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Tropical fruit by-products water extracts as sources of soluble fibres and phenolic compounds with potential antioxidant, anti-inflammatory, and functional properties

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อ่านคำสำคัญ (Keywords) ก่อน จะเป็นตัวบอกว่าเป็นงานวิจัยเกี่ยวกับอะไร ส่วนบทคัดย่อ (Abstract) เก็บไว้อ่านทีหลัง หรือเอาไว้อ่านสุดท้ายเลยก็ได้ เพราะบางครั้งอาจจะเข้าใจยาก ถ้าเราไม่มีพื้นความรู้เชิงลึกในเรื่องนั้น ๆ มาก่อน

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ABSTRACT

Fibre content, phenolic content, antioxidant and anti-inflammatory activities were evaluated for water extracts of fruit by-products (passion fruit, orange, acerola, and mango). The impact of these extracts on microbial growth, folate production, and adhesion ability of *Streptococcus thermophilus* TH-4 and *Lactobacillus rhamnosus* LGG was investigated. Mango water extract (MWE) presented the highest phenolic content and antioxidant activity. Orange water extract (OWE) and MWE showed the best anti-inflammatory potential by decreasing the highest nitric oxide levels. When TH-4 and LGG were grown together, folate production was only stimulated by MWE. Passion fruit water extract and OWE increased the TH-4 adhesion whereas acerola water extracts and MWE improved LGG adhesion when strains were used individually. These results showed that fruit by-product water extracts (FWE), especially from mango, presented potential beneficial biological and functional properties. These FWE could be used to develop new functional antioxidant foods and natural pharmaceutical ingredients.

คำสำคัญ ของแต่ละบทความ มักจะมี 4-6 คำ คำสำคัญที่ดีจะบอกได้ว่างานวิจัยนั้น ๆ ทำอะไรและได้อะไรเป็นผลงาน

Keywords:

Probiotic

Fruit by-products

Dietary fibre

Phenolic compounds

Anti-inflammatory effect

Folate

บทความวิจัย มักประกอบด้วยหัวข้อหลัก ๆ ดังนี้

1. **บทนำ** (Introduction) บอกที่มา วัตถุประสงค์ และผลที่คาดว่าจะได้รับของงานวิจัย
2. **อุปกรณ์และวิธีการ** (Materials and methods) บอกวิธีการว่าใช้วิธีอะไร ใช้อุปกรณ์เครื่องมืออะไรบ้าง
3. **ผลการทดลอง** (Results) แยกเป็นหัวข้อ สอดคล้องกับวิธีการ อาจนำเสนอในรูปตาราง แผนภูมิ ภาพถ่าย ตามความเหมาะสม การนำเสนอที่ดีจะสามารถดูแล้วเข้าใจโดยไม่จำเป็นต้องไปอ่านตัวข้อความบรรยายในบทความอีก
4. **อภิปราย** (Discussion) เป็นการนำเสนอข้อคิดเห็นของผู้วิจัย ว่าผลที่ได้ในคราวนี้สอดคล้องหรือขัดแย้งกับงานวิจัยอื่น ๆ ที่มีมาก่อนหรือไม่ อย่างไร และอะไรที่ถือเป็นการค้นพบใหม่ในครั้งนี้ ในอนาคตควรปรับปรุงหรือพัฒนาอย่างไรต่อไป
5. **สรุป** (Conclusion)

ลำดับการอ่านบทความให้เข้าใจภาพรวม หากมีเวลาน้อย ควรทำตามลำดับดังนี้

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ส่วน อุปกรณ์และวิธีการ หากเราไม่ได้จะทำการทดลองแบบเดียวกัน อาจไม่ต้องอ่าน

5. Conclusion

This study showed that tropical fruit by-products water extracts, especially from mango by-products, are a source of biologically active compounds with anti-inflammatory potential and also can stimulate folate production by lactic acid bacteria. These water extracts could be used in the development of novel foods that can improve the health of the consumers. Taken together, the results highlight the use of mango by-product water extract as the most promising natural water extract from tropical fruit by-products considering the physicochemical, biological, and functional parameters evaluated. Further studies are required to characterize the different bioactive compounds and elucidate the mechanisms of action that allow them to modulate innate inflammatory processes and improve folate production.

