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INSTITUTO DE CIÊNCIA E TECNOLOGIA DE ALIMENTOS

**PROGRAMA DE PÓS-GRADUAÇÃO EM CIÊNCIA E TECNOLOGIA DE
ALIMENTOS**

**Composição físico-química e de compostos bioativos de diferentes espécies de
maracujá, estabilidade do suco e aproveitamento da farinha da casca de maracujá
laranja**

LUZIA CAROLINE RAMOS DOS REIS

Porto Alegre, 2018.

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TESE
COMPOSIÇÃO FÍSICO-QUÍMICA E DE COMPOSTOS BIOATIVOS DE
DIFERENTES ESPÉCIES DE MARACUJÁ, ESTABILIDADE DO SUCO E
APROVEITAMENTO DA FARINHA DA CASCA DE MARACUJÁ LARANJA

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pelo carinho, por sempre me
incentivar a estudar e pela confiança
que depositam em mim.

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RESUMO

O cultivo do maracujá, uma fruta nativa do Brasil, tem apresentado acentuada expansão, proporcionando grande popularização entre os diferentes segmentos de consumo. O maracujá é uma fruta com alto teor de compostos bioativos, dentre eles se destacam principalmente os carotenoides, compostos fenólicos e flavonoides com capacidade antioxidante; sendo que tais compostos têm sido relatados por diminuir o risco de diversas doenças. Por estes fatores, este estudo teve como objetivo avaliar as características físico-químicas e teor de compostos bioativos da polpa, casca e semente de maracujá amarelo, roxo e laranja; além da polpa de quatro novas cultivares de maracujá azedo desenvolvidas pela EPAGRI e também avaliar a estabilidade do suco do maracujá laranja, bem como a produção de farinha a partir da casca da fruta para uso como ingrediente alimentar na formulação de produtos de panificação. Em relação aos maracujás amarelo, roxo e laranja: o maracujá amarelo foi o que apresentou maiores valores de pectina nas cascas; alto teor de β -caroteno (1334 $\mu\text{g}/100\text{ g}$) na polpa e maiores valores de cinzas e fibra alimentar total nas sementes (65,60 g/100 g). Por sua vez, o maracujá roxo mostrou um alto valor de antocianinas nas cascas e sementes e o laranja níveis mais elevados de cinzas, β -caroteno (716,32 $\mu\text{g}/100\text{ g}$) e campferol (229,27 mg/100 g) nas cascas, maiores teores de licopeno (4405 $\mu\text{g}/100\text{ g}$), luteína, zeaxantina, carotenoides totais e fenólicos nas polpas. Em relação à pesquisa com o suco pasteurizado recomenda-se o consumo até o quarto dia mantido sob refrigeração, devido à maior retenção de compostos bioativos, como fenólicos, quercetina, β -criptoxantina, carotenoides totais, licopeno e uma melhor capacidade antioxidante. No estudo com a polpa de quatro novas cultivares de maracujá observou-se que a cultivar ‘BRS Gigante Amarelo’ apresentou maior teor de fenólicos, epigallocatequina,

quercetina, todos os carotenoides (exceto zeaxantina) e provitamina A (367 $\mu\text{g}/100\text{ g}$). A cultivar ‘BRS Rubi do Cerrado’ exibiu maiores concentrações de campferol e cobre; a cultivar ‘BRS Sol do Cerrado’ apresentou maiores concentrações de todos os minerais (exceto cobre) e zeaxantina e as cultivares ‘SCS437 Catarina’ e ‘BRS Gigante Amarelo’ apresentaram maior capacidade antioxidante ABTS. Ao avaliar a farinha da casca do maracujá e a análise sensorial de bolos e pães produzidos com a mesma, observou-se que esta pode ser utilizada como ingrediente para o enriquecimento de produtos de panificação contribuindo com constituintes como fibra alimentar, minerais e compostos bioativos (principalmente β -caroteno), além de ter um índice de aceitação superior a 80%.

Palavras-chave: β -caroteno; licopeno; fibra alimentar total; minerais; análise sensorial; compostos fenólicos; flavonoides; pectina.

ABSTRACT

Passion fruit, a native fruit from Brazil, has presented a marked expansion, providing great popularization among the different segments of consumption. Passion fruit is a fruit with a high content of bioactive compounds, among which are mainly carotenoids, phenolic compounds and flavonoids with antioxidant capacity. These compounds have been reported to decrease the risk of various diseases. This study had the objective of evaluating the physicochemical characteristics and content of bioactive compounds of the pulp, peel, and seed of yellow, purple and orange passion fruit and the pulp of four new passion fruit cultivars developed by EPAGRI. In addition, it was evaluated the stability of orange passion fruit juice, as well as the production of flour from the peel of orange passion fruit for use as a food ingredient in the formulation of bakery products, since there is no data in the literature with this specie of passion fruit. The yellow passion fruit presented the highest values of pectin in the peels; high content of β -carotene (1334 $\mu\text{g}/100\text{ g}$) in the pulp and higher values of ash and total dietary fiber (65.60 $\text{g}/100\text{ g}$) in the seeds; the purple passion fruit showed a high value of anthocyanins in the peels and seeds and the orange passion fruit reported higher levels of ashes, β -carotene (716.32 $\mu\text{g}/100\text{ g}$), and kaempferol (229.27 $\text{mg}/100\text{ g}$) in the peel, higher lycopene (4405 $\mu\text{g}/100\text{ g}$), lutein, zeaxanthin, total carotenoids and phenolic content in the pulps. Regarding the research with pasteurized juice, if consumed until the fourth day under refrigeration, it is recommended because it retained higher content of bioactive compounds, such as phenolic, quercetin, β -cryptoxanthin, total carotenoids, lycopene, and better capacity antioxidant. Regarding of four new passion fruit cultivars, it was observed that the cultivar 'BRS Gigante Amarelo' presented higher phenolic content, epigallocatechin, quercetin, all carotenoids (except zeaxanthin), provitamin A

(367 $\mu\text{g}/100\text{ g}$). The 'BRS Rubi do Cerrado' cultivar showed higher concentrations of kaempferol and copper; the cultivar 'BRS Sol do Cerrado' presented higher concentrations of all minerals (except copper) and zeaxanthin and 'SCS437 Catarina' and 'BRS Gigante Amarelo' showed higher ABTS antioxidant capacity. When evaluating passion fruit peel flour and sensorial analysis with the same, it was observed that this can be used as an ingredient for the enrichment of baking products with constituents such as dietary fiber, minerals and bioactive compounds (mainly β -carotene) and sensory analysis showed that bread and cake formulations presented an acceptance rate higher than 80%.

Keywords: β -carotene; lycopene; total dietary fiber; minerals; sensory analysis; phenolic compounds; flavonoids; pectin.

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CAPÍTULO 1

1. Introdução

O maracujá, uma fruta nativa do Brasil, é uma espécie com elevada importância para as indústrias de alimentos e para os consumidores, sendo utilizado na alimentação na forma *in natura* (polpa) ou como suco ou néctares (principais formas de industrialização). Outros produtos industrializados obtidos a partir do maracujá são doces, geleias, batidas, gelados, licores, entre outros; o que torna essa fruta uma importante matéria-prima com relevância econômica para o país. Além disso, devido ao desenvolvimento de novas cultivares, o maracujá pode ser produzido em todas as épocas do ano.

Em relação ao valor nutricional e funcional, os constituintes essenciais e os compostos bioativos, presentes tanto na polpa quanto na casca da fruta (albedo e flavedo), estão relacionados à atividade biológica na saúde humana (MALACRIDA; JORGE, 2012; ALVES, 2013). Os compostos bioativos presentes no maracujá, flavonoides, carotenoides e compostos fenólicos, possuem propriedades benéficas à saúde e têm sido relacionados à redução do risco de doenças degenerativas, câncer, aterosclerose, degeneração macular relacionada ao envelhecimento, entre outros (DINIZ; ASTARITA; SANTAREM, 2007).

Maracujá é um nome popular das várias espécies do gênero *Passiflora* que pertence à família Passifloraceae, na qual existem mais de 500 espécies distribuídas em regiões de clima tropical e subtropical do mundo. A variedade *Passiflora edulis* Sims fo. *flavicarpa*, conhecido como maracujá azedo, é a mais produzida e comercializada,

representando 95% dos pomares. Seu cultivo é primariamente com foco na indústria de suco e polpa, especialmente devido a sua maior acidez e rendimento.

No entanto, as espécies *Passiflora edulis* Sims fo. *edulis* e *Passiflora caerulea*, conhecidos como maracujá roxo e laranja, respectivamente, têm o sabor mais doce, e são consumidos principalmente na forma *in natura* (ZERAİK; YARIWAKE, 2010).

A quantidade de resíduos gerados a partir da casca na indústria de processamento de suco de maracujá é significativa. As cascas (albedo e flavedo) são constituídas por carboidratos, proteínas, pectinas e apresentam alto teor de compostos bioativos. Sendo assim, há o potencial de uso das cascas (albedo e flavedo) no desenvolvimento de novos produtos tais como a farinha que pode ser usada tanto como aditivo alimentar ou servir de fonte de fitoquímicos específicos para aplicação em suplementos nutricionais e produtos farmacêuticos. O aproveitamento destes resíduos do processo agroindustrial tem impacto positivo nos setores industrial, econômico e ambiental (OLIVEIRA et al., 2002; AYALA-ZAVALA et al., 2011).

Por fim, a Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina (EPAGRI-SC) tem desenvolvido novas cultivares de maracujá com o intuito de apresentar uma maior produtividade e maiores níveis de resistência às principais doenças comuns no maracujazeiro (vírus, bactérias e fungos). Estas cultivares apresentam casca mais robusta, com maior resistência ao transporte. Porém, há a necessidade de estudos que possam indicar a composição físico-química, centesimal, parâmetros de cor, teores de minerais e de compostos bioativos destes frutos.

CAPÍTULO 2

2. Objetivos

2.1 Objetivo Geral

Avaliar as características físico-químicas e teor de compostos bioativos da polpa, casca e semente de maracujá amarelo, roxo e laranja; além da polpa de quatro novas cultivares de maracujá azedo desenvolvidas pela EPAGRI. Com o maracujá laranja também objetivou-se avaliar a estabilidade do suco, bem como a produção de farinha a partir da casca da fruta para uso como ingrediente alimentar em produtos de panificação.

2.2 Objetivos Específicos

- Determinar a composição físico-química, centesimal e de minerais da polpa, casca e sementes de diferentes espécies de maracujá: amarelo (*Passiflora edulis* Sims fo. *flavicarpa*), roxo (*Passiflora edulis* Sims fo. *edulis*) e laranja (*Passiflora caerulea*);
- Avaliar o rendimento da polpa de diferentes espécies de maracujá (amarelo, roxo e laranja);
- Avaliar o teor de pectina da casca de diferentes espécies de maracujá (amarelo, roxo e laranja);
- Identificar e quantificar o perfil de compostos fitoquímicos, tais como compostos fenólicos totais, flavonoides, carotenoides, antocianinas em polpa, casca e semente das diferentes espécies de maracujá (amarelo, roxo e laranja);

- Avaliar a capacidade antioxidante da polpa, casca e semente de diferentes espécies;
- Analisar a estabilidade do suco *in natura* e pasteurizado do maracujá laranja durante armazenamento sob refrigeração (8 °C): 0-4 dias para o fresco e 0-15 dias para o pasteurizado mediante análise dos parâmetros de cor, sólidos solúveis totais (SST), pH e acidez total titulável ATT);
- Analisar o teor de compostos bioativos como compostos fenólicos, flavonoides e carotenoides durante o armazenamento dos sucos de maracujá laranja *in natura* e pasteurizado;
- Avaliar a capacidade antioxidante dos sucos de maracujá laranja *in natura* e pasteurizado durante o armazenamento;
- Analisar a composição físico-química, parâmetros de cor, composição centesimal, teor de minerais, atividade antioxidante e teor de compostos bioativos da polpa das cultivares de maracujá azedo (polpa) produzidas pela EPAGRI-SC (Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina) ‘SCS437 Catarina’, ‘BRS Gigante Amarelo’, ‘BRS Rubi do Cerrado’ e ‘BRS Sol do Cerrado’;
- Produzir farinha a partir da casca do maracujá laranja utilizando diferentes temperaturas de secagem;
- Determinar a composição centesimal, parâmetros de cor, capacidade de retenção de água, capacidade de retenção de óleo e teor de minerais da farinha obtida a partir da casca do maracujá laranja;
- Identificar e quantificar o perfil de compostos fitoquímicos, tais como compostos fenólicos totais, flavonoides e carotenoides da farinha obtida da casca do maracujá laranja;

- Avaliar a capacidade antioxidante da farinha obtida da casca do maracujá laranja pelo método de desativação do radical ABTS;
- Realizar análise sensorial de pães e bolos acrescidos de farinha da casca do maracujá laranja e mucilagem de chia.

CAPÍTULO 3

3. Revisão Bibliográfica

3.1. Maracujá

Maracujá é um nome popular dado a várias espécies do gênero *Passiflora*, do qual há mais de 500 espécies distribuídas por regiões de clima tropical e subtropical do globo, sendo o Brasil o maior produtor com mais de 79 espécies. A produção nacional anual equivale a 923 mil toneladas, com destaque para as regiões produtoras Nordeste e Sudeste. A variedade *Passiflora edulis* flavicarpa, conhecida como maracujá azedo ou amarelo, é o mais produzido e comercializado, e representa 95% dos pomares. Seu cultivo está basicamente voltado para a indústria de sucos e polpas, em especial devido ao seu sabor mais ácido e maior rendimento. Enquanto as espécies *Passiflora edulis* flavicarpa edulis e *Passiflora caerulea*, mais conhecidas como maracujá roxo e laranja, respectivamente, apresentam sabor mais doce, portanto são mais consumidas em forma de suco ou como fruta (IBGE, 2012; ZERAIK; YARIWAKE, 2010; MELETTI; BRUKNER, 2001).

A polpa do maracujá apresenta grande quantidade de compostos antioxidantes, entre eles estão principalmente os compostos fenólicos, flavonoides, carotenoides e provitamina A. A casca do maracujá é composta pelo flavedo, que corresponde à camada externa de coloração amarela, roxa ou laranja, que é rica em fibras insolúveis e o albedo, que corresponde à camada interna branca, que é rica em fibra solúvel, em especial a pectina, com pequenas quantidades de mucilagens. Além disso, a casca (albedo e flavedo) do maracujá também apresenta em sua composição compostos

fenólicos, flavonoides e carotenoides com capacidade antioxidante (JANEIRO et al., 2008).

Diante do exposto, é de extrema importância analisar a composição físico-química, centesimal, teor de minerais, compostos fenólicos, flavonoides, carotenoides e a capacidade antioxidante da polpa e casca (albedo e flavedo) desta fruta, devido a associação dos seus benefícios à saúde dos seres humanos.

3.2 Compostos Bioativos Presentes no Maracujá

O maracujá é uma fruta climatérica de consumo frequente devido à cor e sabor que são atrativos. Esta fruta apresenta boa fonte de vitamina C, compostos fenólicos, tais como os ácidos fenólicos, flavonoides e carotenoides, também apresenta capacidade antioxidante. O maracujá também é rico em alguns minerais essenciais como potássio, fósforo, magnésio, cálcio e ferro. Existem muitos estudos afirmando que o consumo de maracujá tem um impacto positivo na saúde do ser humano (ZERAİK; YARIWAKE, 2010; KUSKOSKI et al., 2006).

3.2.1 Compostos Fenólicos

Os compostos fenólicos representam a maior categoria de fitoquímicos (mais de 8000 compostos já identificados) e estão amplamente distribuídos no reino vegetal. Os três grupos mais importantes entre os compostos fenólicos são os ácidos fenólicos, flavonoides e taninos. Os flavonoides representam o maior grupo de fenólicos presentes nas plantas e são os mais estudados. Os ácidos fenólicos formam um grupo diversificado que inclui os ácidos benzoicos e derivados (hidroxibenzoico, gálico, elágico, etc.) e os ácidos cinâmicos e derivados (cumárico, caféico, ferúlico, clorogênico etc.). Os polifenóis, que também podem ser chamados de taninos, são polímeros de alto

peso molecular e se dividem em duas classes: os taninos hidrolisáveis que incluem o ácido gálico e o elágico, onde podem ser encontrados em frutas como maracujá, uva e nozes e os taninos condensados, como catequinas e epicatequinas (KING; YOUNG, 1999).

A quantidade de compostos fenólicos no alimento pode variar conforme vários fatores como fertilização, tipo de solo, temperatura, genótipo, maturação, luz e água. No entanto, no tocante a absorção, metabolismo e biodisponibilidade no organismo, também podem ocorrer variações dessas classes de polifenóis, devido às suas diferenças estruturais (CELEP; RASTMANESH; MAROTTA, 2014).

Pesquisas têm destacado múltiplas funções e vários mecanismos importantes dos compostos fenólicos no organismo, os quais não estão relacionados somente a sua atividade antioxidante direta, mas também a habilidade destas substâncias de se ligarem a proteínas. Isto inclui a ligação à receptores celulares e transportadores de membrana e capacidade de influenciar a expressão gênica, sinalização e adesão celular (KROOW; WILLIAMSON, 2005). Em função disto, os compostos fenólicos podem também exercer outras funções de extrema importância no organismo humano (GIADA; FILHO, 2006).

3.2.1.1 Flavonoides

Os flavonoides representam o grupo mais comum de polifenóis vegetais e fornecem muito sabor e cor às frutas e hortaliças. Estes compostos são encontrados em frutas, leguminosas, nozes, sementes, ervas, especiarias, caules, flores, bem como chás e vinhos tinto. Os flavonoides são polifenóis bioativos de baixo peso molecular, são solúveis em água com 15 átomos de carbono e podem ser visualizados como dois anéis de benzeno que são unidos em conjunto com uma cadeia curta de três carbonos. As seis

principais subclasses de flavonoides incluem flavonois, flavononas, flavonas, flavanois, flavan-3-óis e isoflavonas, de acordo com as posições dos substitutos presentes na molécula (TANWAR; MODGIL, 2012).

Os flavonoides têm apresentado grande interesse científico por causa de seus efeitos benéficos sobre a saúde humana. Eles têm sido associados à atividade antiviral, antialérgica, antiplaquetária, anti-inflamatória, imunomoduladora, antitumoral e antioxidante. Alguns estudos sustentam um efeito protetor do consumo de flavonoides no diabetes, depressão, úlcera, artrite reumatoide, doença cardiovascular e câncer. Os efeitos destes compostos parecem estar relacionados com as várias atividades biológicas e farmacológicas (TANWAR; MODGIL, 2012; GONZÁLEZ-GALLEGO et al., 2014).

3.2.1.2 Antocianinas

As antocianinas são corantes naturais às quais há um interesse crescente devido à sua ampla gama de cores e efeitos benéficos à saúde. Elas são responsáveis por diversas cores nas plantas, como azul, roxo, violeta, magenta e vermelho. As principais fontes são encontradas em algumas frutas como: amora, uva, mirtilo, açai, ameixa, cereja, framboesa, maçã, morango e acerola e nas hortaliças, como o repolho-roxo, batata-roxa e berinjela (DAMODARAM; PARKIN; FENNEMA, 2010; CASTAÑEDA-OVANDO et al., 2009).

As antocianinas pertencem ao grupo dos flavonoides, devido a sua característica do esqueleto carbônico $C_6C_3C_6$. A cor das antocianinas vai depender da presença e do número de substituintes ligados à molécula. A estrutura básica das antocianinas é o 2-difenilbenzopirona do sal *flavylium*. As antocianinas ocorrem como glicosídeos de poli-hidroxi e/ou polimetoxi derivados do sal. Os açúcares mais comuns que as antocianinas estão ligadas são a glicose, ramnose, galactose, arabinose, xilose di e trissacarídeos

formados a partir desses açúcares. Elas compõem o maior grupo de pigmentos solúveis em água e existem cerca de 400 antocianinas diferentes, sendo que as principais antocianinas encontradas são a pelargonidina, cianidina, delphinidina, peonidina, petunidina e malvidina (DAMODARAM; PARKIN; FENNEMA, 2010).

As antocianinas estão associadas com uma modulação favorável da microbiota e marcadores inflamatórios, o que resulta na melhoria da capacidade da barreira intestinal em reduzir a translocação de lipopolissacarídeos na circulação. Também tem efeito protetor contra doenças neurodegenerativas e crônicas e atuam como inibidores de mutagênese e carcinogênese, devido ao seu poder antioxidante, atuando contra os radicais livres, apresentando propriedades farmacológicas sendo utilizadas para fins terapêuticos (MORAIS et al., 2016; CASTAÑEDA-OVANDO et al., 2009).

3.2.2 Carotenoides

Os carotenoides são pigmentos lipossolúveis presentes nas frutas e hortaliças. As propriedades antioxidantes destes compostos estão ligadas com a sua capacidade de desativar os radicais livres e o *oxigênio singlete*. Os carotenoides reagem com uma vasta gama de radicais fortemente oxidantes via mecanismo de transferência de elétrons que produzem o radical cátion. Também tem sido relatado que com radicais fracamente oxidantes, a reação prossegue através de um processo de abstração de hidrogênio. De um modo geral, existem três características principais que determinam a taxa e o tipo de mecanismo das reações dos carotenoides com os radicais livres: estrutura e potencial redox de um carotenoide e polaridade do meio (JOMOVA; VALKO, 2013).

Os carotenoides também são compostos que apresentam propriedades benéficas muito importantes para a saúde humana. Além de suas propriedades antioxidantes, os carotenoides desempenham uma função importante no organismo, como a sua atividade

pró-vitamínica A. Estudos epidemiológicos e populacionais têm demonstrado a importância destes fitoquímicos na diminuição do risco de muitas doenças como câncer, doenças cardiovasculares (catarata, hipertensão), osteoporose, infertilidade, enfisema, doenças neuro-degenerativas como Parkinson e Alzheimer, entre outros (RAO; RAO, 2007).

Alguns carotenoides merecem atenção especial, pois já são ligados na diminuição do risco de algumas doenças. Por exemplo, o aumento da ingestão de licopeno pode reduzir a progressão do câncer de próstata. A luteína e a zeaxantina ajudam na manutenção da saúde da visão (WOODSIDE et al., 2015). A criptoxantina também tem sido investigada e vários estudos relatam que este carotenoide impede hipertrofia dos adipócitos e obesidade; também reduz moderadamente o colesterol LDL (Lipoproteína de Baixa Densidade), colesterol total e o risco de osteoporose quando combinado com fitoesteróis (GRANADO-LORENCIO et al., 2014). Da mesma forma, o α -caroteno inibe a progressão de determinados tipos de câncer e o β -caroteno neutraliza danos dos radicais livres, pois tem a maior atividade de pró-vitamina A de todos os carotenoides (TAKAYANAGI; MUKAI, 2014; DOWNHAM; COLLINS, 2000; BAUERNFEIND, 1972).

3.2.3 Teor de Compostos Bioativos no Maracujá

A Tabela 1 apresenta o teor de compostos fenólicos de diferentes espécies e partes da fruta maracujá obtidos da literatura. Como se pode observar, há bastante variação no teor destes compostos entre os estudos realizados e isso pode ser explicado devido às diversidades de clima, região, tipo de solo e metodologia de análise escolhida para a realização do teor de compostos fenólicos. Cohen et al. (2008b) foram os autores que detectaram menores teores de compostos fenólicos em polpa de maracujá amarelo

doce (*Passiflora nitida*) - 17 mg GAE/100g em amostra fresca e Souza et al. (2012) encontraram maiores teores na polpa de maracujá amarelo doce (*Passiflora alata* Dryand) - 1545 mg GAE/100g em amostra seca. Observou-se que estes autores mencionados acima utilizaram métodos diferentes: Cohen et al. (2008b) realizaram testes com espectrofotometria e apresentaram os dados em amostra fresca e Souza et al. (2012) testes por Cromatografia Líquida de Alta Eficiência (CLAE), o melhor método de análise dos compostos fenólicos e apresentaram os dados em amostra seca.

Tabela 1. Teor de compostos fenólicos em diferentes espécies e partes de maracujá obtidas de diversos estudos.

Autor	Parte da fruta	Nome Científico	Nome Popular	Matéria*	Metodologia	Compostos Fenólicos
Kuskoski et al. (2006)	Polpa	<i>Passiflora sp</i>	Maracujá amarelo	Fresca	Espectrofotometria	20 mg GAE/100 g
Cohen et al. (2008a)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo híbrido do cerrado	Fresca	Espectrofotometria	36 mg GAE/100 g
Cohen et al. (2008b)	Polpa	<i>Passiflora alata</i>	Maracujá amarelo	Fresca	Espectrofotometria	46 mg GAE/100g
		<i>Passiflora nitida</i>	doce			17 mg GAE/100g
Janzantti et al. (2012)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo	Fresca	Espectrofotometria	529 mg GAE/100 mL
		Sims f. flavicarpa Deg				
Souza et al. (2012)	Polpa	<i>Passiflora alata</i>	Maracujá amarelo	Seca	CLAE**	1545 mg GAE/100g
		Dryand	doce			
Nachbar (2013)	Polpa	<i>Passiflora alata</i>	Maracujá amarelo	Fresca	Espectrofotometria	35 mg GAE/100 g
		Dryand	doce			36 mg GAE/100 g
		<i>Passiflora edulis</i>	Maracujá amarelo			36 mg GAE/100 g

		Sims f. flavicarpa	azedo			
		Deg				
Pongener et al. (2013)	Polpa	<i>Passiflora edulis</i> Sims	Maracujá roxo	Fresca	Espectrofotometria	89 mg GAE/100 mL
Silva et al. (2014)	Polpa	<i>Passiflora edulis</i> Sims	Maracujá amarelo	Seca	Espectrofotometria	765 mg GAE/100 g
	Semente					451 mg GAE/100 g
Septembre-Malaterre et al. (2016)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo	Fresca	Espectrofotometria	286 mg GAE/100 g

*Quando dados foram apresentados em matéria fresca os autores não informaram o teor de umidade das frutas analisados.

**CLAE: Cromatografia Líquida de Alta Eficiência.

A Tabela 2 apresenta os teores de flavonoides de diferentes espécies e partes da fruta maracujá obtidos da literatura. Como pode-se observar, Cohen et al. (2008a) obtiveram os menores teores de flavonoides totais: 3,28 mg /100 g de polpa de maracujá amarelo (matéria fresca) (*Passiflora edulis*). Já López-Vargas et al. (2013) encontraram as maiores concentrações de equivalente a rutina: 1363 mg/100 g de polpa e semente de maracujá amarelo (matéria fresca) (*Passiflora edulis* var. *Flavicarpa*). As diferenças observadas entre os autores mencionados acima podem ser justificadas pelo tipo de amostra, solvente e padrão utilizado para determinar o teor de flavonoides totais em maracujá.

Li et al. (2011) analisaram a composição de flavonoides em *Passiflora edulis* 'edulis' e *Passiflora edulis* 'flavicarpa' mais conhecidos como 'roxo' e 'amarelo'. Os cromatogramas revelaram que os seis flavonoides principais obtidos a partir de *Passiflora edulis* 'flavicarpa' não foram detectados em *Passiflora edulis* 'edulis', o que sugeriu que as duas populações são originalmente díspares e que a cor da fruta está intimamente correlacionada com algumas variabilidades das espécies.

Tabela 2. Teor de flavonoides em diferentes espécies e partes de maracujá obtidas de diversos estudos.

Autor	Parte da fruta	Nome Científico	Nome Popular	Matéria*	Metodologia	Flavonoides
Cohen et al. (2008a)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo híbrido do cerrado	Fresca	Espectrofotometria	3,28 mg flavonoides totais/100 g
Zeraik e Yariwake (2010)	Polpa	<i>Passiflora. edulis</i> Sims f. flavicarpa Degener)	Maracujá amarelo	Fresca	CLAE	16 mg isoorientina/L e 158 mg flavonoides totais/L de extrato
López-Vargas et al. (2013)	Polpa e semente Casca (albedo)	<i>Passiflora edulis</i> var. Flavicarpa	Maracujá amarelo	Fresca	Espectrofotometria	1363 mg rutina equivalente/100 g 512 mg de rutina equivalente/100 g
Nachbar (2013)	Polpa	<i>Passiflora alata</i> Dryand <i>Passiflora edulis</i> Sims f. flavicarpa Deg	Maracujá amarelo doce Maracujá amarelo azedo	Fresca	Espectrofotometria	16 mg de quercetina/100 g 20 mg de quercetina/100 g
Silva et al. (2014)	Polpa Semente	<i>Passiflora edulis</i> Sims	Maracujá amarelo	Seca	Espectrofotometria	60 mg flavonoides amarelos/100 g 43 mg flavonoides

Septembre-Malaterre et al. (2016)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo	Fresca	Espectrofotometria	amarelos/100 g 70 mg de quercetina equivalente/100 g
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*Quando dados foram apresentados em materia fresca os autores não informaram o teor de umidade das frutas analisados.

**CLAE: Cromatografia Líquida de Alta Eficiência.

A Tabela 3 apresenta o teor de carotenoides de diferentes espécies e partes da fruta maracujá obtidas da literatura. Como pode ser visto, Pertuzatti et al. (2015) detectaram diversos carotenoides na polpa de maracujá amarelo (*Passiflora edulis*), pois utilizou-se o método de CLAE que separa e, posteriormente, identifica e quantifica o carotenoide presente.

