fonte: Smith, 2023

A graviola é um fruto que apresenta cor verde mesmo quando madura, em formato de coração, que varia de 15 a 20 cm de diâmetro e pesa entre 0,4 e 4 kg dependendo das condições de cultivo, tais como o solo e clima. Possui polpa branca com caroços no seu interior (Agu et al., 2017; Qazi, et al., 2018; Gyesi et al., 2019).

Devido ao seu excelente sabor e ao aroma agradável de sua polpa, é uma fruta muito utilizada no preparo de diversos alimentos processados, como sorvetes, xaropes, néctares, compotas, geleias, doces e bebidas. No entanto, grande parte de sua produção é consumida *in natura*, na forma de suco ou salada de frutas, descartando-se o bagaço e as sementes (Olas, 2023). A fruta *in natura* e a polpa da graviola são ricas em água, carboidratos, vitaminas e minerais (tabela 2). Muitos compostos e metabólitos secundários também estão presentes na planta. Os principais compostos são acetogeninas, alcalóides, flavonóides, ácido p-cumárico, óleos essenciais, carotenoides, amidas e ciclopeptídeos além de taninos (Gajalakshmi et al., 2012; Moraes et al., 2020; Mutakin et al. 2022).

Os compostos fenólicos estão distribuídos nas diferentes partes da fruta. Moraes et al. (2020) encontraram quantidades expressivas de ácido p-cumárico (62,6 μg/g-dw) na polpa liofilizada. Em quantidades menores os autores também encontraram ácido ferúlico (5,39 μg/g-dw), ácido cafeico (7,75 μg/g-dw) e Ácido 3,4-di-hidroxibenzóico (16,5 μg/g-dw) (Moraes et al., 2020). Aguilar-Hernández et al. (2019) encontraram 8.21 mg/g de taninos condensados na polpa da graviola mostrando que também possui quantidades expressivas desse componente.

Tabela 2 - Análise nutricional em 100 g de graviola crua *(in natura)*.

Componente	**Quantidade (100g)**
Energia (Kcal)	62,0
Umidade (%)	82,2
Proteínas (g)	0,8
Lipídios (g)	0,2
Carboidratos (g)	15,8
Fibra alimentar (g)	1,9
Cálcio (mg)	40,0
Fósforo (mg)	19,0
Ferro (mg)	0,2
Potássio (mg)	250,0
Sódio (mg)	4,0
Magnésio (mg)	23,0
Vit. B1 (mg)	0,2
Vit. B2 (mg)	0,1
Vit. C (mg)	19,1

Fonte: Adaptado de Unicamp (2011).

Historicamente, todas as partes da graviola são utilizadas na medicina tradicional para o tratamento de diversas doenças. Os compostos bioativos presentes na *Annona muricata* conferem à planta importantes características farmacológicas como atividade citotóxica, cicatrizante, antimicrobiana, efeito anticancerígeno e genotóxico (Gajalakshmi et al., 2012; Anaya Esparza; Montalvo-González, 2020; Mutakin et al., 2022).

3.6 Conceitualização: resíduo, subproduto e coproduto

São denominados resíduos tudo aquilo que não é aproveitado e então deve ser descartado, como é o caso de resíduos de cozinhas industriais, por exemplo. Os resíduos agroindustriais são produtos gerados como resultado das atividades antrópicas, ou seja, aquelas desenvolvidas pelo ser humano, geralmente envolvendo a transformação de uma matéria-prima em outra. Esses resíduos podem ser derivados da agricultura, pecuária, produção de laticínios, indústria alimentícia e de bebidas, etc. Embora sejam menos tóxicos que os químicos, os resíduos agroindustriais podem levar a sérios problemas ambientais devido ao tratamento e destinação inadequados. Devido ao seu alto conteúdo orgânico e alta atividade de água, estes produtos são favoráveis ao crescimento de bactérias e fungos, podendo gerar danos à saúde humana, além de provocar poluição ambiental e liberação de metano na atmosfera (Federici et al., 2009; Fierascu et al., 2019; Shirahigue; Ceccato-Antonini, 2020; Kumar et al., 2022; Nair et al., 2022).

Um subproduto é definido, segundo Chapoutot et al. (2018), como uma substância ou objeto resultante de um processo cujo este não é o produto final a que se visa produzir, mas que se pode ter um destino, ou não. Normalmente, os subprodutos contêm carboidratos, lipídios e proteínas e outros compostos que podem ser explorados para a produção de biohidrogênio, por exemplo. Quando o subproduto é descartado, torna-se um resíduo. A valorização econômica do subproduto é parcial, permanecendo com baixo valor agregado.

O coproduto também é definido como produtos secundários do processamento de uma determinada matéria-prima, entretanto, o coproduto pode ser transformado em outros produtos de valor agregado sendo vantajoso para a empresa o seu aproveitamento. Além disso, pode ser utilizado para recuperação de compostos bioativos como fibra alimentar, polifenóis e compostos antioxidantes (Lopez-Marcos et al., 2015; Chapoutot et al., 2018).

3.7 Farinha de coprodutos agroindustriais: valorização dos recursos

O processamento de alimentos, especialmente as indústrias produtoras de sucos e polpas de frutas, gera cerca de 0,5 bilhão de toneladas de resíduos em todo o mundo (Banerjee et al.,

2017; Munekata et al., 2023), sendo grande parte desse material é descartado de alguma forma no meio ambiente. Já os subprodutos industriais de frutas, constituídos principalmente por cascas, bagaços, frações de polpas e sementes (Coman et al., 2020), contêm quantidades consideráveis de energia e nutrientes, podendo ser reaproveitados em matérias-primas para alimentação, seja ela humana ou animal. Assim, esses subprodutos podem reentrar na cadeia de abastecimento alimentar tornando-se um produto com valor agregado (Sun et al., 2024). Entretanto, a maioria desses subprodutos ainda não possuem destino específico e, devido ao alto conteúdo de umidade e alta carga microbiana, acabam por gerar uma intensa poluição ambiental ao final do processamento na indústria de polpas (Banerjee et al., 2017).

Os subprodutos agroindustriais, de forma geral, têm chamado a atenção da comunidade científica por serem uma fonte disponível, econômica e sustentável de polissacarídeos, fibras alimentares, compostos aromatizantes e fitoquímicos com alta capacidade antioxidante (Coman et al., 2020; Reguengo et al., 2022) tornando-se coprodutos com alto valor agregado. De todos os produtos secundários gerados nas agroindústrias, os derivados de frutas são os mais estudados. As frutas latino-americanas têm demonstrado forte relevância na área de pesquisa dos seus coprodutos não apenas por serem altamente disponíveis em seus países de origem, mas também por seu rico perfil fenólico (Sayago-Ayerdi et al., 2021; Reguengo et al., 2022).

Uma das formas recorrentes de aproveitamento de subprodutos é a transformação em farinhas, processo realizado com auxílio de estufa ou forno com circulação de ar, seguido de trituração para transformação em farinha, realizado em moinho de facas ou mesmo em liquidificadores e/ou processadores domésticos (Lucas-González et al., 2017), dessa forma um subproduto se torna um coproduto. De forma geral, os coprodutos derivados de frutas apresentam uma composição centesimal parecida, com baixo teor de gordura, carboidratos e proteínas e alto conteúdo de fibra alimentar. Selani et al. (2016) avaliaram a composição de coprodutos de abacaxi e maracujá e, em ambos, a fibra alimentar representou mais de 50% da amostra. Além disso, os coprodutos, ao passarem por desidratação e transformação em farinhas, tendem a manter baixo pH, baixa atividade de água concomitantemente com uma alta acidez, indicando que, o processamento de coproduto em farinha é eficiente para evitar deterioração (Selani et al., 2016).

Xavier et al. (2022) determinou a composição da farinha de coproduto de umbu (*Spondias tuberosa* Arr. Câmara). Os valores de umidade (7,64%), cinzas (2,63%), proteínas (5,70%), carboidratos (15,39%), fibras alimentares (61,21%), lipídios (7,53%) e valor calórico (152 kcal/100g) mostram um material com potencial inserção em preparações culinárias. Análises em bagaço de laranja apresentaram cerca de 28,8g/100g de fibra insolúvel, composto

majoritariamente por celulose (12,4g/100g) e lignina (8,9g/100g), além de pectina (12,3%) (Mantovan et al., 2023). Estudos realizados com bagaço de maçã também mostraram uma composição de 47,5g/100g de fibras, 8,9% de umidade e 3,4g/100g de cinzas (Guerrero et al., 2014).

Embora os coprodutos contenham quantidades significativas de nutrientes e compostos bioativos, as informações sobre utilização na produção de ingredientes/alimentos funcionais e o impacto na qualidade sensorial dos produtos ainda são escassos (Kandemir et al., 2022; Dimou et al., 2019). Amini Khoozani et al. (2019) avaliaram a inserção de farinha de casca de banana em produtos de panificação e confeitaria mostrando que uma substituição de 10% da farinha convencional por farinha de casca de banana promoveu aumento significativo no conteúdo fenólico total, de cinzas e de fibra alimentar total, mostrando-se eficaz na produção de um pão com alegação funcional.

Lopez-Vargas et al. (2013) avaliaram as propriedades tecnológicas da farinha do mesocarpo do maracujá, coproduto rico em fibra alimentar que apresentou aplicações potenciais como ingredientes em produtos que requerem hidratação, além do desenvolvimento de viscosidade e preservação de frescor, como alimentos assados ou produtos cárneos cozidos. Esse potencial foi atribuído ao seu alto teor de fibra alimentar total e boas propriedades tecnológicas, especialmente suas capacidades de retenção de água.

Estudos também focam na recuperação de compostos fenólicos dessas matrizes. Frutas como maçã, uva, romã, camu-camu, ameixa, jabuticaba, abacate, maracujá e goiaba são frequentemente citados devido à quantidade de antioxidantes existente nos coprodutos onde estudos *in vivo* e *in vitro* já demonstraram efeitos positivos na saúde, como redução de glicose sanguínea e melhora do perfil lipídico (Fidelis et al., 2019).

O aproveitamento de subprodutos de frutas, como o bagaço da graviola e do umbu-cajá transformados em farinhas, surge como uma alternativa para garantir a oferta contínua desses compostos bioativos de forma prática e sustentável. O fortalecimento de políticas públicas voltadas para a inserção de alimentos regionais e o aproveitamento de coprodutos pode contribuir significativamente para a melhoria da qualidade da alimentação escolar, com reflexos positivos na saúde e no bem-estar das crianças, além de valorizar a biodiversidade brasileira e os princípios de sustentabilidade

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4 RESULTADOS

ARTIGO 1

Impact of processing on polyphenols content in food: A nutritional and statistical analysis of Brazilian menus

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Artigo original publicado na revista *Food Research International*

DOI: https://doi.org/10.1016/j.foodres.2024.115115

ABSTRACT

Fresh and minimally processed foods are recognized as important natural sources of phenolic compounds, while industrial processing tends to reduce their concentrations. This *in silico* study investigated the effect of food processing on the presence of phenolic compounds in Brazilian menus, using linear regression models. The research examined menus from 319 schools in 75 counties in the state of Sergipe, Brazil, analyzing the caloric content, nutrients and polyphenols. These variables were grouped based on similarity and subjected to cluster analysis using Euclidean distance and Ward's method. The foods were classified by the degree of processing, based on NOVA classification, with modifications. The polyphenol content in menus was estimated using the Phenol-Explorer database. Cluster analysis revealed three distinct groupings and the results indicated that cluster 2 offered the highest macro and micronutrient values. Linear regression highlighted that the presence of regional foods and culinary ingredients significantly influenced the concentration of flavonoids and phenolic acid in the school menus analyzed. Fresh and minimally processed foods were positively associated with flavonoids without hydrolysis and phenolic acid with hydrolysis. Ultra-processed foods, on the other hand, showed negative associations with flavonoids with hydrolysis. These results provide important insights into the formulation of school menus, with implications for nutrition and public health.

Keywords: Nutrient composition; Dietary assessment; Food classification; Phenol-Explorer database; Cluster analysis; Dietary polyphenols

1 INTRODUCTION

Phenolic compounds (PC) are widely found in most plants and can be classified in different classes according to their structure, which have at least one phenol unit and hydroxyl groups (Lund, 2021). These compounds constitute a significant group of secondary metabolites crucial for plant development and adaptability. They are involved in physiological and biochemical mechanisms, including plant defense against biotic and abiotic stresses. This diverse array of substances encompasses a spectrum of chemical intricacies, spanning from simple phenolic acids to intricate tannins and lignins (Al-Khayri et al., 2023).

The polyphenolic compounds, naturally present in fruits, red wine, teas, vegetables, legumes, and cereals, which have potential health promoting effects, are widely studied for their beneficial activities, including antioxidant, anti-inflammatory, antitumor, and antimicrobial properties (Di Lorenzo et al., 2021; Rocchetti et al., 2022; Niewia domska et al., 2022). Thus, regular ingestion of PC in the diet contributes to reducing the risk of developing chronic diseases, such as diabetes, cardiovascular diseases and some types of cancer (Costa et al., 2017).

Fresh and minimally processed foods (FMPF) derived from plant sources stand out as main sources of natural PC, owing to their minimal processing levels. Conversely, the processing undergone by these foods induces a reduction in their PC concentrations, attributable to the influence of factors such as heat, light exposure, and other physical and chemical treatments employed in industrial settings (Khan et al., 2018). Nonetheless, alterations in dietary patterns have been observed, with high consumption of ultra-processed foods at the expense of FMPF, thereby exacerbating the prevalence of chronic diseases (Cortes et al., 2020, Marino et al., 2021).

Due to the importance of PC for health, tools for making available information on food PC contents have been developed, such as the Phenol-Explorer 3.6 database (Perez-Jimenez et al., 2010, Phenol-Explorer, 2016) that contains food-composition data for polyphenols (flavonoids, phenolic acids, lignans, stilbenes and other minor polyphenols) in foods. These tools facilitate the computation of the content of PC in foods, however, present a large volume of data and diversity of information. In this sense, *in silico* studies, such as the application of regression models, emerge as viable resources to optimize the use of information available in PC databases. Studies have demonstrated the effectiveness of these statistical modeling techniques in estimating food components in different contexts (Cayuela-Sánchez et al., 2019, Leite, 2019, Patra and Lalhriatpuii, 2016).

These *in silico* studies use computational methods and simulations to analyze biological and chemical systems, offering an alternative to traditional experimental approaches. They offer

advantages such as savings, time efficiency and ethical benefits, can integrate diverse data sets, predict outcomes, and explore a range of conditions that may be impractical to test. However, this type of study also has limitations, including reliance on the quality and integrity of input data, and the need for significant computational power for complex models (Madden et al., 2020, Peredo-Lovillo et al., 2022).

Given that statistical modeling techniques can optimize the use of large volumes of data, such as those available for PC in the Phenol-Explorer 3.6 database, and considering protective effects of these PC in a wide range of chronic diseases, the present study aimed to adjust regression models to estimate the PC content in Brazilian menus depending on the occurrence of variables such as different degrees of food processing, according to the NOVA classification: Fresh and minimally processed foods (FMPF), Processed Culinary ingredients (PCI), Processed foods (PF), Ultra-processed foods (UP).

2. MATERIALS AND METHODS

2.1. Obtaining data

The dietary assessment was carried out through the obtained menus and technical preparation sheet (TPS – Appendix A), reviewed by a team of nutritionists from the State Department of Education and Culture of the state of Sergipe (SEDUC), located in the Northeast region of Brazil. The menus served 319 schools located in 75 counties across the state in 2022. The TPS provided information on the per-capita quantities of the ingredients in the served preparations. Thus, the TPS were evaluated to assess the content of polyphenols and nutrients to be researched for each ingredient, according to the respective value of net per-capita. All calculations were carried out considering the content of the ingredients based on their net weight per capita (in grams or milliliters), as described in the TPS for raw food. At the end, for each menu day, the per capita values found for the nutrient and the polyphenol that we wanted to analyze were added, looking for the daily supply in grams or milligrams of each compound offered to a student.

Approximately 190,000 meals were served per day, serving 163,184 students enrolled in public education. The menus are planned considering the teaching modality, stratified as Elementary School 1 (6–10 years old), Elementary School 2 (11–15 years old), High School (16–18 years old) and education for young people and adults (>18 years old). In addition, menus with modifications are also planned for indigenous and quilombola communities.

