Maastricht University

Bioconversion by gut microbiota of predigested mango (Mangifera indica L) 'Ataulfo' peel polyphenols assessed in a dynamic (TIM-2) in vitro model of the human colon

Citation for published version (APA):

Sáyago-Ayerdi, S. G., Venema, K., Tabernero, M., Sarriá, B., Bravo, L. L., & Mateos, R. (2021). Bioconversion by gut microbiota of predigested mango (Mangifera indica L) 'Ataulfo' peel polyphenols assessed in a dynamic (TIM-2) in vitro model of the human colon. *Food Research International*, *139*, Article 109963. https://doi.org/10.1016/j.foodres.2020.109963

Document status and date:
Published: 01/01/2021

DOI:
10.1016/j.foodres.2020.109963

Document Version:
Publisher's PDF, also known as Version of record

Document license:
Taverne

Please check the document version of this publication:

• A submitted manuscript is the version of the article upon submission and before peer-review. There can be important differences between the submitted version and the official published version of record. People interested in the research are advised to contact the author for the final version of the publication, or visit the DOI to the publisher's website.
• The final author version and the galley proof are versions of the publication after peer review.
• The final published version features the final layout of the paper including the volume, issue and page numbers.

Link to publication

General rights
Copyright and moral rights for the publications made accessible in the public portal are retained by the authors and/or other copyright owners and it is a condition of accessing publications that users recognise and abide by the legal requirements associated with these rights.

• Users may download and print one copy of any publication from the public portal for the purpose of private study or research.
• You may not further distribute the material or use it for any profit-making activity or commercial gain
• You may freely distribute the URL identifying the publication in the public portal.

If the publication is distributed under the terms of Article 25fa of the Dutch Copyright Act, indicated by the "Taverne" license above, please follow below link for the End User Agreement:

www.umlib.nl/taverne-license

Take down policy
If you believe that this document breaches copyright please contact us at:

repository@maastrichtuniversity.nl

providing details and we will investigate your claim.

Download date: 20 Apr. 2024

Food Research International 139 (2021) 109963

Contents lists available at ScienceDirect

Food Research International

journal homepage: www.elsevier.com/locate/foodres

Bioconversion by gut microbiota of predigested mango (*Mangifera indica* L) 'Ataulfo' peel polyphenols assessed in a dynamic (TIM-2) *in vitro* model of the human colon

Sonia G. Sáyago-Ayerdi [a,*], Koen Venema [b], Maria Tabernero [c], Beatriz Sarriá [d], L. Laura Bravo [d], Raquel Mateos [d,*]

[a] Tecnológico Nacional de México/Instituto Tecnológico de Tepic, Av. Instituto Tecnológico No 2595, Col. Lagos del Country, CP 63175 Tepic, Nayarit, Mexico
[b] Maastricht University – Campus Venlo, Centre of Healthy Eating & Food Innovation, St. Jansweg 20, 5928 RC Venlo, the Netherlands
[c] IMDEA-Food Institute, CEI (UAM-CSIC), Carretera de Canto Blanco, 8, 28049 Madrid, Spain
[d] Institute of Food Science, Technology and Nutrition (ICTAN-CSIC), Spanish National Research Council (CSIC), José Antonio Nováis 10, 28040 Madrid, Spain

ARTICLE INFO

Keywords:
Mango (*Mangifera indica* L.)
Ataulfo
By-product
Peel
Phenolic compounds
Gut microbiota
In vitro model of the human colon
Bioconversion