สรุป จะมีไม่กี่บรรทัด บางครั้งมีเพียงย่อหน้าเดียว 4-5 ประโยค นับเป็นส่วนที่อ่านเข้าใจง่ายที่สุดในแต่ละบทความ หากส่วนสรุปไม่อยู่ในความสนใจของเรา ก็จบเท่านี้ ไม่ต้องเสียเวลาอ่านต่อ

5. Conclusion

This study showed that tropical fruit by-products water extracts, especially from mango by-products, are a source of biologically active compounds with anti-inflammatory potential and also can stimulate folate production by lactic acid bacteria. These water extracts could be used in the development of novel foods that can improve the health of the consumers. Taken together, the results highlight the use of mango by-product water extract as the most promising natural water extract from tropical fruit by-products considering the physicochemical, biological, and functional parameters evaluated. Further studies are required to characterize the different bioactive compounds and elucidate the mechanisms of action that allow them to modulate innate inflammatory processes and improve folate production.

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บทนำ จะพูดถึงที่มาของงานวิจัย ว่าเพราะอะไรถึงทำงานวิจัยนี้ และมีความสำคัญอย่างไร มักจะแบ่งประเด็นออกเป็นย่อหน้า เราสามารถอ่านเพียง 2-3 บรรทัดของแต่ละย่อหน้าก่อน เพื่อให้ทราบประเด็น และประหยัดเวลาในการอ่าน

1. Introduction

Brazil produces and processes an impressive amount of tropical fruits, such as orange, passion fruit, mango, and acerola. As a result, large amounts of fruit by-products (peel, pulp, and seeds) are generated from the fruit processing industry and represent an important environmental problem (Vieira, Bedani, Albuquerque, Bíscola, & Saad, 2017). In contrast, these residues are a natural source of nutrients and different bioactive molecules. These biocompounds have great potential to be used as functional food ingredients or for application as phytochemical pharmaceutical substances for the prevention or treatment of human diseases (Kowalska, Czajkowska, Cichowska, & Lenart, 2017; O'Shea et al., 2015; Oliveira, Angonese, Gomes, & Ferreira, 2016).

In this context, dietary fibres (DF) are important plant components present in large amounts in fruit by-products. DF are not hydrolysed by the endogenous enzymes in the small intestine of humans (Joint FAO/WHO, 2010) and their consumption is associated with beneficial health effects, including, regulation of the intestinal transit, prevention or treatment of cancer, diabetes, and cardiovascular diseases (Beres et al., 2016; Wang et al., 2015).

Phenolic compounds are known to be widely present in fruits and their by-products, and they are the most important group of natural antioxidants in the diet. These bioactive molecules act as reducing agents and can improve human health. Besides the structural diversity of phenolic compounds, it is crucial to determine their biological properties such as anti-inflammatory activity, microbial growth stimulation, and their impact on the production of beneficial metabolites by different microorganisms, especially by probiotics (LeBlanc et al., 2017; Oh et al., 2012; Rocchetti, Chiodelli, Giuberti, & Lucini, 2018; Santos et al., 2017). Probiotics are "live microorganisms that, when administered in adequate amounts, confer a health benefit on the host" (Hill et al., 2014).

DF from fruits by-products may be good sources of bound phytochemicals including phenolic compounds (Haminiuk, Maciel, Plata-Oviedo, & Peralta, 2012). In some cases, the complex DF-phenolic compounds is considered as being an antioxidant dietary fibre, a category defined as "a dietary fibre concentrate containing significant amounts of natural antioxidants associated with non-digestible compounds" (Quirós-Sauceda et al., 2014).