The menus prepared for all teaching categories followed a biweekly and alternating pattern. In this sense, in a 4-week month, weeks 1 and 3 have the same menu, as do weeks 2

and 4. Each week of school activities is 5 days long, taking place from Monday to Friday. The analyzes include menus intended for part-time and full-time teaching, defined by periods of stay at school of 4 h a day and 8 h a day, respectively. The menus planned for the part-time period included one meal consisting of cereals and tubers (e.g.: pasta, couscous, rice, bread, cassava), vegetables (e.g.: carrots, cabbage), fruits (e.g.: banana, watermelon, apple, orange) and a source of animal protein (e.g. chicken, beef, eggs). The menus planned for the full-time period used similar foods distributed over three meals: one lunch (cereals, legumes, vegetables, fruits, and animal protein) and two light meals (both containing cereals, fruits and animal protein). In addition to these foods, the menus also included, less frequently, options such as cake, processed juice, yogurt, and saltine crackers.

2.2. Nutrient estimation

To estimate the nutrients and polyphenols provided during the school period, the net per capita values of the foods present in the culinary preparations on the menus were used. The assessment of daily caloric and nutrient supply values, including carbohydrates, proteins, lipids, fiber, minerals and vitamins, was carried out in accordance with the parameters of the Brazilian National School Feeding Program (PNAE), using Microsoft Excel (2013). PNAE legislation (Resolução No 6, de 08 de Maio de 2020 — Fundo Nacional de Desenvolvimento da Educação) determines that, for the part-time, 20 % of the daily needs for energy, protein, lipids and carbohydrates must be offered, while the full-time must offer 70 % of energy and the same nutrients. Furthermore, for a more detailed investigation into the composition of menus in different counties, these variables were grouped based on their similarity and subjected to cluster analysis.

2.3. Characterization regarding the degree of processing (NOVA)

Regarding the degree of processing, the foods were classified based on the NOVA classification proposed by Monteiro et al. (2010), with modifications. A subclassification was carried out within the classification of fresh and minimally processed foods, covering typically Brazilian foods, commonly consumed in the municipalities evaluated. All foods included in the menu were listed considering the quantity offered per capita (in grams) and the corresponding kilocalories. Then, the foods were classified into four groups and one subgroup: 1) Fresh and minimally processed foods (FMF); 1.1) Total regional foods (TRF); 2) Culinary Ingredients (CI); 3) Processed Foods (PF) and 4) Ultra-processed Foods (UP) (Fig. 1) (Monteiro et al., 2010, Monteiro et al., 2016).

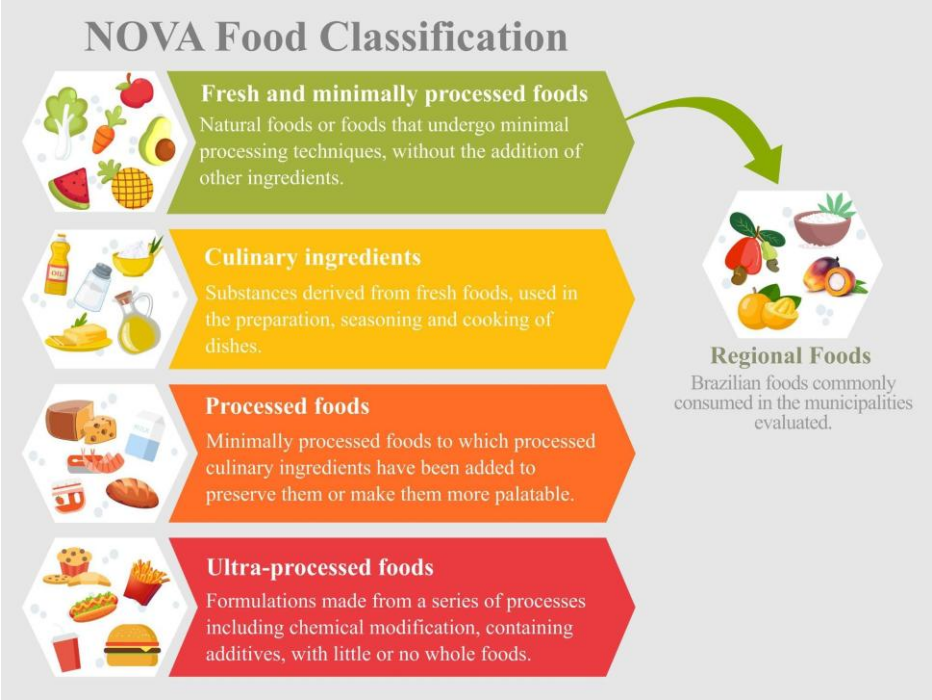

Fig. 1. NOVA food classification according to the degree of processing.

Thus, the first group includes natural foods or foods that undergo minimal processing techniques, such as removal of inedible parts, fermentation, freezing, pasteurization, but without the addition of other ingredients. The second covers substances derived from fresh foods, used in the preparation, seasoning and cooking of dishes. The third group is made up of natural or minimally processed foods to which processed culinary ingredients have been added to preserve them or make them more palatable. The fourth group includes formulations made from a series of processes including chemical modification, containing additives, with little or no whole foods.

2.4. Polyphenol content estimation

To estimate the polyphenol content present in menus, all foods were listed before preparation, and data on flavonoids (FLV), other polyphenols (OP) and total polyphenols (PT) were obtained based on the Phenol Explorer database (https://www.phenol-explorer.eu). Furthermore, it includes data on glycosides and esters. In the case of lignans and hydroxycinnamic acids in certain foods (e.g. cereals and beans), which cannot be released in normal extraction conditions, data corresponding to HPLC with hydrolysis were collected. Additionally, when data obtained by chromatography without hydrolysis were unavailable, polyphenol contents obtained by HPLC with hydrolysis were used. It contains data on a total

of 502 polyphenols (Perez-Jimenez et al., 2010). Thus, the concentration of polyphenols in each food was calculated using the electronic spreadsheet “PhenolCalc”, developed by Souza et al. (2019). Foods that were not present in the database or that did not have botanical equivalents were excluded from the polyphenol analysis. It is highlighted that the Phenol Explorer database is limited on Brazilian regional foods, as tubers, fruits, and vegetables. In this sense, the polyphenol content in the analyzed menus may be underestimated.

2.5. Statistical analysis

The analysis of macro, micronutrients and flavonoids were carried out according to the school period, covering part-time and full-time. Thus, the normality of these data was assessed using the Shapiro-Wilk test and the distribution among school periods was compared using the Mann Whitney test. The median, minimum and maximum of the data was presented, considering a p value ≤ 0.05.

A cluster analysis was conducted, in which the 75 counties were grouped based on the similarity of the supply of macro and micronutrients present in the menus offered. The counties were evaluated according to the variables of interest, resulting in a data matrix with n objects and p variables. This matrix was processed using Euclidean distance and Ward's method.

Multiple linear regression analysis was carried out specifically for the part-time regime, since the full-shift menu did not present sufficient data to carry out further analysis. From this, six models were developed for the response variables: Flavonoids Without Hydrolysis, Flavonoids with Hydrolysis, Phenolic Acid, Phenolic Acid with Hydrolysis, Folin-Ciocalteu Test and Lignans. The independent variables of each model were categorized into Total Regional Foods, Culinary Ingredients, Fresh and Minimally Processed Foods and Ultra-processed Foods. The Backward Elimination method was used to select the variables in the models, with the retention criterion defined as a total correlation greater than or equal to 0.85. In the Backward method, all variables are initially entered into the model and then removed sequentially based on the lowest partial correlation with the dependent variable. If a variable meets the removal criteria, it is deleted from the model. The process continues until there are no more variables in the equation that meet the removal criteria. In inferential statistical procedures, the significance level adopted was 5 % ($p \leq 0.05$). The software used to conduct statistical analysis was GraphPad Prism version 8.0.1, NCSS version 9.0.9, and Minitab version 6.3.0.

3. RESULTS

3.1. Nutritional characterization of menus

This study showed that the full-time period offers higher amounts of all the nutrients analyzed (Table 1). This characteristic can be attributed to the fact that full-time offers more meals compared to part-time.

Table 1. Daily supply of energy, macronutrients, fiber and micronutrients referring to school menus, according to the school period.

Variables	Part-time n=6330		Full-time n=2400		*p*
	Median	**Min-Max**	**Median**	**Min-Max**	
Energy (Kcal)	407.30	216.15 - 866.26	537.29	289.60 - 903.03	<0.001
Carbohydrates (g)	66.25	31.18 - 138.43	94.01	30.70 - 150.65	<0.001
Proteins (g)	17.47	8.05 - 37.91	22.89	6.97 - 43.50	<0.001
Lipids (g)	9.49	3.28 - 28.74	11.44	4.88 - 22.33	<0.001
Saturated lipids (g)	2.37	0.75 - 6.79	3.17	0.87 - 9.14	<0.001
Trans fat (g)	0.23	0.01 - 1.59	0.14	0.00 - 1.73	0.016
Fibers (g)	3.35	1.76 - 12.98	5.48	2.41 - 14.41	<0.001
Calcium (mg)	42.39	14.98 - 262.69	84.50	19.20 - 284.56	<0.001
Magnesium (mg)	50.26	16.26 - 154.62	56.07	21.88 - 174.23	<0.001
Iron (mg)	2.46	0.69 - 6.03	3.23	0.23 - 23.32	<0.001
Zinc (mg)	1.57	0.45 - 6.54	2.11	0.50 - 8.78	<0.001
Retinol (mcg)	3.80	0.72 - 125.11	4.96	0.00 - 766.62	<0.001
Vitamin C (mg)	21.68	0.32 - 112.24	22.27	0.00 - 140.60	<0.001
Sodium (mg)	589.80	285.98 - 1355.80	428.66	104.26 – 1671.21	<0.001

For a more in-depth analysis of the composition of the menu in different counties, the variables in Table 1 were considered, which were used for the cluster analysis, which grouped these variables based on their similarity, revealing three distinct groupings, as evidenced in the dendrogram presented in Fig. 2.

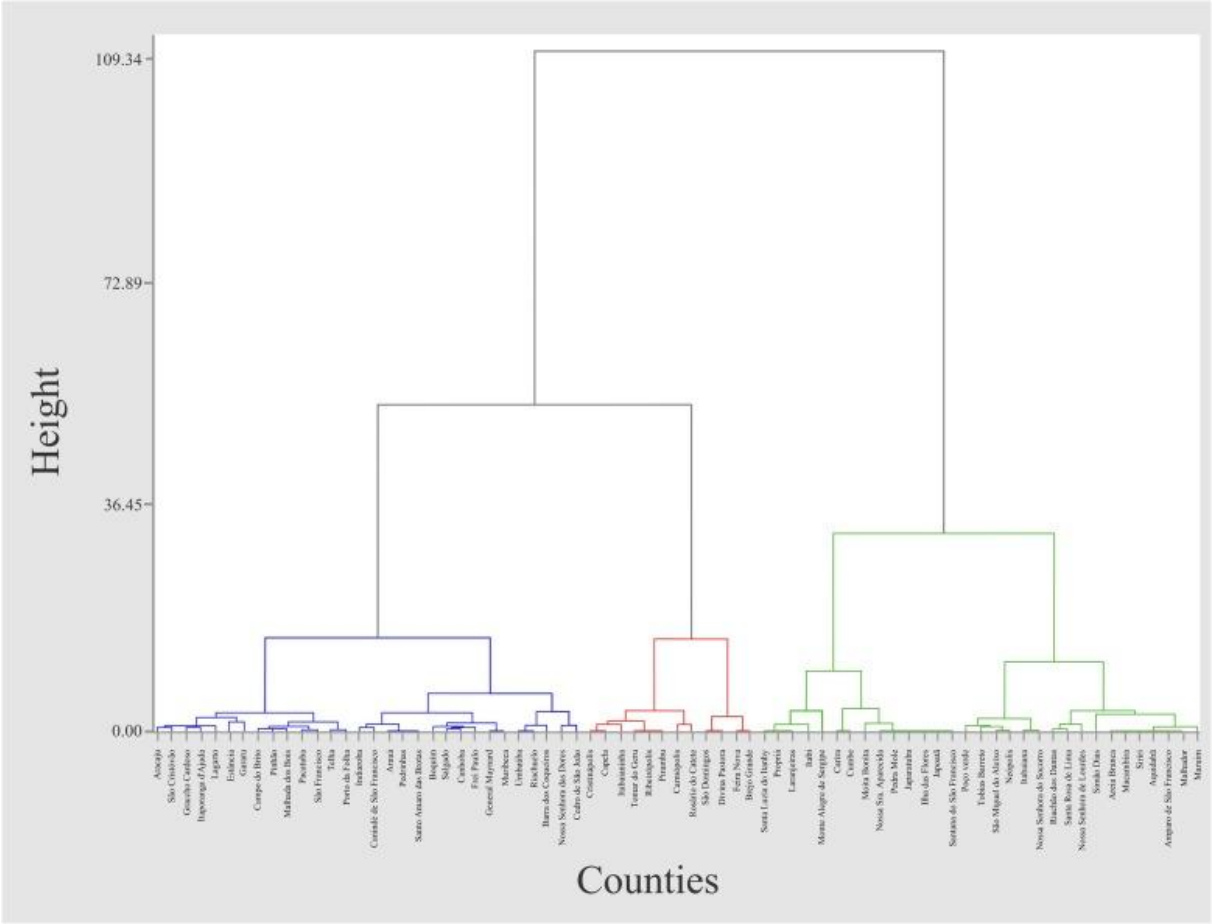

Fig. 2. Cluster analysis for the Brazilian municipalities of Sergipe grouped according to similarity of school menus.

Group 1, identified by the blue color in the dendrogram, consists of 30 counties. Group 2, represented by red, is made up of 12 counties, while group 3, green, is made up of 31 counties. Two counties did not fit into any group and were excluded from subsequent analyses.

In the block analysis, as determined by the clusters formed, it was possible to examine the distribution of each nutrient. There was a significant difference between the clusters when analyzing the amounts of proteins, carbohydrates, lipids, saturated fat, trans fat and fiber (Fig. 3). For all these nutrients mentioned, cluster 2 exhibited the highest values, followed by clusters 1 and 3, which also demonstrated differences between them, with cluster 3 being the lowest among them.

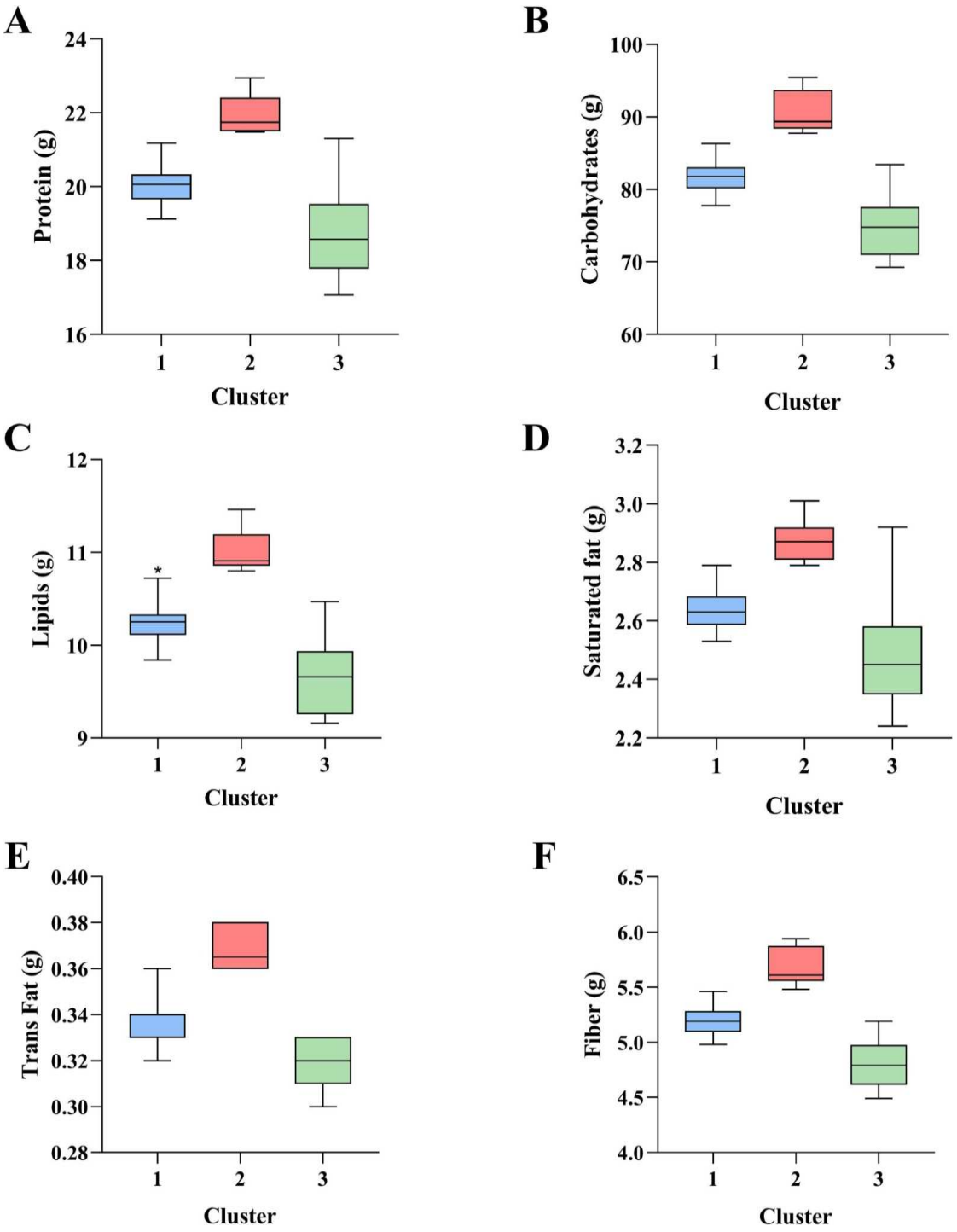

Fig. 3. Supply of proteins (A), carbohydrates (B), lipids (C), saturated fats (D), trans fats (E) and fiber (F) in school menus by cluster.