ABSTRACT

Gut microbiota bioconversion of polyphenols in predigested mango 'Ataulfo' peel was studied using a validated, dynamic *in vitro* human colon model (TIM-2) with faecal microbial inoculum. Dried peels were predigested with enzymatic treatment, followed by TIM-2 fermentation (72 h). Samples were taken at 0, 24, 48 and 72 h and analyzed by HPLC-QToF. Derivatives of hydroxyphenylpropionic, hydroxyphenylacetic and hydroxybenzoic acids, as well as, pyrogallol were the main polyphenols identified. These metabolites might derivate from flavonoid (flavanols and flavonols), gallate and gallotannin biotransformation. Despite the high content of ellagic acid in mango peel, low amounts were detected in TIM-2 samples due to transformation into urolythins A and C, mainly. Xanthone and benzophenone derivatives, specific to mango, remained after the colonic biotransformation, contrary to flavonoids, which completely disappeared. In conclusion, microbial-derived metabolites, such as xanthone and benzophenone derivatives, among others, are partially stable after colonic fermentation, and thus have the potential to contribute to mango peel bioactivity.

1. Introduction

Mango (*Mangifera indica* L) is one of the most popular tropical fruits worldwide. Asia is the major producer with 72.9% of the world production, followed by Africa and America with 15.4% and 11.5%, respectively, of the total production (FAOSTAT, 2016). There are hundred of mango varieties and, Mexico produces different varieties that are highly accepted in the international market, but nowadays the 'Ataulfo' variety has proven to be the most successful because of its unique sensory properties, such as firm consistency, sweet drupe, and intense aroma. Mango fruit pulp can be consumed as a mature fruit, or otherwise it can be used to obtain juices or concentrates. About 50–55% of the fruit, corresponding to seeds, paste, and peel, are discarded (Santos, 2002). The interest in mango by-products is growing because of the increasing demand by consumers for natural ingredients with potential health properties. Mango peel has shown to be a good source of dietary fiber and bioactive compounds, such as polyphenols (Blancas-Benitez et al., 2015). Different polyphenols have been identified in mango peels, such as gallates, gallotannins, flavonoids, ellagic acid and related compounds, mangiferin, and maclurin derivatives (Dorta, González, Lobo, Sánchez-Moreno, & de Ancos, 2014; López-Cobo et al., 2017), among other more complex hydrolyzable polyphenols, comprising gallotannins from penta to trideca-galloyl-glycosides (Sáyago-Ayerdi et al., 2013).

It is pertinent to study the behavior of polyphenols during digestion and fermentation, rather than only to quantify the polyphenols in the food matrix, because some phenolic compounds can be released during gastrointestinal digestion in the small intestine (bioaccesibility) (González-Aguilar, Blancas-Benitez, & Sáyago-Ayerdi, 2017), but most are bound to dietary fiber and can only be released and biotransformed by the colonic microbiota. These resulting microbial metabolites may be responsible for the health effects associated to dietary polyphenols (Crozier, Jaganath, & Clifford, 2009), in agreement with the new definition of "three P for intestinal health" that includes probiotics (bacteria), prebiotics (dietary fiber), and polyphenols at the same biological

* Corresponding authors.
E-mail addresses: ssayago@ittepic.edu.mx (S.G. Sáyago-Ayerdi), raquel.mateos@ictan.csic.es (R. Mateos).

https://doi.org/10.1016/j.foodres.2020.109963
Received 16 April 2020; Received in revised form 8 October 2020; Accepted 27 November 2020
Available online 8 December 2020
0963-9969/© 2020 Elsevier Ltd. All rights reserved.

level (Espín, González-Sarrías, & Tomás-Barberán, 2017). To understand the microbial transformations that take place during colonic fermentation, an *in vitro* dynamic system has been developed by the Dutch Organization for Applied Scientific Research (TNO) (nick-named TIM-2, for TNO *in vitro* model of the colon) with the following advantages: it can simulate the kinetic conditions in the colonic tract, has a dialysis membrane that enables to remove metabolites from the lumen and prevents them from accumulating, thereby leading to inhibition or even death of the microbes (Venema, Nuenen, Smeets-Peeters, Minekus, & Havenaar, 2000) and it allows simultaneous screening of different metabolites, for instance short chain fatty acids, ammonia, polyphenols, minerals, etc. (Koenen, Cruz Rubio, Mueller, & Venema, 2016). Bearing this in mind, the aim of this work was to study the biotransformation of mango peel polyphenols by gut microbiota using a predigested fraction of mango peels and the human colon dynamic *in vitro* model.