According to Ajila and Rao (2013), DF associated with polyphenols may enhance the antioxidant effect of these bound phenolic compounds in the human intestine when compared to the non-bound ones. Considering that the DF-phenolic complexes are not hydrolysed by the human digestive enzymes (Velderrain-Rodríguez et al., 2016), they may exert a positive impact on the gut microbiota modulation (Tomás-Barberán, Selma, Espín, 2016). In addition, they contribute to maintain a balanced oxidant status in the intestinal environment, in that manner, protecting the intestinal epithelium against inflammatory processes, protecting the intestine against the colonization by pathogens, and increasing the adhesion of probiotics microorganisms to intestinal cells (Ajila & Rao, 2013; Dueñas et al., 2015).

Inflammatory processes may start in the gut due to an unbalanced intestinal microbiota promoting the action of pathogens and improvement of inflammation since the Toll-like receptors of macrophages are able to recognize lipopolysaccharides (LPS), an endotoxin produced by pathogenic microorganisms (Ambriz-Pérez, Leyva-López, Gutierrez-Grijalva, & Heredia, 2016). Different mechanisms of action might be related to the anti-inflammatory properties of the phenolic compounds. Among them, up/downregulation of transcriptional factors (e.g, NF-kβ), inhibition of pro-inflammatory mediators (e.g, interleukin IL-6), inhibition of activated immune cells (e.g, macrophages), and inhibition of the inducible nitric oxide synthase (iNOS) and cyclooxygenase-2 (COX-2) are discussed (Ambriz-Pérez et al., 2016).

The use of fruit by-products with prebiotic potential to improve the growth of beneficial microorganisms and the production of beneficial metabolites such as the B-group vitamins by probiotic and gut commensal microorganisms has been investigated (Albuquerque, Bedani, Vieira, LeBlanc, & Saad, 2016; LeBlanc et al., 2017; Vieira, Bedani, Albuquerque, Biscola, & Saad, 2017). According to Gibson et al. (2017), prebiotic is defined as "a substrate that is selectively utilized by host microorganisms conferring a health benefit" extending the prebiotic effect to other substrates such as non-carbohydrate substrates, which could include phenolic compounds.

Folate is a B-group vitamin required by humans for their metabolic activities such as DNA replication, repair, and methylation and for the biosynthesis of nucleic acids. As humans are not able to produce folate, this vitamin must be obtained from the diet or from supplements. In addition, some beneficial microorganisms such as probiotics, present the ability to produce folate *de novo* which could be used to improve folate content in foods or in the gut through the fermentation of vegetable substrates by the human microbiota (Albuquerque et al., 2016).

The aim of this study was to determine the dietary fibre fractions, phenolic content and composition, and the antioxidant activity of four different fruit by-products water extracts (FWE) (from passion fruit, orange, acerola, and mango) and evaluate their anti-inflammatory potential. Additionally, this work aimed to evaluate the impact of each FWE on the growth and folate production by *Streptococcus thermophilus* TH-4 and *Lactobacillus rhamnosus* LGG and on the *in vitro* adherence abilities of both microorganisms to intestinal human epithelial cells.

3. Results

3.1. Fibre and total phenolic contents, antioxidant activity, and phenolic composition of each FWE

Fruit by-product water extracts (FWE) were used as sources of dietary fibre associated with phenolic compounds. The total dietary fibres, soluble fibres, insoluble fibres, total phenolic content, phenolic composition, and the antioxidant activity of all FWE were evaluated. Insoluble fibres were not detected in any of the FWE since all hot boiled mixtures described in Section 2.2 were filtered. Thus, the total dietary fibres amount did not differ from the soluble fibres content. Regarding Table 1, significant differences between the soluble fibres values of the FWE ($p < 0.05$) were observed. Mango water extract (MWE) presented the highest soluble fibres content (0.47 ± 0.01 g 100 mL^{-1}) while passion fruit water extract (PFWE) (0.12 ± 0.0 g 100 mL^{-1}) presented the lowest content.