Wondracek et al. (2011) analisaram a composição de carotenoides por CLAE em Passifloras do cerrado: *Passiflora cincinnata* (maracujá do cerrado), *P. nitida* (maracujá-suspiro), *P. setacea* (maracujá do sono) e *P. edulis* (maracujá amarelo e maracujá roxo). As polpas de maracujá apresentaram neoxantina, violaxantina, *cis*-violaxantina, anteraxantina, luteína, zeaxantina, β -criptoxantina, poli-*cis*-caroteno, prolicopeno, *cis*- ζ -caroteno, *trans*- ζ -caroteno, *trans*- β -caroteno, 13-*cis*- β -caroteno e fitoflueno. Em geral os teores de carotenoides entre as espécies foram significativamente diferentes (Tabela 4). O maracujá amarelo apresentou maior atividade pró-vitamina A em comparação com as demais espécies.

Tabela 3. Teor de carotenoides em diferentes espécies e partes de maracujá obtidas de diversos estudos.

Autor	Parte da fruta	Nome Científico	Nome Popular	Matéria	Metodologia	Carotenoides
Silva e Mercadante (2002)	Polpa	<i>Passiflora flavicarpa</i>	<i>edulis</i> Maracujá amarelo	Fresca	CLAE	β -criptoxantina: 2,65 $\mu\text{g/g}$, prolicopeno: 3,02 $\mu\text{g/g}$, cis- ζ - caroteno: 7,38 $\mu\text{g/g}$, ζ -caroteno: 12,86 $\mu\text{g/g}$, β -caroteno: 13,35 $\mu\text{g/g}$ 8250 $\mu\text{g } \beta$ - caroteno/100g e 5478 μg licopeno/100 g
Souza et al. (2012)	Polpa	<i>Passiflora Dryand</i>	<i>alata</i> Maracujá amarelo doce	Seca	CLAE	1467 $\mu\text{g}/100 \text{ mL}$
Pongener et al. (2013)	Polpa	<i>Passiflora edulis</i>	<i>edulis</i> Maracujá roxo	Fresca	Espectrofotometria	111,6 μg carotenoides totais/100 g
Nachbar (2013)	Polpa	<i>Passiflora alata</i> Dryand <i>Passiflora edulis</i> Sims f. flavicarpa	Maracujá amarelo doce Maracujá amarelo azedo	Fresca (autor não informou umidade)	Espectrofotometria	269,1 μg carotenoides totais/100 g

Deg						
Silva et al. (2014)	Polpa	<i>Passiflora edulis</i> Sims	Maracujá amarelo	Seca	Espectrofotometria	1362 µg β-caroteno/100 g
	Semente					Licopeno: ND
Pertuzatti et al. (2015)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo	Seca	CLAE	58 µg β-caroteno/100 g
	Semente					Licopeno: ND
Septembre-Malaterre et al. (2016)	Polpa	<i>Passiflora edulis</i>	Maracujá amarelo	Fresca (autores não informaram umidade)	Espectrofotometria	4,9 µg luteína + zeaxantina, 122800 µg β-criptoxantina, 137 µg licopeno/100 g
	Semente					3829 µg β-caroteno equivalente/100 g

CLAE: Cromatografia Líquida de Alta Eficiência; ND: Não Detectado.

Tabela 4. Composição de carotenoides ($\mu\text{g/g}$) em polpas de maracujá de diferentes espécies.

Carotenoide	<i>Passiflora cincinnata</i>	<i>Passiflora nitida</i>	<i>Passiflora setácea</i>	<i>P. edulis flavicarpa</i>	<i>P. edulis edulis</i>
Neoxantina	NQ	-	-	-	-
Anteraxantina	NQ	NQ	NQ	NQ	NQ
Luteína	NQ	-	NQ	-	-
Zeaxantina	-	-	NQ	NQ	-
Fitoflueno	-	-	-	NQ	NQ
<i>trans</i> - violaxantina	0,02	-	-	0,5	-
<i>cis</i> - violaxantina	-	-	0,18	-	-
β - criptoxantina	-	-	-	0,24	0,20
Prolicopeno	-	-	-	3,03	5,90
Poli- <i>cis</i> - caroteno	-	-	-	1,30	3,40
<i>cis</i> - ζ -caroteno	-	-	-	6,28	12,10
<i>trans</i> - ζ - caroteno	-	-	-	5,40	10,95
<i>trans</i> - β - caroteno	0,05	0,005	0,66	2,84	2,60
13- <i>cis</i> - β - caroteno	-	-	0,08	0,38	-
Vitamina A ($\mu\text{g}/100\text{ g}$)	0,49	0,04	5,50	23,70	21,40

Fonte: Wondracek *et al.* (2011).

NQ: detectado, mas não quantificado.

3.3 Ação Antioxidante *In Vitro* de Extratos de Maracujá

O estresse oxidativo ocorre quando há um desequilíbrio entre a geração de Espécies Reativas de Oxigênio (EROS) e a sua remoção. As EROS incluem radicais livres como hidroxila, peróxido, ânion superóxido e outras espécies oxidativas como peróxido de hidrogênio e a molécula isolada de oxigênio gerada como consequência dos processos naturais das células aeróbias; assim, eles são comumente presentes na vida. Embora as células exibam sistemas defensivos sobre a neutralização de EROS, vários fatores como o tabaco, a ingestão excessiva de álcool, a poluição do ar, solventes orgânicos, vírus, infecções, pesticidas, entre outros, contribuem para o desequilíbrio entre a produção e desativação destas espécies reativas de oxigênio (SOARES, 2002; HALLIWELL; GUTTERIDGE, 2007; DAMODARAM; PARKIN; FENNEMA, 2010).

A ingestão de frutas e hortaliças, que são alimentos que contém grandes quantidades de compostos antioxidantes, podem auxiliar na remoção das EROS (DAMODARAM; PARKIN; FENNEMA, 2010).

O princípio da atividade antioxidante é baseado na disponibilidade de elétrons para neutralizar os radicais livres. Além disso, esta ação está relacionada com o número e a natureza do padrão de hidroxilação no anel aromático. A capacidade para atuar com o hidrogênio doador e a inibição da oxidação são reforçados pelo aumento do número de grupos hidroxila no anel fenol (GULÇIN, 2012).

Kuskoski et al. (2006) avaliaram a atividade antioxidante de polpa de maracujá amarelo (*Passiflora* sp) utilizando o equivalente de Trolox e encontraram 0,9 µmol de Trolox/g de amostra fresca.

Souza et al. (2012) avaliaram a atividade antioxidante ABTS (2,2'-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) em polpa de maracujá amarelo doce (*Passiflora alata* Dryand) e encontraram 10,84 µmol de trolox equivalente/g de polpa fresca.

Nachbar (2013) avaliou a atividade antioxidante de duas espécies de maracujá amarelo: *Passiflora alata* Dryand (maracujá doce) e *Passiflora edulis* Sims f. *flavicarpa* Deg (maracujá azedo). Os resultados de atividade antioxidante encontrados para a espécie *Passiflora alata* Dryand apresentaram média de 50,14 % de atividade de sequestro do radical livre DPPH (radical 2,2-difenil-1-picrilhidrazil), enquanto a espécie *Passiflora edulis* Sims f. *flavicarpa* Deg. apresentou atividade de sequestro do radical livre DPPH de 53,70 %.

López-Vargas et al. (2013) também avaliaram a capacidade antioxidante através do radical DPPH em polpa, casca (albedo) e semente de maracujá-amarelo (*Passiflora edulis* var. *Flavicarpa*). Eles concluíram que as amostras de casca (albedo) mostraram uma capacidade superior para inibir o radical DPPH do que as amostras de polpa e sementes.

Alves (2013) avaliou efeito bloqueador de radicais livres de DPPH em extratos obtidos a partir de polpa, casca (albedo e flavedo) e semente de maracujá-roxo (*Passiflora edulis* Sims *edulis*). As matrizes que apresentaram melhor capacidade de inibição, por ordem decrescente, foram as cascas (albedo e flavedo) e as sementes, atingindo EC50 (concentração efetiva a 50 % de inibição de radicais de DPPH) 0,35 e 0,41mg/mL de extrato, respectivamente. O menor efeito bloqueador de radicais livres de DPPH foi obtido na polpa, onde se obteve um EC50 = 12,09 mg/mL de extrato.

Cazarin et al. (2014) analisaram a atividade antioxidante da farinha de casca (albedo e flavedo) de maracujá amarelo (*Passiflora edulis*) através das técnicas DPPH (radical 2,2-difenil-1-picrilhidrazil), FRAP (capacidade antioxidante de redução do ferro) em extrato aquoso e ORAC (capacidade de absorção do radical oxigênio), metanólico/acetona e etanólico. Para o extrato aquoso, as atividades antioxidantes DPPH, FRAP e ORAC foram: 46,35%, 36,56 e 40,83 μ mol Trolox equivalentes/g de amostra, respectivamente. Para o extrato metanólico/acetona as atividades antioxidantes

DPPH, FRAP e ORAC foram de 32,5 %, 38,65 e 68,58 μmol Trolox equivalentes/g de amostra. Para o extrato etanólico as atividades antioxidantes DPPH, FRAP e ORAC foram de 29,6 %, 34,95 e 63,48 μmol Trolox equivalentes/g de amostra. Observou-se que para cada extrato utilizado/testado, os radicais comportaram-se com aspectos divergentes: para o DPPH, o extrato aquoso conseguiu maior extração da atividade antioxidante na fruta, para o FRAP não houve muita variação no tipo de solvente utilizado na extração da atividade antioxidante e para o ORAC, tanto os extratos metanólico/acetona quanto o etanólico, apresentaram maior atividade antioxidante comparados com o extrato aquoso.

Souza et al. (2012) avaliaram a capacidade antioxidante de várias frutas e em ordem crescente de capacidade antioxidante nas polpas foram listadas o jenipapo, o maracujá doce, a graviola, o murici e o marolo.

Septembre-Malaterre et al. (2016) observaram que a maior capacidade antioxidante foi encontrada na polpa do maracujá (*Passiflora edulis*) (64% de redução do DPPH) quando comparado com outras frutas, incluindo manga, abacaxi, banana e lichia que exerceram valores inferiores no sequestro do radical (45-58 %). De acordo com dados dos ensaios DPPH, o maracujá exerceu também a maior capacidade antioxidante através da capacidade de absorção dos radicais oxigenados (ORAC) (14,08 mM Trolox equivalente).

3.4 Minerais

Como os macronutrientes, as vitaminas e as enzimas, os minerais também são nutrientes essenciais para a vida. Os minerais são substâncias inorgânicas, presentes em todos os tecidos e fluidos corporais e a sua presença é necessária para a manutenção de certos processos físico-químicos que são essenciais para vida (SOETAN; OLAIYA; OYEWOLE, 2010).

O ciclo de vida desses nutrientes começa no solo, que é sua fonte principal. O solo fornece minerais para plantas e através delas, os minerais vão para os animais e seres humanos. Embora os produtos de origem animal também sejam fonte de minerais para o organismo humano, os vegetais contêm quase todos os minerais estabelecidos como essenciais para a nutrição. Os minerais, principalmente o cálcio, fornecem grande parte da estrutura esquelética, por exemplo, ossos e dentes, além de serem fundamentais para os processos fisiológicos, com atuação como co-fatores essenciais de uma série de enzimas. Os seres humanos não podem utilizar a maioria dos alimentos, sem a presença dos minerais e das enzimas, pois estes são responsáveis pela digestão e absorção. Em determinadas situações ou condições fisiológicas (como o estresse, tabagismo, poluição, drogas e consumo de álcool), a necessidade destes micronutrientes são ainda maiores, sendo necessária uma maior ingestão através da dieta (GUPTA; GUPTA, 2014; CILLA, 2014).

Gondim et al. (2005) determinaram a concentração de minerais em casca (albedo e flavedo) de maracujá amarelo (*Passiflora edulis*), utilizando espectrômetro de absorção atômica. Os autores relataram valores de 44,51 mg de cálcio, 0,89 mg de ferro, 43,77 mg de sódio, 27,82 mg de magnésio, 0,32 mg de zinco, 0,04 mg de cobre e 178,40 mg de potássio em 100 g de amostra fresca.

Felipe et al. (2006) analisaram o conteúdo de minerais (cálcio, sódio, potássio, cobre, ferro, zinco e manganês) em pó de casca (albedo e flavedo) de maracujá amarelo. Para a obtenção do pó, as cascas foram desidratadas em estufa a vácuo sob temperatura de 65 °C. Para a quantificação dos minerais de cálcio, sódio e potássio foi empregado a fotometria de chama enquanto que para os minerais de ferro, manganês, cobre e zinco, foi empregada a técnica de espectrofotometria de absorção atômica. Os resultados foram os seguintes: 58,65 mg de cálcio/100 g, 504,43 mg de sódio/100 g, 690,02 mg de

potássio/100 g, 1,41 mg de cobre/100 g, 13,52 mg de ferro/100 g, 1,82 mg de zinco/100 g e 1,26 mg de manganês/100 g.

Reolon, Braga e Salibe (2009) analisaram o conteúdo de minerais em casca (albedo e flavedo) de maracujá amarelo (*Passiflora edulis* f. *flavicarpa*) variedade 'AC275 Maravilha'. Para a determinação dos minerais, utilizou-se espectrofotômetro UV-Visível (420 nm) para o fósforo, segundo método de colorimetria do metavanadato de amônio. Os demais minerais foram determinados em espectrômetro de absorção atômica, modalidade chama. Os resultados encontrados em matéria seca foram de 0,14 mg de fósforo/100 g 44,95 mg de cálcio/100 g, 1,17 mg de magnésio/100 g, 0,15 mg de cobre/100 g, 0,97 mg de zinco/100 g, 1,29 mg de manganês/100 g e 66,37 mg de ferro/100 g.

Souza et al. (2012) avaliaram o teor de minerais (fósforo, potássio, cálcio, magnésio e ferro) em polpa de maracujá doce (*Passiflora alata* Dryand). A quantificação dos elementos foi realizada por espectrofotometria usando uma curva padrão para cada mineral: para determinar a concentração de cálcio, ferro e manganês, foi utilizado um espectrofotômetro de absorção atômica com acetileno; um fotômetro de chama foi utilizado para determinação do potássio (768 nm) e um espectrofotômetro de luz visível foi usado para determinação do fósforo (420 nm). Os autores encontraram valores de 34,95 mg para o fósforo, 375,42 mg para o potássio, 4,76 mg para o cálcio, 19,82 mg para o manganês e 1,06 mg para o ferro em 100 g de polpa fresca.

As diferenças encontradas no teor de minerais apresentadas acima podem ser justificadas: pela diferença de espécies e cultivares, tipo de solo, clima, fertilização da planta, diferentes métodos e solventes utilizados na extração destes minerais, apresentação dos resultados em matéria fresca ou seca.

3.5 Pectina

A pectina é um heteropolissacarídeo obtido a partir de paredes celulares primárias das plantas terrestres, que é uma matéria-prima muito importante para produtos alimentares e farmacêuticos. Este carboidrato possui uma capacidade única de formar géis espalháveis, na presença de açúcar e ácido ou na presença de íons cálcio, sendo usado principalmente nesses tipos de aplicações (produtos alimentares e farmacêuticos). A estrutura principal e fundamental de todas as moléculas de pectina é uma cadeia linear de unidades de ácido α -D-galacturônico unidas por ligações α -1,4 e ligadas a resíduos de L-ramnose α -1,2 (SEIXAS et al., 2014; DAMODARAM; PARKIN; FENNEMA, 2010).

A pectina é encontrada em frutas, especialmente na casca, como é o exemplo da casca (albedo) de maracujá que representa aproximadamente 50 % do peso total da fruta. A casca (albedo) do maracujá apresenta 10 a 20 % de pectina de qualidade semelhante a da laranja. A pectina do maracujá é constituída de 76 a 78 % de ácido galacturônico, 9 % do grupo metoxila, baixas concentrações de galactose e arabinose; tem propriedades geleificantes e pode ser comparada à pectina dos citros, sendo utilizada como ingrediente funcional na formulação de geleias e sobremesas (MANICA, 1981).

A pectina é uma fibra solúvel sendo muito importante o seu consumo, pois ajuda na regulação do trato gastrointestinal e pode evitar distúrbios relacionados ao intestino como é o caso da diverticulite e hemorroidas. Também protege contra o câncer de cólon ao fazer as fezes passarem mais rapidamente pelo intestino, com redução do contato com agentes cancerígenos; pode reduzir os níveis de colesterol no sangue que consequentemente reduz o risco de doenças cardiovasculares decorrentes de arteriosclerose (formação de placa de gordura nas artérias); pode ajudar a controlar os níveis de glicose no sangue. As fibras solúveis dão a sensação de saciedade e ajudam a

reduzir ou controlar o peso (DAMODARAM; PARKIN; FENNEMA, 2010; VITOLO, 2008; SCHWARCZ; BERKOFF, 2006).

A Figura 1 apresenta a estimativa das aplicações da pectina no mercado global. Observa-se que a maioria das pectinas (37 %) é utilizada para produção de compotas, seguida por bebidas de leite acidificadas (21 % do total). Já os preparados de fruta e bebidas de fruta correspondem a 12 e 9 % do total da utilização, respectivamente, uma importante aplicação na indústria alimentar e que apresenta rápido crescimento. A indústria farmacêutica e de produtos de higiene pessoal são responsáveis pelo consumo de 6 % da produção de pectinas, confeitaria, produtos lácteos e outros produtos alimentares com 5 % e, por último, 2 % para a indústria de panificação (COIMBRA, 2010).

Liew et al. (2014) avaliaram a extração da pectina da casca (albedo e flavedo) de maracujá amarelo (*Passiflora edulis* f. *flavicarpa*) com ácido cítrico. Os autores encontraram 14,60 % de rendimento de pectina da casca do maracujá.

Kulkarni e Vijayanand (2010) também realizaram um estudo para avaliar o teor de pectina em casca (albedo e flavedo) de maracujá (*Passiflora edulis* f. *flavicarpa* L.) variedade amarela. As cascas de maracujá foram desidratadas para os experimentos da extração da pectina. As condições otimizadas para a extração da pectina a partir da casca do maracujá amarelo promoveram um rendimento de 14,80 g/100 g de cascas secas.

Em outra pesquisa realizada por Seixas et al. (2014), foi investigada a extração de pectina (albedo e flavedo) a partir das cascas secas de maracujá-amarelo (*Passiflora edulis* f. *flavicarpa*), sendo que o maior rendimento encontrado foi de 18,20 %.

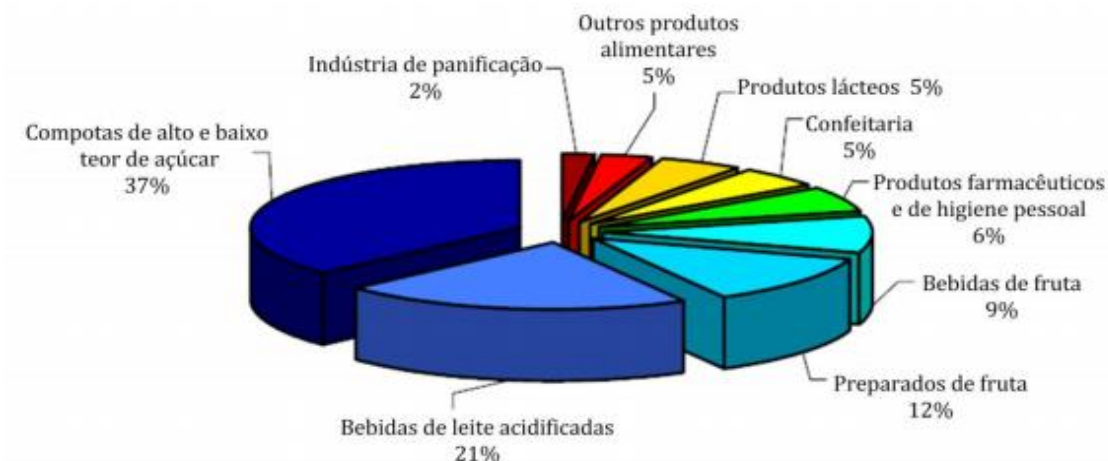


Figura 1. Estimativa das aplicações da pectina no mercado global.

*Adaptado de Coimbra (2010).

3.6 Suco de maracujá

Uma ampla variedade de sucos de frutas está disponível no mercado para atrair os consumidores. O processamento do suco ocorre logo após a colheita, sendo a pasteurização muito importante, pois a combinação adequada de temperatura e tempo permite a destruição de micro-organismos e inativação de enzimas, com aumento da vida de prateleira do produto. No entanto, em condições impróprias, o binômio tempo/temperatura podem comprometer a cor, o conteúdo de compostos bioativos e outras características nutricionais do suco (ASHURST, 2016; FELLOWS, 2006).

O suco integral de fruta é definido pela legislação brasileira, Instrução Normativa nº 01/00, como “bebida não-fermentada e não-diluída, obtida da parte comestível, no caso do maracujá (*Passiflora*, spp.), por meio de processo tecnológico adequado”. Deverão apresentar características de odor e sabor próprios da fruta. A coloração do suco de maracujá varia da cor amarela à alaranjada. Para o suco integral de maracujá, a legislação brasileira define os seguintes limites: mínimo 2,5 g/100 g de acidez total em ácido cítrico; mínimo de 11 de sólidos solúveis (°Brix a 20 °C); e máximo de 18 g/100 g de açúcares totais, naturais do maracujá (BRASIL 2000).

Saron et al. (2007) estudaram a estabilidade das características sensoriais e físico-químicas de suco de maracujá pronto para beber acondicionado em latas de aço, com três sistemas de revestimento orgânico interno e condicionado a 25 e 35 °C durante 360 dias. Ocorreu um acentuado decréscimo do conteúdo de ácido ascórbico até os 180 dias, mantendo-se estável até 360 dias em todas as condições estudadas. Em relação à cor, observou-se um escurecimento do suco até os 120 dias e posteriormente sua descoloração, entre os 300 e 360 dias, tanto a 25 quanto a 35 °C. As principais alterações verificadas no produto ao longo da estocagem foram associadas às alterações intrínsecas à bebida e não à interação suco/embalagem. A análise sensorial utilizando os atributos de aroma, perda de qualidade, sabor residual, estranho, metálico, aguado, oxidado, amargo, doce, ácido e maracujá, indicou que após os 360 dias de estocagem, o suco apresentou boas condições de consumo, para todas as variáveis de temperatura e latas avaliadas, demonstrando que os três sistemas podem ser utilizados para acondicionamento de suco de maracujá pronto para beber, garantindo que, do ponto de vista de qualidade sensorial, a bebida apresente uma vida-de-prateleira mínima de um ano.

Nogueira (2011) avaliou a estabilidade de ácido L-ascórbico em suco de maracujá (*Passiflora edulis* f. *flavicarpa*) durante 26 horas de armazenamento em temperatura de 5 a 7 °C. Houve uma redução significativa de 21,5 % de ácido L-ascórbico após o período de armazenamento (26 horas).

Cândido Filho e Bergamasco (2014) avaliaram a estabilidade de armazenamento do néctar de maracujá (*Passiflora edulis* f. *flavicarpa*) enriquecido com β -ciclodextrina (para reduzir a perda de qualidade do néctar, pois apresenta uma superfície externa hidrofílica e uma cavidade hidrofóbica, capaz de formar complexo de inclusão com uma variedade de moléculas, como é o caso, o maracujá), durante 30 dias à temperatura ambiente. Os resultados mostraram que o tempo de armazenamento afetou a qualidade

nutricional do néctar de maracujá e que a adição de β - ciclodextrina não teve um efeito negativo nos parâmetros analisados. O teor de sólidos solúveis totais e a luminosidade das amostras se manteve constante durante o período de armazenamento. Porém houve uma redução no teor de provitamina A, acidez titulável, pH e nos parâmetros de cor a^* e b^* durante 30 dias de armazenamento.

Borges et al. (2017) avaliaram o efeito da forma de armazenamento na estabilidade físico-química do suco de maracujá amarelo (*Passiflora edulis*) durante 48 horas após sua elaboração em ambiente refrigerado (4 ± 1 °C) e em temperatura ambiente (22 ± 1 °C), ambas com e sem luminosidade. O tempo de armazenamento influencia negativamente na qualidade do suco de maracujá-amarelo provocando a diminuição dos teores de ácido cítrico e vitamina C, aumento do pH e conteúdo de sólidos solúveis, além de promover alterações na cor, principalmente na luminosidade. Entretanto se armazenado em ambiente refrigerado e sem luz o suco conserva suas características iniciais por mais tempo (observado no armazenamento sob refrigeração a 4 °C).

3.7 Cultivares de maracujá desenvolvidas pela EPAGRI

A estação experimental EPAGRI-SC (Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina) desenvolveu em 2015 a cultivar de maracujá azedo 'SCS437 Catarina', e a EMBRAPA (Empresa Brasileira de Pesquisa Agropecuária) desenvolveu as cultivares 'BRS Gigante Amarelo', 'BRS Rubi do Cerrado' e 'BRS Sol do Cerrado'. As cultivares SCS437 Catarina, BRS Gigante Amarelo e BRS Sol do Cerrado possuem características de casca de cor amarela, polpa amarelo-alaranjada e sementes pretas. Já a cultivar BRS Rubi do Cerrado apresenta 50% de frutas de casca vermelha ou arroxeadas, polpa amarelo-alaranjada e sementes pretas (EMBRAPA 2014).

Estas Empresas de Pesquisa, com o intuito de avaliar e desenvolver essas novas cultivares, tem o objetivo de apresentar frutas para o comércio e indústrias com maior rendimento em polpa, maior produtividade e também apresentar maior resistência durante o desenvolvimento da fruta ainda no maracujazeiro contra as doenças que podem vir a se instalar na fruta.

Portanto torna-se necessário também avaliar o potencial nutricional e funcional de tais cultivares para fornecer maiores informações ao consumidor sobre a quantidade e o tipo de composto presente, podendo este escolher a cultivar que lhe apresentar maior interesse.

3.8 Farinha da casca do maracujá

Os subprodutos (cascas) obtidos através do processamento de frutas são excelentes fontes de antioxidantes devido à presença de compostos bioativos, sendo que a exploração desses recursos renováveis abundantes e de baixo custo podem ser utilizados nas indústrias farmacêutica e de alimentos, no desenvolvimento de novos produtos nutracêuticos e/ou farmacêuticos, o que pode reduzir os resíduos industriais e os custos, tendo como resultado um impacto econômico e ambiental positivo (SILVA et al. 2014).

A farinha da casca de maracujá tem um enorme potencial para ser incluída na dieta como fonte de fibra, com possibilidade de uso para o enriquecimento de produtos como pães, biscoitos e barras de cereais. O uso de tal ingrediente pode melhorar as qualidades nutricionais e tecnológicas dos produtos finais, além de reduzir os subprodutos da indústria alimentícia (de SOUZA et al. 2008).

Martínez et al. (2012) avaliaram a composição centesimal de co-produtos concentrados de fibra alimentar (casca, polpa e semente) de maracujá (*Passiflora edulis* L., cv. Flocarpa) e relataram valores de 9,3 g/100 g de umidade, 6,5 g/100 g de

carboidratos, 0,8 g/100 g de gordura, 5 g/100 g de cinzas, 4 g/100 g de proteínas e 81,5g/100 g de fibra dietética total (dados expressos em matéria seca). Em relação à capacidade de retenção de água e óleo da farinha, os valores foram de 13,5 g/g e 0,9, respectivamente. Para compostos fenólicos, os resultados foram de 150 mg de ácido gálico/100 g.

Os autores ainda determinaram a capacidade antioxidante *in vitro* e encontraram valores de 5,5 e 5,1 μM TE (equivalentes Trolox)/g para ABTS e DPPH, respectivamente. Os concentrados de fibra obtidos a partir da fruta mostraram-se ricos em compostos fenólicos contendo uma variedade de grupos hidroxila fenólicos que podem ser responsáveis pela capacidade antioxidante e pela atividade sequestradora de radicais livres.

Morais et al. (2015) relataram baixos níveis de fenólicos (86,74 mg equivalentes de ácido gálico/100 g) e flavonoides (35,13 mg equivalentes de quercetina/100 g) em casca de maracujá (*Passiflora edulis*) seca em forno com circulação de ar a 60 °C por 48 horas. Também realizaram a análise de capacidade antioxidante utilizando-se o IC 50, obtendo o valor de 37 mg/100 mL de IC 50 (concentração de extrato necessária para inibir 50 % do radical DPPH).

Santos et al. (2015) avaliaram a farinha de casca de maracujá e obtiveram 5,73% de umidade, 6,62 % de cinza, 8,93 % de proteínas, 1,45 % de lipídios, 77,27 % de carboidratos e 53,94 % de fibra alimentar total.

Lima et al. (2016) analisaram a composição centesimal da farinha de casca de maracujá (*Passiflora edulis*) e encontraram 7,42 % de umidade, 6 % de cinzas, 3,39 % de lipídios, 8,87 % de proteínas, 74,32 % de carboidratos e 60,08 % de fibra alimentar total. Também encontraram 37 e 308 mg/100 g para ácido fítico e taninos, respectivamente.

Coelho et al. (2017) avaliaram os parâmetros de cor na farinha de casca de maracujá e encontraram 36,71 para luminosidade (L^*), 7,53 para componente vermelho-verde (a^*) e 24,57 para componente amarelo-azul (b^*). O L^* indica a luminosidade e quanto mais próximo do 100 o valor, mais clara é a amostra. A coordenada vermelho/verde (a^*), quanto mais próxima o valor de 60, indica a cor vermelha e quanto menor o valor, indica a cor verde. Já para a coordenada b^* , quanto mais próximo o valor de 60, indica a cor amarela e quanto menor o valor, refere-se a cor azul.