In relation to micronutrients, it was possible to observe, through block analysis, that cluster 2 presented the highest values for calcium, magnesium, iron, zinc, retinol and vitamin C, followed by clusters 1 and 3, with cluster 3 presenting the smallest quantities. As for sodium,

it was observed that cluster 3, which had the lowest amounts of micronutrients, had the highest amount of this mineral, while cluster 2 had the lowest amount of sodium (Fig. 4).

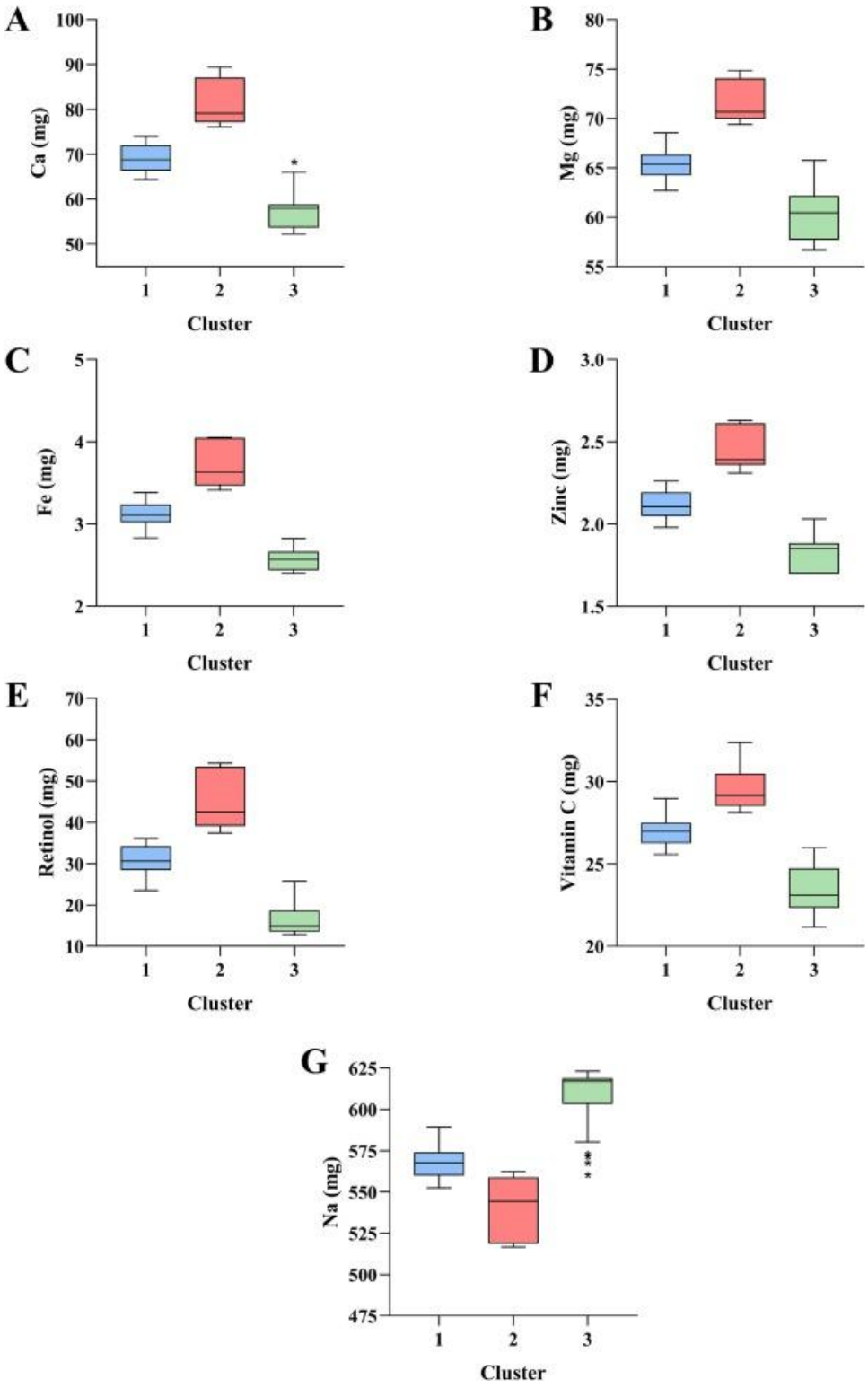

Fig. 4. Supply of calcium (A), magnesium (B), iron (C), zinc (D), retinol (E), vitamin C (F) and sodium (G) in school menus by cluster.

3.2. Offer of polyphenols on menus

After analyzing the macronutrients and micronutrients, the average daily supply of polyphenols on the menus was analyzed (Table 2). The analysis was carried out considering the two school periods, partial and full-time.

Table 2. Supply of polyphenols on school menus, according to school periods

Variables (mg/100g)	**Part-time n=6330**		**Full-time n=2400**		***p***
	Median	**Min-Max**	**Median**	**Min-Max**	
Flavonoids without hydrolysis	1.09	0.00 – 68.30	1.09	0.00 – 69.02	0.001
Flavonoids with hydrolysis	1.13	0.33 – 43.02	1.27	0.00 – 82.64	0.195
Phenolic acids without hydrolysis	2.16	0.18 – 222.78	3.16	0.00 – 47.49	<0.001
Phenolic acids with hydrolysis	7.45	0.63 – 245.67	4.55	0.00 – 218.15	<0.001
Lignans	2.43	0.10 – 17.43	3.35	0.00 – 27.42	0.004
Folin assay	109.00	0.00 – 2088.37	109.00	0.00 – 2592.59	<0.001

The concentration of polyphenols on menus varied according to the school period, with a general tendency for them to be highlighted on full-time menus. Except for phenolic acids with hydrolysis, which showed no difference between periods, all other polyphenol variables showed greater prominence in this period. Among the polyphenols analyzed, phenolic acids with hydrolysis were the most abundant, while flavonoids without hydrolysis were the least found.

3.3. Statistical modeling

To develop the linear regression models, first the food data were categorized according to the degree of processing and subclassified into fresh and minimally processed foods, covering regional foods (Table 3) and then the regression model was carried out considering the concentration of polyphenols offered (Table 4). It was observed that when categorizing the

foods, none were classified in the "processed" category. Foods categorized as fresh and ultra-processed presented larger portions in the full period, while culinary ingredients presented larger portions in the partial period.

Table 3. Classification of foods according to the degree of processing and portion offered in different school periods.

Classification	**Foods**	**Portion (g)/ Period**		
		Part-time Median (Min-Max)	**Full-time Median (Min-Max)**	***p***
In natura and minimally processed foods	Pumpkin, lettuce, garlic, rice, rolled oats, banana, sweet potato, potato, coffee powder, sun meat, beef, onion, chives, carrot, chayote, coriander, colorific, cumin, cabbage, cassava flour, carioca beans, chicken, yam, orange pear, powdered milk, fuji apple, pasta, cassava, watermelon, corn, egg, green pepper, cabbage, tangerine, tomato.	196.37 (78.77 – 478.16)	259.60 (60.00 – 549.90)	<0.001
Total Regional Foods	Acerola, banana, cocoa, cajá, cajarana, cashew, ciriguela, coconut, dendê, breadfruit, soursop, juá, papaya, passion fruit, pitomba, sapodilla, tamarind, umbu, watercress, jurubeba, major-gomes, gherkin, palm, okra, vinegar, mesquite, string beans, pigeon pea, arrowroot, sesame, yam, sedge, sorghum, tapioca flour, chives and coriander	2.00 (0.00 - 181.82)	21.16 (0.00 - 136.36)	0.126
Culinary ingredients	Soybean oil, salt, sugar.	4.00 (0.50 – 9.00)	1.50 (0.00 – 4.00)	<0.001
Ultra-processed foods	Savory biscuit, sweet biscuit, cake, cornbread, flavored yogurt, margarine, porridge mix, tomato sauce, nectar, hot dog bread.	65.00 (0.00 – 305.00)	100.00 (0.00 – 300.00)	<0.001

Table 4. Multiple Linear Regression Models to estimate the supply of polyphenols in menus based on the degree of food processing.

Variable Response	Model	R^2
Flavonoids without Hydrolysis	11.1646 - 0.4587 x TRF - 5.5287 x CI + 0.1530 x IN	0.98
Flavonoids with Hydrolysis	-6.1024 + 0.0802 x IN - 0.0871 x UP	0.98
Phenolic Acid	-135.4814 + 42.7235 x CI	0.99
Phenolic Acid with Hydrolysis	-98.1720 + 26.4696 x CI + 0.1876 x IN	0.99
Follin Assay	-310.1832 + 144.5790 x CI	0.95
Lignans	-0.4807 + 0.0425 x TRF + 0.0103 x IN	0.97

TRF – Total Regional Foods; CI – Culinary Ingredients; IN – *In Natura* and minimally processed foods; UP – Ultra-processed Foods.

Multiple linear regression models were used to understand the relationship between different food categories and the presence of different polyphenols in part-time menus, as full-time menus did not contain all the necessary information.

The results revealed that total regional foods, culinary ingredients and *in natura* foods had a significant impact on flavonoids without hydrolysis. For flavonoids with hydrolysis, it was observed that there was an influence of *in natura* and ultra-processed foods. As for phenolic acid and the Folin assay, they were impacted by culinary ingredients, while phenolic acid with hydrolysis was influenced by culinary ingredients and *in natura* foods. Finally, lignans were affected by total regional foods and *in natura* foods (Table 4).

For each model, the R2 value and the Mean Squared Error were observed. All of them presented R2 values above 0.94 and Mean Squared Errors below 0.3, except for the Follin Assay model, which presented a higher Mean Squared Error value. However, the model presented a high R2 value of 0.95 and, therefore, presented a good prediction measure. These results provide valuable insights into how different types of foods can influence the presence of specific phenolic compounds on menus, which could have important implications for the nutritional quality of the meals offered.

4. DISCUSSION

Tools like the Phenol-Explorer 3.6 database provide information on PC content in foods, but the volume of data pose challenges for your use. In this sense, statistical techniques, particularly regression models, offer a promising approach to optimize the utilization of PC databases.

The statistical modeling approach that was used in this study has shown to be a viable solution in different areas of food and nutrition sciences, as for nutritional labeling (Cayuela-Sánchez et al., 2019), diet and body composition, food quality (Shumilina et al., 2018) and phytochemical ingestion and absorption (Selby-Pham et al., 2018). However, this statistical tool is still underused for estimating nutrients and bioactive compounds in planned menus.

This study used multiple linear regression models to estimate the polyphenol content in Brazilian school menus considering different levels of food processing. It is already well established that fresh and minimally processed foods represent the main sources of phenolic compounds (Khan et al., 2018). Accordingly, the results obtained by linear regression demonstrate its influence on the different response variables, that is, classifications of phenolic compounds. The results revealed significant associations between certain food categories and the presence of specific polyphenols on menus. The linear regression equations showed high predictability, with coefficients of determination (R2) varying between 0.95 and 0.99.

Thus, it was possible to observe that the presence of FMPF on menu increased the supply of all PC classes analyzed, except phenolic acids and total phenolics by Folin. The presence of UP, in turn, reduced the supply of flavonoids without hydrolysis. The IC led to an increase in the supply of phenolic acids, phenolic acids without hydrolysis and total phenolics per Folin. On the other hand, they reduced the supply of flavonoids without hydrolysis.

These results provide valuable insights into how different types of foods can influence the presence of specific phenolic compounds on food, which may have significant implications for the nutritional quality of the meals offered in different menus. Understanding these associations can guide strategies to optimize the composition of school menus, aiming to provide food options richer in polyphenols and, thus, promote healthier eating among students. Future research can further explore these relationships and investigate how targeted interventions can improve the provision of polyphenols in schools.

In the analysis of menus provided to schools the results revealed significant differences in the nutritional composition of meals among the different school periods. Notably, it was observed that the full-time period offers higher amounts of all analyzed nutrients compared to the part-time. This can be attributed to the fact that the full-time periods provide more meals

throughout the day (3 meals a day), including a complete meal such as lunch, thus allowing for higher nutrient intake.

Furthermore, cluster analysis identified three distinct clusters based on the similarity of nutrient composition in the offered meals. Block analysis provided a more detailed view of the distribution of macro and micronutrients among the clusters. Cluster 2 was characterized by higher values of macronutrients such as proteins, carbohydrates, and lipids, and also presented the highest values of micronutrients such as calcium, magnesium, iron, zinc, retinol, and vitamin C. On the other hand, Cluster 3 showed the lowest amounts of these nutrients and had the highest amount of sodium, suggesting a less healthy dietary pattern in these municipalities.

Disparities in food provision among different periods and municipalities can have significant implications for children's health and development. Therefore, interventions aimed at improving the nutritional quality of school meals, especially in municipalities belonging to Cluster 3, may be necessary to promote healthy eating and prevent nutritional deficiencies among students.

Since many chronic diseases result, in part, from long and silent oxidative and inflammatory processes (Steven et al., 2019), early and daily consumption of PC-rich foods is an important strategy for the prevention of these diseases. Daily ingestion of processed food has grown over the years and changes in the food system continue to promote obesity. This is linked to significant increases in chronic diseases in different populations of different ages and groups, especially children, whose development is compromised by the early unfolding of chronic diseases (Cortes et al., 2020, de Deus Mendonca et al., 2016). Thus, considering the growth in UF consumption among children, child nutrition requires even more attention to ensure the regular dietary consumption of foods that are sources of PC and by that to potentially reduce the risks of chronic diseases also in adulthood.

The average daily content of polyphenols offered in school menus was found to be higher during full-time periods compared to part-time periods, following the trend observed for other nutrients. This discrepancy suggests the provision of foods richer in bioactive compounds during this period. Furthermore, a notable variance in polyphenol distribution among meals within the same school period was observed, which can be attributed to the diversity of foods available in the menus, the presence or absence of naturally polyphenol-rich foods, as well as the level of processing and seasonality of the foods, resulting in fluctuations in polyphenol concentrations among meals served in the same school period.

In addition to the concentration of phenolics in foods, it is crucial to consider the impact of processing on the presence of antinutrients and the bioavailability of these bioactive

compounds. Antinutrients can interfere with the absorption of essential nutrients, while processing techniques such as cooking and fermentation can both increase or decrease the availability of phenolics. Some reports indicate that heating can increase the extractability of phenolic compounds from plant matrices by disrupting cell walls and membranes, thereby facilitating their release (Minatel et al., 2017). Similarly, extrusion processing has been shown to improve the bioaccessibility of phenolics, likely due to the combined effects of high temperature, pressure, and shear (Ribas-Agustí et al., 2018). However, more severe processing conditions such as canning can result in losses of water-soluble and heat-sensitive phenolic compounds. Therefore, for a comprehensive assessment of the health benefits associated with phenolics, it is essential not only to measure their concentration but also to understand how processing practices influence their final nutritional quality and potential impact on human health.