The Folin-Ciocalteu assay was used to determine the total phenolic content (TPC) of FWE and the results are shown in Table 1. MWE presented the highest TPC (1.31 ± 0.03 mg GAE mL^{-1} FWE) whereas

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Table 1
Soluble fibre, total phenolic content, phenolic composition, and antioxidant activity of fruit by-products water extracts (FWE).

	Fruit by-products water extracts			
	PFWE	OWE	AWE	MWE
Soluble fibres (g/100 mL)	0.12 (0.0) [D]	0.39 (0.01) [B]	0.26 (0.01) [C]	0.47 (0.01) [A]
Total phenolics (mg GAE mL^{-1} FWE)	0.14 (0.00)[D]	0.52 (0.01)[C]	0.95 (0.06)[B]	1.31 (0.03)[A]
Phenolic profile (mg L^{-1} FWE)				
Phenolic acids				
Vanillic acid	1.0	8.0	2.0	0
Siringic acid	1.0	16.0	0	0
Gallic acid	1.0	38.0	0	199.0
Protocatechuic acid	0	4.0	0	1.0
Flavonoids				
Rutin	7.0	17.0	8.0	29.0
Quercitin	4.0	10.0	0	4.0
Epicatechin	0	0	0	2.0
Antioxidant activity				
ORAC (μmol Trolox L^{-1} FWE)	46.95 (0.65)[B]	13.62 (0.48)[C]	47.03 (1.86)[B]	61.56 (2.16)[A]
DPPH (μmol Trolox L^{-1} FWE)	304.7 (36)[D]	898.9 (94)[CD]	4431.7 (27 4)[B]	18708.7 (1987)[A]

PFWE: passion fruit water extract, OWE: orange water extract, AWE: acerola water extract, MWE: mango water extract. [A,B] Different capital letters in the same row denote significant differences between fruit by-products ($P < 0.05$).

water extractions yielded higher amounts of total phenolics than those with ethanol. From an ecological and economical point of view the use of water to perform extractions would be better and safer than the use of organic solvents.

The method employed for extracting phenolics used in our study was chosen based on a previous study (Beres et al., 2016). Temperature, solute:solvent ratio, and particle size were reported to influence the extraction of antioxidant fibers, which can be defined as polysaccharides bounded to phenolic compounds (Saura Calixto et al., 1998). Our data suggests that the FWE may be a good source of soluble fibres associated to phenolic compounds. Since the extraction of FWE were conducted at 100 °C during 1 h, free phenolics might be degraded, and phenolic compounds bound to polysaccharides could be protected during this treatment and be extracted from the matrix.

In fact, Beres et al. (2016) found that the variable that most influenced the extraction of antioxidant fibers was the temperature. The authors reported a positive relation between higher temperatures and extraction efficiency. Free phenolics are easily extracted from different matrix using conventional extraction methods. However, according to Chamorro, Viveros, Alvarez, Vega, and Brenes (2012), these compounds are not bio-accessible in the human intestine or even metabolized by the gut microbiota and, in this case, the benefits shown *in vitro* would not be reproducible *in vivo*. The main focus of our study was to obtain FWE that can be used by humans, considering that the bio-accessibility

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A B S T R A C T

Fibre content, phenolic content, antioxidant and anti-inflammatory activities were evaluated for water extracts of fruit by-products (passion fruit, orange, acerola, and mango). The impact of these extracts on microbial growth, folate production, and adhesion ability of *Streptococcus thermophilus* TH-4 and *Lactobacillus rhamnosus* LGG was investigated. Mango water extract (MWE) presented the highest phenolic content and antioxidant activity. Orange water extract (OWE) and MWE showed the best anti-inflammatory potential by decreasing the highest nitric oxide levels. When TH-4 and LGG were grown together, folate production was only stimulated by MWE. Passion fruit water extract and OWE increased the TH-4 adhesion whereas acerola water extracts and MWE improved LGG adhesion when strains were used individually. These results showed that fruit by-product water extracts (FWE), especially from mango, presented potential beneficial biological and functional properties. These FWE could be used to develop new functional antioxidant foods and natural pharmaceutical ingredients.