Diante do exposto, a farinha apresenta potencial para uso como ingrediente em diferentes formulações, enriquecidas principalmente em fibras, minerais e compostos bioativos.

CAPÍTULO 4**ARTIGO 1****Antioxidant potential and physicochemical characterization of yellow, purple and orange passion fruit**

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Antioxidant potential and physicochemical characterization of yellow, purple and orange passion fruit

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Abstract This study evaluated yellow, purple and orange passion fruit in pulp, peel, and seed for physicochemical characteristics, proximate composition, minerals, antioxidant capacity (DPPH and ABTS), phenolic compounds, carotenoids, flavonoids and anthocyanins. Yellow passion fruit presented higher concentrations of pectin (37.37 g/100 g) in peels; high cryptoxanthin, α -carotene, β -carotene, provitamin A, quercetin, and kaempferol in pulps and higher values of ash and total dietary fiber in seeds. The purple fruit was highlighted by a great value of anthocyanins (103.68 mg/100 g) in peels and seeds and the orange fruit reported higher levels of ash, carotenoids (mainly β -carotene with 21,274 μ g/100 g), kaempferol in peels, higher contents of total soluble solids, lycopene (4405 μ g/100 g), lutein, zeaxanthin, total carotenoids in

pulps and phenolics in general. This research revealed that the pulp of passion fruit and his residues have a significant content of bioactive compounds, differing in type according the species analyzed.

Keywords Pectin · Total dietary fiber · Lycopene · Quercetin · Anthocyanin

Introduction

Passion fruit is a popular name given to several species of the genus *Passiflora* that belongs to *Passifloraceae* family, which there are more than 500 species distributed in regions of tropical and subtropical climate of the world. The variety *Passiflora edulis* Sims fo. *flavicarpa*, known as sour or passion fruit, is the most produced and marketed, and represents 95% of its fruit farm. Its cultivation is primarily focusing on the juice and pulp industry, especially due to its higher acidity and pulp yield. However, the species *Passiflora edulis* Sims fo. *edulis* and *Passiflora caerulea*, known as purple and orange passion fruit, respectively, have the sweetest flavor, so are best consumed as juice or as fresh pulp (Zeraik and Yariwake 2010). These fruits are of interest not only due to the pulps but also due to its peels and seeds which contain high levels of bioactive compounds. Furthermore, regarding species *Passiflora caerulea*, no research has been reported about bioactive compounds content.

The ideal condition for the development of passion fruit occurs in places where the tropical climate prevails, and it is emphasizing that the water content in the soil is one of the factors that most influences the flowering of passion fruit crop. The regions that this fruit is widely using are American and European countries, due to the favorable

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weather. The best variety known and more used is *Passiflora edulis*, not only because of its pulp but also due of the infusions made with the leaves (Silva et al. 2013).

The common consumption of fruit and vegetable in the diet can protect our organism of Manu chronic diseases such as cancer, neurological diseases, cardiovascular diseases, obesity, inflammations and infections (Volp et al. 2009). However, passion fruit appears to be an excellent source of nutrients as carbohydrates, vitamins, and minerals that are essential nutrients for life. The fruit has a high content in nutraceuticals, as phenolic acids, where anthocyanins and flavonoids are the majoritarian compounds of this group; carotenoids and β -carotene appear to be the principal component, with consequently increased provitamin A activity. These nutraceutical compounds have biological activities in the health, protective effect against degenerative and chronic diseases and act as mutagenesis and carcinogenesis inhibitors. Also, these compounds have been associated with antiviral, anti-allergic, antiplatelet and anti-inflammatory activities (Morais et al. 2016; Castañeda-Ovando et al. 2009; Tanwar and Modgil 2012; González-Gallego et al. 2014).

A major problem of passion fruit juice in the manufacturing industry is the amounts of waste generated come from discarded that are the peels and seeds. These residues are excellent sources of nutrients, bioactive compounds and studies about your potential can even indicate a future application as food ingredients in the formulation of new products, encouraging the reutilization of food and offering a nutritious alternative diet at low cost.

Based on this, the aim of this study was to evaluate the physicochemical characteristics, color parameters, pectin, proximate composition, minerals, the content of bioactive compounds and antioxidant capacity in pulp, peel, and the seed of yellow, purple and orange passion fruit to future use of parts as functional ingredients.

Materials and methods

Sample preparation

The yellow passion fruit (*Passiflora edulis* Sims fo. *flavicarpa*) samples were obtained CEASA (Central Supply of Rio Grande do Sul), Caxias do Sul, RS, Brazil. The purple (*Passiflora edulis* Sims fo. *edulis*) and orange passion fruit (*Passiflora caerulea*) samples were collected from farmers in the Criuva District (Caxias do Sul), the Rio Grande do Sul, Brazil. The fruits were harvested when fully ripe, with their skin color (yellow, purple or orange). After the crop, the fruits were transported immediately to the laboratory where they were cleaned in running water. Then the fruits were cut in half and made a separation of the pulp, peel

(albedo and flavedo) and seed, the parts were packed in plastic bags and stored in a freezer at $-18\text{ }^{\circ}\text{C}$ until analysis.

Physicochemical and chemical analysis

Total soluble solids (TSS) were determined in pulps with a Brix refractometer. The pH was measured using a pH meter (Quimis, model Q-400A). The yield was calculated by the percentage ratio (%) of the weight of the fruit with and without seed. The titratable acidity was determined by titration method using standardized 0.1 M NaOH solution, and the results analyzing in g/100 g of citric acid following the Analytical Standards Instituto Adolfo Lutz (2008). All of these analyses were made in triplicate.

Yield

The yield calculation of the pulp was calculated by weight of whole fruit (one by one) and weight of the fruit without peel and seed (pulp). The result was expressed as percentage (%). The average number of passion fruit analyzed was around 30 units.

Color parameters

The color was analyzed using a portable colorimeter (Konica Minolta Model CR 400, Singapore) by the *Commission Internationale de l'Eclairage* (CIELAB system) by determining the values of L^* (lightness), a^* (component red-green) and b^* (yellow-blue component). All of these analyses were made in triplicate.

Proximate composition

All analyses were performed according to AOAC (2000). Protein content was determined by the Kjeldahl method with a conversion factor of 5.75. Lipids were obtained by cold extraction and ash was determined in a muffle furnace at $550\text{ }^{\circ}\text{C}$. Total dietary fiber was determined by the enzymatic-gravimetric method, moisture by gravimetry. Carbohydrates were estimated by the difference of 100 per cent of the sum between proteins, lipids, water and ash. The results were expressed as % and the data presented is the average of triplicate analysis.

Determination of pectin

The pectin of peels was extracted using a method of Canteri-Schemin et al. (2005) and Seixas et al. (2014) with some modifications. The extraction process was carried out in beakers (600 mL), followed by heating in a microwave

oven (Electrolux, ME21S 800 W). Each 2 g of passion fruit peel flour was added to 50 mL of distilled water in a beaker. Then, an addition of a tartaric acidic solution (10%), was added to maintain a final pH of 2 for the solutions. The beaker was put in a microwave heating for 3 min. The solution (still warm), was vacuum filtered on a filter paper and the filtrate (containing the soluble pectin) was cooled to 4 °C.

To isolate the soluble pectin from the filtrate, the solution was slowly added under magnetic stirring to two volumes of absolute ethyl alcohol. This mixture was stirred for 10 min, after which it was allowed to rest for 30 min to facilitate the flotation of the pectin. The pectin was separated by vacuum filtration on a filter paper. The extracted pectin in a gel form was immersed in absolute ethyl alcohol for about 12 h and then was partially dehydrated by immersion in acetone. The drying pectin was put in an air-circulated oven at 50 °C until constant weight (approximately 8 h). The results were expressed as g of pectin/100 g dry peels (triplicate).

Determination of minerals

All the samples (pulp, peel, and seed) of yellow, purple and orange passion fruit were lyophilized (Liotop, L101, Brazil) before mineral analysis. The analysis of minerals in the passion fruit samples were made in Plant Soil Laboratory Faculty of Agronomy, Federal University of Rio Grande do Sul (UFRGS), according to the methodology of atomic emission spectrometry with inductively coupled plasma source (ICP-OES) described by Tedesco and Gianello (2004) This method is compiling in Table 1S (supplementary material). The results were expressed as % and the data presented is the average of duplicate analysis.

Determination of carotenoid profile and provitamin A content

The profile of carotenoids was determined according to Mercadante and Rodriguez-Amaya (1998). The extraction of pigments was with acetone and the saponification in a KOH solution (10% in methanol) overnight. The extract was rotary evaporated (Fisatom, Model 801) ($T < 25$ °C) and stored in a freezer (-18 °C) for quantification by high-performance liquid chromatography (HPLC).

For HPLC (High Performance Liquid Chromatography) analysis, the samples stored in a freezer were diluted with methyl *tert*-butyl ether (MTBE-JT Baker, CAS. Number 1634-04-4, purity 99.96%), sonicated (Unique, Model USC 1400) for 1 min and filtered (Millex LCR 0.45 μ m, 13 mm) for injection into the HPLC (Agilent 1100 Series, Santa Clara, CA, USA), a UV-visible detector and with a quaternary system.

The column used was a C30 polymeric reverse phase (250 \times 4.6 mm ID, 3 μ m, YMC, model CT99SO3-2546WT). The mobile phase gradient (water:methanol:MTBE) (JT Baker, CAS Number 04.04.1634, 99.96% purity) commenced at 5:90:5, reaching 0:95:5 at 12 min, 0:89:11 at 25 min, 0:75:25 at 40 min, and finally 00:50:50 at 60 min. The temperature of column was 33 °C and a flow rate of 1 mL/min (Spectra were obtained at a fixed wavelength of 450 nm for carotenoids).

Compounds were identified by comparing the sample retention time's with the retention times obtained for controls. For quantification, a standard curve was constructed for carotenoids over the following ranges: lutein 1–65 μ g/mL ($\geq 95\%$, Sigma-Aldrich); zeaxanthin 1–40 μ g/mL ($\geq 95\%$, Sigma-Aldrich); cryptoxanthin 4–100 μ g/mL ($\geq 97\%$, Sigma-Aldrich); α -carotene 2–25 μ g/mL ($\geq 95\%$, Sigma-Aldrich); β -carotene 5–50 μ g/mL ($\geq 97\%$, Sigma-Aldrich) and lycopene ($\geq 85\%$ Sigma-Aldrich).

The limits of detection (LOD) and quantification (LOQ) were calculated by injecting 10 times the blank of the sample at very low level were used for measurement of LOD and LOQ which were determined as follows: LOD = mean value + 3 standard deviation (SD) LOQ = mean value + 10 SD where, mean value is zero (Ertas et al. 2007). Lutein: 6.9×10^{-3} and 1.15×10^{-2} μ g/g; zeaxanthin: 9.56×10^{-2} and 1.59×10^{-2} μ g/g; cryptoxanthin: 2.11×10^{-2} and 3.51×10^{-2} μ g/g; α -carotene: 1.97×10^{-2} and 3.28×10^{-2} μ g/g; β -carotene: 6.53×10^{-2} and 10.89×10^{-2} μ g/g and lycopene were 7×10^{-3} and 33×10^{-3} μ g/g.

Provitamin A activity was calculated by the bioconversion factor following Institute of Medicine (2001), yielding a value of 12 mg of β -carotene with 1 mg of Retinol Activity Equivalent (RAE). The results were expressed as % and the data presented is the average of triplicate analysis.

Determination of phenolic compounds

Samples (5 g) were homogenized by exhaustive extraction with 20 mL of ethanol and centrifuged at 15 °C (Cientec, CTR—5000R, Brazil) at 5000 \times g for 20 min. Then, 20 μ L of supernatant was added to 1.58 mL of water and 100 μ L of Folin-Ciocalteu (0.4 mol/L). After reaction (3 min), 300 μ L of Na_2CO_3 was added, and the mixture kept at room temperature for 2 h. The absorbance was then read at 765 nm on a UV-visible spectrophotometer (Shimadzu, UV-1700 PharmaSpec, Japan). The ethanol was used as the blank and gallic acid was used for calibration of the standard curve (0–0.50 mg/L). The results were expressed as mg of gallic acid equivalent (GAE) per 100 g of dry sample (triplicates) (Swain and Hillis 1959).

Determination of quercetin and kaempferol

Quercetin and kaempferol contents were analyzed according to Zeraik and Yariwake (2010) with modifications. The sample (10 g) were homogenised in an Ultra-Turrax (T25, IKA, China) with 30 mL of methanol at room temperature. The extracts were centrifuged at $15,000 \times g$, 4°C for 20 min and after in rotary evaporator giving 2 mL of extract. The resulting aqueous solution was filtered through a $0.45 \mu\text{m}$ Millex-HV PVDF membrane (Millipore, New Bedford, MA, USA) before HPLC analysis. The samples were prepared and analyzed in triplicate and the results were expressed as %.

The HPLC-/DAD analyses were carried out on a Waters Alliance 2695 (Milford, MA, USA) liquid chromatograph connected to a model 2996 (DAD) diode array detector and controlled by Waters Empower software. The separation was performed using a C18 polymer column ($250 \text{ mm} \times 4.6 \text{ mm id}$, $5 \mu\text{m}$ Vydac, 218TP). The samples were injected automatically ($10.0 \mu\text{L}$). A flow rate of 0.8 mL/min was applied, using a linear gradient of 0.2% formic acid in water (solvent A) and 0.2% formic acid in acetonitrile (solvent B). The gradients were: 0–10 min, 15% B in 85% A and 10–30 min, 20% B in 80% A. The chromatogram was monitored at 330 nm, and UV spectra of individual peaks were recorded in the range of 200–400 nm.

The contents of quercetin and kaempferol were determined by comparison with an external standard, injecting a new standard daily at 30 mg/mL for Quercetin ($\geq 98\%$, Sigma-Aldrich) and 4 mg/mL for kaempferol ($\geq 99\%$, Sigma-Aldrich).

Determination of anthocyanins

The anthocyanins were analyzed according to Zanatta et al. (2005), so 5 g of sample were homogenized in an Ultra-Turrax (T25, IKA, China) with acidified methanol (HCl 1%) and then quantified by HPLC (Agilent 1100 Series, Santa Clara, CA, USA) equipped with a quaternary pump system solvent and a UV-visible detector was used with a C18Shim-PakCLC-ODScolumn ($5 \mu\text{m}$, $250 \times 4.6 \text{ mm}$).

The mobile phase was 5% aqueous formic acid/methanol 85:15 (v/v) to 20:80 over 25 min and this isocratic ratio was maintained for 15 min. The mobile phase flow were 0.8 mL/min , the injection volume was $5 \mu\text{L}$, and 29°C was the temperature of column. The chromatograms were processed at a fixed wavelength of 520 nm.

The standards were from Sigma-Aldrich (USA): cyanidin-3-glucoside (CAS 7084-24-4, $\geq 95.0\%$), cyanidin-3,5-glucoside (CAS 2611-67-8, $\geq 90.0\%$), delphinidin-3- β -glucoside (CAS 6906-38-3, $\geq 97\%$), pelargonidin-3-glucoside (CAS 17334-58-6, $\geq 90\%$), aglycone delphinidin

(CAS 528-53-0, $\geq 95\%$), aglycone cyaniding (CAS 528-58-5, $\geq 95\%$), malvidin-3,5-diglucoside (CAS 643-84-5, $\geq 95\%$) and aglycone pelargonidin (CAS 17334-58-6, $\geq 90\%$). The quantification and identification of compounds were performed by comparing peak areas and retention times with their respective standards under the same chromatographic conditions. The results were expressed as % and the data presented is the average of triplicate analysis.

Determination of antioxidant capacity

The methodology used to determine antioxidant capacity was based on the sequestration of DPPH (2,2-diphenyl-1-picryl-hydrazyl) and ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) radicals according to Brand-Williams et al. (1995). Samples (5 g) were placed in 20 mL of ethanol and then centrifuged at 15°C at $5000 \times g$ (Cientec, CTR—5000R, Brazil) for 20 min. The liquid was diluted in three concentrations (10, 30, and 100%). For the DPPH assay, aliquots of each concentration were treated with 2 mL of DPPH (0.06 mM). The absorbance was read at 517 nm (Shimadzu, UV-1700 PharmaSpec, Japan). The results were presented as IC (Effective Concentration of 50% radical inhibition) 50 (mg/100 mL).

For the ABTS assay, aliquots of each concentration were treated with 2 mL of ABTS (7 mM). The absorbance was read at 734 nm (Shimadzu, UV-1700 PharmaSpec, Japan). The results were presented as IC 50 (mg/100 mL).

Statistical analysis

Data were analyzed by ANOVA and Tukey's mean comparison test with a significance level of 5%, followed by a principal component analysis (PCA) using the software Statistica 12.0 (Statsoft Inc, São Paulo, Brasil). Pearson's correlation was to the results of phenolic compounds and antioxidant capacity using Statistica 12.0 (Statsoft, São Paulo, Brazil).

Results and discussion

Physicochemical analysis

The results of the physicochemical analysis in pulps of different species of passion fruit were: 9.10% of TSS (total soluble solids), 9.06% of titratable acidity (citric acid) and a pH of 2.66 in yellow passion fruit pulp. For purple passion fruit the results were 11.60% of TSS, 2.83% of titratable acidity and a pH of 2.72 and for orange passion fruit pulp, we found 12.30% of TSS, 2.21% of

titratable acidity and a pH of 3.97. So, the pulp of orange passion fruit is sweeter because it showed the higher content of total soluble solids (TSS), pH and consequently, lower content of acidity. The pulp of yellow passion fruit showed higher acidity and lower pH and TSS and then it is more acid. Some factors such as soil type, fertilization, climate, irrigation and genetic traits of the cultivar can explain the variations between the physicochemical parameters.

Kishore et al. (2011) evaluated the physicochemical attributes as TSS and titratable acidity in purple passion fruit pulp (*Passiflora edulis* Sims) who the values were 15.30 and 3.80%, respectively. These results were higher when compared to purple passion fruit pulp of this research. In another study realized by Souza et al. (2012), who analysed the pH, titratable acidity and total soluble solids in sweet passion fruit pulp (*Passiflora alata* Dryand), the results were 3.31, 2.00, and 13.33%, respectively. These values were more similar to specie *Passiflora caeurea* analysed by us that was the passion fruit sweeter. Janzantti et al. (2012) did a research with yellow passion fruit pulp (*P. edulis* Sims f. *flavicarpa* Deg.) and the higher values were 4.32% for titratable acidity, 14.71% soluble solids and 3.53 for pH, different values found by us when compared to yellow passion fruit. López-Vargas et al. (2013) evaluated the pH in pulp and seed of yellow passion fruit (*Passiflora edulis* var. *flavicarpa*) and the results were 3.75. They found higher values of pH in relation to the same variety (yellow passion fruit) of our research. Pongener et al. (2013) found the highest total soluble solids of 16.2° Brix in purple passion fruit pulp (*Passiflora edulis* Sims) and titratable acidity of 2.34 g citric acid/100 mL extract. These values were different both for total soluble solids and titratable acidity when compared to purple passion fruit pulp analysed in this study.

The yield of different species of passion fruit was evaluated, and the results are shown in Table 1. As expected, pulp and peel of yellow passion fruit presented higher yield when compared to purple and orange passion fruit, but for seed, orange passion fruit showed higher yield. Thus, it appears that the residues that represent most of the fruit should be characterized to indicate the possible applications and uses. For example, replacing wheat flour in some formulations such as pasta, breads, cakes, biscuits, which will have a higher content of fibers and bioactive compounds.

PCA (Principal Component Analysis) is a statistical technique used to reduce the dimensionality of a data set containing a large number of inter-related variables. The analysis is performed to maintain the maximum variance present in the data. This reduction produces a new reduced and uncorrelated set of variables, called principal components. These components are then chosen to ensure that the

Table 1 Yield color parameters and pectin in different species of passion fruit (mean and standard deviation)

	Yellow	Purple	Orange
Yield (%)			
Pulp	27.71 ± 1.00 ^a	25.23 ± 1.02 ^c	26.54 ± 0.98 ^b
Peel	64.05 ± 2.03 ^a	62.11 ± 1.90 ^b	57.28 ± 2.05 ^c
Seed	8.24 ± 0.05 ^c	12.66 ± 0.12 ^b	16.18 ± 0.08 ^a
Color L*			
Pulp	58.05 ± 0.64 ^b	77.65 ± 0.57 ^a	26.90 ± 0.17 ^c
Peel	43.16 ± 1.47 ^a	21.58 ± 2.15 ^c	36.94 ± 0.45 ^b
Seed	33.86 ± 0.80 ^a	28.16 ± 0.78 ^b	26.96 ± 0.49 ^b
Color a*			
Pulp	8.44 ± 0.01 ^b	0.29 ± 0.08 ^c	10.78 ± 0.11 ^a
Peel	2.29 ± 0.25 ^b	2.20 ± 0.22 ^b	13.23 ± 0.49 ^a
Seed	1.91 ± 0.09 ^b	0.30 ± 0.08 ^c	8.25 ± 0.24 ^a
Color b*			
Pulp	42.83 ± 1.28 ^a	17.18 ± 1.08 ^b	6.08 ± 0.07 ^c
Peel	31.33 ± 0.53 ^b	3.83 ± 1.75 ^c	35.78 ± 1.64 ^a
Seed	19.36 ± 0.45 ^a	5.90 ± 0.21 ^c	8.89 ± 0.55 ^b
Pectin			
Peel	37.67 ± 0.97 ^a	32.85 ± 1.20 ^b	21.55 ± 0.55 ^c

^{a,b,c}Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$)

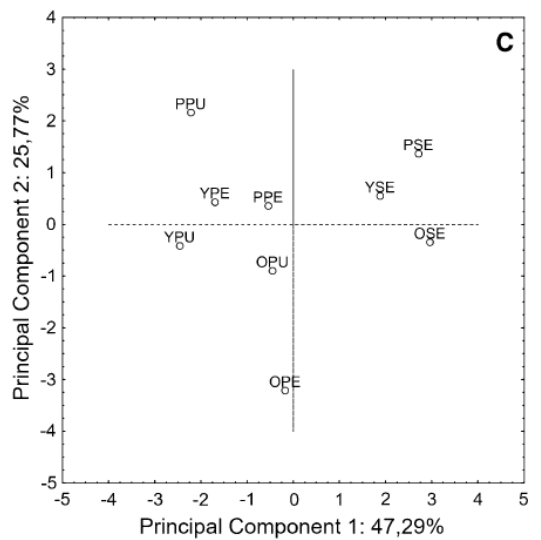
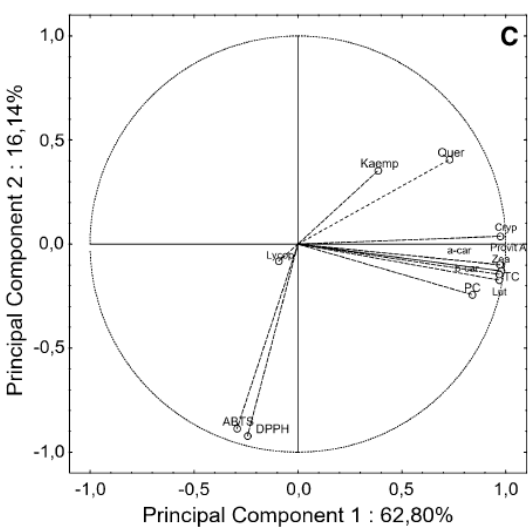
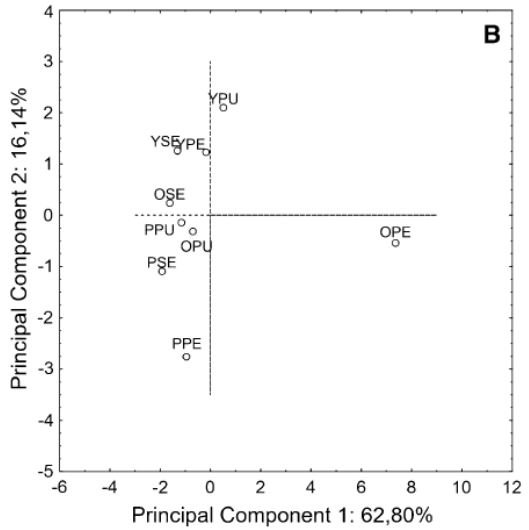
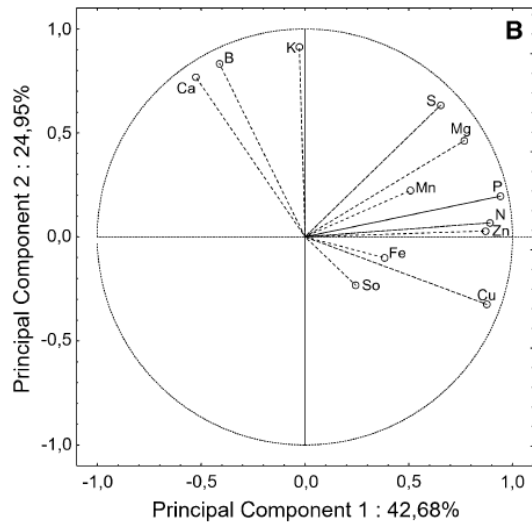
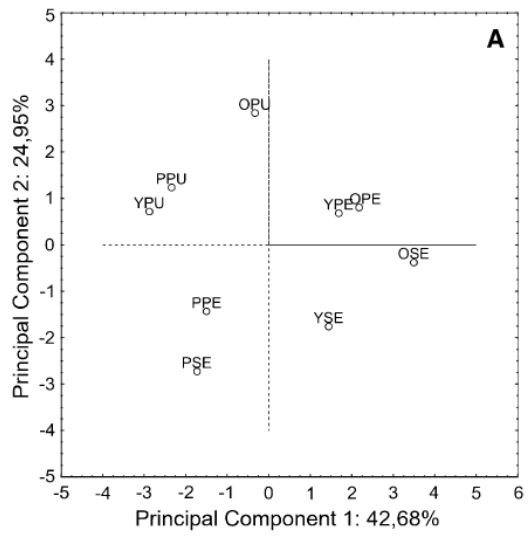
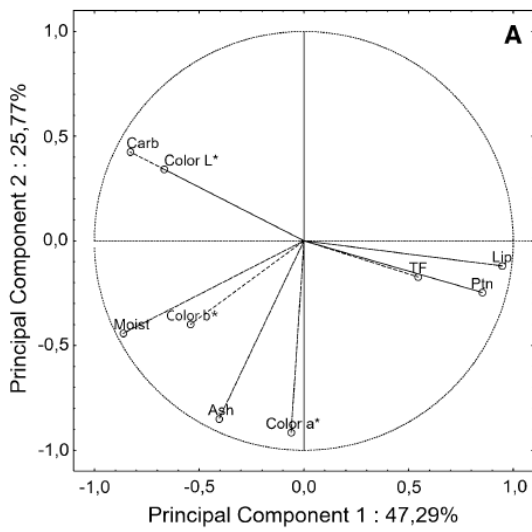
former retain the greater part of the variance present in the original variables.

The three passion fruits were evaluated for the color parameters, proximate composition, minerals, carotenoids, flavonoids, phenolic compounds, antioxidant capacity and the results are showed in Fig. 1.

By analyzing the components, the variance of the data was accounted for the significant contributions of 47.29% for the first principal component representing variable ash, color b and color L and 25.77% for the second principal components representing other variables (Fig. 1a).

Color

The passion fruit was evaluated within the context of color parameters (Fig. 1a). It was observed that for color L*, the pulp of purple passion fruit showed higher brightness. For color a* that indicates the intensity of colors red-green, orange passion fruit showed higher levels for pulp, peel, and seed when compared to yellow and purple passion fruit. For color b*, which indicates the intensity of colors yellow-blue, both for pulp and seed, yellow passion fruit presented more intensity, and for the peel, orange passion fruit showed more intensity when compared to other species of passion fruit. The differences in the color



◀ **Fig. 1** Principal component analysis of yellow, purple and orange passion fruit in pulp, peel, and seed: proximate composition and color parameters (a); minerals (b); carotenoids, phenolic compounds and flavonoids (c). Legend: YPU: yellow passion fruit pulp; YPE: yellow passion fruit peel; YSE: yellow passion fruit seed; PPU: purple passion fruit pulp; PPE: purple passion fruit peel; PSE: purple passion fruit seed; OPU: orange passion fruit OPE: orange passion fruit peel; OSE: orange passion fruit seed; Moist: moisture; Ptn: protein; Lip: lipids; Carb: carbohydrates; TF: total fibre; N: nitrogen; P: phosphorus; K: potassium; Ca: calcium; Mg: magnesium; S: sulphur; Cu: copper; Zn: zinc; Fe: iron; Mn: manganese; B: boron; So: sodium; Lut: lutein; Zea: Zeaxanthin; Cryp: Cryptoxanthin; a-car: α -carotene; b-car: β -carotene; Lycop: lycopene; Provit A: provitamin A; TC: total carotenoids; PC: phenolic compounds; Quer: quercetin; Kaemp: kaempferol

parameters can be explained mainly the species, soil and harvesting period are different.

Proximate composition

By principal component analysis (Fig. 1a) can be seen that the orange passion fruit stands out for its red coloration in the peel, high ash content present in the pulp and peel and the high content of proteins and lipids in the seeds. The yellow passion fruit, in turn, showed a high content of carbohydrates in the peel, probably due to the high content of pectin (37.37 g/100 g) in this species. The total fiber content in the pulp was greater in this species compared with others. The purple passion fruit highlights to have a high content of carbohydrates in the pulp compared with orange and yellow passion fruit (Table 2).