Standardizing menus can ensure a more consistent and balanced food provision in terms of nutritional content, including the presence of polyphenol-rich foods, promoting greater uniformity in meal composition over time. Prioritizing the offering of fresh and minimally processed foods emerges as an effective strategy to promote a more nutrient-balanced food provision, benefiting the health and well-being of children.

Given that the statistical models developed in our study are based on the menus of one Brazilian state, their application may face challenges in other regions, due to the variability in composition between menus. However, these models can still be applied in other contexts with similar menus. Thus, a strength of our study is the possibility of applying these models to estimate polyphenols in Brazilian menus planned in the national public school system, for example. These menus follow the PNAE guidelines, varying only between regional foods. Future studies can use our protocol to develop new statistical models for estimating polyphenols in other types of menus.

5. CONCLUSION

Based on the data obtained, it was observed that the presence of fresh and minimally processed foods on menus is associated with a greater concentration of total polyphenols and flavonoids. These compounds have the potential to promote health benefits, including cognitive development and promoting healthy eating habits. On the other hand, the inclusion of ultra-processed foods on menus resulted in a decrease in the supply of total polyphenols and flavonoids. Therefore, it is recommended to review and replan menus, prioritizing the reduction or elimination of these ultra-processed foods.

Furthermore, adjusted regression models can be a potential alternative with good ability to predict the contents of polyphenols and flavonoids in menus. This tool can be useful for evaluating and monitoring the nutritional composition of menus, contributing to the promotion of healthier and more balanced food choices.

CRediT authorship contribution statement

Valéria Silva de Lana: Writing – original draft, Methodology, Investigation, Data curation, Conceptualization. Thais Barcelos de Castro: Writing – original draft, Methodology, Investigation, Data curation, Conceptualization. Livya Alves Oliveira: Writing – original draft, Investigation, Data curation. Stephanie Michelin Santana Pereira: Writing – review & editing, Investigation, Data curation. Kelly Aparecida Dias: Writing – review & editing, Investigation, Data curation. Rafaela Neto dos Santos Rodrigues: Writing – review & editing, Investigation. Ceres Mattos Della Lucia: Writing – review & editing, Investigation. Fernando Frei: Writing – review & editing, Supervision, Methodology, Data curation. Izabela Maria Montezano de Carvalho: Writing – review & editing, Supervision, Resources, Project administration, Methodology, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This work was supported by the “Conselho Nacional de Desenvolvimento Científico e Tecnológico - CNPq” (CNPq/MCTI/FNDCT N° 18/2021, grant 422847/2021-6) and “Fundação Coordenação de Aperfeiçoamento de Pessoal de Nível Superior” (CAPES).

Appendix A. Supplementary material

The following are the Supplementary data to this article: Supplementary Data 1 https://ars.els-cdn.com/content/image/1-s2.0-S0963996924011852-mmc1.xlsx

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ARTIGO 2

Nutritional and technological potential of umbu-caja and soursop co-product flours

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Artigo original publicado na revista *Food Research International*

DOI: https://doi.org/10.1016/j.foodres.2024.115520

ABSTRACT

Umbu-caja and soursop from the Northeast region of Brazil are rich in nutrients and bioactive compounds and are widely processed by the fruit agroindustry. However, there is a lack of research examining the composition and nutritional/technological potential of these co-product fruits. The present study evaluated the nutritional and technological characteristics of umbu-caja and soursop co-product flours (UCF and SCF, respectively), in addition to cytotoxicity in healthy cells. The results demonstrated that they are rich in dietary fiber (approximately 53 %), low in protein (approximately 8.0 %), and have minimal moisture content (<15 %). The mineral composition of the flours exhibited a notable presence of copper, iron, zinc, manganese, and boron. The evaluation of antioxidant capacity using the DPPH, ABTS, and FRAP methods demonstrated the presence of antioxidants that resisted processing, indicated by a high antioxidant capacity. Furthermore, the flours were found to contain phenolic compounds, predominantly rutin (UCF) and p-coumaric acid (SCF). The cytotoxicity test demonstrated that both co-product flours did not exert detrimental effects on healthy cells according to the MTT assay. The technological analyses highlighted low pH values (2.38 and 3.61 for UCF and SCF, respectively), which is favorable for a greater shelf life and suggests applications in fermented products. In addition, the flours have good water and oil holding capacity and low foaming, and they could be incorporated into food products that require these properties. The results demonstrated promising qualities of the UCF and SCF for incorporation into the human diet and product development, mainly due to their high fiber content, antioxidant capacity and low cytotoxicity.

Keywords

Annona muricata; *Spondias bahiensis*; Brazilian regional fruits; Sustainability; Antioxidants; Peel

1. INTRODUCTION

The production and marketing of tropical fruits has grown in recent years, with Brazil standing out as the third largest fruit producer in the world. Its annual production of over 40 million tons has played a crucial role in income generation and national agricultural development (FAO, 2020). Although the consumption of fresh fruit is highly valued for its nutritional properties, much of the fruit produced is destined for agro-industrial processing. The industrial processing of fruit generates a large amount of co-products rich in bioactive compounds, which can be used for different purposes but do not yet have a specific destination (Araújo et al., 2024, Viuda-Martos et al., 2012).

The term “co-product” is used to describe a secondary product or derivative that is obtained during the manufacturing or production process of a primary product. In the context of the agro-industry, co-products are the parts of fruit that are not used directly in the main product (such as pulp, juice, or jam) and include peels, seeds, pomace, and other components, which generally exceed one-third of the total weight of the fruit (Araújo et al., 2024, da Lima et al., 2023, Serena and Knudsen, 2007).

Co-products contain a variety of compounds of biotechnological interest, including phenolic compounds (Lucas-González et al., 2018, Selani et al., 2016) and dietary fiber (Gurak et al., 2014, López-Marcos et al., 2015). Depending on the technology, they can be converted into products of commercial interest as well as raw materials for new products (Viuda-Martos et al., 2012). It has been demonstrated that co-products can be employed as substrates in fermentation processes and in the formulation of novel ingredients, thereby enhancing the functional properties of this raw material and increasing its value (Araújo et al., 2024, Muñoz-Bas et al., 2024, Muñoz-Tebar et al., 2023). From a technological functionality perspective, co-products can be incorporated into foods to improve texture, act as a sugar-reducing agent, replace fats because they are rich in fiber, add color and act as a natural antioxidant. Furthermore, co-products possess an excellent antioxidant capacity (Klosterhoff et al., 2018, Sabino et al., 2020), which has attracted the attention of researchers (Lima et al., 2023). The bioactive compounds present can promote numerous health benefits, including intestinal health, weight regulation, cardiovascular health, and general well-being (Viuda-Martos et al., 2012).

A number of native fruit trees in Brazil remain relatively understudied (Camacam & Messias, 2022). In this context, the umbu-caja (*Spondias bahiensis*) and the soursop (*Annona muricata*), which are typical fruits of the Northeast region of the Brazilian territory, deserve particular mention (Carvalho & Conte-Junior, 2021). Both fruits are widely cultivated and appreciated in this region, with approximately 500 tons of umbu-caja and 8,000 tons of soursop

being produced annually. These fruits are supplied to both the fresh fruit market and the pulp processing industries (Freitas et al., 2013). Umbu-caja fruits are a rich source of several nutrients and bioactive compounds, including vitamin C, retinol, carotenoids, and vitamin B6. Additionally, they contain flavonoids, which have been linked to various health benefits. Soursop is a rich source of B vitamins and minerals, including potassium, magnesium, phosphorus, and calcium (Brasil, 2011, dos Santos et al., 2010).

The pulp production of these fruits generates a considerable amount of co-products. Despite the importance of native fruits in local food diversity, there is still a lack of studies on the nutritional profile of products derived from the processing of these fruits. The potential for new discoveries to encourage the agro-industrial sector to value these co-products as value-added ingredients for the food industry (Duarte et al., 2017) should remain a topic of interest. In light of the necessity to characterize this material, the present study aims to characterize the nutritional, antioxidant, and technological composition of the flours, as well as to assess the toxicity of the co-products of umbu-caja (*Spondias bahiensis*) and soursop (*Annona muricata*) from the Northeast region of Brazil.

2. MATERIAL AND METHODS

2.1. Chemicals

6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylicacid (Trolox), 2,2-diphenyl-1-picrylhydrazil (DPPH), 2,2′-azino-bis-(3-ethylbenzothiazoline-6-sulfonic acid)-diammonium salt (ABTS), 2,4,6-Tris(2-piridil)-s-triazina (TPTZ), pancreatin from porcine pancreas and trypsin from bovine pancreas were purchased from Sigma-Aldrich (St. Louis, MO, USA). The Resistant Starch Assay Kit and Total Dietary Fiber Assay Kit were purchased from Megazyme (Neogen, 620 Lesher Place, Lansing, MI 48912 USA). Ferric chloride and Phenolphthalein were purchased from Vetec (Merck KGaA, Darmstadt, Alemanha). Sodium hydroxide was purchased from CRQ química (Diadema − SP) and potassium persulfate was purchased from Êxodo Científica (Sumaré − SP).

2.2. Flours characterization

2.2.1. Raw materials

The umbu-caja (UCF) and soursop (SCF) co-products were supplied by Agroindústria Pomar Polpa de Frutas located in Aracaju, state of Sergipe. Umbu-caja and soursop fruits are native to the municipalities of Igaci (9°32′12.7″ S, 36°37′45.0″ W), state of Alagoas and Aracaju (10°55′40.0″ S, 37°4′13.9″ W), state of Sergipe, respectively. Three batches of each

co-product were collected from the 2023 harvest, between June and August. Immediately after collection, the co-products were taken in refrigerated thermal boxes to the Dietary Technique Laboratory of the Federal University of Sergipe. After separating the seeds, the UCF and SCF co-products (peels and pomace) were dried in an air circulation oven at 60 ± 2 °C for 7.5 h and 8 h, respectively. These drying conditions were defined considering previous drying tests so that the samples reached a moisture percentage of less than 15 %. This value is established by Brazilian legislation for the characterization of vegetable flours (Brasil, 1978). The dried co-products were then ground in a knife mill (Marconi, MA-090CFT, São Paulo/Brazil) and passed through a 20 mesh sieve. The UCF and SCF flours were stored at −20 °C until further analyses. The subsequent analyses were performed at the Experimental Nutrition Laboratory and the Food Analysis Laboratory, both located in the Department of Nutrition and Health at the Federal University of Viçosa.

2.2.2. Centesimal composition

The moisture, protein, total ash and lipid contents were determined according to the analytical standards of the Association of Official Analytical Chemists (2012). Total dietary fiber, soluble and insoluble fraction, were determined based on AOAC Method 991.43 and AACC Method 32–07.01 using the Megazyme® Enzymatic Kit. Carbohydrates were calculated by difference, using the equation (1) (Eq.1). The total energy value was calculated by considering 4 Kcal/g for carbohydrates and protein, and 9 Kcal/g for lipids (FAO, 1991).

[1]

[100-(%moisture+%lipids+%proteins+%total dietary fiber+%ash)]

2.2.3. Mineral content

The minerals were quantified using nitric-perchloric digestion in a 4:1 v/v. Briefly, the flours were pre-digested by adding 1 mL of 65 % nitric acid for 12 h. Then, an additional 3 mL of nitric acid was added, and the digestion began with a gradual increase in temperature to approximately 95 °C. The digestion volume was halved, and 2 mL of 70 % perchloric acid was added, with the temperature gradually increased to 150 °C until the flours in the digestion tube were clear. The tubes were then removed from the block, cooled to room temperature, and the digested flours (about 2 mL) was diluted with distilled water to a final volume of 25 mL. The digested flours were then analyzed, in duplicates, using an Inductively Coupled Plasma Atomic Emission Spectroscopy − ICP-OES (PerkinElmer, OPTIMA 8300, USA).

2.2.4. *In vitro* protein digestibility

In vitro digestibility was carried out, in duplicate, using the "pH Drop" methodology proposed by Hsu et al. (1977) and modified by Queiroz Mendes et al. (2016). Initially, a suspension solution was prepared by diluting the flours to a concentration of 6.25 mg of protein/mL in 50 mL of distilled water. The diluted flours were then placed in a metabolic bath with stirring at 37 °C (Marconi, MA093, São Paulo/Brazil). Once the temperature was reached, the pH was adjusted to 8 using a 0.1 N NaOH or 0.1 N HCl solution. To each replicate, 5 mL of an enzyme solution (2.5 mg/mL trypsin and 1.6 mg/mL pancreatin, 25 BAEE units and 12.8 USP specifications, respectively) was added. The decrease in pH was measured every minute for 10 min using a digital potentiometer (BEL engineering, 966-PLUS, São Paulo/Brazil). Digestion was indicated by a drop in pH occurring approximately 10 min after the enzyme solution was added. Decrease in pH after 10 min and parameter equations were used to describe in vitro digestibility. The degree of digestibility was calculated based on the following standard equation proposed by Mendes et al. (2016): % D = 93.1359 [1 − e3.4138 × (8 − pH)], R2 = 0.8848.

2.3. Physicochemical composition

2.3.1. Phytates content

Phytates were analyzed, in triplicate, using the Phytase Assay Kit from Megazyme®. This method involves the hydrolysis of phytic acid by phytase and the quantitative measurement of the released phosphate. Briefly, 0.2 mL aliquots of phytic acid were distributed into test tubes and pre-incubated at 40 °C for 5 min. The properly diluted enzyme was also pre-incubated at 40 °C for 5 min in a separate container. Then, 0.2 mL of the pre-incubated enzyme extract was added to each tube containing the phytic acid solution. The mixture was homogenized and incubated at 40 °C for 10 min. After incubation, 0.1 mL of stop reagent was added, and the contents were shaken. A 0.1 mL aliquot was taken from the reaction mixture to detect free phosphate. The results were measured using a UV–VIS microplate reader (Thermo Fisher Scientific, Multiskan go, Finland) at 360 nm. The Phytic acid (g/100 g) content was calculated using the spreadsheet available at www.megazyme.com.

2.3.2. Condensed tannins

The concentration of tannins was determined, in triplicate, using the vanillin/HCl reaction method, according to Burns (1971), with modifications by Maxson and Rooney (1972) and Prince, Van Scoyoc, and Butler (1978). A 0.2 g of each flour was added to 10 mL of a 1 %

HCl solution in methanol in Falcon tubes. The extract was left to stir for over a night in a metabolic bath (Marconi, MA093, São Paulo/Brazil) at room temperature to extract the tannins. After this period, the tubes were centrifuged at 3000 rpm for 20 min and the supernatant was separated. For the analysis, 1 mL of the supernatant was combined with 2.5 mL of a 1 % vanillin solution in methanol and 2.5 mL of an 8 % HCl solution in methanol in a test tube. The tubes were left to stand for 20 min, after which the absorbance was measured at 500 nm against the blank (1 mL of the extract and 5 mL of 4 % HCl in methanol), in which the vanillin solution was omitted. A catechin standard curve (y = 0.1697x + 0.0469, R2 = 0.9943) was constructed and the results are expressed in mgEC/g.

2.3.3. Resistant starch

Resistant starch was measured using the AOAC 202.02/AACC 32–40.01 direct method with the Megazyme® enzyme kit. For the procedure, 0.1 g of each flour was weighed in duplicate. Four milliliters of pancreatic alpha-amylase containing amyloglucosidase was added to the Falcon tubes, which were then mixed in a vortex and shaken in a water bath (Marconi, MA 093, São Paulo/Brazil). at 37 °C with continuous agitation for approximately 16 h to digest the non-resistant starch. The enzymes were inactivated, and the non-resistant starch components were extracted using 50 % and 99 % ethanol. The resistant starch from the flours was dissolved with 2 M KOH, neutralized with sodium acetate (pH 3.8), and hydrolyzed with amyloglucosidase. The resulting D-glucose was quantified using the Glucose Determination Reagent (GOPOD) and measured with a spectrophotometer (Thermo Fisher Scientific, Multiskan go, Finland) at 510 nm. The blank and standard were prepared using water and glucose, respectively. The resistant starch content was calculated using the spreadsheet available at https://www.megazyme.com.