Souza et al. (2012) analyzed the proximate composition in sweet passion fruit pulp (*Passiflora alata* Dryand), and the results were (dry weight, except for moisture): 84.12% of moisture, 8.50% proteins, 0.63% lipids, 82.17% carbohydrates, 4.40% dietary fiber and 4.28% ash. The results of proteins and carbohydrates were similar with yellow passion fruit analyzed in this study, but for moisture, lipids, dietary fiber, and ash we found higher values.

Thereby, the peels and seeds of yellow, purple and orange passion fruit showed to be richer in total dietary fiber, proteins (seeds) and lipids, which should have an unsaturated origin.

Pectin

As it can be seen in Table 1, the peel of yellow passion fruit showed higher levels of pectin when compared to the purple and orange passion fruit (the latter presented the lowest content).

Liew et al. (2014) evaluated the production of pectin yellow passion fruit peel (*Passiflora edulis f. flavicarpa*) in extraction with citric acid. The authors found 14.60% yield

Table 2 Proximate composition of different species of passion fruit (g/100 g of dry weight—except for moisture) with mean and standard deviation

	Yellow	Purple	Orange
Moisture			
Pulp	90.06 ± 0.00 ^a	83.44 ± 0.99 ^b	88.18 ± 0.87 ^a
Peel	87.14 ± 3.29 ^b	87.02 ± 1.22 ^b	94.25 ± 0.26 ^a
Seed	57.09 ± 2.36 ^a	45.91 ± 0.97 ^b	58.46 ± 0.38 ^a
Proteins			
Pulp	8.57 ± 0.10 ^b	6.53 ± 0.23 ^c	9.90 ± 0.35 ^a
Peel	3.40 ± 0.06 ^c	6.47 ± 0.04 ^b	11.60 ± 0.44 ^a
Seed	13.07 ± 0.12 ^b	13.23 ± 0.48 ^b	15.84 ± 0.15 ^a
Lipids			
Pulp	1.11 ± 0.04 ^b	1.09 ± 0.04 ^b	2.92 ± 0.09 ^a
Peel	4.20 ± 0.03 ^c	4.89 ± 0.07 ^b	10.25 ± 0.12 ^a
Seed	12.31 ± 0.78 ^c	14.94 ± 0.41 ^b	19.64 ± 0.30 ^a
Ash			
Pulp	6.94 ± 0.01 ^a	2.95 ± 0.14 ^b	7.31 ± 0.22 ^a
Peel	6.62 ± 0.24 ^c	7.93 ± 0.05 ^b	13.29 ± 0.41 ^a
Seed	3.56 ± 0.05 ^a	1.85 ± 0.06 ^c	3.23 ± 0.18 ^b
Carbohydrates			
Pulp	83.37 ± 0.00 ^b	89.42 ± 0.00 ^a	79.87 ± 0.00 ^c
Peel	85.78 ± 0.00 ^a	80.71 ± 0.00 ^b	64.86 ± 0.00 ^c
Seed	71.07 ± 0.00 ^a	69.98 ± 0.00 ^a	61.38 ± 0.00 ^b
Total fibre			
Pulp	7.15 ± 0.07 ^a	1.40 ± 0.18 ^c	2.17 ± 0.11 ^b
Peel	61.16 ± 1.02 ^a	61.68 ± 1.31 ^a	62.14 ± 2.62 ^a
Seed	65.60 ± 0.52 ^a	55.06 ± 0.35 ^b	51.47 ± 0.60 ^c

^{a,b,c} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$)

of pectin from passion fruit peel. Kulkarni and Vijayanand (2010) also conducted a study to evaluate the pectin content of passion fruit peel (*Passiflora edulis f. flavicarpa* L.) yellow variety. The passion fruit peels were dehydrated for pectin extraction experiments. The conditions for the extraction of pectin from the passion fruit peel promoted a yield of 14.80 g/100 g of the dried peel. In another research carried out by Seixas et al. (2014), pectin extraction was investigated from the passion fruit peel (*Passiflora edulis f. flavicarpa*). The highest yield was found to be 18.20%. Different species of passion fruit of this study showed better results compared to the yield of pectin compared to all kinds of passion fruit analyzed.

Minerals

The minerals content of passion fruit is shown in Fig. 1B. By analyzing the components, the variance of the data was accounted for the significant contributions of 42.66% for

the first and 24.95% for the second principal components. It can be observed that pulps of three species have high calcium, potassium, and boron. The yellow and orange passion fruit peels showed high zinc, manganese, copper, phosphorus, and sulfur. As for the purple passion fruit peel, these minerals had low content. Regarding seeds, orange passion fruit presented a considerable amount of copper followed by yellow passion fruit (Table 3).

Gondim et al. (2005) determined the mineral concentration in yellow passion fruit peels (*Passiflora edulis*). They found 44.51 mg of calcium, 0.89 mg of iron, 43.77 mg sodium, 27.82 mg magnesium, 0.32 mg zinc, 0.04 mg copper and 178.40 mg potassium in 100 g of fresh sample. These values were lower (except for sodium) when compared to the yellow passion fruit peel analyzed in this research due to they did not present the values in dry basis.

Souza et al. (2012) evaluated the mineral content (phosphorus, potassium, calcium, magnesium and iron) in passion fruit pulp (*Passiflora alata* Dryand). The authors found values of 34.95 mg for phosphorus, 375.42 mg potassium, 4.76 mg calcium, 19.82 mg magnesium and 1.06 mg of iron in 100 g of fresh pulp. They found significant amounts of calcium and potassium in pulp compared to our pulps studied.

As example, in adults above 19 years old, a portion of 100 g of orange passion fruit pulp presents 81 and 59% of recommended daily intake of zinc in women and men, respectively; 70 and 55% for magnesium in women and men; 111% for copper; 55% for phosphorus and 33% for calcium. In orange passion fruit peel (100 g), potassium presents 85% of recommended daily intake and manganese 405 and 317% in women and men. The pulp of yellow passion fruit (100 g) gives 30 and 69% of iron recommended daily intake for women and men; the concentrations of boron and sodium of all parts in passion fruit (portion of 100 g) is according to tolerable upper intake levels from 1 year old; sulphur and nitrogen not appears in recommended daily intake and tolerable upper intake levels.

Thus, it can be seen that from the nutritional matrix, we can ingest the recommended daily amount of minerals through the passion fruit, noting that the pulps have higher concentrations of minerals than peels.

Carotenoid profile and provitamin A content

The content of carotenoids and provitamin A are shown in Fig. 1c. By analyzing the components, the variance of the data was accounted for the significant contributions of 62.80% for the first and 16.14% for the second principal components. For peel, orange passion fruit stands out to all carotenoids evaluated and provitamin A about other species of passion fruit, where the majoritarian carotenoid was

Table 3 Mineral composition of different species of passion fruit (mg/100 g of dry weight) with mean and standard deviation

	Yellow	Purple	Orange
Zinc			
Pulp	5.20 ± 0.10 ^b	2.10 ± 0.05 ^c	6.50 ± 0.12 ^a
Peel	1.00 ± 0.02 ^b	0.90 ± 0.01 ^c	5.80 ± 0.05 ^a
Seed	4.10 ± 0.09 ^c	4.60 ± 0.05 ^b	8.90 ± 0.04 ^a
Iron			
Pulp	5.50 ± 0.03 ^a	2.90 ± 0.02 ^c	3.20 ± 0.01 ^b
Peel	3.20 ± 0.04 ^c	4.60 ± 0.03 ^a	3.90 ± 0.02 ^b
Seed	5.20 ± 0.02 ^a	4.30 ± 0.03 ^c	4.50 ± 0.03 ^b
Boron			
Pulp	0.70 ± 0.02 ^a	0.20 ± 0.01 ^b	0.70 ± 0.02 ^a
Peel	1.30 ± 0.03 ^c	1.40 ± 0.03 ^b	1.60 ± 0.04 ^a
Seed	0.40 ± 0.01 ^b	0.50 ± 0.02 ^a	0.50 ± 0.02 ^a
Manganese			
Pulp	1.20 ± 0.05 ^a	0.40 ± 0.01 ^b	1.20 ± 0.01 ^a
Peel	0.50 ± 0.0 ^c	0.70 ± 0.02 ^b	7.30 ± 0.02 ^a
Seed	2.20 ± 0.05 ^c	2.30 ± 0.03 ^b	8.90 ± 0.01 ^a
Copper			
Pulp	0.60 ± 0.02 ^b	0.20 ± 0.01 ^c	1.00 ± 0.02 ^a
Peel	0.10 ± 0.01 ^c	0.20 ± 0.01 ^b	0.30 ± 0.01 ^a
Seed	0.90 ± 0.02 ^b	0.70 ± 0.02 ^c	1.30 ± 0.03 ^a
Phosphorus			
Pulp	380 ± 1.98 ^b	150 ± 1.70 ^c	390 ± 2.76 ^a
Peel	140 ± 1.30 ^b	70.00 ± 1.12 ^c	240 ± 1.71 ^a
Seed	310 ± 2.05 ^b	63.00 ± 1.19 ^c	390 ± 2.90 ^a
Sulfur			
Pulp	170 ± 2.00 ^b	90.00 ± 0.85 ^c	330 ± 3.92 ^a
Peel	70.00 ± 0.40 ^c	160 ± 1.35 ^b	280 ± 2.80 ^a
Seed	150 ± 1.23 ^b	32 ± 0.10 ^c	230 ± 2.09 ^a
Sodium			
Pulp	1.40 ± 0.02 ^c	5.30 ± 0.04 ^b	9.40 ± 0.20 ^a
Peel	2.20 ± 0.02 ^c	7.30 ± 0.12 ^b	11.50 ± 0.15 ^a
Seed	3.46 ± 0.07 ^c	4.80 ± 0.03 ^a	4.40 ± 0.05 ^b
Magnesium			
Pulp	200 ± 1.23 ^b	120 ± 0.95 ^c	220 ± 1.30 ^a
Peel	120 ± 0.90 ^c	130 ± 0.97 ^b	140 ± 1.34 ^a
Seed	150 ± 1.10 ^c	290 ± 1.80 ^a	200 ± 1.22 ^b
Nitrogen			
Pulp	2400 ± 20.0 ^a	1100 ± 8.0 ^c	1700 ± 12.0 ^b
Peel	620 ± 9.00 ^c	920 ± 7.50 ^b	940 ± 7.00 ^a
Seed	1800 ± 15.0 ^b	380 ± 1.50 ^c	2200 ± 18.5 ^a
Potassium			
Pulp	3800 ± 25.5 ^a	1600 ± 16.0 ^c	2900 ± 18.5 ^b
Peel	2600 ± 15.7 ^c	2800 ± 16.3 ^b	4000 ± 32.0 ^a
Seed	760 ± 6.40 ^b	112 ± 3.00 ^c	1000 ± 7.80 ^a
Calcium			
Pulp	50.00 ± 0.40 ^a	20.00 ± 0.12 ^c	30.00 ± 0.10 ^b
Peel	250 ± 1.98 ^b	310 ± 1.69 ^a	30.00 ± 0.11 ^c
Seed	30.00 ± 0.35 ^b	6.00 ± 0.02 ^c	330 ± 1.18 ^a

^{a,b,c} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$)

β -carotene because his peel color is orange. This species also presented in the pulp the highest content of lycopene (Table 4).

Souza et al. (2012) analyzed the content of β -carotene and lycopene in sweet passion fruit pulp (*Passiflora alata* Dryand), and they found 8249 and 5478 $\mu\text{g}/100\text{ g}$ dry weight, confirming that these results were higher than our study for both β -carotene and lycopene. Pongener et al. (2013) evaluated the total carotenoids in purple passion fruit, and the higher value was 1467 $\mu\text{g}/100\text{ mL}$, being higher when compared to purple passion fruit analyzed in this research, but lower values compared to yellow and orange pulps.

Silva et al. (2014) evaluated the content of β -carotene and lycopene in pulp and peel of yellow passion fruit (*Passiflora edulis* Sims). They found 1362.07 and 57.93 μg β -carotene/100 g (dry basis), respectively and lycopene was not detected both in pulp and peel. The values of β -carotene were similar to our research for pulp (1333.97 $\mu\text{g}/100\text{ g}$ dry basis), but for the peel, we detected more content of this compound (272.52 $\mu\text{g}/100\text{ g}$ dry basis).

However, Pertuzatti et al. (2015) analyzed the carotenoids profile in yellow passion fruit (*Passiflora edulis*) and found: for lutein + zeaxanthin 1 $\mu\text{g}/100\text{ g}$, β -cryptoxanthin 24,990 $\mu\text{g}/100\text{ g}$, lycopene 28 $\mu\text{g}/100\text{ g}$ and total carotenoids 25,100 $\mu\text{g}/100\text{ g}$. The values of β -cryptoxanthin and total carotenoids were much higher than our study, but for lutein + zeaxanthin, we found higher concentrations in all varieties and for lycopene, the variety 'orange' were higher too.

In another research realized by Septembre-Malaterre et al. (2016) where the content of β -carotene in passion fruit pulp (*Passiflora edulis*) was investigated, the values found were 3829.20 μg β -carotene equivalent/100 g. These

results were higher when compared to all varieties analyzed in this study.

As an example (Institute of Medicine 2002), pulp of yellow passion fruit (100 g) presents 15 and 11% of DRI (dietary reference intakes) of vitamin A in women and men (> 14 years old), respectively. A portion of 100 g of orange passion fruit peel could contribute with 233 and 181% for women and men (> 14 years old), respectively according to recommended a daily intake of vitamin A.

Phenolic compounds

The presence of phenolic compounds in passion fruit makes this fruit an excellent candidate to evaluate several effects in vivo. As can be seen in Fig. 1c, the pulps and peels revealed the higher content of phenolic compounds, where the orange passion fruit stands out due to the higher concentrations. Therefore, the peels are residues that can be used and applied in formulations that would enrich the food due to the presence of these compounds (Table 5).

Silva et al. (2014) measured the total phenolic content in yellow passion fruit (*Passiflora edulis* Sims). They found 765.09 mg gallic acid equivalent/100 g (dry basis) in pulp and 451.06 mg gallic acid equivalent/100 g in peel (dry basis), similar values to our purple pulp. Already Septembre-Malaterre et al. (2016), found 286.6 mg gallic acid equivalent/100 g in passion fruit pulp (*Passiflora edulis*), different values about our research.

In summary, genotype, geographic effect, crop year, maturation and storage conditions are some characteristics that can influence the content of phenolic compounds, anthocyanins, flavonoids, carotenoids and other bioactive compounds in all fruit (Souza et al. 2008; Cardeñosa et al. 2016).

Table 4 Analysis of carotenoids in different species of passion fruit ($\mu\text{g}/100\text{ g}$ dry weight; mean and standard deviation)

	Pulp			Peel		
	Yellow	Purple	Orange	Yellow	Purple	Orange
Lutein	44.28 \pm 2.33 ^b	10.68 \pm 0.11 ^c	105.36 \pm 3.24 ^a	504.97 \pm 24.77 ^b	366.88 \pm 17.89 ^b	2881 \pm 148.7 ^a
Zeaxanthin	65.51 \pm 0.86 ^b	7.49 \pm 0.05 ^c	91.22 \pm 1.89 ^a	65.61 \pm 0.22 ^b	48.70 \pm 2.85 ^b	323.98 \pm 11.11 ^a
Cryptoxanthin	254.38 \pm 3.32 ^a	30.85 \pm 0.07 ^b	nd	75.31 \pm 0.05 ^b	74.56 \pm 0.12 ^b	617.23 \pm 37.71 ^a
α -carotene	86.43 \pm 4.59 ^a	67.65 \pm 2.16 ^b	nd	nd	37.19 \pm 1.29 ^b	420.07 \pm 15.02 ^a
β -carotene	1334 \pm 78.8 ^a	171.88 \pm 2.12 ^c	744.60 \pm 15.47 ^b	272.52 \pm 11.77 ^b	716.32 \pm 30.65 ^b	21,274 \pm 676 ^a
Lycopene	nd	nd	4405 \pm 135.1 ^a	nd	nd	nd
Provitamin A*	111.16 \pm 6.57 ^a	14.32 \pm 0.18 ^c	62.05 \pm 1.29 ^b	22.71 \pm 0.98 ^b	59.69 \pm 2.55 ^b	1773 \pm 56.4 ^a
Total carotenoids	1785 \pm 81.5 ^b	288.56 \pm 0.03 ^c	5346 \pm 145.4 ^a	918.41 \pm 36.81 ^b	1244 \pm 52.5 ^b	25,516 \pm 561.9 ^a

^{a,b,c}Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$)

nd not detected

*Expressed as μg RAE (Retinol Activity Equivalent)

Table 5 Analysis of phenolic compounds (mg/100 g dry weight), flavonoids (mg/100 g dry weight) and anthocyanins ($\mu\text{g}/100$ g dry weight) in different species of passion fruit with mean and standard deviation

	Pulp			Peel			Seed		
	Yellow	Purple	Orange	Yellow	Purple	Orange	Yellow	Purple	Orange
Phenolic compounds	1297.31 \pm 13.43 ^b	788.93 \pm 3.99 ^c	1559.15 \pm 5.33 ^a	1061.87 \pm 25.00 ^c	1570.80 \pm 26.76 ^b	2584.91 \pm 96.67 ^a	346.69 \pm 6.58 ^b	325.69 \pm 1.18 ^c	429.33 \pm 0.19 ^a
Quercetin	506.45 \pm 23.79 ^a	229.79 \pm 10.99 ^b	16.28 \pm 0.19 ^c	760.21 \pm 32.07 ^a	nd	800.13 \pm 24.18 ^a	nd	nd	120.41 \pm 2.82
Kaempferol	199.66 \pm 1.10 ^a	12.35 \pm 0.08 ^b	nd	nd	74.70 \pm 1.44 ^b	229.27 \pm 8.90 ^a	375.32 \pm 13.50	nd	Nd
Cyanin	nd	nd	nd	nd	1477.47 \pm 20.85	nd	nd	nd	Nd
Delphinidin-3,5-Glu	nd	nd	nd	nd	8679.60 \pm 341.32	nd	nd	nd	Nd
Cyanidin-3-Glu	nd	nd	nd	nd	2852.92 \pm 177.93	nd	nd	nd	Nd
Pelargonidin-3-Glu	nd	nd	183.95 \pm 6.52	nd	1551.94 \pm 239.03	nd	nd	nd	Nd
Aglycone Delphinidin	nd	nd	nd	nd	90,998.72 \pm 5218.53	nd	nd	nd	Nd
Aglycone Cyanidin	nd	nd	nd	nd	1237.73 \pm 37.68	nd	nd	nd	159.18 \pm 5.92
Malvidin 3,5-Di	nd	nd	nd	nd	nd	nd	4598.70 \pm 119.73 ^b	8232.41 \pm 6.54 ^a	Nd
Aglycone Pelargonidin	nd	nd	nd	nd	nd	nd	nd	nd	134.18 \pm 0.84
Total anthocyanins	nd	nd	183.95 \pm 6.52	nd	103,686.48 \pm 542.11	nd	4598.70 \pm 119.73	8232.41 \pm 6.54 ^a	293.36 \pm 6.75
DPPH*	0.20 \pm 0.03 ^a	3.32 \pm 0.02 ^c	2.41 \pm 0.01 ^b	1.69 \pm 0.03 ^a	6.98 \pm 0.20 ^c	2.45 \pm 0.03 ^b	1.18 \pm 0.03 ^a	6.30 \pm 0.08 ^c	2.68 \pm 0.03 ^b
ABTS*	0.82 \pm 0.03 ^a	4.59 \pm 0.01 ^c	3.72 \pm 0.05 ^b	2.22 \pm 0.01 ^a	9.37 \pm 0.05 ^c	2.95 \pm 0.02 ^b	3.84 \pm 0.08 ^a	4.76 \pm 0.03 ^b	3.87 \pm 0.00 ^a

^{a,b,c}Different superscript letters in the same row and to the same part indicate statistically significant difference ($p < 0.05$)

nd not detected

*IC50 (g/100 mL): Effective Concentration of 50% radical inhibition

Flavonoids

Flavonoids were analyzed in all parts of different species of passion fruit, but the anthocyanins were analyzed only in seeds of all species of passion fruit, peel of purple passion fruit (because his color is purple) and pulp of orange passion fruit (because his color is red). In the other parts, we did not analyze because contains a negligible amount of these compounds.

The pulp of yellow passion fruit presented to be richer in quercetin and kaempferol than other species; for the peel, orange passion fruit showed more retention of kaempferol and for seed, only the species of yellow passion fruit detected this component.

Zeraik and Yariwake (2010) evaluated the total flavonoids, expressed as rutin and isoorientin in yellow passion fruit pulp (*Passiflora edulis* Sims f. *flavicarpa* Degener) and found 158.03 mg/L and 16.22 mg/L, respectively, suggesting that *P. edulis* fruits may be comparable with other flavonoid food sources such as orange juice or sugarcane juice.

Li et al. (2011) analyzed the flavonoid composition of *Passiflora edulis* 'edulis' and *Passiflora edulis* 'flavicarpa' more known as 'purple' and 'yellow.' The chromatograms revealed that the six major flavonoids obtained from *Passiflora edulis* 'flavicarpa' had not been detected in *Passiflora edulis* 'edulis' which suggested that the two population are originally disparate and that the fruit color is closely correlated with some other variabilities of the species.

Silva et al. (2014) measured the content of yellow flavonoids in pulp and peel of yellow passion fruit (*Passiflora edulis* Sims). They found 60.37 and 43.08 mg/100 g (dry basis), respectively, different values when compared to our study, because they analyzed by spectrophotometry and by high-performance liquid chromatography.

In another research realized by Septembre-Malaterre et al. (2016) in which the content of total flavonoid content in passion fruit pulp (*Passiflora edulis*) was investigated, the values found were: 70.10 mg quercetin equivalent/100 g. Our study found different values to yellow (506.45 mg/100 g), purple (229.79 mg/100 g) and orange (16.28 mg/100 g).

Consequently, these fruit residues and pulp of passion fruit analyzed in this study can be regarded as a natural source of flavonoids and the part (pulp, peel, seed) where the color will indicate the concentration and type of flavonoid present in this matrix.

Anthocyanins

A large variety of anthocyanins were found in each part of different species passion fruit. As expected, the peel of

purple passion fruit reveals a lot of anthocyanins, among them: cyanin, delphinidin-3,5-glucoside, cyanidin-3-glucoside, pelargonidin-3-glucoside, aglycone delphinidin (the majoritarian anthocyanin) and aglycone cyanidin. They were also found pelargonidin-3-glucoside in pulp of orange passion fruit (his color is red), aglycone cyanidin and aglycone delphinidin in seed of orange passion fruit and malvidin 3,5-diglucoside in seeds of yellow and purple passion fruit, which are the same species (*Passiflora edulis*), the purple showed higher content of this anthocyanin when compared to yellow passion fruit (Table 5).

As no data were found in the literature about the content of anthocyanins in purple passion fruit peel, we compared with fruits that have purple color in the peel. In a study realized by Todaro et al. (2009), delphinidin-3-rutinoside was extracted and identified as the major anthocyanin in eggplant peel (*Solanum melongena* var. *esculentum* peel). Leite-Legatti et al. (2012) analyzed the content of anthocyanins in jaboticaba peel (*Myrciaria jaboticaba*). They found delphinidin 3-glucoside and cyanidin 3-glucoside (634.75 and 1963.57 mg/100 g, respectively) as anthocyanins. Cyanidin-3-O-glucoside was the dominant anthocyanin with 75.6% of the total anthocyanins.

Our results indicate promising perspectives for the exploitation of pulps of passion fruit species and their residues with significant levels of bioactive substances mainly peels of purple passion fruit which is rich in several anthocyanins.

Antioxidant capacity

The DPPH radical has purple color and ABTS the green color. With the addition of the radical fruit extract, the DPPH and ABTS are reduced, presenting yellow color, with consequent disappearance of absorption. From the results, it determines the percentage of antioxidant activity and scavenging of free radicals.

In the antioxidant capacity, pulp, peel and seed of yellow passion fruit has a greater power to scavenge the free radicals DPPH and ABTS (Table 5), which means that this fruit has a higher antioxidant capacity when compared to purple and orange passion fruit, except for seeds of yellow and orange passion fruit in ABTS analysis that no significant difference was detected.

Souza et al. (2012) evaluated the antioxidant capacity of different fruit, and they concluded that the smallest antioxidant capacity was observed in the jenipapo pulps, followed by sweet passion fruit, soursop, murici and marolo.

López-Vargas et al. (2013) also evaluated the antioxidant capacity DPPH in pulp and seed and albedo of yellow passion fruit (*Passiflora edulis* Sims fo. *flavicarpa*). They concluded that the albedo showed a higher ability to inhibit

DPPH radical than the pulp and seed samples. Our study showed different results because we had higher antioxidant capacity in pulps compared with the peels, mainly due to the presence of high concentrations of β -carotene, lycopene, and quercetin in pulps.

Septembre-Malaterre et al. (2016) concluded that the highest antioxidant capacity was found in passion fruit (*Passiflora edulis*) pulp (64% of DPPH reduced) when compared to other fruits including mango, pineapple, banana, and litchi exerted lower free radical-scavenging activities (45–58%). In agreement with data of DPPH assays, passion fruit exercised the highest free radical scavenging capacity too through oxygen radical absorbance capacity (ORAC) (14.08 μ M Trolox equivalent).

The antioxidant properties of yellow, purple and orange passion fruit are not affected by phenolic compounds by the Pearson's Correlation analysis. So, the antioxidant capacity measured in passion fruit samples is certainly related to the content of carotenoids and anthocyanins of these samples, mainly in the content of lycopene (as can be seen in the Fig. 1c).

Conclusion

This research showed that pulp of passion fruit and its residues as peels and seeds have a significant content of bioactive compounds, differing in type according to the species examined, as example, peel of purple passion fruit has more anthocyanins; pulp of orange passion fruit present a good source of lycopene and his peels a large content of β -carotene and phenolic compounds; the pulp of yellow passion fruit present more content of quercetin and antioxidant capacity ABTS and DPPH. These residues can be added to formulations that can enrich the foods and have therapeutic effects to the human health and too reduce the waste promoting a positive environmental and economic impact.

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Supplementary material:**Table 1S:** Methodology used for quantification of mineral in plant tissues.

Determinations	Methodology applied/limit od detection
Nitrogen (TKN) (%)	Kjeldahl/0.01 %
Phosphorus (%)	Wet digestion nitric perchloric/ICP-OES/0.01 %
Potassium (%)	Wet digestion nitric perchloric/ICP-OES/0.01 %
Calcium (%)	Wet digestion nitric perchloric/ICP-OES/0.01 %
Magnesium (%)	Wet digestion nitric perchloric/ICP-OES/0.01 %
Sulfur (%)	Wet digestion nitric perchloric/ICP-OES/0.01 %
Copper (mg/100 g)	Wet digestion nitric perchloric/ICP-OES/0.3 mg/kg
Zinc (mg/100 g)	Wet digestion nitric perchloric/ICP-OES/1 mg/kg
Iron (mg/100 g)	Wet digestion nitric perchloric/ICP-OES/2 mg/kg
Manganese (mg/100 g)	Wet digestion nitric perchloric/ICP-OES/2 mg/kg
Sodium (mg/100 g)	Wet digestion nitric perchloric/ICP-OES/10 mg/kg
Boron (mg/100 g)	Dry digestion/espec. abs. mol./1 mg/kg

CAPÍTULO 5**ARTIGO 2**

Stability of functional compounds and antioxidant activity of fresh and pasteurized orange passion fruit (*Passiflora caerulea*) during cold storage

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Stability of functional compounds and antioxidant activity of fresh and pasteurized orange passion fruit (*Passiflora caerulea*) during cold storage

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ABSTRACT

This research aimed to evaluate differences in the stability of physicochemical and color parameters, phenolic compounds, flavonoids, carotenoids and antioxidant capacity in fresh and pasteurized juice of orange passion fruit, respectively cold stored (8 °C) during 0–4 or during 0–15 days. The results showed that in the physicochemical analysis, no significant differences were observed comparing pasteurized and fresh juice during storage. The pasteurized juice showed higher concentrations of color parameters, phenolic compounds (15% more of retention for days 0 and 4), epigallocatechin gallate (40% in day 0 and 27% in day 4), lycopene (142% for day 0 and 39% for day 4), total carotenoids (114% in day 0 and 8% in day 4) and antioxidant capacity (12% in day 0 and 7% in day 4); already fresh juice retained more values of quercetin (79% in day 0 and 245% in day 4), α-carotene (57% in day 4), β-carotene and provitamin A (80% of retention in day 4). Therefore, the pasteurization processing was positive in orange passion fruit juice and improved the accessibility of most bioactive compounds.

1. Introduction

The world's tropical regions encompass extensive biodiversity, opening up an extensive range of exotic plants that produce fruit. Most of the fruit obtained are edible and gained popularity because of their sensory quality, unique taste, and nutraceutical values. The fruit can be consumed fresh or as juice, among other ways. Developing countries are the main producers of tropical fruits, like Brazil that is the third biggest country in fruit production, while developed countries are the main importers (Bhat & Paliyath, 2016; Ding, 2017).

The Brazil is the world's largest producer of passion fruit and the plantations reached 62 thousand ha, so in the same year yielded around 920 thousand tons of fruit, reaping 75% of the passion fruit produced in the world. The most popular passion fruit is intended mainly for the juice production industry (FAO, 2012).

The orange passion fruit (*Passiflora caerulea*), also known as *maracujá laranja* or *maracujá do mato*, belongs to the Passifloraceae family with occurrence in the Brazil, Mexico, Bermudas, Guiana, Peru, Paraguay, Argentina, and Uruguay. The fruit is ovoid or subglobose, orange when ripe, with 4–6 × 3.5–4 cm, the plant occurs in oaks, fields,

roadsides and forest edges. The pulp is sweet and has a red coloration (Mondin, Cervi, & Moreira, 2011).

The orange passion fruit juice has not been previously studied and it can present a huge potential for juice production, mainly because it is an easy-to-drink product that may contain many bioactive compounds that can contribute to human health. It is consumed as juice or fresh.

Many substances present in passion fruit, also called bioactive compounds, can contribute to beneficial effects in our body, they are phenolic compounds, flavonoids such as quercetin and catechins; carotenoids, being the β-carotene the majoritarian carotenoid in most species; and vitamins C and provitamin A (due to the presence of β-carotene). These compounds present antioxidant capacity, which will neutralize the free radicals present in many pathological processes: decrease of the risk of cardiovascular diseases such as arteriosclerosis and hypertension; neurodegenerative diseases such as Alzheimer's, decreased rate of glucose and cholesterol in blood, inflammatory and infectious processes, act as carcinogenesis and mutagenesis inhibitors, among others (Castañeda-Ovando, Pacheco-Hernández, Páez-Hernández, Rodríguez, & Galán-Vidal, 2009; González-Gallego, García-Mediavilla, Sánchez-Campos, & Tuñón, 2014; Zeraik, Pereira, Zuin, & Yariwake, 2010).

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The dietary antioxidants mode of action is interesting in understanding the potential health benefits. The main classes of antioxidants are present in fruit, beverages, vegetables and herbs. Antioxidants extraction, identification and quantification are being studied (Oroian and Escriche, 2015).

A wide variety of fruit juices is available in the market to attract consumers. The juice processing takes place right after harvest, being the pasteurization very important, since the appropriate combination of temperature and time allows the destruction of microorganisms and inactivation of enzymes, thereby increasing the shelf life of the product. However, in improper conditions, the binomial time/temperature can compromise the color, bioactive compounds content and other nutritional characteristics of the juice (Ashurst, 2016; Fellows, 2006).

This research aimed to evaluate the orange passion fruit juice during cold storage (8 °C): 0–4 days for fresh juice and 0–15 days for pasteurized juice in the stability of physicochemical and color parameters, phenolic compounds, flavonoids, carotenoids and antioxidant capacity.

2. Materials and methods

2.1. Sample and juice preparation

The orange passion fruit was acquired in the south of Brazil, in the city of Caxias do Sul – RS (Criuva District) between the geographic coordinates latitude 28° 52' 33.65" S, longitude 50° 58' 36.72" W and an average height of 860 M. The fruit (approximately 10 kg) was collected in the spring-summer of 2016/2017 when they were ripe with the color of peel orange. The orange passion fruit was placed in plastic bags and stored in a freezer at –18 °C. After the completely harvest, the fruit (frozen) were transported to the Laboratory of Bioactive Compounds of UFRGS (The Federal University of Rio Grande do Sul). The passion fruit was washed in running water and opened for the removal of the juice (that it is the pulp). A fabric to make cheese was used to separate the seed from the juice and then it was squeezed until the total juice was removed.

The orange passion fruit juice was divided into two parts: fresh juice and pasteurized juice. The fresh juice was packed in amber glass bottles of 300 mL (day 0, day 1 and day 4) and stored in a refrigerator (8 °C). The pasteurized juice was made as follows: the juice was placed in a beaker in a water bath until it reached at 88 °C for 15 s. The pasteurized juice was packed in amber glass bottles of 300 mL (day 0, day 1, day 4, day 6, day 13 and day 15) and stored in a refrigerator (8 °C), for analysis.

2.2. Physicochemical analysis

The total titratable acidity (TTA) was determined by titration method using standardized 0.1 M NaOH solution, and the results analyzing in % of citric acid following the Analytical Standards Instituto Adolfo Lutz (2008). The Total soluble solids (TSS) were determined with a Brix refractometer (Atago, Pocket Refractometer, Model Pal-1, Brazil) and the results were expressed in percentage (%). The pH was measured utilizing a digital bench pH meter (Model DM-22 – Digimed®, Brazil). All these analysis were made in triplicate.

2.3. Color parameters

The color of orange passion fruit juice was analyzed using a portable colorimeter (Konica Minolta, Model CR 400, Singapore) by the Commission Internationale de l'Eclairage (CIELAB system) by determining the values of L^* (lightness), a^* (component red-green) and b^* (yellow-blue component). All these analysis were made in triplicate. The delta E* (ΔE^*) was calculated through equation: $[\Delta L^*2 + \Delta a^*2 + \Delta b^*2]^{1/2}$.

2.4. Phenolic compounds

The juice (2.5 g) was extracted with 10 mL of ethanol and centrifuged at 15 °C (Cientec, CTR – 5000R, Brazil) at 5000 × g for 20 min. Then, 20 µL of supernatant was added to 1.58 mL of water and 100 µL of Folin-Ciocalteu (0.4 mol/L). After reaction (3 min), 300 µL of Na₂CO₃ was added, and the mixture kept at room temperature for 2 h. The absorbance was then read at 765 nm on a UV-visible spectrophotometer (Shimadzu, UV-1700 PharmaSpec, Japan). A standard curve was constructed to quantify phenolic compounds, using gallic acid at concentrations of 0 to 0.50 mg/mL. All these analysis were made in triplicate. The results analyzing in mg Gallic acid/100 g of fresh sample (Dranca and Oroian (2016); Dranca and Oroian (2017)).

2.5. Flavonoids

Epigallocatechin gallate and quercetin contents were analyzed according to Zeraik and Yariwake (2010) with some modifications. The juice (3 g) was extracted with 10 mL of methanol at room temperature. The extracts were centrifuged (Cientec, CTR – 5000R, Brazil) at 15,000 × g, 4 °C for 20 min, after which the supernatant was evaporated to 2.0 mL in a rotary evaporator (Fisatom, Model 801, Brazil). The resulting aqueous solution was filtered through a 0.45 µm Millex-HV PVDF membrane (Millipore, New Bedford, MA, USA) before HPLC analysis. The samples were prepared and analyzed in triplicate.

The HPLC-/DAD analyses were carried out on a Waters Alliance 2695 (Milford, MA, USA) liquid chromatograph connected to a model 2996 (DAD) diode array detector and controlled by Waters Empower software. The separation was performed using a C18 polymer column (250 mm × 4.6 mm id, 5 µm Vydac, 218TP). The samples were injected automatically (20.0 µL). The column was thermostatically controlled at 35 °C, and a 0.8 mL min⁻¹ flow rate was applied, using a linear gradient of 0.2% formic acid in water (solvent A) and 0.2% formic acid in acetonitrile (solvent B). The optimized gradients employed in passion fruit extracts were: 0–10 min, 15% B to 85% A and 10–20 min, 20% B in 80% A. The chromatogram was monitored at 330 nm, and UV spectra of individual peaks were recorded in the range of 200–400 nm.

The contents of epigallocatechin gallate and quercetin were determined by comparison with an external standard, injecting a new standard daily at 10 mg/mL for epigallocatechin gallate (≥97.0%, Sigma-Aldrich) and 20 mg/mL for Quercetin (≥98%, Sigma-Aldrich).

2.6. Carotenoid profile and provitamin A content

The profile of carotenoids in orange passion fruit juice was determined according to Mercadante and Rodriguez-Amaya (1998). The extraction of pigments was with acetone and the saponification in a KOH solution (10% in methanol) overnight. The extract was rotary evaporated (Fisatom, Model 801, Brazil) (T < 25 °C) and stored in a freezer (–18 °C) for quantification by high-performance liquid chromatography (HPLC). All these analysis were made in triplicate.

For HPLC analysis, the samples stored in a freezer were diluted with methyl *tert*-butyl ether (MTBE-JT Baker, CAS. Number 1634-04-4, purity 99.96%), sonicated (Unique, Model USC 1400) for 1 min and filtered (Millex LCR 0.45 µm, 13 mm) for injection into the HPLC (Agilent 1100 Series, Santa Clara, CA, USA), a UV-visible detector and with a quaternary system.

The column used was a C30 polymeric reverse phase (250 × 4.6 mm ID, 3 µm, YMC, model CT99SO3-2546WT). The mobile phase gradient (water:methanol:MTBE) (JT Baker, CAS Number 04.04.1634, 99.96% purity) commenced at 5:90:5, reaching 0:95:5 at 12 min, 0:89:11 at 25 min, 0:75:25 at 40 min, and finally 00:50:50 at 60 min. The temperature of the column was 33 °C and a flow rate of 1 mL/min (Spectra were obtained at a fixed wavelength of 450 nm for carotenoids).

Compounds were identified by comparing the sample retention times with the retention times obtained for controls. For quantification,

a standard curve was constructed for carotenoids over the following ranges: lutein 1 to 65 µg/mL ($\geq 95\%$, Sigma-Aldrich); zeaxanthin 1 to 40 µg/mL ($\geq 95\%$, Sigma-Aldrich); β -cryptoxanthin 4 to 100 µg/mL ($\geq 97\%$, Sigma-Aldrich); α -carotene 2 to 25 µg/mL ($\geq 95\%$, Sigma-Aldrich); β -carotene 5 to 50 µg/mL ($\geq 97\%$, Sigma-Aldrich) and lycopene ($\geq 85\%$ Sigma-Aldrich).

The limits of detection (LOD) and quantification (LOQ) were as follows. Lutein: 6.9×10^{-3} and 1.15×10^{-2} µg/g; zeaxanthin: 9.56×10^{-2} and 1.59×10^{-2} µg/g; β -cryptoxanthin: 2.11×10^{-2} and 3.51×10^{-2} µg/g; α -carotene: 1.97×10^{-2} and 3.28×10^{-2} µg/g; β -carotene: 6.53×10^{-2} and 10.89×10^{-2} µg/g and lycopene were 7×10^{-3} and 33×10^{-3} µg/g.

Provitamin A activity was calculated by the bioconversion factor following [Institute of Medicine \(2001\)](#), yielding a value of 12 mg of β -carotene with 1 mg of Retinol Activity Equivalent (RAE).

2.7. Antioxidant capacity

The method used to determine antioxidant capacity in pulps of yellow passion fruit was based on the sequestration of ABTS (2,2'-azobis(3-ethylbenzothiazoline-6-sulphonic acid) radical according to [Llorach, Tomás-Barberán, and Ferreres \(2004\)](#), with some modifications. Samples (2.5 g) were placed in 10 mL of ethanol and then centrifuged at 15 °C at 5000 \times g (Cientec, CTR – 5000R, Brazil) for 20 min. The supernatant was diluted to three concentrations (15%, 20%, and 25%). The aliquots of each concentration (300 µL) were treated with 1.7 mL of ABTS (7 mM). The absorbance was read at 734 nm (Shimadzu, UV-1700 PharmaSpec, Japan). All these analysis were made in triplicate. The results were presented as equivalents Trolox (µmol/g fresh weight).

2.8. Statistical analysis

Data were analyzed by ANOVA and Tukey's mean comparison test at a significance level of 5% using Statistica 12.0 (StatSoft, São Paulo, Brazil).

2.8.1. Principal component analysis

The physicochemical parameters and bioactive concentrations have been submitted to Principal Component Analysis and this is a statistical technique used to reduce the dimensionality of a data set containing a large number of interrelated variables. The analysis is performed to maintain the maximum variance present in the data. This reduction produces a new reduced and uncorrelated set of variables, called principal components. These components are then chosen to ensure that the former retains the greater part of the variance present in the original variables.

The fresh and pasteurized juices were analyzed in the TSS, pH, TTA, color parameters, flavonoids, carotenoids, and antioxidant capacity and the results are shown in [Fig. 1](#). By analyzing the components, the variance of the data were accounted for the significant contributions of 52.70% for the first principal component and 29.17% for the second principal components.

3. Results and discussion

The total soluble solids (TSS) and the pH in orange passion fruit fresh juice increased after the time of storage, but for total titratable acidity (TTA), no significant changes were observed. In the pasteurized juice, the TSS and the pH decreased after the storage (15 days) and for TTA no significant changes were found.

Comparing the pasteurized and fresh juice in the same time of storage, the results indicate that no significant differences were observed ([Fig. 1](#) and [Table 1](#)). Similar results were found by [Jachna, Hermes, Flôres, and Rios \(2016\)](#) that analyzed the pindo palm (*Butia capitata*) fresh and pasteurized juice and concluded that for pH, TTA and TTS

neither the storage days nor the pasteurization process caused changes in these physicochemical parameters.

Low variations of color were detected in orange passion fruit juices ([Table 2](#)). In the fresh juice, no significant changes were found. For the pasteurized juice, it was observed that day 0 presented lower values of color L^* , a^* and b^* according to the storage days. It was evaluated too the Delta E* (ΔE^*), that indicates the total color difference between L^* , a^* and b^* and we can observe that the values stayed below of 1,37. In this study was observed too that the pasteurized juice increased the color parameters L^* , a^* and b^* compared to fresh juice; Similar results found by [Mena et al. \(2013\)](#) that evaluated the pomegranate (*Punica granatum* L.) juice after pasteurization processing. It is can be explained because the pasteurization processing can suspend particles in juice with a partial precipitation ([Genovese, Elustondo, & Lozano, 1997](#)).

[Jachna et al. \(2016\)](#) found similar results too, is that the color parameters of pindo palm (*Butia capitata*) juice increased after fresh and pasteurized juices. These increases in color parameters can be elucidated by the fact that the products of β -carotene oxidation (known as apocarotenoids and epoxides) and other carotenoids have red coloration.

No decreases of phenolic compounds in the storage of orange passion fruit fresh juice were observed (days 0–4), but for pasteurized juice, significant differences were found after day 4 of storage. Even with a decrease in the phenolic compounds, the pasteurized juice retained higher concentrations of these compounds when compared to the same days of storage in fresh juice. Different results were found by [Mena et al. \(2013\)](#) who concluded that after pasteurization at 90 °C, no significant changes were observed in relation to pomegranate fresh juice. [Saeeduddin et al. \(2015\)](#) found that after pasteurization (65 and 95 °C) of pear juice (*Pyrus bretschneideri* Read.) the content of phenolic compounds decreased. [Odrizola-Serrano, Soliva-Fortuny, and Martín-Belloso \(2008\)](#) found the highest content of phenolic compounds in fresh strawberry juice compared to pasteurization processing (90 °C for 60 s) and with the storage days. [Jachna et al. \(2016\)](#) showed that pasteurization processing did not affect the phenolic compounds compared to pindo palm fresh juice, but after four days of storage, the phenolic compounds in pasteurized juice decreased (similar results compared to this research). Therefore, the pasteurized juice of orange passion fruit presented higher concentrations compared to the fresh due to the rupture of the vegetal tissue with the heating being stable for 4 days ([Fig. 2](#)).

The flavonoids detected in orange passion fruit juices (epigallocatechin gallate and quercetin) decreased after the days of storage ([Table 3](#)), so for epigallocatechin gallate, we found higher concentrations in pasteurized juice when compared to fresh juice in the same days of storage. [Odrizola-Serrano et al. \(2008\)](#) concluded that no significant differences were found in the content of myricetin, quercetin, and kaempferol after the thermal processing in strawberry juice (*Fragaria ananassa* Duch, cultivar *Camarosa*), but the content of these flavonoids decreased significantly in the storage days (from the seventh day). For quercetin, the contrary was observed: higher values in the fresh juice. [Saeeduddin et al. \(2015\)](#) found similar results (65 and 95 °C) in the pear juice (*Pyrus bretschneideri* Read.): the content of total flavonoids were higher for fresh juices (expressed as catechin), that is, the flavonoids decreased after pasteurization.

The amounts of flavonoids in orange passion fruit varied according to the type analyzed and the thermal processing. These changes can be explained by the oxidative stability of its constituents, extraction/hydrolysis and analytical methods used ([Jongberg, Tomgren, Gunvig, Skibsted, & Lund, 2013](#); [Kosar, Kafkas, Paydas, & Baser, 2004](#)).

These flavonoids (epigallocatechin gallate and quercetin) are good for health and the juice of orange passion fruit analyzed is an interesting source, but for quercetin, the pasteurization processing is not the most appropriate, being preferable the consumption of fresh juice.

In relation to carotenoids profile ([Table 4](#)), we observed that the majoritarian carotenoid was the lycopene. Several changes were

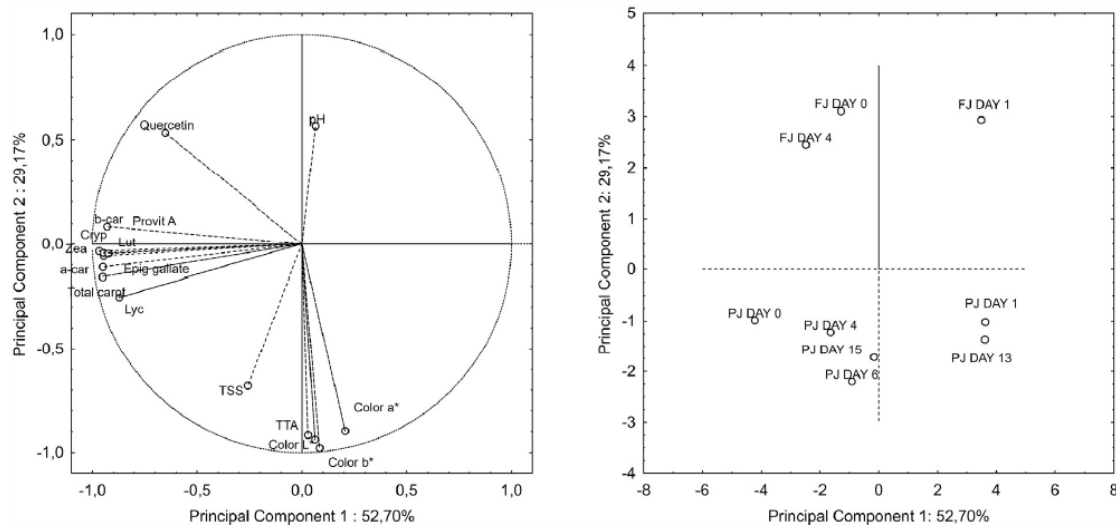


Fig. 1. Principal component analysis of fresh and pasteurized juices: TSS, TTA, pH, color parameters, carotenoids and flavonoids.

Legend:

Lut: lutein; Zea: zeaxanthin; Cyp: β -cryptoxanthin; a-car: α -carotene; b-car: β -carotene; Lyc: lycopene; Provit A: provitamin A; TC: total carot; Lyc: lycopene; Epig gallate: epigallocatechin gallate.

detected during the storage: in fresh juice the α -carotene, β -carotene, lycopene, total carotenoids and provitamin A increased after four days of storage; in pasteurized juice the lutein remained constant during 0–15 days of storage, lycopene remained constant during 0–6 days; β -cryptoxanthin remained constant only 0–4 days of storage; the carotenoids zeaxanthin, α -carotene, β -carotene, total carotenoids and provitamin A decreased after the days of storage.

Comparing the fresh and pasteurized juices, the carotenoids lutein and zeaxanthin did not present changes in four days of storage; the fresh juice presented lower concentrations of β -cryptoxanthin, lycopene, and total carotenoids; so α -carotene, β -carotene e provitamin A were higher after four days of storage compared to pasteurized juice. These differences can be explained by the fact that shortly after heating (in this case, the pasteurization) the plant membrane is broken and the compounds become more available; on the other hand, with the plant membrane ruptured, the compounds are more exposed to degradation as the storage time. The food processing such as mechanical homogenization and pasteurization modifies the chromoplast structure having a beneficial effect on the bioaccessibility and bioavailability of carotenoids in different food matrices (Miglio, Chiavaro, Visconti, Fogliano, & Pellegrini, 2008; Stinco et al., 2012; Van Buggenhout et al., 2010; Yeum & Russell, 2002).

Different results were found by Achir et al. (2016) that evaluated *cis*-violaxanthin, lutein, zeaxanthin, *cis*-antheraxanthin and β - β -cryptoxanthin in orange juice (*Citrus sinensis* L. Osbeck) and concluded that after pasteurization at 70 °C for 150 min, these carotenoids decreased; so they analyzed too the grapefruit (*Citrus paradisi* Macf) juice and no

Table 2

Color parameters L^* , a^* , b^* and ΔE^* in orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage.

	Color L^*	Color a^*	Color b^*	ΔE^*
FJ day 0	29.23 \pm 0.59 ^{ab}	15.02 \pm 0.65 ^{ab}	5.14 \pm 0.24 ^{ab}	1.37
FJ day 1	27.60 \pm 1.33 ^{ab}	14.40 \pm 0.76 ^{ab}	4.85 \pm 0.17 ^{bb}	1.31
FJ day 4	27.47 \pm 1.01 ^{ab}	15.11 \pm 0.67 ^{ab}	5.06 \pm 0.03 ^{abb}	0.32
PJ day 0	30.74 \pm 0.03 ^{ba}	15.57 \pm 0.05 ^{ba}	6.88 \pm 0.01 ^{ba}	0.04
PJ day 1	31.31 \pm 0.36 ^{ba}	17.74 \pm 0.47 ^{ba}	7.70 \pm 0.32 ^{ba}	1.26
PJ day 4	31.91 \pm 0.20 ^{ba}	17.91 \pm 0.22 ^{ba}	7.94 \pm 0.17 ^{ba}	0.59
PJ day 6	32.35 \pm 0.29 ^a	18.55 \pm 0.14 ^a	8.12 \pm 0.23 ^a	0.70
PJ day 13	32.18 \pm 0.56 ^a	18.08 \pm 0.23 ^a	7.78 \pm 0.25 ^a	1.19
PJ day 15	31.94 \pm 0.75 ^a	17.76 \pm 0.60 ^a	8.25 \pm 0.05 ^a	1.11

The samples were treated by ANOVA, followed by Tukey's test ($P < .05$). Upper-case letters highlight significance for different treatments employed for each day, lower-case for days of storage.

significant differences were found for the carotenoids tested (lycopene and β -carotene) after pasteurization processing.

Lee and Coates (2003) concluded that after the pasteurization (90 °C for 30 s) of Valencia orange juice, the content of some carotenoids decreased, because the heating losses the most pigments of 5,6-epoxide carotenoids (such as violaxanthin (−46.40%), *cis*-violaxanthin (−19.70%), and antheraxanthin (−24.80%)). Violaxanthin is one of the most labile carotenoids and is easily isomerized in the presence of acid to luteoxanthin and then to auroxanthin. However, for 5,8-epoxide carotenoids, it was observed slight increases in mutatoxanthin and

Table 1

Evaluation of TSS, pH and TTA of orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage (with mean and standard deviation).

		Day 0	Day 1	Day 4	Day 6	Day 13	Day 15
FJ	TSS	12.15 \pm 0.07 ^{cb}	12.70 \pm 0.17 ^{ba}	13.17 \pm 0.06 ^{aa}			
	pH	4.64 \pm 0.00 ^{ba}	4.69 \pm 0.04 ^{ba}	4.70 \pm 0.02 ^{ba}			
	TTA	2.79 \pm 0.01 ^{ab}	2.90 \pm 0.04 ^{aa}	2.91 \pm 0.09 ^{aa}			
PJ	TSS	14.03 \pm 0.06 ^{sa}	13.37 \pm 0.38 ^{ba}	13.43 \pm 0.32 ^{ba}	13.07 \pm 0.06 ^b	13.30 \pm 0.00 ^b	13.30 \pm 0.00 ^b
	pH	4.58 \pm 0.01 ^{bc}	4.67 \pm 0.02 ^{sa}	4.70 \pm 0.00 ^{ba}	4.60 \pm 0.01 ^b	4.57 \pm 0.01 ^{bc}	4.57 \pm 0.01 ^c
	TTA	3.16 \pm 0.07 ^{sa}	3.19 \pm 0.03 ^{sa}	3.06 \pm 0.07 ^{sa}	3.18 \pm 0.05 ^a	3.06 \pm 0.03 ^a	3.10 \pm 0.06 ^a

The samples were treated by ANOVA, followed by Tukey's test ($P < .05$). TTA is expressed as percent of acid citric. Upper-case letters highlight significance for columns, lower-case for rows.

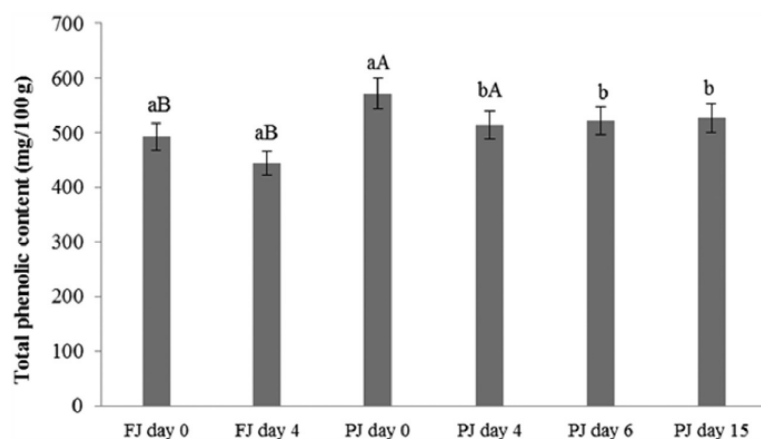


Fig. 2. Total phenolic content in orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage. The samples were treated by ANOVA, followed by Tukey's test ($P < .05$). Upper-case letters highlight significance for different treatments employed for each day, lower-case for days of storage.

Table 3
Flavonoids in orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage.

	Epigallocatechin gallate	Quercetin
FJ day 0	2.20 ± 0.06 ^{aB}	375.97 ± 0.88 ^{aA}
FJ day 4	1.52 ± 0.07 ^{bB}	190.07 ± 2.12 ^{bA}
PJ day 0	3.09 ± 0.02 ^{aA}	209.79 ± 3.98 ^{aB}
PJ day 4	1.93 ± 0.09 ^{bA}	55.07 ± 0.63 ^{bB}
PJ day 6	1.86 ± 0.06 ^b	40.77 ± 2.04 ^c
PJ day 15	1.88 ± 0.02 ^b	37.64 ± 0.39 ^c

The samples were treated by ANOVA, followed by Tukey's test ($P < .05$). Upper-case letters highlight significance for different treatments employed for each day, lower-case for days of storage.

luteoxanthin, after pasteurization processing.

Gama and Sylos (2007) found losses in violaxanthin and lutein due to the pasteurization processing of Brazilian Valencia orange juices, nevertheless, for zeaxanthin (which protects against cataracts and age-related macular degeneration), β -carotene, α -carotene, and β - β -cryptoxanthin (that have provitamin A activity) no significant decreases were detected after pasteurization. Jachna et al. (2016) concluded that the β -carotene decreased after the pasteurization of pindo palm juice already to the β - β -cryptoxanthin, the contrary was observed (this carotenoid increased after the pasteurization).

In the antioxidant capacity measured with ABTS radical (Fig. 3), the fresh juice presented lower antioxidant capacity compared to pasteurized juice, so it did not decrease after four days of storage, likewise that it happened with the pasteurized juice that only after day 6, the antioxidant capacity decreased. Mena et al. (2013) found similar conclusions in pomegranate juice, so the antioxidant capacity ABTS increased after pasteurization processing. This increase may be explained by the

fact that the pasteurization processing increased the extraction of anthocyanins in the pomegranate juice due to the better extraction of these bioactive compounds after the heating.

Already Saeeduddin et al. (2015) concluded that after pasteurization (65 and 95 °C) of pear juice (*Pyrus bretschneideri* Read.) the antioxidant capacity (expressed as ascorbic acid equivalent) decreased due to the fact that phenolic compounds and ascorbic acid are highly thermo-sensitive compounds. Odriozola-Serrano et al. (2008) found decreases in the antioxidant capacity ABTS after the pasteurization and seven days of storage (4 °C) in strawberry juice (*Fragaria ananassa* Duch, cultivar Camarosa) too. Therefore, the pasteurized juice of strawberry decreased in the content of total anthocyanins and vitamin C, which could explain the decrease in antioxidant capacity found in treated samples compared to fresh juice.

4. Conclusions

The findings of this research showed that the pasteurization processing and fresh juice of orange passion fruit did not alter after the days of storage in the pH, TTA, and TSS. The pasteurization processing presented higher values of color parameters during all the storage (fifteenth days). In the content of epigallocatechin gallate, α -carotene, β -carotene and provitamin A, the fresh juice demonstrated more retention. Thus, the pasteurized juice if consumed until the fourth day at cold storage is recommended because it had beneficial effects in the content of bioactive compounds, as phenolic compounds, quercetin, β -cryptoxanthin, total carotenoids, lycopene (that was the majoritarian carotenoid, being that this compound is related to literature as a functional food) and a better antioxidant capacity too, making them more available.

Table 4
Carotenoids profile in orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage.