2.3.4. Total phenolic content

The total amount of phenolic compounds was determined in triplicate using the Folin-Ciocalteu reagent according to the methodology described by Singleton et al. (1999), using the reagent proposed by Folin and Ciocalteau (1927). Two grams of UCF or SCF were mixed with 20 mL of a methanol:water solution (60:40 v/v). The suspension was stirred for 12 h and then centrifuged at 2790 g for 5 min at room temperature. The supernatant was collected for immediate analysis. For the analysis, 500 µL of the extract was mixed with 500 µL of Folin-Ciocalteu solution (20 %) and 500 µL of sodium carbonate solution (7.5 %). The mixture was vortexed and incubated at room temperature for 30 min. Absorbance was measured at 765 nm

using a spectrophotometer (Thermo Fisher Scientific, Multiskan go, Finland). Quantification was performed using an analytical curve (y = 0,0013x + 0,1809, R2 = 0,9928) based on the absorbance of solutions with different concentrations of gallic acid. The results were expressed in milligrams of gallic acid equivalents per gram of flour (mg GAE/g).

2.3.5. Extraction by sonication and identification of phenolic compounds by UPLC-PDA-MS analysis

The extraction of UCF and SCF was performed according to Pizani et al. (2024) with slight modifications. Briefly, 2 g of both flours were weighed directly into 50 mL test tubes and 20 mL of the solvent methanol:water (60:40 w/v) were added to the tubes, making a solute to flour ratio of 1:10 w/v. Following this, the solution was sonicated in an ultrasonic bath (P60H, Elmasonic, Singen, Germany, 2.75 L, 37 kHz, 135 W) for 60 min, starting at room temperature and reaching 50 °C at the end of the extraction. Subsequently, the extracts were centrifugated, filtered in a nylon filter of 0.22 µm, and further injected in an ultra-performance liquid chromatography coupled to a photodiode array and mass spectrometry (UPLC-PDA-MS) (Acquity, Waters Co., Milford, MA, USA). The separation system was performed in an Acquity UPLC BEH C18 50 × 2.1 mm, 1.7 µm analytical column, and the mobile phases consisted of water (A) and acetonitrile (B), both acidified with 0.1 % acetic acid (v/v). The gradient elution was from 5 % to 100 % B in 0 min to 10 min. The chromatography conditions were: flow rate of 0.5 mL/min, column oven at 40 °C, and injection volume of 5 µL. UV spectra were recorded from 210 nm to 400 nm. MS analysis was performed in positive and negative ionization mode (100 – 1000 Da) with cone voltage of 15 V and 30 V, respectively, capillary voltage of 0.8 kV, and probe at 600 °C. The p-coumaric acid (CAS: 501–96-4) and rutin (CAS: 153–18-4) analytical standards were acquired from Sigma-Aldrich (Sigma-Aldrich, San Luis, MO, USA) and injected to guarantee the properly identification of both phenolics. The software Empower 3 was applied for data processing (Waters Alliance, Milford, MA, USA).

2.3.6. DPPH scavenging assay

The antioxidant activity of the flours was determined by their ability to scavenge the free radical 2,2-diphenyl-1-picrylhydrazyl (DPPH). In brief, 1 g of flour was weighed, in duplicate, into a Falcon tube and diluted in 10 mL of 50 % ethanol. The extraction was performed for 10 min in an ultrasonic bath (SolidSteel, model USC-1600A, 40 kHz). After extraction, the mixture was centrifuged (Nüve, NF 1200R, Turkey) at 5000 rpm for 10 min at 5 °C (Arruda et al., 2019). Then, 100 µL of the extract was added to a microtube containing 1.5

mL of 0.1 mM DPPH solution. The solution was vortexed for 1 min and left to rest at room temperature in the dark for 30 min. After incubation, the solution was pipetted into a 96-well microplate and the absorbance was read at 517 nm using a spectrophotometer (Thermo Fisher Scientific, Multiskan go, Finland). Trolox 1.5 mM was used as the antioxidant standard (y = −0,1592x + 0,5218, R2 = 0,9946), and the results were expressed as µmol TE/g (Brand-Williams et al., 1995).

2.3.7. Trolox equivalent antioxidant capacity (TEAC) assay

The TEAC assay was based on the reports of Leite-Legatti et al. (2012) and Pavan et al. (2014). Briefly, 1 g of flour was measured, in triplicate, into a falcon tube. The flours were diluted with 10 mL of 50 % ethanol and placed in an ultrasonic bath (SolidSteel, model USC − 1600A, 40 kHz) for 10 min at room temperature. They were then centrifuged (Nüve, NF 1200R, Turkey) at 5000 rpm for 10 min at 5 °C. The radical cation ABTS•+ was chemically generated using 88 µL of potassium persulfate (140 mmol/L) and 5 mL of ABTS (7 mmol/L), leaving it to react for 12 h at room temperature in the dark. The radical was diluted with ethanol until reaching an absorbance of 0.70 ± 0.02 at 734 nm. In brief, 250 µL of ABTS•+ solution was mixed with 50 µL of extract. After 6 min of reaction at room temperature in the dark, the absorbance measurements were carried out at 734 nm against blank on a Microplate reader (Thermo Fisher Scientific, Multiskan go, Finland). The ABTS•+ radical scavenging activity was expressed as a percentage calculated from the absorbance of the control (Abs Control) and the extract (Abs Extract), as shown in the equation (2). Trolox 200 μMol was used as the antioxidant standard (y = 0,6395x − 0,3452, R2 = 0,9993), and the results were expressed as µmol TE/g.

[2]

$$\%\ inhibition\ = \left(\frac{Abs_{Control} - Abs_{Extract}}{Abs_{Control}}\right) x\ 100$$

2.3.8. Ferric reducing antioxidant power (FRAP) assay

The FRAP assay followed the method of Oyaizu (1986) with some modifications. The method is based on the ability of an antioxidant agent to reduce Fe3+ to Fe2+. The FRAP reagent was prepared at the time of analysis (25 mL of 0.3 M acetate buffer, 2.5 mL of 10 mM TPTZ solution, and 2.5 mL of 20 mM aqueous ferric chloride solution). For the blank, 20 µL of ethanol was used instead of the extract, and a five-point 800 µmol trolox curve (varying the

concentration from 5 µmol/L to 30 µmol/L) was constructed. The extract was made using the same conditions described previously (topic 2.3.7). The plate was incubated at 37 °C for 30 min and the absorbance at 595 nm was recorded using a microplate reader (Thermo Fisher Scientific, Multiskan go, Finland). Trolox 800 μMol was used as the antioxidant standard (y = 0,002x + 0,0804, R2 = 0,9958), and the results were expressed as µmol TE/g.

2.3.9. Total flavonoid content

The total flavonoid content was quantified using the method of Zhishen et al. (1999) adapted for microplates. First, the extract was made using the same conditions described previously (topic 2.3.7). Next, in the microplate, 25 µL of each extract was pipetted in triplicate, and 100 µL of distilled water and 7.5 µL of NaNO2 (5 %) were added. After 6 min, 7.5 µL of AlCl3 (10 %) was added, and, after further 6 min, 100 µL of NaOH (4 %) and 10 µL of distilled water were pipetted into the mixture. The absorbance at 510 nm was read from the plate reader (Thermo Fisher Scientific, Multiskan go, Finland). For the blank, 25 µL of distilled water was used instead of the flour extract. The results were expressed in micrograms of catechin equivalent which is determined only from a catechin curve (12.5 µg/mL to 400 µg/mL, y = 0,001x + 0,0852, R2 = 0,9984).

2.4. Methyl-Thiazolyl-Tetrazolium (MTT) cytotoxicity assay

2.4.1. Processing the tested solutions

A 50,000 µg/mL stock solution of each flour was prepared by diluting a volume equivalent to 0.02 g of the flour in 400 µL of dimethyl sulfoxide (DMSO). A working solution with a concentration of 200 µg/mL of flour was prepared from the stock solution. For this, 8 µL of the stock solution was diluted in 2 mL of complete Dulbeccos's Modified Eagle Medium (DMEM), supplemented with 10 % Fetal Bovine Serum and 1 % antibiotic (penicillin 10,000 U/mL; streptomycin 10000 mg/mL). Serial dilutions were prepared from the working solution in decreasing concentrations from 200 to 6.25 µL/mL.

2.4.2. Cell lineage and cell preparation

The L929 strain (fibroblast) was seeded in 96-well culture plates (1 × 104 cells/well) and cultured in DMEM. The cells were subjected to different concentrations of UCF and SCF (6.25, 12.5, 25, 50, 100 and 200 µg/mL) for 24 h in an incubator at 37 °C and 5 % CO2. The dilution vehicle DMSO 0.1 % in DMEM was used as a negative control for cell death. Cell viability was assessed by the colorimetric method using MTT.

2.4.3. MTT assay

The plates were incubated for 24 h at 37 °C in a 5 % CO_2 atmosphere. At the end of the incubation time, the culture medium was removed and the cells were washed twice with PBS. A solution of MTT (0.025 g diluted in 50 mL of PBS) was placed in contact with the cells, then incubated at 37 °C for 3 h. Following the removal of the MTT, DMSO was added for 10 min to solubilize the formazan crystals formed from the reduction of the MTT salt by the metabolic activity of the viable cells after the treatments. The optical density (OD) was then read in an automated microplate reader (Agilent, BioTek Synergy H1, USA) at a wavelength of 570 nm. The result is expressed as the percentage cell viability obtained using equation (3).

[3]

$$Cell\ viability\ (\%) = \left(\frac{Treated\ cells}{Untreated\ cells}\right) x\ 100$$

Tests for cytotoxicity were performed in quadruplicate and the graphs were plotted using the GraphPad Prism software version 8.0 (Dogmatics, Boston, Massachusetts, USA).

2.5. Functional and technological capability

2.5.1. Water and oil holding capacity

To determine the water and oil holding capacity, the methods proposed by Diniz and Martin (1997) and Haque and Mozaffar (1992) were used with slight modifications. A total of 0.1 g of the flours was weighed in triplicate into a microtube. Subsequently, 1 mL of water or oil was added to the microtube. The tubes were homogenized in a vortex for one minute and then left to stand for 30 min at room temperature. Finally, the microtubes were centrifuged (Hermle, Z 216 MK, Germany) at room temperature at 12,000 rpm for 20 min. The supernatants were then carefully discarded, and the edge of the microtube was dried on absorbent paper until all residual supernatants had been drained off. The Water Holding Capacity (WHC) and Oil Holding Capacity (OHC) were reported as the amount of water/oil absorbed per gram of flour. The equation (4) were used for the calculation. Where M1 is the mass (g) of the tube with the wet flour after discarding the residual water or oil supernatant; Mt is the mass (g) of the microtube and M0 is the initial mass (g) of the flour.

[4]

$$WHC\ or\ OHC\ (\frac{g}{g}flour) = \left(\frac{M1 - Mt - M0}{M0}\right)$$

2.5.2. Foaming property

The method proposed by Silva et al. (2022) was used to determine the foaming capacity and stability. A total of 1.5 g of the flours was weighed in triplicate into a beaker. Subsequently, 60 ml of distilled water was added, and the beaker was transferred to a magnetic stirrer (Fanem, 258, São Paulo/Brazil). The pH of the dispersion was then adjusted to 7.0 using 0.1 N NaOH or 0.1 N HCl. A 15-milliliter of the solution was transferred to a 50-milliliter graduated cylinder and homogenized using an Ultra-Turrax (Solab, SL-114, São Paulo/Brasil) for two minutes. The volume of the foam was then measured in the cylinder itself at 0 min, 10 min, 30 min, and 60 min.

2.5.3. Soluble solids, pH, and titratable acidity

The soluble solids (SS) content was measured using a refractometer (BEL Engineering, RMT, São Paulo/Brazil). A small amount of flour was diluted in distilled water, filtered through qualitative filter paper, and dripped on the prism of the refractometer in sufficient quantity to cover the surface. The pH analysis was carried out by dispersing 1 g of the flour in 10 mL and determined directly in the solution using a potentiometer with a glass membrane electrode (Gehaka, PG2000, São Paulo/Brazil). Titratable acidity was measured by weighing 1 g of the flour, in triplicate, in a 125 mL flask and dispersing it in 50 mL of distilled water. Phenolphthalein solutions was added to each flask, and homogenized manually. The volume of NaOH used was noted down, and the calculations were carried out according to the equation (5). The results are expressed as a percentage of malic acid, according to AOAC (2005).

[5]

$$\%\ malic\ acid = \frac{Vol.\ used\ of\ 0.1M\ NaOH\ x\ 0.1M\ NaOH\ correction\ factor\ x\ 0.06705\ (Malic\ acid\ factor) x\ 10}{Weight\ of\ flour\ (g)}$$

2.6. Statistical analysis

Student's t-test for independent samples was used to analyze the data. Results were considered statistically significant at p-values of 0.05. Statistical analyses were carried out using GraphPad Prism software version 8.0 (Dogmatics, Boston, Massachusetts, USA). Results were expressed as mean ± standard deviation. Pearson's correlation test was used to assess the correlation between the variables total phenolic compounds, total flavonoids and the in vitro antioxidant tests (DPPH, ABTS and FRAP). The correlation test was carried out using the R

version 4.4.0.

3. RESULTS AND DISCUSSION

3.1. Chemical composition and physicochemical characteristics

The chemical composition and antioxidant analysis of the UCF and SCF on a dry basis (g/100 g dry matter) are shown in Table 1. As for their proximal composition, few differences were found between the flours, with SCF having a higher moisture and soluble dietary fiber content, while UCF had a higher insoluble dietary fiber and ash content.

Table 1. Centesimal composition and phytochemical composition of umbu-caja and soursop coproduct flours

Variables	UCF	SCF
Moisture (g/100g)	7,3±0,35[b]	10.5±0.33[a]
Carbohydrates (g/100g)	23.3± 0.46[a]	24.0±0.39[a]
Proteins (g/100g)	8.5±0.01[a]	7.9±0.49[a]
Lipids (g/100g)	1.8±0.23[a]	1.3±0.35[a]
Dietary fibers (g/100g)	53.0±1.23[a]	53.3±1.23[a]
Soluble dietary fiber (g/100g)	3.7±0.07[b]	9.8±1.17[a]
Insoluble dietary fiber (g/100g)	49.3±1.14[a]	43.5±0.05[b]
Ash (g/100g)	6.1±0.03[a]	3.2±0.02[b]
In vitro protein digestibility (%)	45.83±6.83[b]	92.34±0.19[a]
Total calories (kcal/100g)	143.9±1.52[a]	138.2±1.38[a]
Total phenolics (mg EqGA/g)	36.1 ±2.2[a]	15.8± 2.8[b]
DPPH (µmol TE/g)	23683.6±230.43[a]	22261.7± 190.17[b]
ABTS (µmol TE/g)	21.8±0.79[a]	9.27±0.07[b]
FRAP (µmol TE/g)	1347.0±98.64[a]	383.4±5.26[b]
Total flavonoids (µg CE/mL)	35.3±0.87[a]	12.9±0.49[b]
Resistant starch (g/100g)	1.85±0.36[a]	1.72±0.11[a]
Phytic acid (g/100g)	0.00±0.00[a]	0.05±0.01[a]
Condensed tannins (mg CE/g)	5.47±2.92[b]	17.65±0.65[a]
pH	2.38±0.09[b]	3.61±0.22[a]

Values expressed on a dry basis as the mean of replicates ± standard deviation. Means followed by the same letter in the same row do not differ by Student's t-test at a 5% probability level. Caption: UCF = Umbu-caja coproduct flour and SCF = Soursop coproduct flour. Kcal = kilocalories. mg EqGA/g = milligrams of gallic acid equivalent per gram of flour. µmol TE/g = micromol of trolox equivalent per gram of flour. µg CE/mL = micrograms of catechin equivalent per milliliter of extract. mg CE/g = catechin equivalent milligrams per gram of flour.