	FJ day 0	FJ day 4	PJ day 0	PJ day 4	PJ day 6	PJ day 15
Lutein	8.59 ± 0.41 ^{aA}	8.75 ± 0.21 ^{aA}	9.33 ± 0.39 ^{aA}	8.79 ± 0.45 ^{aA}	8.45 ± 0.28 ^a	8.18 ± 0.06 ^a
Zeaxanthin	10.20 ± 0.25 ^{ab}	9.40 ± 0.27 ^{aA}	11.82 ± 0.09 ^{aA}	10.28 ± 0.20 ^{bA}	8.50 ± 0.29 ^c	8.65 ± 0.23 ^c
β -cryptoxanthin	35.33 ± 1.60 ^{aA}	34.65 ± 0.42 ^{ab}	37.35 ± 1.31 ^{aA}	37.99 ± 0.26 ^{aA}	31.78 ± 0.35 ^b	32.11 ± 0.98 ^b
α -carotene	7.02 ± 0.26 ^{bb}	10.05 ± 0.02 ^{aA}	19.78 ± 0.36 ^{aA}	6.41 ± 0.08 ^{bb}	6.14 ± 0.07 ^b	6.28 ± 0.23 ^b
β -carotene	37.98 ± 1.78 ^{bb}	99.63 ± 1.65 ^{aA}	103.59 ± 1.39 ^{aA}	55.27 ± 2.73 ^{bb}	34.27 ± 0.53 ^c	34.08 ± 1.43 ^c
Lycopene	108.39 ± 3.29 ^{bb}	188.97 ± 5.37 ^{bb}	262.02 ± 4.81 ^{aA}	263.07 ± 4.37 ^{aA}	276.33 ± 0.54 ^a	80.43 ± 4.02 ^b
Total carotenoids	207.51 ± 0.00 ^{bb}	351.45 ± 4.63 ^{bb}	443.89 ± 5.58 ^{aA}	381.80 ± 7.40 ^{bA}	365.47 ± 0.16 ^b	169.73 ± 6.82 ^c
Provitamin A*	3.17 ± 0.15 ^{bb}	8.30 ± 0.14 ^{aA}	8.63 ± 0.12 ^{aA}	4.61 ± 0.23 ^{bb}	2.86 ± 0.04 ^c	2.84 ± 0.12 ^c

The samples were treated by ANOVA, followed by Tukey's test ($P < .05$). Upper-case letters highlight significance for different treatments employed for each day, lower-case for days of storage.

* Expressed as μ g RAE (Retinol Activity Equivalent).

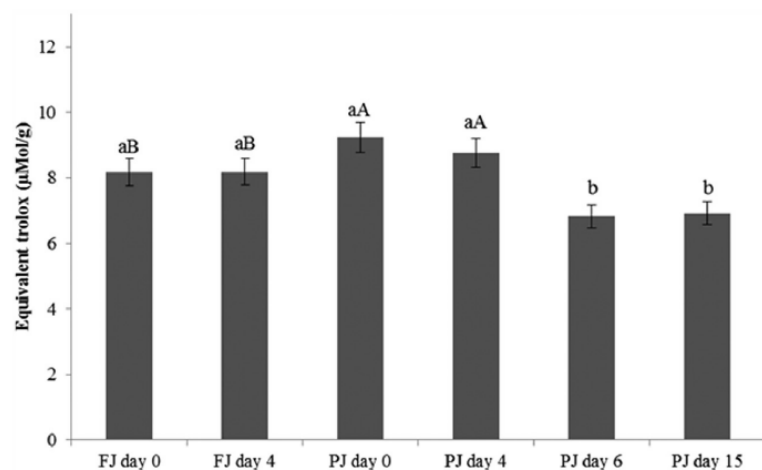


Fig. 3. Antioxidant capacity measured with ARTS radical in orange passion fruit fresh juice (FJ) and pasteurized juice (PJ) during cold storage.

The samples were treated by ANOVA followed by Tukey's test ($P < .05$). Upper-case letters highlight significance for different treatments employed for each day, lower-case for days of storage.

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CAPÍTULO 6**ARTIGO 3****Physicochemical properties, minerals and bioactive compounds in four cultivars of
sour passion fruit pulp**

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Physicochemical properties, minerals and bioactive compounds in four cultivars of sour passion fruit pulp

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Abstract

The sour passion fruit is a native fruit from Brazil, being the largest producer and consumer in the world. This research aimed to evaluate the physicochemical properties, color parameters, proximate composition, minerals, bioactive compounds and antioxidant capacity of four cultivars of sour passion fruit: ‘SCS437 Catarina’, ‘BRS Gigante Amarelo’, ‘BRS Rubi do Cerrado’ and ‘BRS Sol do Cerrado’. The results were: ‘BRS Rubi do Cerrado’ is sweeter and the ‘BRS Sol do Cerrado’ is more acid; the ‘BRS Gigante Amarelo’ showed more content of moisture, total fat, ash, proteins and total fiber, total phenolic content, epigallocatechin gallate, quercetin, carotenoids and provitamin A; the ‘BRS Rubi do Cerrado’ showed higher content of kaempferol; for minerals tested the ‘BRS Sol do Cerrado’ exhibit higher concentrations of all minerals,

except for copper. 'BRS Gigante Amarelo' presented higher bioactive compounds and 'BRS Sol do Cerrado' higher minerals content. The cultivars 'SCS437 Catarina' and 'Gigante Amarelo' showed a higher antioxidant capacity.

Index terms: β -carotene, provitamin A, total fiber, quercetin, potassium.

Propriedades físico-químicas, minerais e compostos bioativos em quatro cultivares de polpa de maracujá azedo

Resumo

O maracujá azedo é uma fruta nativa do Brasil, sendo o maior produtor e consumidor no mundo. Esta pesquisa teve o objetivo de avaliar quatro cultivares de maracujá azedo: 'SCS437 Catarina', 'BRS Gigante Amarelo', 'BRS Rubi do Cerrado' and 'BRS Sol do Cerrado' nas propriedades físico-químicas, parâmetros de cor, composição centesimal, minerais, compostos bioativos e capacidade antioxidante. Os resultados foram: 'BRS Rubi do Cerrado' é o mais doce e o 'BRS Sol do Cerrado' é mais ácido; o 'BRS Gigante Amarelo' apresentou maior conteúdo de umidade, lipídeos totais, cinzas, proteínas e fibra total, compostos fenólicos totais, epigallocatequina galato, quercetina, carotenoides e provitamina A; o 'BRS Rubi do Cerrado' apresentou maior retenção de camferol; para os minerais testados o 'BRS Sol do Cerrado' exibiu maiores concentrações de todos os minerais, exceto o cobre. 'BRS Gigante Amarelo' apresentou maiores teores de compostos bioativos e o 'BRS Sol do Cerrado' maior conteúdo de minerais. As cultivares 'SCS437 Catarina' e 'Gigante Amarelo' apresentaram uma maior capacidade antioxidante.

Termos de indexação: β -caroteno, provitamina A, fibra total, quercetina, potássio.

Introduction

The plants of the genus *Passiflora*, belonging to the family Passifloraceae Juss. Ex DC, are known as passion fruit. It is a highly diverse genus, presenting about 520 species with distribution in the Americas, mainly. Brazil and Colombia are centers of diversity in particular, where 30 % of the species are found in these countries, approximately (Cerqueira-Silva et al., 2014).

The yellow passion fruit or passion fruit sour (*Passiflora edulis* Sims) is a native fruit from Brazil, being the largest producer and consumer in the world. The selections with yellow peel are preferable of the consumer, and these are the most cultivated. It is an important species for family farming in Brazil. The food industry is the main source of utilization of the sour passion fruit pulp. Therefore the pulp is applied for the packing and commercialization of the juice for all the country. The production of sour passion fruit in Brazil begins in early summer and runs through autumn in southeast and south, and in the north, northeast and midwest produce all year round (Bernacci et al., 2008; Silva et al., 2013; Zeraik & Yariwake, 2010; Souza et al., 2012).

The experimental station EPAGRI-SC (Agricultural Research and Rural Extension Company of Santa Catarina) developed in 2015 the cultivar of sour passion ‘SCS437 Catarina’, and the EMBRAPA (Brazilian Company of Agricultural Research) developed the cultivars BRS Gigante Amarelo, BRS Rubi do Cerrado and BRS Sol do Cerrado. The cultivars SCS437 Catarina, BRS Gigante Amarelo and BRS Sol do Cerrado have characteristics of yellow colored peel, yellow-orange pulp, and black seeds. Already, the BRS Rubi do Cerrado cultivar presents 50% of fruits of red or purplish peel, yellow-orange pulp and black seeds (EMBRAPA, 2014).

The launch of new cultivars of different breeding programs has provided farmers with better results regarding yield and quality of fruit offered to consumers. Besides, due to the high genetic variability of the Passifloras, the different cultivars have presented different degrees of adaptation to the different regions where they are

cultivated. In addition, these differences influence the quality of fruit produced, both in the visual characteristics of the fruit and in the chemical composition of the pulp, peel, and seeds (Bruckner et al., 2002; Meletti et al., 2005; Meletti 2011).

The major factors that limit productivity are the absence of productive varieties that are adapted to local conditions, a lack of resistance to major diseases, and the use of seeds obtained by open pollination from plants in the planted area, which generates low-quality fruit and inconsistent production (Cerqueira-Silva et al., 2014). So, these new cultivars have higher levels of resistance to the main diseases of sour passion fruit (viruses, bacterial blight, anthracnose, and scab) and high levels of productivity. These cultivars present more robust peel, showing a greater resistance to the transport and causing the fruit to reach the markets and food industry without damages/beats. The longer shelf life and good pulp yield are also worthy of note (EMBRAPA, 2014).

The sour passion fruit pulp exhibits a high acceptance by consumers, for having a very pleasant aroma and flavor. Therefore these are not their main adjectives, as it has also been shown in the literature that these pulps have high levels of bioactive compounds, being mainly phenolic compounds, flavonoids, carotenoids, provitamin A and vitamin C with antioxidant capacity. These compounds have antioxidant power and have been described by numerous health benefits such as the neutralization of free radicals that accelerate aging and the incidence of some diseases (Janzantti et al., 2012, López-Vargas et al., 2013, Sergent et al., 2010, Tanaka et al., 2012).

Some researchers have already been carried out with passion fruit pulp and a number of its bioactive compounds were identified and quantified. Janzantti et al. (2012), Souza et al. (2012) and Septembre-Malaterre et al. (2016) found values of phenolic compounds that varied between 529, 1545 and 286 mg GAE/100 g. Cohen et al. (2008) obtained the lowest levels of total flavonoids: 3.28 mg/100 g and López-Vargas et al. (2013) found the highest routine concentrations: 1363 mg/100 g. Silva et

al. (2013) found 1362 μg of β -carotene/100 g and Souza et al. (2012) detected 111.6 μg of total carotenoids/100 g passion fruit pulp.

Thereby, this research aimed to evaluate four new cultivars of sour passion fruit in the physicochemical properties, color parameters, proximate composition, minerals, phenolic compounds, flavonoids, carotenoids and antioxidant capacity.

Material and methods

Plant materials and sample preparation

The four new cultivars of sour passion fruit pulp (*Passiflora edulis* flavicarpa) were obtained by EPAGRI-SC (Agricultural Research and Rural Extension Company of Santa Catarina) and EMBRAPA (Brazilian Company of Agricultural Research). They are: SCS437 Catarina, BRS Gigante Amarelo, BRS Rubi do Cerrado and BRS Sol do Cerrado. The fruits were harvested when they were ripe, that is, when the color of the skin was 1/3 yellow ('SCS437 Catarina', 'BRS Gigante Amarelo' and 'BRS Sol do Cerrado') or purple ('BRS Rubi do Cerrado'). After harvesting, the fruits were packed in black plastic bags and transported to the Laboratory of Bioactive Compounds of UFRGS (Federal University of Rio Grande do Sul). Then the fruits were cut in half and made a separation of the pulp that were packed in plastic bags and stored in a freezer at - 18 °C until analysis.

Physicochemical analysis and yield

The titratable acidity was determined by titration method using standardized 0.1 M NaOH solution, and the results analyzing in g/100 g of citric acid following the Analytical Standards Instituto Adolfo Lutz (IAL, 2008). The Total soluble solids (TSS) were determined with a Brix refractometer, and the results were expressed in percentage

(%). The yield was calculated by the percentage ratio (%) of the weight of the full fruit and the fruit without the peel and seeds.

Color parameters

The color of four new cultivars of sour passion fruit was analyzed using a portable colorimeter (Konica Minolta Model CR 400, Singapore) by the *Commission Internationale de l'Eclairage* (CIELAB system) by determining the values of L^* (lightness), a^* (component red-green) and b^* (yellow-blue component).

Proximate Composition

All analyses were performed according to AOAC (2000). Lipids were obtained by cold extraction and ash was determined in a muffle furnace at 550 °C. Protein content was determined by the Kjeldahl method with a conversion factor of 5.75. Total dietary fiber was determined by the enzymatic-gravimetric method, moisture by gravimetry. Carbohydrates were estimated by difference.

Minerals

All the samples of sour passion fruit pulp were lyophilized (Liotop, L101, Brazil) before mineral analysis. The minerals quantification were performed according to the methodology of atomic emission spectrometry with inductively coupled plasma source (ICP-OES) described by Tedesco & Gianello (2004). The potassium, phosphorus, magnesium, calcium, sulfur, zinc, iron, manganese and copper we used with wet digestion nitric-perchloric and boron was applied with dry digestion. The Nitrogen (TKN) was applied by methodology Kjeldahl.

Total phenolic content

Samples (2.5 g) were extraction with 10 mL of ethanol and centrifuged at 15 °C (Cientec, CTR – 5000R, Brazil) at 5000 x g for 20 minutes. Then, 20 µL of supernatant was added to 1.58 mL of water and 100 µL of Folin-Ciocalteu (0.4 mol/L). After reaction (3 minutes), 300 µL of Na₂CO₃ was added, and the mixture kept at room temperature for 2 hours. The absorbance was then read at 765 nm on a UV-visible spectrophotometer (Shimadzu, UV-1700 PharmaSpec, Japan). A standard curve was constructed to quantify phenolic compounds, using gallic acid at concentrations of 0 to 0.50 mg/mL. The results analyzing in mg gallic acid/100 g of dry sample (Swain & Hillis, 1959).

Flavonoids

Epigallocatechin gallate, kaempferol and quercetin contents were analyzed according to Zeraik & Yariwake (2010) with modifications. The pulps (10 g) were extracted with 30 mL of methanol at room temperature. The extracts were centrifuged at 15000 x g, 4 °C for 20 min, after which the supernatant was evaporated to 2.0 mL in a rotary evaporator. The resulting aqueous solution was filtered through a 0.45 µm Millex-HV PVDF membrane (Millipore, New Bedford, MA, USA) before HPLC analysis. The samples were prepared and analyzed in triplicate.

The HPLC-/DAD analyses were carried out on a Waters Alliance 2695 (Milford, MA, USA) liquid chromatograph connected to a model 2996 (DAD) diode array detector and controlled by Waters Empower software. The separation was performed using a C18 polymer column (250 mm × 4.6 mm id, 5 µm Vydac, 218TP). The samples were injected automatically (20.0 µL). The column was thermostatically controlled at 35 °C, and a 0.8 mL min⁻¹ flow rate was applied, using a linear gradient of 0.2% formic acid in water (solvent A) and 0.2% formic acid in acetonitrile (solvent B). The

optimized gradients employed in passion fruit extracts were: 0–10 min, 15 % B to 85 % A and 10–20 min, 20 % B in 80 % A. The chromatogram was monitored at 330 nm, and UV spectra of individual peaks were recorded in the range of 200–400 nm.

The contents of epigallocatechin gallate, kaempferol and quercetin were determined by comparison with an external standard, injecting a new standard daily at 10mg/mL for epigallocatechin gallate (≥ 97.0 %, Sigma-Aldrich), 5 mg/mL for kaempferol ($> 99\%$, Sigma-Aldrich) and 20 mg/mL for Quercetin ($> 98\%$, Sigma-Aldrich).

Carotenoid profile and provitamin A content

The profile of carotenoids in sour passion fruit pulps was determined according to Mercadante & Rodriguez-Amaya (1998). The extraction of pigments was with acetone and the saponification in a KOH solution (10 % in methanol) overnight. The extract was rot evaporated (Fisatom, Model 801) ($T < 25$ °C) and stored in a freezer (-18 °C) for quantification by HPLC (High Performance Liquid Chromatography).

For HPLC analysis, the samples stored in a freezer were diluted with methyl *tert*-butyl ether (MTBE-JT Baker, CAS. Number 1634-04-4, purity 99.96%), sonicated (Unique, Model USC 1400) for 1 minute and filtered (Millex LCR 0.45 μ m, 13 mm) for injection into the HPLC (Agilent 1100 Series, Santa Clara, CA, USA), a UV-visible detector and with a quaternary system. The column used was a C30 polymeric reverse phase (250×4.6 mm ID, 3 μ m, YMC, model CT99SO3-2546WT). The mobile phase gradient (water:methanol:MTBE) (JT Baker, CAS Number 04.04.1634, 99.96% purity) commenced at 5:90:5, reaching 0:95:5 at 12 minutes, 0:89:11 at 25 minutes, 0:75:25 at 40 minutes, and finally 00:50:50 at 60 minutes. The temperature of the column was 33 °C and a flow rate of 1 mL/min (Spectra were obtained at a fixed wavelength of 450 nm for carotenoids).

Provitamin A activity was calculated by the bioconversion factor following Institute of Medicine (2001), yielding a value of 12 mg of β -carotene with 1 mg of Retinol Activity Equivalent (RAE).

Antioxidant capacity

The method used to determine antioxidant capacity in pulps of sour passion fruit was based on the sequestration of ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) radical according to Brand-Williams et al. (1995), as adapted by Embrapa Agroindústria Tropical. Samples (2.5 g) were placed in 10 mL of ethanol and then centrifuged at 15 °C at 5000 x g (Cientec, CTR – 5000R, Brazil) for 20 minutes. The supernatant was diluted to three concentrations (20 %, 40 %, and 60 %). The aliquots of each concentration (300 μ L) were treated with 1.7 mL of ABTS (7 mM). The absorbance was read at 734 nm (Shimadzu, UV-1700 PharmaSpec, Japan). The results were presented as equivalent Trolox (μ Mol/g dry weight).

Statistical Analysis

Data were analyzed by ANOVA and Tukey's mean comparison test at a significance level of 5% using Statistica 12.0 and Principal Component Analysis (PCA) (StatSoft, São Paulo, Brazil).

Results and discussion

The cultivar that presented a higher pulp yield in sour passion fruit was the SCS437 Catarina (Table 1).

PCA (Principal Component Analysis) is a statistical technique used to reduce the dimensionality of a data set containing a large number of interrelated variables. The analysis is performed to maintain the maximum variance present in the data. This

reduction produces a new reduced and uncorrelated set of variables, called principal components. These components are then chosen to ensure that the former retains the greater part of the variance present in the original variables.

The pulps of four cultivars of passion fruits were analyzed in the physicochemical, color parameters, proximate composition, minerals, total phenolic content, flavonoids, carotenoids and antioxidant capacity and the results are shown in figure 1 and 2 and in Table 1.

By analyzing the components of physicochemical, color parameters, proximate composition and minerals, the variance of the data were accounted for the significant contributions of 54.05 % for the first principal component and 32.92 % for the second principal components (Figure 1).

The sour passion fruit pulp BRS Rubi do Cerrado cultivar is sweeter and the cultivar BRS Sol do Cerrado is more acid. In relation to color parameters, the luminosity (L^*) was higher in SCS437 Catarina cultivar, however no differences in color a^* and b^* was found in cultivars tested, except for the BRS Gigante Amarelo that showed fewer results in relation to other cultivars studied. However. This cultivar, showed a higher content of moisture, total fat, ash, proteins and total fiber, indicating that the content of carbohydrates is mainly given a number of fibers found (Table 1 and Figure 1).

Table 1. Physicochemical, yield, color parameters and proximate composition (g/100g dry weight – except for moisture) in different cultivars of passion fruit pulp (mean and standard deviation).

	SCS437	BRS Gigante	BRS Rubi do	BRS Sol do
	Catarina	Amarelo	Cerrado	Cerrado

TSS (°Brix)	10.62 ± 0.20 ^c	11.88 ± 0.08 ^b	12.50 ± 0.20 ^a	12.12 ± 0.18 ^{ab}
Acidity (% citric acid)	3.65 ± 0.05 ^d	4.40 ± 0.10 ^b	3.91 ± 0.01 ^c	4.84 ± 0.06 ^a
Yield in pulp (%)	45.90 ± 0.54 ^a	44.30 ± 0.35 ^c	31.95 ± 0.97 ^d	45.26 ± 0.60 ^b
Color <i>L</i> *	42.63 ± 0.86 ^a	40.27 ± 0.71 ^b	40.95 ± 0.67 ^{ab}	39.63 ± 0.28 ^b
Color <i>a</i> *	7.10 ± 0.55 ^a	4.82 ± 0.07 ^b	8.15 ± 0.81 ^a	7.23 ± 0.65 ^a
Color <i>b</i> *	32.56 ± 1.37 ^a	28.23 ± 1.36 ^b	32.29 ± 1.09 ^a	29.22 ± 0.60 ^b
Moisture	85.10 ± 1.21 ^b	87.19 ± 0.10 ^a	83.74 ± 0.34 ^c	86.28 ± 0.47 ^{ab}
Total fat	1.70 ± 0.05 ^c	7.14 ± 0.28 ^a	5.28 ± 0.22 ^b	6.55 ± 0.24 ^a
Ash	4.52 ± 0.18 ^b	4.88 ± 0.17 ^a	3.96 ± 0.01 ^c	4.74 ± 0.04 ^{ab}
Proteins	6.44 ± 0.09 ^c	9.66 ± 0.02 ^a	5.88 ± 0.06 ^d	7.87 ± 0.17 ^b
Carbohydrates	86.32 ± 0.00 ^a	81.67 ± 0.00 ^d	85.94 ± 0.00 ^b	83.74 ± 0.00 ^c
Total fiber	2.26 ± 0.10 ^b	2.64 ± 0.06 ^a	1.78 ± 0.01 ^c	2.34 ± 0.09 ^b

^{a-d} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$).

According to USDA (2012) the values of proximate composition in yellow passion fruit pulp (dry matter) were: 84.21 % moisture, 4.24 % proteins, 1.13 % total fat, 91.51 % carbohydrates, 1.26 % total fiber. Other results were found by Rodriguez-Amaya (2012) in yellow passion fruit pulp (dry matter): 85.62 % moisture, 2.71 % proteins, 0.35 % total fat, 94.57 % carbohydrates, 1.39 % total fiber and 2.36 % ash. These results differ from the results of our study because there are many factors that can interfere with the proximate composition, such as climatic conditions, soil and plant cultivar. The samples from this research had higher levels of proteins and fiber than other studies mentioned above.

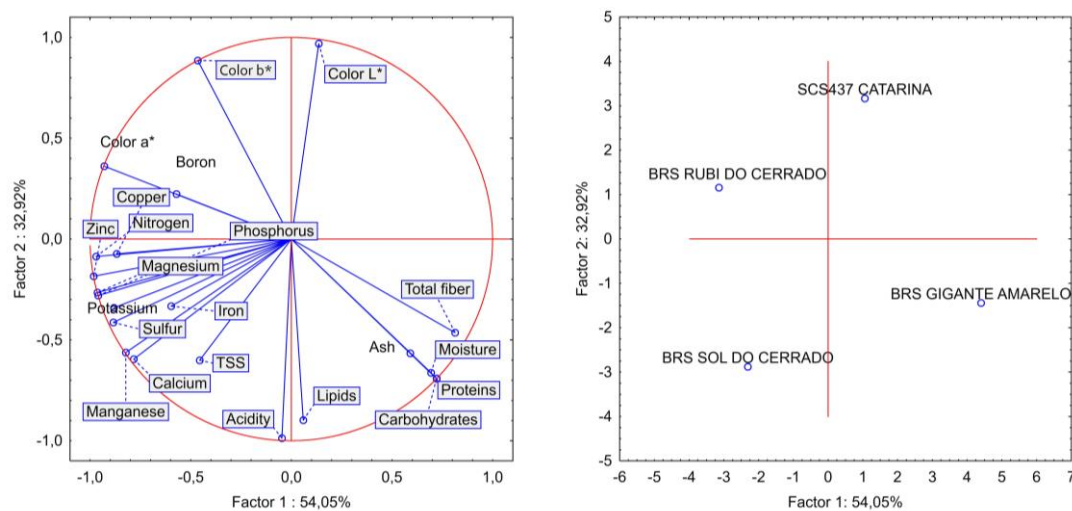


Figure 1. Principal Component Analysis of physicochemical, color parameters, proximate composition and minerals in different cultivars of sour passion fruit pulp.

The minerals tested (Table 2 and Figure 1) in this research were: calcium, phosphorus, magnesium sulfur, potassium, nitrogen, iron, manganese, copper, boron, and zinc. In general, the BRS Sol do Cerrado cultivar exhibit higher concentrations of these minerals, except for copper, that the BRS Rubi do Cerrado cultivar was the one that showed higher levels. The major minerals found in all cultivars studied were similar to the existing literature: phosphorus, potassium, nitrogen and magnesium.

Table 2. Minerals in different cultivars of passion fruit pulp (mg/100g dry weight) with mean and standard deviation.

	SCS437 Catarina	BRS Gigante Amarelo	BRS Rubi do Cerrado	BRS Sol do Cerrado
Calcium	20.00 ± 0.00 ^c	20.00 ± 0.00 ^c	25.00 ± 1.41 ^b	30.00 ± 0.00 ^a
Phosphorus	155.00 ± 7.07 ^b	135.00 ± 7.07 ^b	217.50 ± 3.54 ^a	225.50 ± 9.19 ^a
Magnesium	90.00 ± 0.00 ^b	80.00 ± 0.00 ^b	116.00 ± 5.66 ^a	120.00 ± 0.00 ^a
Sulfur	70.00 ± 0.00 ^b	72.00 ± 4.24 ^b	92.50 ± 4.95 ^a	92.45 ± 4.88 ^a
Potassium	1720.00 ± 42.43 ^{bc}	1550.00 ± 70.71 ^c	1875.00 ± 106.07 ^{ab}	2085.00 ±

49.50^a

Nitrogen	900.00 ± 42.43 ^{ab}	815.00 ± 7.07 ^b	960.00 ± 28.28 ^a	985.00 ± 7.07 ^a
Iron	1.00 ± 0.00 ^b	0.2 ± 0.00 ^d	0.65 ± 0.04 ^c	1.95 ± 0.07 ^a
Manganese	0.50 ± 0.00 ^b	0.51 ± 0.04 ^b	0.66 ± 0.04 ^a	0.76 ± 0.04 ^a
Copper	0.65 ± 0.01 ^c	0.65 ± 0.01 ^c	0.86 ± 0.03 ^a	0.76 ± 0.01 ^b
Boron	0.50 ± 0.00 ^a	0.30 ± 0.00 ^c	0.40 ± 0.00 ^b	0.50 ± 0.00 ^a
Zinc	1.90 ± 0.00 ^b	1.35 ± 0.07 ^c	3.00 ± 0.14 ^a	3.05 ± 0.14 ^a

^{a-d} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$).

Similar studies have been discussed with the content of minerals in the yellow passion fruit pulp (*Passiflora edulis* Sims f. *Flavicarpa Degener*) by Pacheco et al. (2017) that found 0.30 mg of zinc in 100 g of yellow passion fruit pulp, 0.37 mg iron, 0.10 mg copper, 114.13 mg nitrogen, 32.25 mg phosphorus, 337.84 mg potassium, 2.53 mg calcium, 6.34 mg magnesium and 11.89 mg sulfur; and by Rodriguez-Amaya (2012) that found 27.82 mg of calcium/100 g yellow passion fruit pulp (dry matter), 1.67 mg iron, 118 mg magnesium, 90 mg phosphorus, 1933 mg potassium 0.34 mg zinc and 0.37 mg copper. All these values low when compared to our results.

By analyzing the components total phenolic content, flavonoids, carotenoids, and antioxidant capacity the variance of the data was accounted for the significant contributions of 59.93 % for the first principal component and 22.17 % for the second principal components (Figure 2).

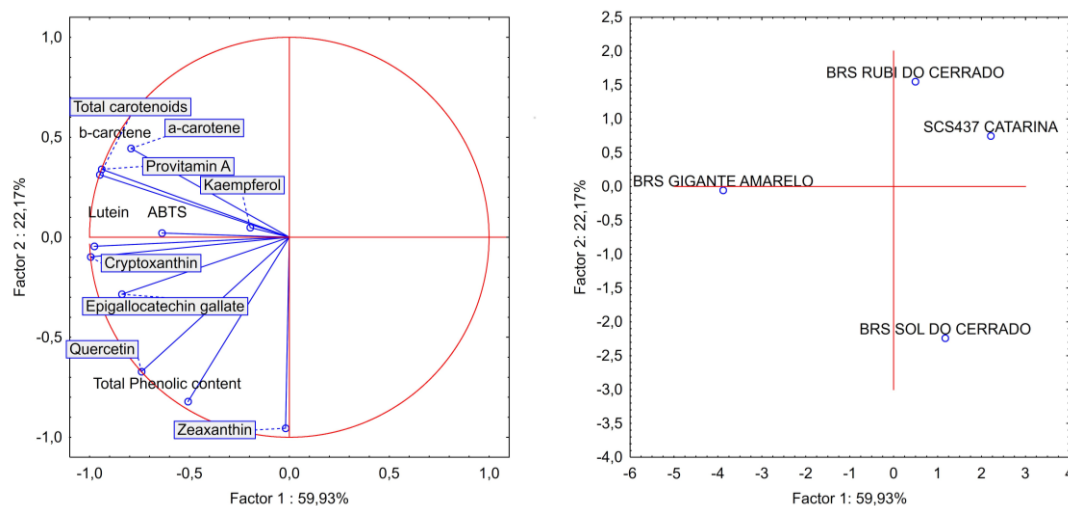


Figure 2. Principal Component Analysis of bioactive compounds (total phenolic content, flavonoids, carotenoids and antioxidant capacity) in different cultivars of sour passion fruit pulp.

The ‘BRS Gigante Amarelo’ and ‘BRS Sol do Cerrado’ showed higher levels of total phenolic content; and the ‘BRS Gigante Amarelo’ presented higher levels of quercetin and epigallocatechin gallate, confirming that these flavonoids make part of the total phenol content. However, the ‘BRS Rubi do Cerrado’ showed higher concentrations of kaempferol. Phenolic compounds are natural antioxidants in fruits and vegetables that eliminate free radicals in the body, avoiding damage to cells and DNA, and, therefore, prevent the onset of cancer. The foods considered functional by the presence of quercetin have anti-inflammatory and antihistamine action that help protect against heart disease and relieve some allergic symptoms and it can reduce intracellular lipid accumulation (Snyder et al., 2016). Studies with some fruit revealed large amounts of quercetin as Liaudanskas et al. (2014) that evaluated the content of quercetin in apples (*Malus domestica* Borkh.) and found 583.7 and mg/100 g; Celli et al. (2011) evaluated the red and purple cultivars of cherry (*Eugenia uniflora* L.) and detected 116 and 375 mg of quercetin/100 g. These studies indicated above showed lower levels of quercetin compared to the cultivars of passion fruit tested.

Some researchers had already analyzed the pulp of different species of yellow passion fruit, as Janzanti et al. (2012) (*P. edulis* Sims f. *flavicarpa* Deg.); Silva et al. (2013) (*Passiflora edulis* Sims) and Zeraik & Yariwake (2010) (*Passiflora edulis* fruit pulp) and all found lower values of phenolic compound quantified by gallic acid equivalent.

In relation to all carotenoids tested (Table 3 and Figure 2) the BRS Gigante Amarelo cultivar showed higher concentrations of lutein, cryptoxanthin, α -carotene, β -carotene, provitamin A and total carotenoids, corresponding to the higher total fat content found in this cultivar. Nevertheless, the BRS Sol do Cerrado cultivar showed higher values of zeaxanthin. For all cultivars tested, the majoritarian carotenoid was the β -carotene, indicating a good source of provitamin A. According to Institute of Medicine (2002), for example, the ingestion of 100 g of sour passion fruit pulp BRS Gigante Amarelo, supplies 73 % of provitamin A for children between 7-12 months; 122 % for children between 1-3 years old; 92 % for children between 4-8 years old; 40 % and 52 % for men and women above 14 years old.

In a research performed by Rodriguez-Amaya (2012) with the content of β -carotene and provitamin A in yellow passion fruit pulp was evaluated, it was found values of 2913 μg of β -carotene in 100 g of dry pulp and 250 μg of provitamin A in 100 g of dry pulp. The USDA analyzed the yellow passion fruit pulp too and discover 1792 μg of β -carotene and 283 μg of provitamin A (dry matter).

Fruits and vegetables are sources of antioxidants that alleviate the damaging effect of oxidative stress. Carotenoids are a group of bioactive compounds responsible for different food colors ranging from yellow to red. They play an important role in preventing human health and also contribute to vitamin A from the diet. There is scientific evidence of these compounds in the prevention of various chronic diseases (Rao and Rao 2007). Lima et al., (2005) detected 4060 μg of β -carotene/100 g in acerola

fruit (*Malpighia emarginata* D.C.), similar content when compared to BRS Gigante Amarelo passion fruit. Already Laura Rodriguez-Urbe et al. (2012) evaluated the *Capsicum annuum* fruit red cultivar (“NuMex Garnet”) and found 3600 µg of β-carotene, 1902 µg of β-cryptoxanthin, 2408 µg of zeaxanthin, 643 µg of violaxanthin, 11625 µg of capsanthin and 11702 µg of unidentified carotenoids, so the content of carotenoids of this research were much higher when compared to passion fruit cultivars.

For ABTS assay (Table 3 and Figure 2), we observed that the sour passion fruit pulps SCS437 Catarina (10.99 µmol/g) and BRS Gigante Amarelo (11.71 µmol/g) showed a higher antioxidant capacity when compared to BRS Rubi do Cerrado (9.83 µmol/g) and BRS Sol do Cerrado (10.13 µmol/g) cultivars. It can also be emphasized that the antioxidant capacity is related to the content of α-carotene, β-carotene, total carotenoids and provitamin A (Fig 2).

Table 3. Total phenolic content, flavonoids (mg/100 g of dry weight), carotenoids profile (µg/100 g of dry weight) and antioxidant capacity (equivalent trolox - µMol/g of dry weight) in different cultivars of passion fruit pulp with mean and standard deviation.

	SCS437 Catarina	BRS Gigante Amarelo	BRS Rubi do Cerrado	BRS Sol do Cerrado
Total phenolic content	1721.74 ± 60.64 ^b	2097.83 ± 35.57 ^a	1532.12 ± 26.03 ^b	2079.00 ± 62.69 ^a
Epigallocatech in gallate	9.68 ± 0.01 ^c	33.72 ± 0.84 ^a	24.39 ± 1.28 ^b	25.47 ± 0.71 ^b
Kaempferol	6.62 ± 0.06 ^d	11.86 ± 0.52 ^c	17.37 ± 0.32 ^a	13.45 ± 0.03 ^b
Quercetin	553.98 ± 1.27 ^c	1201.09 ± 33.53 ^a	583.16 ± 9.84 ^c	1034.33 ± 39.82 ^b
Lutein	83.68 ± 0.56 ^b	107.19 ± 2.73 ^a	84.99 ± 0.64 ^b	85.10 ± 0.39 ^b
Zeaxanthin	65.23 ± 1.04 ^d	77.94 ± 1.16 ^b	69.13 ± 0.04 ^c	103.38 ± 0.35 ^a
Cryptoxanthin	388.24 ± 20.98 ^b	575.17 ± 18.08 ^a	441.99 ± 11.71 ^b	437.85 ± 1.71 ^b
α-carotene	150.41 ± 8.44 ^b	287.00 ± 0.93 ^a	271.66 ± 2.71 ^a	161.27 ± 1.23 ^b

β -carotene	1851.05 \pm 101.02 ^c	4413.91 \pm 98.40 ^a	2729.25 \pm 71.99 ^b	1407.43 \pm 8.23 ^d
Provitamin A	154.25 \pm 8.42 ^c	367.83 \pm 8.20 ^a	227.44 \pm 6.00 ^b	117.29 \pm 0.69 ^d
Total carotenoids	2538.60 \pm 128.84 ^c	5461.22 \pm 77.83 ^a	3597.02 \pm 81.59 ^b	2195.04 \pm 8.75 ^c
Antioxidant capacity	10.99 \pm 0.08 ^a	11.71 \pm 0.17 ^a	9.83 \pm 0.40 ^b	10.83 \pm 0.18 ^b

^{a-d} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$).

In a research performed by Janzanti et al. (2012) they found 112.21 μ mol Trolox in total antioxidant capacity/100 mL for sour passion fruit pulp (*P. edulis* Sims f. *flavicarpa* Deg.); Rotili et al. (2013) observed an antioxidant capacity in yellow passion fruit pulp (*Passiflora edulis* fo. *flavicarpa* Degener) of 62 μ g equivalent Trolox/100 mL extract. Souza et al. (2012) found 10.84 μ mol Trolox equivalent/g (fresh weight) in sweet passion fruit (*Passiflora alata* Dryand), so the last work cited found an antioxidant capacity similar to our research.

Fu et al. (2011) evaluated the antioxidant capacities FRAP (ferric reducing antioxidant power) and TEAC (Trolox equivalent antioxidant capacity) of 62 fruits between them the passion fruit and they concluded that the fruits had very large variation and diverse antioxidant capacities, so guava, pomegranate, sweetsop, plum Chinese wampee and persimmon presented highest antioxidant capacity.

These differences in the content of bioactive compounds and antioxidant capacity can be explained because many factors are involved that range from plant growth on land such as soil, climate, fertilization, type of cultivation and cultivar and also the technique used in the laboratory for the extraction of these compounds (mainly the type of organic solvent).

Conclusions

1. The four cultivars of passion fruit pulps analyzed in this research show that all of these have good quantities of bioactive compounds.
2. The cultivar SCS437 Catarina, showed a higher luminosity (L*), and no differences in color a* and b* was found in cultivars tested, except for the BRS Gigante Amarelo.
3. The 'BRS Gigante Amarelo' presents a higher content of bioactive compounds in relation to other cultivars, as total phenolic content, epigallocatechin gallate, quercetin, all carotenoids tested (except for zeaxanthin), provitamin A.
4. The 'BRS Rubi do Cerrado' exhibit higher concentrations of kaempferol and copper.
5. The 'BRS Sol do Cerrado' showed higher concentrations of all minerals tested (except for copper) in relation to other cultivars and the carotenoid zeaxanthin.
6. Finally, the cultivars SCS437 Catarina and BRS Gigante Amarelo showed a higher antioxidant capacity in relation to other cultivars utilizing the ABTS radical.

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CAPÍTULO 7**ARTIGO 4****Characterization of orange passion fruit peel flour and its use as an ingredient in
bakery products**

Enviado para Publicação na Revista Journal of Culinary Science and Technology

Characterization of orange passion fruit peel flour and its use as an ingredient in bakery products

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Abstract

The cultivation of passion fruit, has presented a great expansion, providing great popularization among the different segments of consumption. This research studied the effect of drying temperature in the content of bioactive compounds such as phenolic compounds, carotenoids, flavonoids and antioxidant capacity of orange passion fruit peel flour; as well as the application of this flour as an ingredient in the formulation of bakery products (bread and cake). In methodology it was evaluated in flours: water holding capacity, oil holding capacity, color parameters, proximate composition, minerals, bioactive compounds and antioxidant capacity; for the formulations of cakes and breads we evaluated the color parameters, proximate composition and sensorial analysis. As noted, the drying at 80 °C retained higher concentrations of phenolic compounds and all carotenoids tested and provitamin A. However regarding quercetin, it was observed that temperature at 60 °C showed higher levels and the drying at 70 °C had a greater effect on the radical sequester ABTS. Independent of temperature employed, the flours evaluated can be used as ingredients for the enrichment of bakery products with constituents as dietary fiber, minerals, and bioactive compounds. In the sensorial analysis, the attributes of appearance, color, flavor, texture, taste and global acceptance showed that the bread and cakes formulations had a minimum acceptable rate of 70 %.

Keywords: carotenoids; minerals; antioxidant capacity; proximate composition; sensorial analysis.

Introduction

The usual intake of fruits, vegetables or even ingredients with a high content of bioactive compounds can reduce the effects caused by oxidative stress, and consequently minimize the risk of various diseases, such as cardiovascular disease and some cancers. This protective effect is attributed to a variety of antioxidant substances such as flavonoids and carotenoids (Gawlik-Dziki, 2012).

Passion fruit is a tropical plant that belongs to the family *Passifloraceae* and Brazil are the largest producers of yellow passion fruit in the world. The pulp and peel are rich in bioactive compounds as carotenoids, flavonoids, phenolic compounds, minerals; the peel, normally not used by consumers is an important source of fibers soluble and insoluble. Health benefits of the peel, pulp, and seeds of passion fruit have been recognized by researchers and recommended for inclusion in the human diet (Wijeratnam, 2016). Passion fruit is most popularly consumed as a drink/juice, however after processed in the form of pulp its used to obtain jellies, mousses, and other products. Unfortunately, during processing of the juice by the industry, only 30 % of all fruit weight is harnessed, and the by-products as the peels that represent 50 % of the total weight are discarded (Nascimento, Ascheri, Carvalho & Galdeano, 2013). The production of large volumes of residues is a problem caused by the fruit and vegetable processing industry because generates economic and environmental impacts worldwide. These residues are used as fertilizer or for animal feed once has a significant amount of fiber; besides, have bioactive compounds. Thus, the exploitation of these abundant and low-cost renewable resources could contribute to obtained new ingredients to food

industries and provide a lower economic and environmental impact (Marín, Soler-Rivas, Benavente-García, Castillo & Pérez-Alvarez, 2007; Djilas, Čanadanović-Brunet & Cetković, 2009; Silva et al. 2014).

The orange passion fruit peel flour can be used in the enrichment of products such as bread, cakes, biscuits, and cereal bars, improving their nutritional qualities and technology (de Souza, Ferreira & Vieira, 2008).

This research aimed to evaluate the conditions of time and temperature for obtainment flour of orange passion fruit peel and evaluate the content of bioactive compounds (phenolic compounds, carotenoids, and flavonoids) and antioxidant capacity of this new ingredient. Later it was used in the formulation of cakes and bread along with the chia mucilage to increase fiber and decrease the addition of fat, being evaluated the analyzes of proximate composition, texture, color parameters and sensorial analysis.

Materials and methods

Obtention of orange passion fruit peel flour

The orange passion fruit peels were collected in the Criuva District (Caxias do Sul), Rio Grande do Sul, Brazil (latitude 28° 52' 33.65" S, longitude 50° 58' 36.72" W and an average height of 860 m). The fruit was selected and collected when were with orange color in the peel that indicates the fruit is ripe, the size of ≈ 5 cm, the absence of damages and injuries. The fruit was cleaned with running water, cut and the pulp and seeds were removed. Then, the peels (albedo and flavedo) were cut with a knife into pieces of 1 cm and then placed in the oven.

Drying of the samples (5 kg) was performed in an air circulating oven (Nova Ética, 400/3ND, Brazil) at three different temperatures (60 °C, 70 °C and 80 °C) to obtain flour with moisture between 10-15%. A curve of drying (Fig. 1) was constructed

by removal of 5g of each sample at 1-hour intervals, dried at 105 °C (Nova Ética, 400/3ND, Brazil) for 24 hours. After the binomial time/temperature the optimum to achieve the desired moisture (10-15 %), the samples were dried under ideal conditions. The material dried was individually milled in a mill (Arbel model MCF55, Brazil), sieved to obtain flour, using sieves for particle size analysis (Bertel, Brazil) and the particles were less or equal than 125 µm (115 mesh). Afterward, the flour was packed in plastic pots, closed and stored in the dark at room temperature (≈ 25 °C).

The characterization of the peels were previally studied and the results are shown in a paper (dos Reis et al. 2018).

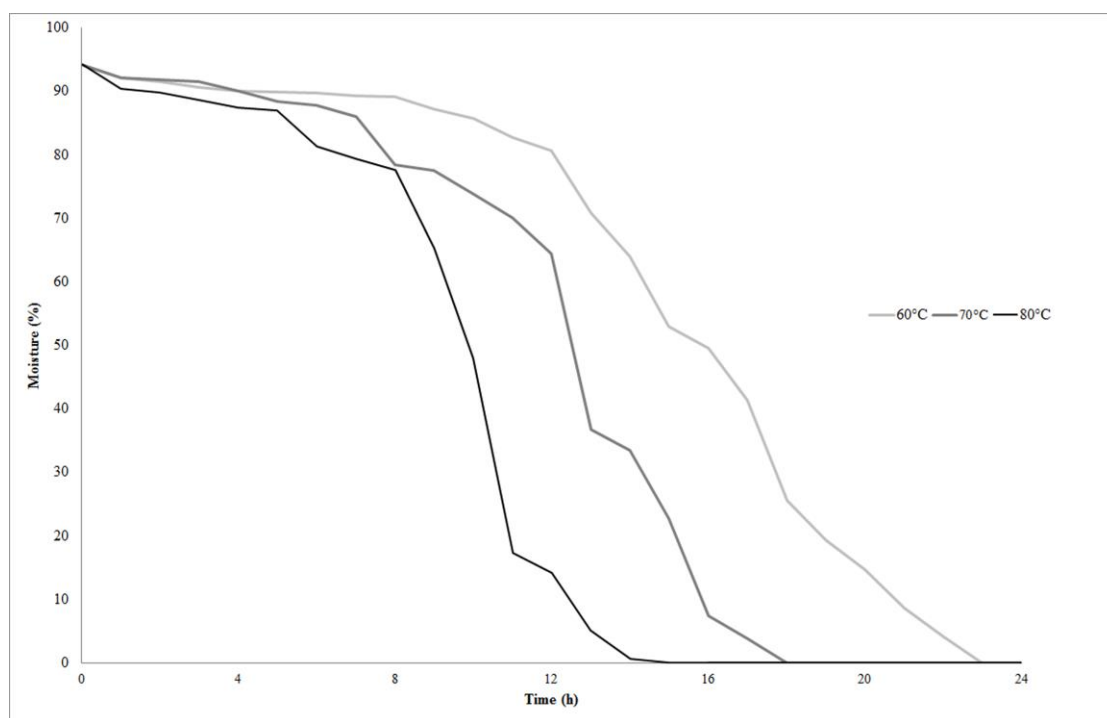


Fig. 1. Moisture curve of orange passion fruit flour in different temperatures.

Water holding capacity

The water holding capacity (WHC) analysis of the flours was performed according to Fernández-López et al. (2009) with minor modifications. A portion of distilled water (30 mL) was added to 1 g of the flours. The suspension was

homogenized in a vortex (Quimis, Model Q920-A2, Brazil) for 1 min and left at room temperature for 24 h. After centrifugation (3000 x g for 20 min, Cientec, CTR – 5000R, Brazil) the supernatant was removed, and the residue weighed. The water retention capacity was expressed in grams of water per gram of dry sample.

Oil holding capacity of flour

The oil holding capacity (WHC) analysis of the flours was performed according to Fernández-López et al. (2009) with minor modifications. A portion of oil (30 mL) was added to 1 g of the flours. The suspension was homogenized in a vortex (Quimis, Model Q920-A2, Brazil) for 1 min and left at room temperature for 24 h. After centrifugation (3000 x g for 20 min, Cientec, CTR – 5000R, Brazil) the supernatant was removed, and the residue weighed. The oil retention capacity was expressed in grams of oil per gram of dry sample.

Color parameters of flour

The color was analyzed using a portable colorimeter (Konica Minolta Model CR 400, Singapore) by the Commission Internationale de l'Eclairage (CIELAB system) by determining the values of L^* (lightness), a^* (component red-green) and b^* (yellow-blue component).

Minerals content in flour

The macro and micronutrients analysis of the flours were performed in Plant Soil Laboratory Faculty of Agronomy, The Federal University of Rio Grande do Sul (UFRGS), according to the methodology of atomic emission spectrometry with Inductively Coupled Plasma Source (ICP-OES) described by Tedesco and Gianello (2004). The phosphorus, potassium, calcium, magnesium, sulfur, copper, zinc, iron,

manganese, and sodium we used with wet digestion nitric-perchloric and boron was applied with dry digestion.

Carotenoid profile and provitamin A content in flour

The carotenoid content was determined according to Mercadante and Rodriguez-Amaya (1998) by extracting pigments with acetone and saponifying in a KOH solution (2 mol/L of KOH in distilled water) overnight at room temperature. The extract was concentrated by rotary evaporation (Fisatom, Model 801) ($T < 25\text{ }^{\circ}\text{C}$) and stored at $-18\text{ }^{\circ}\text{C}$ for quantification by high-performance liquid chromatography.

For HPLC (high performance liquid chromatography) analysis, the extract was diluted with methyl *tert*-butyl ether (MTBE-JT Baker, CAS. Number 1634-04-4, purity 99.96%), sonicated (Unique, Model USC 1400) for 30 seconds and filtered (Millex LCR 0.45 μm , 13 mm) for injection into an Agilent 1100 Series HPLC (Santa Clara, CA, USA) equipped with a quaternary system and a UV-visible detector.

The column used was a $250 \times 4.6\text{ mm ID}$, 3 μm , C30 polymeric reverse phase column (YMC, model CT99SO3-2546WT). The mobile phase gradient (water:methanol:MTBE) (JT Baker, CAS Number 04.04.1634, 99.96% purity) commenced at 5:90:5, reaching 0:95:5 at 12 minutes, 0:89:11 at 25 minutes, 0:75:25 at 40 minutes, and finally 00:50:50 at 60 minutes. The flow rate was 1 mL/min at $33\text{ }^{\circ}\text{C}$. Spectra were obtained between 250 and 600 nm (or at a fixed wavelength of 450 nm for carotenoids).

Compounds were identified by comparing the sample retention times with the retention times obtained for controls. For quantification, a standard curve was constructed for carotenoids over the following ranges: lutein 1 to 65 $\mu\text{g/mL}$ ($\geq 95\%$, Sigma-Aldrich), zeaxanthin 1 to 40 $\mu\text{g/mL}$ ($\geq 95\%$, Sigma-Aldrich), cryptoxanthin 4 to 100 $\mu\text{g/mL}$ ($\geq 97\%$, Sigma-Aldrich), α -carotene 2 to 25 $\mu\text{g/mL}$ ($\geq 95\%$, Sigma-

Aldrich), β -carotene 5 to 50 $\mu\text{g/mL}$ ($\geq 97\%$, Sigma-Aldrich) and lycopene ($\geq 85\%$ Sigma-Aldrich).

The limits of detection (LOD) and quantification (LOQ) were as follows. Lutein: 6.9×10^{-3} and 1.15×10^{-2} $\mu\text{g/g}$, zeaxanthin: 9.56×10^{-2} and 1.59×10^{-2} $\mu\text{g/g}$, cryptoxanthin: 2.11×10^{-2} and 3.51×10^{-2} $\mu\text{g/g}$, α -carotene: 1.97×10^{-2} and 3.28×10^{-2} $\mu\text{g/g}$, β -carotene: 6.53×10^{-2} and 10.89×10^{-2} $\mu\text{g/g}$ and lycopene were 7×10^{-3} and 33×10^{-3} $\mu\text{g/g}$.

Provitamin A activity was calculated by the bioconversion factor following Institute of Medicine (2001), yielding a value of 12 mg of β -carotene with 1 mg of retinol.

Phenolic compounds in flour

Samples (1 g) were homogenized by exhaustive extraction with 20 mL of ethanol and centrifuged at 15 °C (Cientec, CTR – 5000R, Brazil) at 6000 rpm for 20 minutes. Then, 20 μL of supernatant was added to 1.58 mL of water and 100 μL of Folin-Ciocalteu (0.4 mol/L). After the reaction (3 minutes), 300 μL of Na_2CO_3 was added, and the mixture kept at room temperature for 2 hours. The reading absorbance was made at 765 nm on a UV-visible spectrophotometer (Shimadzu, UV-1700 PharmaSpec, Japan). A standard curve was constructed to quantify phenolic compounds, using gallic acid at concentrations of 0 to 0.50 mg/mL. The results are presented in mg gallic acid/100 g of dry sample (Swain & Hillis, 1959).

Quercetin and Kaempferol content in flour

Quercetin and kaempferol contents were analyzed according to Zeraik and Yariwake (2010) with modifications. The sample extracts (10 g) were homogenized in an Ultra-Turrax (T25, IKA, China) with and 30 mL of methanol at room temperature. The extracts were centrifuged at 15000 x g, 4 °C for 20 min, after which the supernatant

was evaporated to 2.0 mL in a rotary evaporator. The solution was filtered with a 0.45 μm Millex-HV PVDF membrane (Millipore, New Bedford, MA, USA) before HPLC analysis. The samples were analyzed and prepared in triplicate.

The HPLC-UV/DAD analyses were carried out on a Waters Alliance 2695 (Milford, MA, USA) liquid chromatograph connected to a model 2996 (DAD) diode array detector and controlled by Waters Empower software. The separation was performed using a C18 polymer column (250 mm \times 4.6 mm id, 5 μm Vydac, 218TP). The samples were injected automatically (10.0 μL). The column (controlled at 35 $^{\circ}\text{C}$), and a 0.8 mL min^{-1} flow rate was applied, using a linear gradient of 0.2% formic acid in water (solvent A) and 0.2% formic acid (CAS Number: 64-18-6) in acetonitrile (CAS Number 75-05-8) (solvent B). The optimized gradients employed in passion fruit extracts were: 0–10 min, 15 % B in 85 % A and 10–30 min, 20 % B in 80 % A. The chromatogram was monitored at 330 nm, and UV spectra of individual peaks were recorded in the range of 200–400 nm.

The contents of quercetin (CAS Number 117-39-5) and kaempferol (CAS Number 520-18-3) were determined by comparison with an external standard, injecting a new standard daily at 5 mg/mL for Quercetin (\geq 98%, Sigma-Aldrich) and 1 mg/mL for kaempferol (\geq 99%, Sigma-Aldrich).

Antioxidant capacity of flour

The method used to determine antioxidant capacity was based on the sequestration of ABTS (2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) radicals according to Brand-Williams, Cuvelier & Berset (1995), as adapted by Embrapa Agroindústria Tropical. Samples (1 g) were placed in 20 mL of ethanol and then centrifuged at 15 $^{\circ}\text{C}$ at 3000 x g (Cientec, CTR – 5000R, Brazil) for 20 minutes. The

supernatant was diluted to three concentrations. For the ABTS assay, aliquots of each concentration were treated with 2 mL of ABTS (7 mM). The absorbance was read at 734 nm (Shimadzu, UV-1700 PharmaSpec, Japan). The results were presented as IC 50 (g/100 mL) (Effective Concentration of 50 % radical inhibition).

Obtention of chia mucilage

The chia seeds (acquired in the local market) were suspended in distilled water at the ratio of 1:30 (seed: water). This mixture was stirred for 4 hours at the temperature of 80 °C on a shaker. After this step, the liquid was separated from the chia seed with the aid of a cheese desorption cloth. The liquid was then placed in a glass dish and taken to an oven at 55 °C, where it remained for 48 hours until complete drying to obtain dry and dehydrated mucilage (Campos, Ruivo, Madrona & Bergamasco, 2015). The chia mucilage showed a yield of 5.52 % (± 0.04).

Bread preparation

The bread formulations are shown in Table 2. Butter (in the BF formulation) was replaced by chia mucilage at 50 % of substitution. Before bread elaboration, chia mucilage was prepared by hydrating it with distilled water in a concentration of 1.5 g of chia mucilage/ 100 g water according to preliminary tests, followed by mixing and resting for 30 min. The orange passion fruit peel flour was mixed with wheat flour at a proportion of 85:15 (wheat flour:orange passion fruit peel flour in the breads). The ingredients were initially mixed using a planetary mixer (Arno, model BPA, Brazil). The dry ingredients were mixed (wheat flour, orange passion fruit peel flour (in the BF, sugar, sodium chloride, dry yeast, ascorbic acid) and after it was added the fat (butter) and suspension of chia mucilage. Lastly, the mixture was passed on the bread-making machine (Arke, Brazil) and put for 10 minutes until the gluten network had completely

developed and placed in a rectangular shape 15cm x 10cm in an electric oven preheated at 200 °C for 30 minutes.

Cake preparation

The cakes formulations are shown in Table 1. Butter (in the CF formulation) was replaced by chia mucilage at 50 % of substitution. Before cake elaboration, chia mucilage was prepared by hydrating it with milk in a concentration of 5 g of chia mucilage/50 g milk according to preliminary tests, followed by mixing and resting for 30 min. The orange passion fruit peel flour was mixed with wheat flour at a proportion of 50:50 (wheat flour:orange passion fruit peel flour in the cakes). The ingredients were initially mixed using a planetary shaper (Arno, model BPA, Brazil). The ingredients were mixed in this sequence: dehydrated whole egg, sugar, butter, suspension of chia mucilage, chocolate powder, wheat flour, orange passion fruit peel flour, and baking powder. Lastly, the mixture was placed in a rectangular shape 30cm x 15cm in an electric oven preheated at 200 °C for 45 minutes.

Table 1 Formulation (g) of controls cake and bread and with the addition of orange passion fruit peel flour and chia seed mucilage.

	CC	CF	BC	BF
Wheat flour	50	25	100	85
Orange passion fruit peel flour	-	25	-	15
Fat (butter)	10	5	3	1.5
Milk	50	50	-	-

Water	-	-	60	60
Sugar	38.4	38.4	5	5
Dehydrated whole egg	20	20	-	-
Chocolate powder	10	10	-	-
Sodium chloride	-	-	2	2
Dry yeast	-	-	2	3.5
Baking powder	1.7	1.7	-	-
Ascorbic acid	-	-	0.009	0.009
Suspension of chia mucilage	-	5	0.00	1.5

CC: cake control; CF: cake added of orange passion fruit peel flour; BC: bread control; BF: bread added of orange passion fruit peel flour.

CC and CF: formulations with 0 and 50 % substitution of wheat by orange passion fruit peel flour and vegetable fat by chia mucilage gel.

BC and BF: formulations with 0 and 15 % substitution of wheat by orange passion fruit peel flour and vegetable fat by chia mucilage gel.

Proximate Composition and caloric value

The proximate composition was used to characterize both the flour and the baked products developed. All analyses were performed according to AOAC (2000). Proteins were analysed by the Kjeldahl method with a conversion factor of 6.25. Lipids were obtained by cold extraction and ash was determined in a muffle furnace at 550 °C. The total dietary fiber was determined by the enzymatic-gravimetric method, moisture by gravimetry. Carbohydrates were estimated by the difference of proteins, lipids, ash, and moisture.