The moisture content for both flours was under 15 % which is in accordance with Brazilian legislation that standardizes moisture values for flour from vegetable sources (Brasil, 2022, Xavier et al., 2022). Similar results of moisture were observed in umbu (*Spondias tuberosa*) waste flour (7.64 %) (Xavier et al, 2022) and soursop seeds flour (12.63 %) (Menezes et al., 2019). This moisture content, combined with the acidic pH (3.61 and 2.38 for SCF and UCF, respectively), provides stability to the product and low susceptibility to the growth of microorganisms such as fungi and bacteria, allowing storage at room temperature and extended shelf life (Rana et al., 2024, Xavier et al., 2022).

Among macronutrients, dietary fiber was the most abundant in both flours (53 g/100 g), making SCF and UCF excellent sources of fiber. Comparable results have been found in banana (65.55 g/100 g) (He et al., 2021), umbu (61.21 g/100 g) (Xavier et al., 2022), and yellow mombin (68.85 g/100 g) (Oliveira et al., 2024) co-product flours. Flours derived from co-products tend to be highly fibrous due to their composition, attributable to the fact that these materials are predominantly composed of peels and fibrous fractions of the fruit pulp (Villacís-Chiriboga et al., 2023).

Several beneficial effects on human health are related to fiber consumption through mechanisms, which help in preventing and treating chronic diseases, as well as reducing serum lipid levels, controlling weight and postprandial glycemia, increasing satiety and improving intestinal function (He et al., 2022, Ötles and Ozgoz, 2014, Snauwaert et al., 2023). Also, soluble dietary fibers can increase intestinal viscosity, reducing the contact of the intestinal mucosa with toxic and carcinogenic compounds and reducing the absorption of bile acids and cholesterol (McRorie and McKeown, 2017, Passos et al., 2024).

Furthermore, a study with fruit and vegetable co-product flours demonstrated the ability of this matrix to be fermented by intestinal microbiota, increasing the production of short-chain fatty acids such as butyrate and stimulating the development of Bifidobacterium and Lactobacillus (Andrade et al., 2020). Additionally, in the technological context, extracted fibers can also be included in food matrices such as meat products (Alarcón García et al., 2015, Nieto et al., 2021), cookies, cakes, yogurts, and jams (Guimarães et al., 2023), contributing to

increasing indirect fiber intake in the population.

In our study, the SCF and UCF flours had equal concentrations of protein. However, SCF had a higher digestibility than UCF, which could be explained by the SF and IF content present in the flours, since the IF content in UCF is higher than that of SCF, which can reduce the accessibility of enzymes to the protein matrix (Ye & Yu, 2024). In parallel, according to Joye (2019), protein digestibility can be higher or lower depending on extrinsic and intrinsic factors in the matrix. Intrinsic factors include the amino acid profile and the level of protein folding. Extrinsic factors are related to environmental conditions such as pH, temperature and the presence of secondary molecules. An electrophoresis test was performed to identify the proteins in these flours (Fig. S7), but no prominent proteins were found that could explain this digestibility difference.

3.2. Mineral profile

In terms of mineral content (Table 2), UCF had a higher content of several minerals such as iron, calcium and aluminum compared to SCF. This higher content may be associated with the more acidic pH of UCF, which increases the solubility of minerals, improving their retention in this matrix (Gharibzahedi & Jafari, 2017). Considering both UCF and SCF, 100 g of the flour would be enough to meet 100 % of the iron RDA for an adult man. Meanwhile, for adult women, it would provide 53.9 % and 46.7 % of the RDA, respectively (Padovani et al., 2006). UCF and SCF are viable and potentially sustainable alternatives for increasing iron intake in the population. This is particularly important when we consider the fact that the Northeast region has the highest prevalence of anemia in adults in Brazil (Machado et al., 2019). However, the bioavailability of iron in vivo needs to be assessed.

Table 2. Mineral composition of umbu-caja and soursop coproduct flours

Mineral (mg/100g)	**UCF**	**SCF**
Nitrogen	1150.00±10.00^{a}	1081.00±1.00^{b}
Phosphorus	138.09±1.38^{a}	104.22±1.50^{b}
Potassium	2061.68±9.48^{a}	1009.05±9.42^{b}
Calcium	590.81±0.39^{a}	167.91±0.08^{b}
Magnesium	160.21±1.55^{a}	104.88±0.41^{b}
Sulfur	104.06±0.13^{a}	99.79±0.19^{b}
Copper	0.31±0.00^{b}	0.54±0.00^{a}
Iron	9.57±0.10^{a}	8.41±0.05^{b}

Zinc	1.16±0.01[b]	1.69±0.01[a]
Manganese	1.66±0.02[a]	0.70±0.02[b]
Sodium	5.16±0.22[b]	65.31±0.53[a]
Boron	2.24±0.01[a]	1.12±0.02[b]
Selenium	n.d.	n.d.
Nickel	0.11±0.00[b]	0.31±0.00[a]
Lead	n.d.	n.d.
Cadmium	n.d.	n.d.
Chromium	0.15±0.00[b]	0.62±0.00[a]
Aluminum	2.49±0.03[a]	0.19±0.00[b]
Barium	7.42±0.07[a]	0.19±0.00[b]
Titanium	n.d.	n.d.
Molybdenum	n.d.	n.d.
Cobalt	n.d.	n.d.

Values expressed as mean of replicates ± standard deviation. Means followed by the same letter in the same row do not differ by Student's t-test at a 5% probability level. Caption: mg/100g = milligram per 100g of flour. n.d. = not detected; UCF = Umbu-caja coproduct flour; SCF = Soursop coproduct flour

A high aluminum content was observed in the UCF, which may be caused by contamination during the pulp processing, either from mineral residues released from equipment or water contaminated with heavy metals. Contamination can also be derived from the soil, due to pesticides containing aluminum in their composition, which can accumulate in the peels and leaves of plants (Alasfar and Isaifan, 2021, Hassan, 2019, Mello et al., 2023). In addition, highly weathered tropical soils can contain iron and aluminium oxides and hydroxides, which may partly explain the high concentration of these minerals in the flours (Christoffoleti et al., (2088)., Nguyen et al., 2020). However, even with the high amount of aluminum in UCF, the flour can still be consumed by the population. According to the Joint FAO/WHO Expert Committee on Food Additives (JECFA), the permitted weekly intake of aluminum is 2 mg/kg body weight without harm to health (World Health Organization, 2011).

3.3. Antioxidant activity (AA), contents of flavonoids (TF) and total phenolic (TP) in the flours

The use of fruit processing co-products has been studied not only for environmental reasons but also related to their antioxidant characteristics (Albizzati et al., 2021, Ominski et al., 2021). *In vitro* and *in vivo* assays have already demonstrated potent reducing action of oxidative radicals from agro-industrial co-product flours, acting on the inhibition of enzymes

related to lipid and/or glucose metabolism and modulation of biomarkers, genes, and intestinal microbiota (Cádiz-Gurrea et al., 2022). In our study, UCF showed higher antioxidant capacity for all tests, while SCF showed less but yet suggesting a great antioxidant capacity (Table 1). Fig. 1 and Tables S1 and S2 show the correlation between the AA (ABTS, FRAP, DPPH), TP (Total Phenolics) and TF (Total Flavonoids) of the flours.

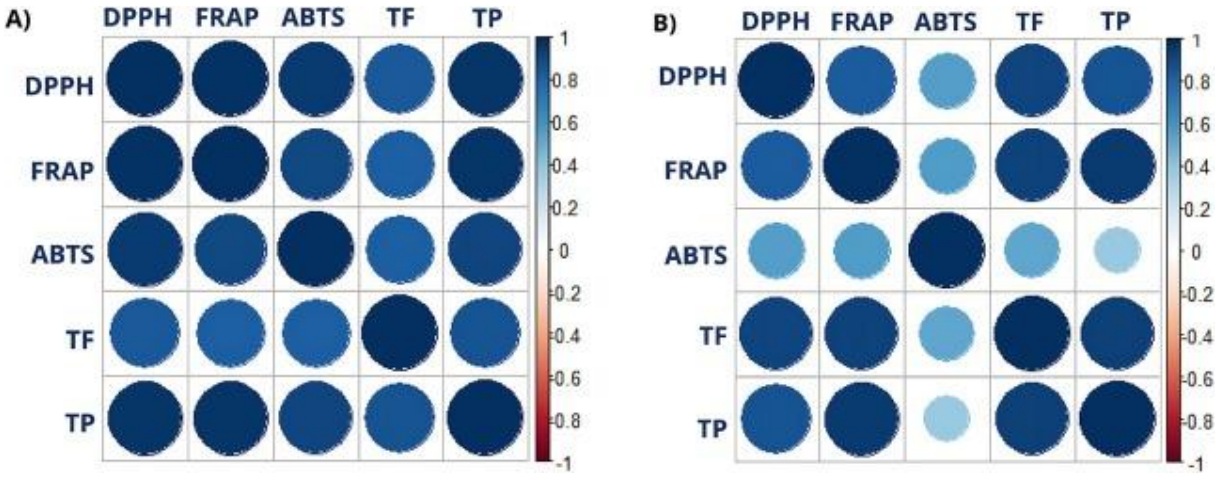

Fig. 1. Correlation between the AA (DPPH, FRAP, ABTS) and TP (Total Phenolic) and TF (Total Flavonoids). Umbu-caja co-product flour (1A) and Soursop co-product flour (1B)

It was noted that in both flours the antioxidant capacity was linked to the amount of total phenolic compounds. For UCF (Fig. 1A) we observed a strongly positive correlation in all the tests. In contrast, for SCF, the ABTS test had a positive but slightly weaker correlation with the other antioxidant tests (Fig. 1B).

The antioxidant tests employed here utilize disparate methodologies for the elimination of free radicals, thereby demonstrating that the compounds present in the flours exhibit a diverse range of actions. In DPPH tests, the 2,2-diphenyl-1-picrylhydrazyl radical is neutralized by accepting a hydrogen atom or electron from an antioxidant species (Bibi Sadeer et al., 2020, Xiao et al., 2020). This assay is suitable for measuring the antioxidant capacity of small molecular polyphenols with lipophilic properties, such as quercetins, resveratrol, and anthocyanins. However, it is less specific for those with little or no lipophilic properties (Lang et al., 2024). ABTS method (2,2-azinobis-(3-ethyl-benzothiazolin-6-sulfonic acid (ABTS + -)) is suitable for both lipophilic and hydrophilic compounds and has a strong correlation with the biological activity of antioxidants, even though this radical is not present in the human body. Finally, the FRAP assay is a method based on single electron transfer (SET) that measures the reduction of the ferric (Fe3+)-tripyridyl triazine-iron complex to the intensely blue ferrous

complex (Fe2+) by antioxidants in acidic pH environments (pH = 3.6) in order to maintain iron solubility. The redox potential of Fe3+ is approximately 0.70 V, which is comparable to the redox potential of ABTS +- (0.68 V) (Bibi Sadeer et al., 2020, Lang et al., 2024, Munteanu and Apetrei, 2021, Pivec et al., 2019, Xiao et al., 2020).

The flavonoid rutin, present in UCF (Table 3), has high antioxidant activity attributed to the composition of their three rings (A, B and C) (Brito et al., 2022, Shen et al., 2022). This is reflected in the antioxidant tests in which they all showed a strong positive correlation with TP and TF, indicating that the main compounds present in UCF are effective in neutralizing the free radicals evaluated.

Table 3. Phenolic compounds tentatively identified by UPLC-PDA-MS in soursop and umbu-caja co-products extracts.

Peak	[1]Rt (min)	% Area	λmax (nm)	Main m/z $[M-H]^-$	Potential identification	Reference
Soursop						
1	1.432	43.46	315.4	325	Coumaric acid hexose derivatives	Jiménez et al. (2014)
2	1.899	50.52	309.4	163.26	*p*-coumaric acid	Jiménez et al. (2014); Du et al. (2021)
3	2.607	6.02	220.4; 292.7; 317.8	300.14	4-Hydroxybenzoic acid 4-*O*-glucoside derivative	Du et al. (2021)
Umbu-caja						
1	1.459	27.10	227.5; 313	165	Coumaric acid derivatives	Ribeiro et al. (2022)
2	2.095	11.28	228.7; 313	279.19/469.32	[2]n.i.	-
3	2.217	53.19	211; 255.9; 353.7	611.40	Rutin	Ribeiro et al. (2022)
4	3.088	8.44	255.9; 354.9	464.73	Isoquercitrin	Ribeiro et al. (2022)

[1]Rt: retention time; [2]n.i.: non identified.

The main polyphenolic compound present in SCF flour, p-coumaric acid (Table 3), also has a strong antioxidant capacity. P-coumaric acid can be esterified with molecules as alcohols,

amines, monosaccharides, oligosaccharides or form glycosides with sugars, forming water-soluble conjugates. In plant cell walls it can be esterified with polysaccharides and lignin, forming water-insoluble complexes (Pei et al., 2016). This may be connected with the solvent used to extract the compound (ethanol:water 50:50 v/v), in which p-coumaric acid may have been less soluble and consequently had less free radical scavenging activity. So, although the samples have undergone industrial processing, and subsequently heating and grinding to produce flours, we found that they preserve a good amount of antioxidant compounds and antioxidant activity.

3.4. Determination of polyphenolic profile

The SCF and UCF extracts were analyzed by UPLC-PDA-MS and their corresponding chromatograms are depicted in Fig. 2. Additionally, the UV and the MS spectra of the identified peaks can be seen in Figs. S1–S5. The combined application of the two high-sensitivity detection methods allows an interesting discussion of potential phenolic compounds present in each flour, especially through the combined evaluation of both λmax and m/z, and the comparison of these findings with databases and literature (Sanches et al., 2022).

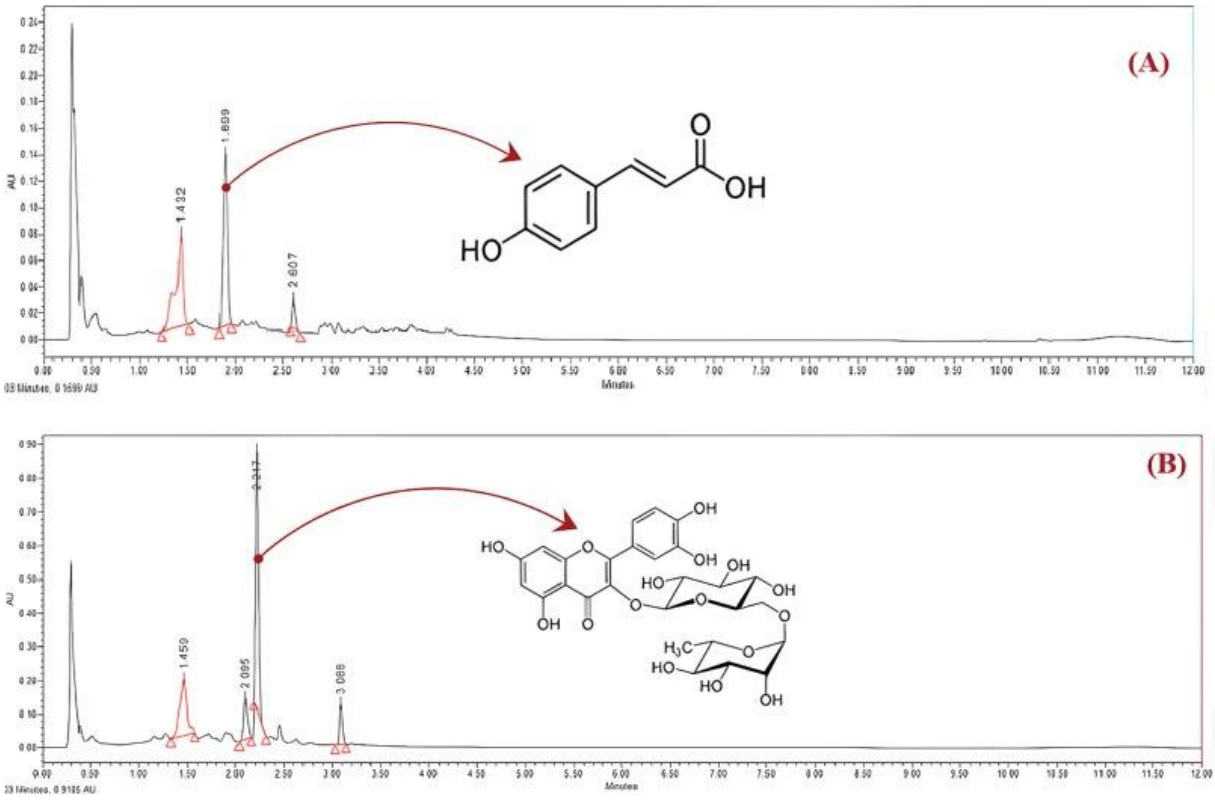

Fig. 2. Chromatogram of soursop (*Annona muricata*) (A) and umbu-caja (*Spondias bahiensis*) at 320 nm, showing their most significant peaks, their respective retention times and the

chemical structure of the phenolic compounds identified at their wider peaks: p-coumaric acid (2A) and rutin (2B).