The caloric value was calculated only for bread (BC – bread control and BF – bread added with orange passion fruit peel flour and chia mucilage) and cakes (CC – cake control and CF - added with orange passion fruit peel flour and chia mucilage)

using the coefficients of Atwater (Wart & Merrill, 1963), corresponding to the carbohydrate, protein and lipid contents:

Caloric value (kcal/100 g) = (g of carbohydrates *4) + (g of protein *4) + (g of lipids *9).

Texture

The measurement of firmness was performed with a digital penetrometer, PCE model FM 200 (PCE Instruments, Albacete, España), and the results are expressed in Newton (N).

Sensorial analysis

A multiple-comparison test was used to assess the following sensory attributes: appearance, color, flavor, texture, taste and global acceptance. The untrained judges received a card with a scale of nine points since nine (9): I liked it very much and one (1): I even disliked very much, and a note should be assigned for each attribute. The index of acceptability (IA) was calculated using the Equation (Fernandes and Salas-Melado 2017):

$$IA (\%) = \frac{\text{Score} * 100}{9}$$

9

In the sensory evaluation of the cakes and bread, most of the panelists were female (80.77 %) and aged between 18 and 35 years (69.23 %), and 19.23 % were male and aged between 18 and 35 years (17.30 %) from a total of 52 panelists.

Statistical Analysis

Data were analyzed by ANOVA and Tukey's mean comparison test at a significance level of 5% using Statistica 12.0 (StatSoft, São Paulo, Brazil).

Results and discussion

The orange passion fruit peel flour was produced from the drying of samples until to obtain a moisture content of 14% since for commercialization of this product the moisture should be up to 15% (ANVISA, 1996).

The time required for the drying process at temperatures of 60 °C, 70 °C and 80 °C were 20 hours, 15.5 hours and 12 hours, respectively. The time of samples drying up to 14 % moisture can be explained by the high pectin content present in the peels, which probably difficult to lose the water; mainly during the first 6 hours of drying, during which time the samples lost only 20 % of the initial moisture content independent of the temperature used (data not shown). After 6 hours of drying the rate of water loss increased, and at 80 °C was necessary 6 hours more drying to reduce moisture from 80 % to 14 %. At the lower temperature used (60 °C), it took another 14 hours to complete the process and obtain the final product with the moisture suitable for commercialization and use in food formulations.

Table 2 shows the proximate composition, water and oil-holding capacity and color parameters of flours in different drying temperatures. The results showed that no significative differences were shown in moisture, proteins, ash and total fibers of flours at 60, 70 and 80 °C. However, for lipids, the temperatures at 70 °C and 80 °C showed smaller values, due to the oxidation of lipids at higher temperatures employed. The content of carbohydrates at 60 °C was lower than 70 and 80 °C because carbohydrates were estimated by the difference of proteins, moisture, lipids, and ash, and the lipids content was higher at this temperature.

Table 2 Proximate composition, Water Holding Capacity (WHC), Oil Holding Capacity (OHC) g/100 g; color parameters; mineral composition (mg/100 g); carotenoid profile ($\mu\text{g}/100\text{ g}$); phenolic compounds (mg/100 g); quercetin (mg/100 g) and antioxidant capacity ABTS of orange passion fruit peel flour (mean and standard deviation).

	60 °C	70 °C	80 °C
Moisture	14.56 \pm 0.27 ^a	14.20 \pm 0.10 ^a	14.08 \pm 0.18 ^a
Proteins	9.25 \pm 0.38 ^a	9.31 \pm 0.03 ^a	9.39 \pm 0.20 ^a
Lipids	5.26 \pm 0.22 ^a	4.20 \pm 0.11 ^b	4.39 \pm 0.04 ^b
Ash	9.31 \pm 0.04 ^a	9.25 \pm 0.52 ^a	9.11 \pm 0.05 ^a
Carbohydrates	61.62 \pm 0.07 ^b	62.95 \pm 0.50 ^a	63.04 \pm 0.02 ^a
Total fiber	45.19 \pm 0.43 ^a	45.01 \pm 1.86 ^a	45.34 \pm 0.34 ^a
WHC	6.87 \pm 0.07 ^b	7.71 \pm 0.04 ^a	7.89 \pm 0.14 ^a
OHC	2.86 \pm 0.02 ^a	2.98 \pm 0.06 ^a	2.96 \pm 0.04 ^a
Color <i>L</i> *	41.22 \pm 1.31 ^a	41.58 \pm 0.62 ^a	40.88 \pm 0.86 ^a
Color <i>a</i> *	5.92 \pm 0.60 ^a	6.03 \pm 0.15 ^a	5.69 \pm 0.48 ^a
Color <i>b</i> *	9.91 \pm 1.03 ^a	10.46 \pm 0.49 ^a	9.98 \pm 1.06 ^a
Calcium	46.00 \pm 0.07 ^a	45.93 \pm 0.01 ^a	45.96 \pm 0.07 ^a
Phosphorus	280 \pm 3.50 ^a	273 \pm 3.54 ^a	276 \pm 7.00 ^a
Magnesium	190 \pm 2.83 ^a	191 \pm 2.80 ^a	192 \pm 3.54 ^a
Sulfur	300 \pm 1.41 ^a	303 \pm 4.24 ^a	309 \pm 2.12 ^a
Sodium	11.50 \pm 0.04 ^a	11.80 \pm 0.02 ^a	11.50 \pm 0.01 ^a
Potassium	4003 \pm 66.00 ^a	4005 \pm 70.00 ^a	4000 \pm 68.00 ^a
Iron	6.90 \pm 0.07 ^a	7.00 \pm 0.01 ^a	6.98 \pm 0.05 ^a

Manganese	8.10 ± 0.04 ^a	8.06 ± 0.06 ^a	8.09 ± 0.02 ^a
Copper	0.50 ± 0.01 ^a	0.50 ± 0.00 ^a	0.50 ± 0.01 ^a
Boron	2.00 ± 0.01 ^a	2.00 ± 0.00 ^a	2.00 ± 0.01 ^a
Zinc	4.00 ± 0.08 ^a	4.03 ± 0.03 ^a	4.10 ± 0.02 ^a
Lutein	1697.05 ± 99.86 ^b	2402.81 ± 86.27 ^a	2797.42 ± 125.82 ^a
Zeaxanthin	532.96 ± 12.03 ^b	839.07 ± 9.70 ^a	827.47 ± 14.16 ^a
Cryptoxanthin	1147.44 ± 58.79 ^c	1650.20 ± 47.96 ^b	2845.89 ± 35.81 ^a
α-carotene	544.02 ± 60.46 ^b	698.99 ± 4.32 ^b	1304.53 ± 32.11 ^a
β-carotene	12692.94 ± 204.68 ^c	21442.71 ± 1132.30 ^b	31073.25 ± 1036.08 ^a
Provitamin A*	1057.74 ± 17.06 ^c	1786.89 ± 94.61 ^b	2589.44 ± 86.34 ^a
Total carotenoids	16614.40 ± 411.77 ^c	27202.10 ± 1156.15 ^b	39890.33 ± 2329.78 ^a
Phenolic compounds	673.41 ± 6.79 ^b	700.01 ± 9.79 ^b	758.09 ± 7.03 ^a
Quercetin	261.76 ± 3.55 ^a	114.02 ± 0.10 ^c	162.83 ± 4.63 ^b
Antioxidant capacity**	2.11 ± 0.00 ^b	1.99 ± 0.00 ^c	2.64 ± 0.01 ^a

^{a,b,c} Different superscript letters in the same row indicate statistically significant difference ($p < 0.05$).

*Expressed as µg RAE (Retinol Activity Equivalent).

**IC50 Effective Concentration of 50 % radical inhibition.

As it can be observed, the produced flour of orange passion fruit peel is nutritionally better, because it showed the higher content of total dietary fiber (between 45 %), positive result compared to the traditional wheat flour that has around 2.70 % of total dietary fiber (USDA, 2002). This can be explained by the fact that the peels of fruit feature higher fiber content when compared to its pulps and even by grains.

The drying at 60 °C presents a lower water holding capacity (WHC) about temperatures at 70 and 80 °C, probably because this has a higher content of lipids.

Regarding oil holding capacity (OHC), no significative differences were found, by the fact that flours have the same moisture content. Martínez et al. (2012) found in passion fruit (*Passiflora edulis* L., cv. Flavicarpa) dietary fibers concentrate co-products (peel, pulp, and seed), 13.5 g/g for WHC and 0.9 for OHC, different values when compared to our study. According to Chau et al. (2004), high values of WHC and OHC suggest that the flour has potential application as a functional ingredient in a reduction of calories in formulations new.

Coelho et al. (2017) evaluated the color parameters in passion fruit peel flour and found 36.71 for luminosity (L*), 7.53 for red-green component (a*) and 24.57 for yellow-blue component (b*), different values to those found in these research.

In general, minerals content (Table 2), did not have a significative difference in various temperatures tested (60, 70 and 80 °C). It was expected that mineral content of the flours do not vary on the drying conditions since minerals are heat stable compounds.

Therefore, this research showed a significant amount of minerals in flour of orange passion fruit peel that may be an ingredient in different formulations, making the food ready for consumption is enriched with these micronutrients. The minerals in greatest abundance found in passion fruit peel flour were potassium, iron, zinc, phosphorus, and magnesium is vital for the proper functioning of the human body.

The drying at 80 °C presented higher values of lutein, zeaxanthin, cryptoxanthin, α -carotene, β -carotene, provitamin A and total carotenoids (Table 2) when compared to drying at 60 and 70 °C. The temperature at 60 °C showed higher losses of these bioactive compounds, due to the long exposure time in the oven. Then, a higher temperature by leaving for a shorter time (12 hours) leads to less loss of carotenoids and

too can be contributed in some way to an increased content of these compounds, as reported by Reis et al. (2015) where they found that processing can become the compounds more available due to the disruption of plant tissue of carotenoids.

The content of provitamin A of orange passion fruit flour (100 g) presents 117 % and 151 % of recommended daily intake of vitamin A in men and women (> 14 years old), respectively (DRI, 2002).

In the content of phenolic compounds, drying at 80 °C showed higher values about other temperatures applied (60 and 70 °C) because heating helps break the membrane plant, leaving an increase of phenolic compounds bioaccessibility. But for quercetin, it was observed that temperature at 60 °C showed higher contents and 70 °C and 80 °C the lower concentrations because quercetin is unstable at high temperatures, which may decrease its level.

Already Martínez et al. (2012), found 150 mg gallic acid/100 g of passion fruit (*Passiflora edulis* L., cv. Flavicarpa) dietary fiber concentrate co-products (peel, pulp, and seed). The gallic acid can capture the total phenolic compounds present in the extract of the fruit, being that a part of the value obtained can be related to quercetin, because this compound is inside of the group of phenolic compounds. During the food processing and storage, many elements such as pH, heat, metal ions, could affect the chemical stability (including degradation and oxidation) of quercetin (Wang et al. 2016). Flour that contains quercetin is an alternative source for the preparation of food with antioxidant properties, such as, bread, pasta, biscuits, among others.

Morais et al. (2015) found low levels of phenolics (86.74 mg gallic acid equivalent/100 g) and flavonoids (35.13 mg quercetin equivalent/100 g) of oven dried peels of passion fruit (*Passiflora edulis*). Lima et al. (2016) found 37 and 308 mg/100 g of *Passiflora edulis* peel flour for phytic acid and tannins, respectively.

The quercetin was analyzed in this study because the passion fruit contains high values of this compound. The intake of quercetin in fruit extracts has been shown to reduce diabetes, inflammation, hepatic complications and obesity because of the high-fat consumption; it is associated to antioxidant, anticancer and antiviral activities (Snyder et al. 2016).

In the antioxidant capacity (Table 1), the drying at 70 °C had the greatest effect on the radical sequester ABTS and the drying at 80 °C that showed the less capacity in radical inhibition.

Martínez et al. (2012) evaluated the *in vitro* antioxidant properties in passion fruit (*Passiflora edulis* L., cv. Flovicarpa) dietary fiber concentrate co-products (peel, pulp, and seed). The results were: 5.5 and 5.1 µM TE (Trolox equivalents)/g for ABTS and DPPH, respectively. They also revealed that phenolic compounds are the primary phytochemicals responsible for the antioxidant activity of vegetables and fruit. The fiber concentrates obtained from fruit were shown to be rich in phenolic compounds containing a variety of phenolic hydroxyl groups that may be responsible for the antioxidant capacity and free radical scavenging activity.

Bread and cakes elaboration

For the bread and cakes formulations, the drying method at 80 °C of orange passion fruit peel was used, since it presented lower losses of bioactive compounds.

The proximate composition and caloric value of bread and cakes can be seen in Table 3. It was observed that the cake added with orange passion fruit peel flour and chia mucilage (CF) and the bread control (BC) showed higher moisture; for proteins the CC (cake control) and the BF (bread added with orange passion fruit peel flour and chia mucilage) showed more values. The bread and cakes control showed higher contents of lipids and carbohydrates; the BF and CF showed more values of ash, and total dietary

fiber and the texture was higher in these formulations too (due to the high fiber content and more consistent batters).

Table 3 Proximate composition (g/100 g) and Caloric value (Kcal/100 g) of cakes and bread control and with the addition of orange passion fruit peel flour and chia seed mucilage.

	CC	CF	BC	BF
Moisture	26.45 ± 0.22 ^b	41.31 ± 0.13 ^a	30.06 ± 0.09 ^a	28.41 ± 0.42 ^b
Proteins	10.92 ± 0.27 ^a	6.56 ± 0.13 ^b	6.65 ± 0.10 ^b	7.65 ± 0.34 ^a
Lipids	11.76 ± 0.31 ^a	7.46 ± 0.06 ^b	3.08 ± 0.11 ^a	2.05 ± 0.03 ^b
Ash	1.72 ± 0.04 ^b	2.47 ± 0.08 ^a	1.57 ± 0.03 ^b	3.54 ± 0.04 ^a
Carbohydrates	49.15 ± 0.10 ^a	42.20 ± 0.09 ^b	58.64 ± 0.12 ^a	58.35 ± 0.12 ^b
Total fiber	2.37 ± 0.12 ^b	9.12 0.05 ^a	3.43 0.05 ^b	8.81 0.03 ^a
Caloric value	347.50	262.10	288.80	282.45

^{a-b} Different superscript letters in the same row indicate the statistically significant difference ($p < 0.05$). CC: cake control; CF: cake added of orange passion fruit peel flour; BC: bread control; BF: bread added of orange passion fruit peel flour.

Miranda, Caixeta, Flávio & Pinho (2013) developed and tested the acceptance of cakes enriched with yellow passion fruit peel (*Passiflora edulis F. Flavicarpa deg*). They were four formulations: control (without yellow passion fruit peel), with the addition of 6.66 %, 8.88 %, and 12.22 %. While the control cake had a soft texture, the cakes added with yellow passion fruit peel flour had firmer texture; with increasing proportions of yellow passion fruit peel flour in relation to wheat flour, there was a reduction of the value of the proportion of carbohydrates, proteins, sodium, and fat in the preparations. In this way, the preparation with the addition of 12.22 % of yellow

passion fruit peel flour, which received the highest concentration of passion fruit flour, presented the lowest levels of carbohydrates, proteins, total fat and sodium and higher fiber contents.

In the color parameters (Table 4), the values of luminosity, color a^* and b^* in the cakes control were higher than the CF both crust and crumb; for bread, in the crust, no differences were found for luminosity, indicating that they were baked equally and standard. The color of the crust in baked products is directly influenced by the caramelization and Maillard Reaction, as major changes gain during cooking. The color a^* and b^* of control was higher than them added with orange passion fruit peel flour and chia mucilage; for a crumb, the luminosity was higher in the bread control, because of the clear coloration of the flour. The color of the crumb is influenced by the wheat flour, the smaller the particle size, the brighter (Popov-Raljic et al. 2009). For color a^* and b^* the values were higher in the BF due to the dark coloration of the flour.

Table 4 Color parameters of crust and crumb and Texture (N) of cakes and bread control and with the addition of orange passion fruit peel flour and chia seed mucilage.

		CC	CF	BC	BF
CRUST	L*	28.47 ± 0.82 ^a	20.25 ± 0.82 ^b	60.96 ± 2.30 ^a	59.75 ± 2.89 ^a
	a*	11.40 ± 0.23 ^a	7.57 ± 0.25 ^b	15.77 ± 0.86 ^a	11.72 ± 0.01 ^b
	b*	8.44 ± 0.06 ^a	4.18 ± 0.19 ^b	30.94 ± 1.02 ^a	28.90 ± 0.40 ^b
CRUMB	L*	28.71 ± 2.38 ^a	25.21 ± 1.18 ^b	75.61 ± 1.49 ^a	47.86 ± 1.62 ^b
	a*	13.34 ± 0.61 ^a	10.31 ± 0.39 ^b	3.60 ± 0.05 ^b	11.98 ± 0.40 ^a
	b*	14.59 ± 0.49 ^a	13.12 ± 0.42 ^b	14.34 ± 0.45 ^b	27.18 ± 0.85 ^a
Texture		2.32 ± 0.06 ^b	3.45 ± 0.05 ^a	8.68 ± 0.08 ^b	17.60 ± 0.18 ^a

^{a-b} Different superscript letters in the same row indicate the statistically significant difference ($p < 0.05$). CC: cake control; CF: cake added of orange passion fruit peel flour; BC: bread control; BF: bread added of orange passion fruit peel flour.

In the sensorial analysis (Table 5), it was observed that the cake control had better notes of appearance, color, flavor, texture, taste, and global acceptance. In relation to BC and BF, remained largely unchanged in relation to appearance, flavor, and texture. Already for color, the BF presented higher grades, and the BC presented higher grades in relation to taste and global acceptance. So, the bread formulations with addition of orange passion fruit peel flour and chia mucilage had an acceptance better than the cakes. All of the formulations tasted have acceptance of 70 % in all sensory parameters evaluated, indicating that the products can be a future market.

Table 5 Sensorial evaluation of cakes and bread control and the with addition of orange passion fruit peel flour and chia seed mucilage.

Attribute	CC	IA	CF	IA	BC	IA	BF	IA
Appearance	7.83 ^a	86.97	6.56 ^b	72.86	7.65 ^a	85.04	7.90 ^a	87.82
Color	7.85 ^a	87.18	6.69 ^b	74.36	7.21 ^b	80.13	8.04 ^a	89.32
Flavour	7.71 ^a	85.68	6.81 ^b	75.64	7.38 ^a	82.05	7.37 ^a	81.84
Texture	8.06 ^a	89.53	6.33 ^b	70.30	7.94 ^a	88.25	7.42 ^a	82.48
Taste	8.00 ^a	88.89	6.54 ^b	72.65	7.81 ^a	86.75	6.62 ^b	73.50
Global acceptance	8.06 ^a	89.53	6.79 ^b	75.43	7.79 ^a	86.54	7.10 ^b	78.85

IA: Index of Acceptability (%).

^{a-b} Different superscript letters in the same row and for the same formulation, indicate the statistically significant difference ($p < 0.05$).

CC: cake control; CF: cake added of orange passion fruit peel flour; BC: bread control; BF: bread added of orange passion fruit peel flour.

Miranda et al. (2013) developed and tested the acceptance of cakes enriched with yellow passion fruit peel to take advantage of their dietary fiber considered functional. They evaluated four formulations: control (without yellow passion fruit peel), with the addition of 6.66 %, 8.88 % and 12.22 % The parameters of color, flavor, aroma, and texture had a score between seven and eight evaluated by fifty untrained panellists and the formulations presented an index of acceptability higher than 70 %.

Figure 1 shows the spider graphic that shows the sensorial profile of the bread (1A) and cakes (1B) analyzed, highlighting their similarities and differences. The center of the figure represents the zero point of the scale, and the intensity increases from the center to periphery. The average of each attribute per sample is marked on the corresponding axis, where the sensory profile is drawn by the connection of the points.

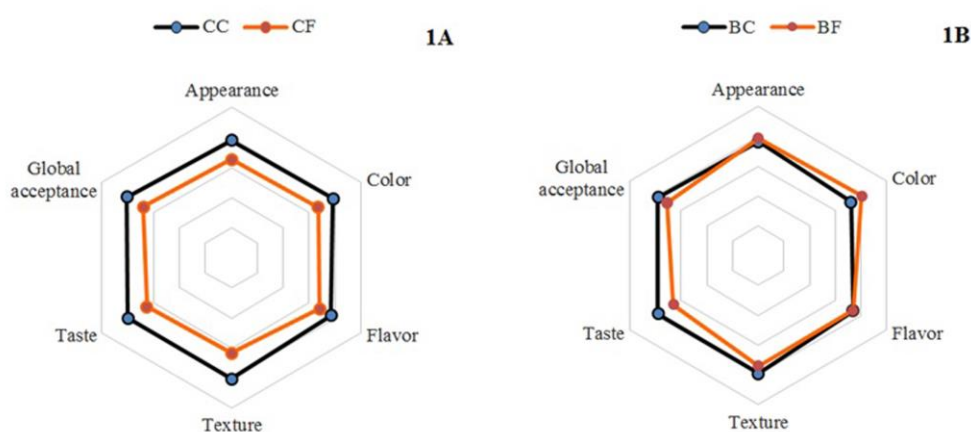


Fig 2. Sensory profile of the CC and CF(A) and BC and BF (B).

CC: cake control; CF: cake added of orange passion fruit peel flour; BC: bread control; BF: bread added of orange passion fruit peel flour.

The addition of orange passion fruit peel flour provided bread and cakes with an acceptance rate of more than 70 %, which demonstrates market potential as a food ingredient since the elaborated products obtained high sensorial acceptance besides adding nutritional value.

Conclusions

The drying at 80 °C was better because it demanded less time and retained higher concentrations of carotenoids evaluated (lutein – 827 µg/100 g, zeaxanthin – 758 µg/100 g, cryptoxanthin – 2797 µg/100 g, α -carotene – 1304 µg/100 g, β -carotene 31073 µg/100 g and phenolic compounds – 758 mg/100 g. For quercetin, it was observed that temperature at 60 °C showed higher concentrations (163 mg/100 g). In the antioxidant capacity, the drying at 70 °C had a greater effect on the radical sequester ABTS, with a value of 1.99 g/100 mL (effective concentration of 50 % radical inhibition).

Despite of temperature employed, the flours made from residues, as is the case of the peel of orange passion fruit can promise ingredients for the enrichment of cakes and bread with total dietary fiber, minerals, and bioactive compounds as with antioxidant potential, that can contribute to reducing of agro-industrial waste too. The sensorial analysis showed that the formulations tested had an acceptance index of at least 70 % to be commercialized in the future.

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CAPÍTULO 8

8. Discussão Geral

A caracterização físico-química e de compostos bioativos de todas as partes de diferentes espécies e cultivares de maracujá é de extrema importância tanto para prospecção do fruto e de novas espécies bem como para aproveitamento integral dos frutos. Pelo presente estudo pode-se identificar que as variedades estudadas têm características diferentes e cada uma se destaca por diversos compostos. O maracujá laranja se destaca por sua coloração vermelha na polpa, alto teor de cinzas presentes na polpa e casca e o alto conteúdo de proteínas e lipídios nas sementes. O maracujá amarelo, por sua vez, mostrou um alto teor de carboidratos e pectina na casca. O maracujá roxo destaca por apresentar um alto teor de carboidratos na polpa em comparação com maracujá laranja e amarelo.

Em relação às polpas, as três espécies possuem alto teor de cálcio, potássio e boro. Por sua vez, as cascas de maracujá amarelo e laranja apresentaram alto teor de zinco, manganês, cobre, fósforo e enxofre. Quanto à casca do maracujá roxo, esses minerais tinham baixo teor. As sementes dos maracujás, muitas vezes descartadas, apresentaram uma quantidade considerável de cobre para o maracujá laranja, seguido do maracujá amarelo.

No teor de compostos bioativos, a casca do maracujá roxo apresenta antocianinas, a polpa do maracujá laranja representa uma boa fonte de licopeno e suas cascas um grande conteúdo de β -caroteno e compostos fenólicos. A polpa do maracujá amarelo apresentou maior conteúdo de quercetina e capacidade antioxidante ABTS e DPPH.

Neste trabalho também foram avaliados quatro cultivares desenvolvidas pela EPAGRI-SC (Empresa de Pesquisa Agropecuária e Extensão Rural de Santa Catarina) onde observou-se que a cultivar ‘BRS Rubi do Cerrado’ mostrou ser mais doce devido ao maior teor de açúcares e a ‘BRS Sol do Cerrado’ mais ácida (maior acidez titulável); a cultivar ‘BRS Gigante Amarelo’ apresentou maior teor de umidade, gordura total, cinzas, proteínas e fibra total, teor fenólico total, epigallocatequina galato, quercetina, carotenoides e provitamina A. A cultivar ‘BRS Rubi do Cerrado’ mostrou maior conteúdo de campferol; para os minerais testados, a cultivar ‘BRS Sol do Cerrado’ exibiu maiores concentrações de todos os minerais, exceto o cobre e as cultivares ‘SCS437 Catarina’ e ‘Gigante Amarelo’ apresentaram maior capacidade antioxidante.

A partir dos dados encontrados com as polpas de maracujá, constatou-se que algumas cultivares desenvolvidas pela EPAGRI apresentaram teor de compostos semelhantes em relação às espécies estudadas no primeiro artigo, tais como: rendimento da polpa, SST, umidade, lipídeos e carboidratos; para alguns minerais como: cobre, fósforo, enxofre, magnésio, potássio e cálcio; para alguns compostos bioativos como: compostos fenólicos, quercetina, campferol, luteína, zeaxantina, β -caroteno, provitamina A e carotenoides totais. Por isso, observou-se muita similaridade entre a composição físico-química, centesimal e de compostos bioativos.

Através destes dois estudos pode-se concluir que o maracujá, de maneira geral, é uma fruta rica nutricionalmente contendo algumas variações na sua composição devido, provavelmente, às diferenças relativas ao solo, clima e temperaturas em que são cultivados.

Nesta pesquisa também foi avaliada a estabilidade do suco do maracujá laranja fresco e pasteurizado. Os resultados mostraram que nas análises físico-químicas, observaram-se diferenças significativas entre o suco pasteurizado e fresco durante o

armazenamento. Para parâmetros de cor, compostos fenólicos, quercetina, criptoxantina, licopeno, carotenoides totais e capacidade antioxidante, o suco pasteurizado apresentou maiores concentrações. Já o suco fresco reteve maiores valores de epigalocatequina, α -caroteno, β -caroteno e provitamina A. Segundo este estudo, o consumo ideal do suco seria após o preparo e, na impossibilidade deste, recomenda-se se consumo em até 4 dias para suco *in natura* ou em até 6 dias para suco pasteurizado de armazenagem sob temperatura refrigerada (8 °C) após processamento do suco, respectivamente.

Pode-se afirmar, que todos os maracujás analisados neste estudo têm potencial para produção de suco, pois o maracujá é um fruto com alta aceitação no mercado devido ao seu sabor e aroma agradáveis. O suco do maracujá laranja foi estudado isoladamente, devido à restrita quantidade de estudos científicos com esta fruta.

Para o aproveitamento integral do maracujá laranja, foi avaliada a secagem das cascas em três temperaturas diferentes 60 °C, 70 °C e 80 °C para a produção e aplicação da farinha em formulações de pão e bolo. Os resultados indicaram que a farinha obtida sob secagem a 80 °C apresentou maiores valores de compostos bioativos, contudo independente da temperatura empregada, todas as farinhas avaliadas podem ser utilizadas como ingredientes para o enriquecimento de produtos de panificação, pois são fontes de fibra alimentar, minerais e compostos bioativos. A análise sensorial mostrou que as formulações de pão e bolo tiveram uma aceitação superior a 80 % nos quesitos de aparência, cor, aroma, textura, sabor e aceitação global.

O consumo de formulações à base de farinha de casca de maracujá (tanto o laranja como outras espécies já estudadas) tem finalidade ambiental e nutricional, pois se utilizam as cascas que seriam descartadas e, ao mesmo tempo, se oferecem alimentos com ingredientes que enriquecem a formulação, principalmente as fibras solúveis e os compostos bioativos.

CAPÍTULO 9

9. Conclusão

Os resultados desta pesquisa mostraram que o maracujá na sua forma integral (polpa, cascas e sementes) é rico nutricionalmente e tem elevado conteúdo de compostos bioativos.

Em relação ao conteúdo de compostos bioativos, o que vai diferir vai ser o tipo de maracujá que vai descrever a quantidade destes em cada espécie ou cultivar. Vale ressaltar que as espécies e cultivares estudadas apresentaram elevado teor de compostos fenólicos, flavonoides e capacidade antioxidante.

O uso tanto da polpa como os produtos originados da casca podem contribuir na composição de uma dieta saudável e balanceada. As cascas são ricas em minerais e pectina, uma fibra solúvel que é capaz de diminuir o colesterol sérico e a glicemia e também auxilia no bom funcionamento do intestino. As sementes são ricas em proteínas, lipídeos e fibras totais. As polpas apresentaram elevada quantidade de carboidratos e minerais.

A utilização do suco pasteurizado (88 °C por 15 segundos) mostrou ser benéfica na proteção contra a degradação de alguns carotenoides analisados, flavonoides, compostos fenólicos e capacidade antioxidante, sendo incentivada à sua utilização.

Por fim, a utilização da casca do maracujá laranja em forma de farinha para o enriquecimento de formulações como pão e bolo apresentou boa aceitação (superior a 80 %) na análise sensorial, e que esta farinha, pode melhorar as qualidades nutricionais dos produtos, contendo fibras, minerais e compostos bioativos com potencial antioxidante, e que o uso das cascas podem contribuir para a redução do desperdício agroindustrial.

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