The chromatogram of SCF extract (Fig. 2A) highlights three main peaks with retention times of 1.432, 1.899, and 2.607 min, as shown in Table 3. The first peak exhibited a λmax of 315.4 nm (Fig. S1A), which, according to Jiménez et al. (2014), can be related to coumaric acid hexose. Moreover, this compound showed a m/z of 325 identified at the MS spectrum (Fig. S2A). Following this, the second peak demonstrated a λmax of 309.4 nm (Fig. S1B), which can be associated to p-coumaric acid. Such compound presents m/z [M−H]- of 163, and this value was properly identified at the MS spectra of the second peak (Fig. S2B). The second peak corresponded to 50.52 % of the total peak areas, hence being the major compound identified in the flour (Table 3). Therefore, the p-coumaric acid analytical standard was also injected aiming at confronting its retention time, UV and MS spectra with those from the extract. Our findings then confirmed the initially proposed identification for the second peak: the same retention time of 1.899 min and the expected UV and MS spectra, as can be seen in Fig. S3.

The p-coumaric acid is a phenolic acid belonging to the hydroxycinnamic acid family, widely present in plants and mushrooms both in its free form and conjugated (Pei et al., 2016). Several studies have demonstrated that p-coumaric acid and especially their conjugated form present many biological activities such as antioxidant, antimicrobial, anti-inflammatory, and neuroprotective effects, as well as diabetes mitigation, and other health promoting effects (Boz, 2015, Ferreira et al., 2019, Kiliç and Yeşiloğlu, 2013, Lou et al., 2012, Pei et al., 2016). Finally, the third peak corresponded to only 6.02 %, and is likely identified as 4-hydroxybenzoic acid 4-O-glucoside derivatives, according to Du et al. (2021).

A similar approach was used for the analysis of UCF extracts, which provided the identification of four peaks (Fig. 2B). It can be seen that the third peak stood out among the other ones since its area covered 53.19 % of the total peak areas. Such peak presented a retention time of 2.217 min (Fig. 2B), λmax in 255.9 and 353.7 (Fig. S4C), and m/z [M−H]- = 611.4 (Fig. S5C), which indicated the possible identification of rutin (Bastos et al., 2007). Considering its widest area, the confirmation of rutin was assessed by confronting its retention time, UV and MS spectra with those from rutin analytical standard (Fig. S6), which finally confirmed the presence of this compound.

Rutin, also known as quercetin 3-O-rutinoside or vitamin P, is a natural flavonoid glycoside and one of the most prevalent secondary metabolites of plants (Semwal et al., 2021). Several biological activities are associated with rutin, including those against cancer, diabetes,

oxidative stress, microbial growth, among others (Negahdari et al., 2021, Ganeshpurkar and Saluja, 2017), and it can be suggested that the key factors for its bioactivity reside on its hydroxyl groups as well as on the rutinoside molecule right attached at the C-3 position (Semwal et al., 2021). According to studies found in the literature, this compound is one of the major compounds in umbu-caja extracts (Bastos et al., 2007, Dutra et al., 2017).

The information of the remaining three peaks obtained from UCF extract have also been analyzed (UV and MS spectra shown in Figs. S4 and S5), allowing an identification attempt based on the literature, as shown in Table 3. It is important to highlight that umbu-caja is an underexplored plant in terms of metabolite profile. Therefore, further studies using high-resolution MS equipment are recommended for more conclusive identification.

3.5. Cytotoxicity in vitro

An *in vitro* cytotoxicity test was performed with the aim of verifying the safety of the flours in fibroblast cells (Fraga et al., 2021). The cytotoxicity results for UCF and SCF are shown in Fig. 3. The test results revealed low variations in the cell viability of the L929 lineage when it tested at different concentrations of the flours extracts (Fig. 3B). According to Cerqueira et al. (2008) cell toxicity is considered when the viability of the exposed cells was under 70 %.

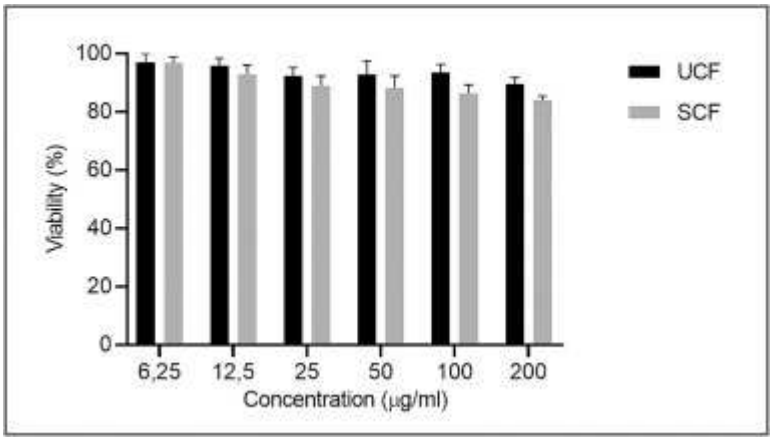

Fig. 3. Cell viability of L929 cells. The effect of the UCF and SCF on cell viability was evaluated using the MTT assay.

Cytotoxicity studies are necessary and can indicate the toxic profile of plant materials. In our analysis, cell viability was higher than 82 % in both flours. Studies evaluating the cytotoxicity of the *Spondias* genus observed high cytotoxic activity of S. mombin against prostate tumor cells (Guedes et al., 2020) and low cytotoxic and antiparasitic action for S. tuberosa, with the exception of the hydroalcoholic extract of the roots which showed around 28

% cytotoxicity against murine fibroblasts at the highest concentration evaluated (1000 μg/mL) (Gomez et al., 2020). Other studies working on the cytotoxicity of Annona fruits showed low impact on healthy cells, but an antiproliferative effect in vitro on various tumor cell lines (HeLa and PC3, MCF7) (Raybaudi-Massilia et al., 2015, Sabapati et al., 2019). In contrast, another study evaluating soursop seed extract showed that it had a dose-dependent cytotoxic effect (1000 μg/mL), so as the concentration increased, cell viability was reduced (Andrade et al., 2022).

These results are encouraging and provide support for the potential use of these flours in diet. Future research could build upon these findings by evaluating additional cell types and conditions of use, as well as in vivo trials. Additionally, the safety of this raw material is reinforced with a view to creating new products from industry-generated products, which is also a potential economically viable source of BCs with high antioxidant potential.

3.6. Functional and technological capability

In terms of oil and water absorption capacity, we observed that both flours have similar capacities ($p < 0.05$) (Fig. 4A). Technological properties such as water and oil absorption capacity and foam formation predict flour quality for incorporation into food products (Chandra et al., 2015, Du et al., 2014). UCF and SCF did not show foam formation within 60 min, which can be attributed to the low solubility and protein content in these food matrices (Rodsamran & Sothornvit, 2018). Particle size can also influence these characteristics, indicating that finer particle flours would contribute to more efficient absorption (Xavier et al., 2022). Ulloa et al. (2017) identified in jackfruit seeds that their foam-forming capacity could be directly related to the types of proteins present, such as globular proteins, which are difficult to denature on the surface and cause low foam capacity.

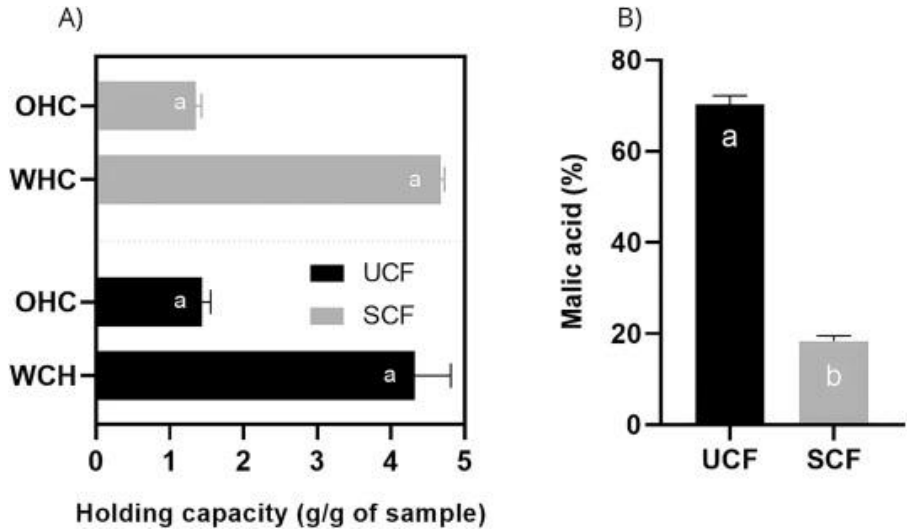

Fig. 4. Oil holding capacity and water holding capacity (4A) and titratable acidity content present in the umbu-caja and soursop co-product flours (4B). Values expressed as the mean of replicates ± standard deviation. Different letters differ by Student's t-test at a 5 % probability. Legend: UCF = Umbu-caja co-product flour; SCF = Soursop co-product flour.

The results of the pH analysis show a pH of 3.61 for SCF and a slightly more acidic pH (2.38) for UCF. Since pH measures the immediate concentration of hydrogen ions, titratable acidity measures the total amount of acids that can be neutralized (Lobit et al., 2002). In this respect, we observed that UCF had a higher titratable acidity (70.3 %) than SCF (18.4 %) ($p < 0.05$) (Fig. 4B). This is probably due mainly to the different chemical composition of the fruit, which interfered differently with the parameters of the flours analyzed (Silva Júnior et al., 2021). The high acidity suggests the application of these ingredients in products where acidification is desirable or necessary. This includes, for example, fermented products, where the acidic pH can favor the development of specific microbial cultures, or in baked goods, where acidity can interact with leavening agents to modify texture and flavor. Additionally, acidity can provide a natural preservative effect, increasing the shelf life of food products by inhibiting the growth of pathogenic and deteriorating microorganisms (Iuga & Mironeasa, 2020; Shiferaw Terefe & Augustin, 2020).

The main solid constituents are sugars (mainly glucose and fructose) and their concentration directly affects density, viscosity and refractive index. The refractive index for the diluted flours remained at 1.334, close to that of water, the dilution vehicle (1.332). This indicates that the flours have no soluble solids in aqueous medium.

4. CONCLUSION AND FUTURE PERSPECTIVES

The umbu-caja and soursop co-product flours showed promising qualities for incorporation into the human diet and product development, mainly due to their high fiber content, antioxidant capacity and low cytotoxicity. Soursop co-product flour stood out for its high content of soluble dietary fiber and condensed tannins, which makes it a promising alternative for future studies on intestinal health and microbiota. On the other hand, the umbu-caja co-product flour showed better antioxidant capacity, probably promoted by rutin in high concentrations, which merits further research into its use in reducing diseases and metabolic alterations with oxidative and inflammatory characteristics. Both flours had considerable amounts of the minerals calcium and iron, which should be explored in the future with the aim of studying the bioavailability of these minerals.

CRediT authorship contribution statement

Valeria Silva de Lana: Conceptualization, Methodology, Validation, Investigation, Formal analysis, Writing - Original Draft, Writing - Review & Editing. Patrícia Nayara Estevam: Conceptualization, Writing - Review & Editing. Thais Barcelos de Castro: Investigation, Writing - Review & Editing. Vinícius Parzanini Brilhante de São José: Conceptualization, Methodology, Writing - Review & Editing. Thais Carvalho Brito-Oliveira: . Pedro Henrique Santos: Writing – review & editing, Investigation. Cristiane Almeida Santos Oliveira: Investigation, Formal analysis. Cristiane Bani Corrêa: Investigation, Formal analysis. Mauricio Ariel Rostagno: Supervision, Methodology, Writing - Review & Editing. Hércia Stampini Duarte Martino: Conceptualization, Methodology, Resources, Project administration, Writing - Review & Editing. Izabela Maria Montezano de Carvalho: Conceptualization, Supervision, Methodology, Resources, Project administration, Funding acquisition, Writing - Original Draft.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This work was supported by the “Conselho Nacional de Desenvolvimento Científico e Tecnológico – CNPq” (CNPq/MCTI/FNDCT N° 18/2021, grant 422847/2021-6) and by the “Fundação de Amparo à Pesquisa do Estado de São Paulo - FAPESP” (grants 2018/14582-5; 2019/13496-0; 2023/11025-6; 2023/03439-5).

Appendix A. Supplementary data

The following are the Supplementary data to this article:

https://ars.els-cdn.com/content/image/1-s2.0-S0963996924015916-mmc1.docx

Supplementary Data 1.

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SUPPLEMENTARY MATERIAL

Nutritional and technological potential of umbu-caja and soursop co-product flours

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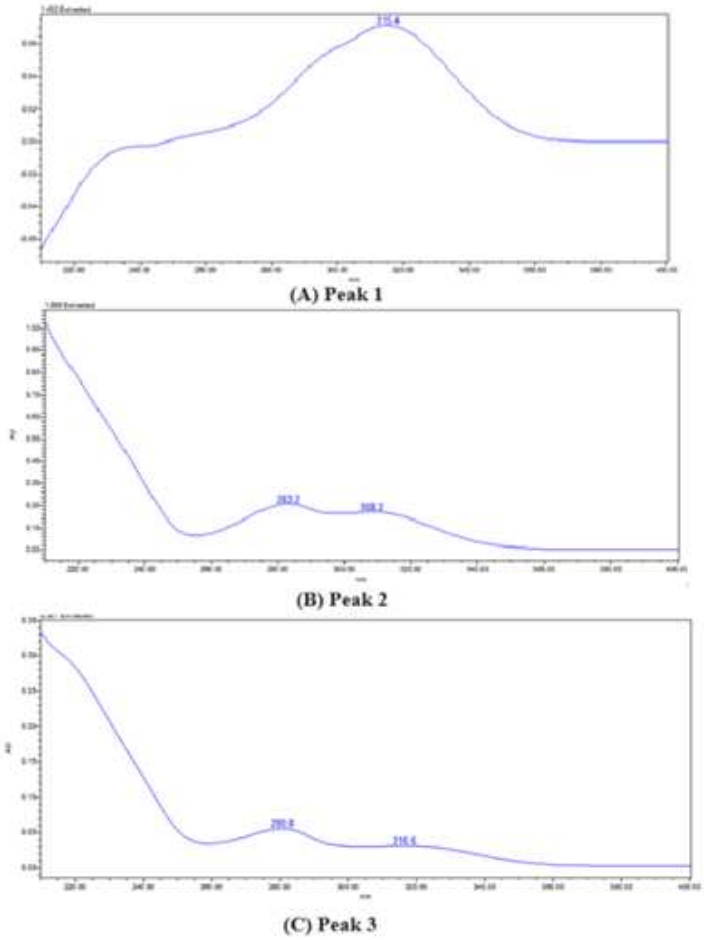
1. CHROMATOGRAMS

Figure S1. UV spectra of the three most significant peaks obtained from the chromatogram of soursop (*Annona muricata*) at 320 nm.

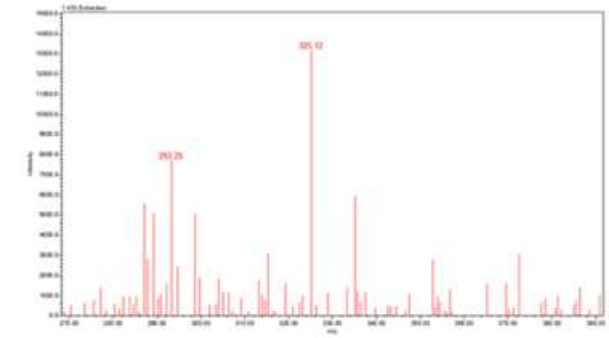


(A) Peak 1

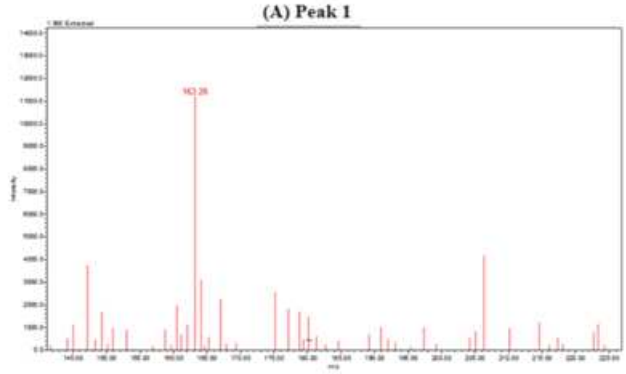
(B) Peak 2

(C) Peak 3

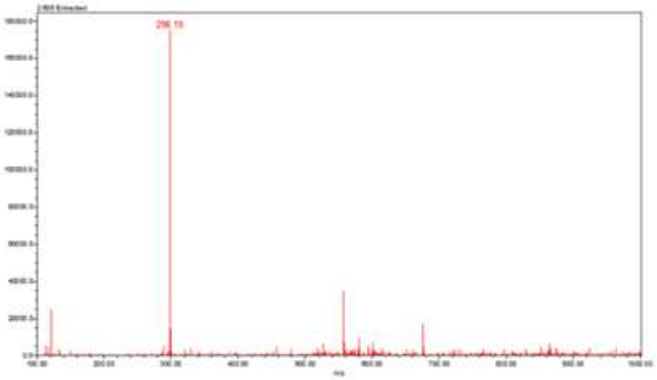
Figure S2. MS spectra of the three most significant peaks obtained from the chromatogram of soursop (*Annona muricata*) at 320 nm.



(A) Peak 1



(B) Peak 2



(C) Peak 3

Figure S3. Chromatogram (A), UV spectrum (B) and MS spectrum (C) of *p*-coumaric acid standard at 320 nm.

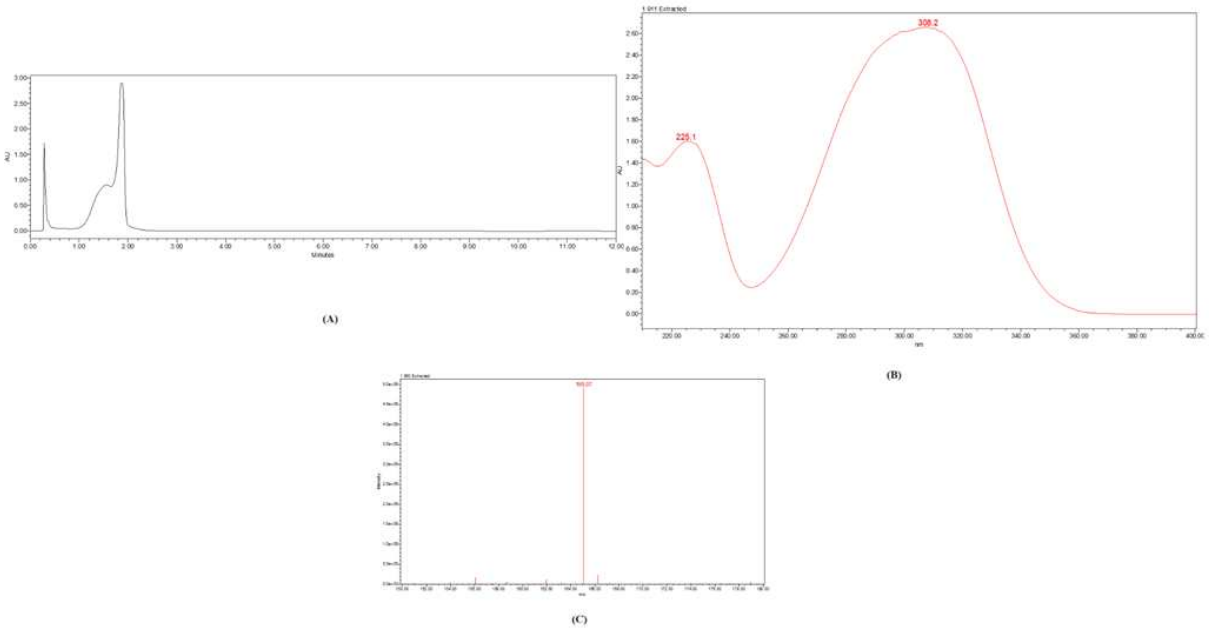


Figure S4. UV spectra of the three most significant peaks obtained from the chromatogram of umbu-cajá (*Spondias bahiensis*) at 320 nm

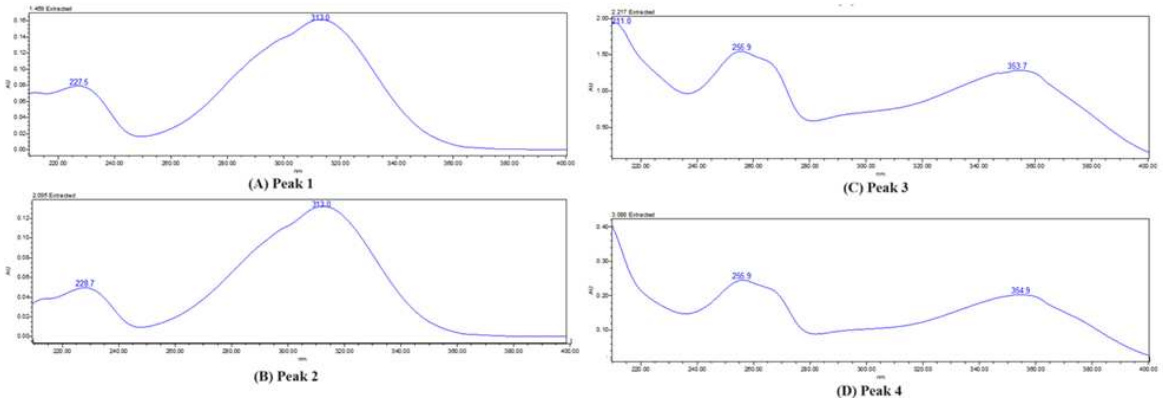


Figure S5. MS spectra of the three most significant peaks obtained from the chromatogram of umbu-cajá (*Spondias bahiensis*) at 320 nm

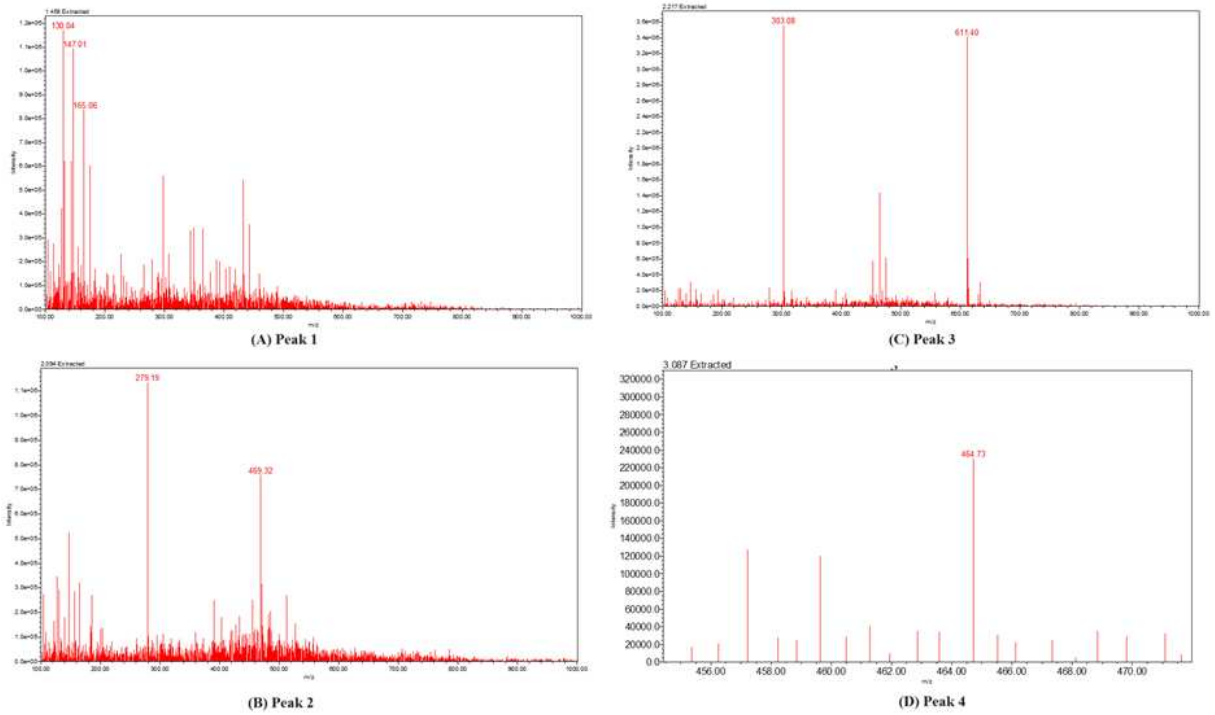
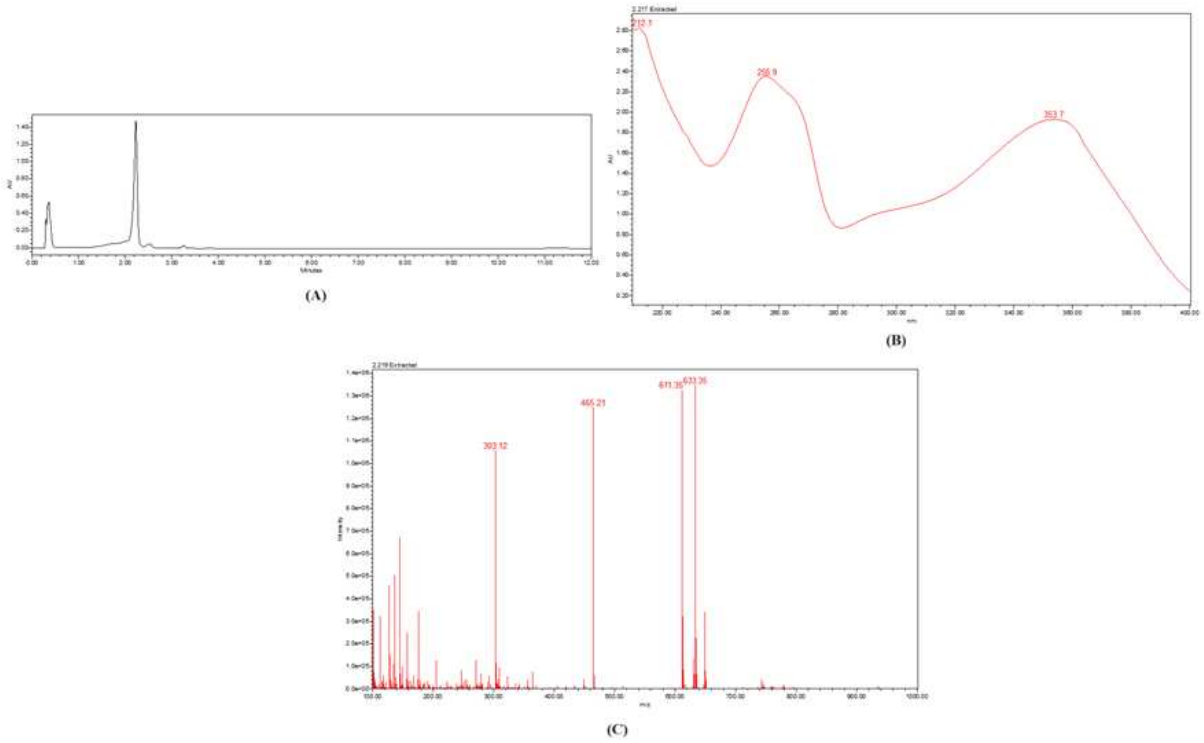


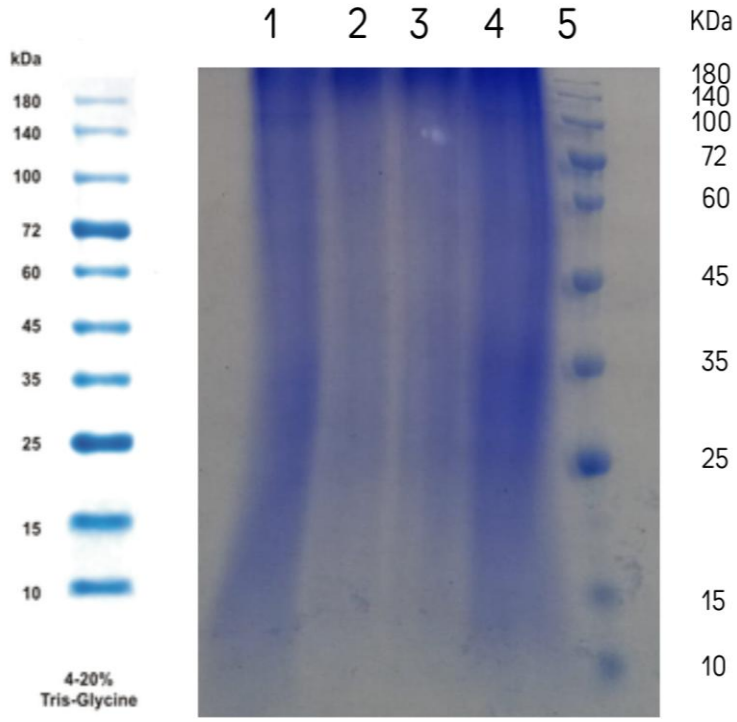
Figure S6. Chromatogram (A), UV spectrum (B) and MS spectrum (C) of Rutin standard at 320 nm.



2. ELECTROPHORESIS OF UMBU CAJA AND SOURSOP PROTEINS

10 mg of powdered extract was dissolved in 200 µL of SDS Sample Loading Buffer (5% Glycerol [v/v]; 2% SDS [w/v]; 0.2% bromophenol blue [w/v]; 4% β-mercaptoethanol [v/v]; 200 mM Tris-HCl pH 6.8) and incubated at 100 °C for 5 min. The samples were centrifuged and the supernatant was applied in a 10% polyacrylamide gel containing SDS. The Coomassie Colloidal staining method was used for protein visualization. The analysis showed dragging along the gel and no protein bands could be visualized, indicating a degraded sample.

Figure S7. Electrophoresis of umbu-caja and soursop proteins



Caption:

1 – 1 mg of soursop soluble extract

2 – 1mg of umbu-cajá soluble extract

3 – 1 mg of umbu-cajá soluble extract

4 – 1 mg of soursop soluble extract

5 – Prestained Protein Marker

5 CONCLUSÃO GERAL

A promoção de uma alimentação escolar rica em polifenóis demanda estratégias inovadoras e sustentáveis, de modo a aproveitar tanto alimentos *in natura* e minimamente processados quanto a valorização de produtos regionais. Os alimentos regionais, que são fontes importantes de polifenóis, contribuem para a diversificação dos cardápios escolares. No entanto, desafios como a ausência de frutas regionais nas refeições oferecidas demonstram a necessidade de ações mais efetivas na construção desses cardápios. Nesse contexto, o aproveitamento de coprodutos surge como uma alternativa viável para ampliar a presença desses frutos e garantir a oferta regular de polifenóis na alimentação escolar. A graviola e o umbu-cajá, frutas escolhidas para este trabalho, podem ser utilizadas na forma de polpa para o preparo de sucos, e os materiais que não são aproveitados (casca e bagaço) podem ser secos e transformados em farinha pelo processo de moagem. Esse material, mesmo após o processamento ainda é considerado como um produto minimamente processado e de fácil incorporação em preparações culinárias. Essa abordagem não apenas contribui para a diversificação e o enriquecimento nutricional dos cardápios, mas também promove a sustentabilidade e o aproveitamento integral dos alimentos, alinhando-se às diretrizes de uma alimentação saudável e ambientalmente responsável associado a uma oferta constante de polifenóis. Estudos futuros sobre a biodisponibilidade dos minerais presentes nas farinhas são importantes para verificar o quanto eles são aproveitados pelos sistemas biológicos. Além disso, devido a quantidade de fibras, tanto solúveis quanto insolúveis, estudos com saúde intestinal e microbiota são interessantes para verificar os efeitos da presença dessa matriz alimentar complexa no intestino.