2. Materials and methods

2.1. Predigestion of mango peels

The mango peel by-product was provided by by MexiFrutas®, S.A. of C.V. in Nayarit, Mexico. The peels were obtained after processing the mango to obtain juice. The fruits had to be in a mature stage, this means with 12–14 ^{o}Brix, and they were collected at the summer season in the Occident of Mexico. Mango peel sample was freeze-dried (Labconco, Freezone 6–7752020, Kansas City, Missouri, United States). The mango peel has 52.61% of total indigestible fraction (Sáyago-Ayerdi, Zamora-Gasga & Venema, 2019), that comprises the dietary fiber (soluble and insoluble), and polyphenols content. On the basis of this percentage the amount of mango peel to be predigest was considered, according to the amount required to feed the TIM-2 system. This sample was submitted to hydrolysis with pepsin (pH 1.5, 37 °C, 1 h), and pancreatin (pH 6.9–7.5, 37 °C, 4 h), centrifuged (20 min, 8000 *g*, Centurion Scientific, K243R, Germany), and the supernatant was diafiltrated (Sureflux, Nipro Europe NV, Zaventum, Belgium) using a peristaltic pump to remove small digestion products and water (Venema, 2015).

2.2. Preparation of fecal inoculum

Fecal samples were collected from donors at Maastricht University - Campus Venlo, The Netherlands. The volunteers (two males and one female with an age range of 28–47 years) were healthy and declared that they did not follow any dietary restrictions, had no gastrointestinal diseases nor had taken antibiotics 3 months previous to donation. Donors put the fecal samples (2 h max after defecation) in a plastic jar containing a gastight bag with an anaerocult strip (AnaeroGen™, Cambridge, UK). The fecal samples (500 g) were mixed with 450 mL of 10 × concentrated dialysis liquid, 2490 mL of demi-water, and 560 g of glycerol; the mixture was afterwards standardized in an anaerobic cabinet (Sheldon Lab – Bactron IV, Cornelius, OR, USA) according to Venema et al. (2000). The fecal material was aliquoted, frozen in liquid nitrogen, and stored at − 80 °C until use for fermentation in the TIM-2 system.

2.3. In vitro colonic fermentation of predigested mango peels in the dynamic TIM-2 system

TIM-2 system contains a dialysis membrane that simulates the uptake of microbial metabolites by the body avoiding accumulation of these in the system (Maathuis, Hoffman, Evans, Sanders, & Venema, 2009). The TIM-2 system was inoculated with 70 mL of the standardized microbiota (described above) with 50 mL of dialysis liquid which contained (per liter): 2.5 g dipotassium hydrogen phosphate trihydrate, 4.5 g sodium chloride, 0.005 g ferrous sulfate heptahydrate, 0.5 g magnesium sulfate heptahydrate, 0.45 g calcium chloride dihydrate, 0.05 g bile and 0.4 g *L*-cysteine hydrochloride, plus 1 mL of the vitamin mixture (containing per liter: 1 mg menadione, 2 mg *D*-biotin, 0.5 mg vitamin B12, 10 mg pantothenate, 5 mg nicotinamide, 5 mg *p*-aminobenzoic acid and 4 mg thiamine). The microbiota was adapted to the model conditions with the simulated ileal effluent medium (SIEM), which simulates the indigestible fraction of a high fiber diet that can reach the colon and be fermented (without polyphenols) (Sáyago-Ayerdi et al., 2019).

Before the addition of the test sample (predigested mango peel), the microbiota was adapted to the model conditions with the SIEM for 20 h, and after that a 4 h starvation period allowed the bacteria to ferment all available carbohydrates in the system. After the starvation period, samples were collected at time-point zero (t_0). As a control sample, SIEM medium was added in one unit of the system. The predigested mango peel (7.5 g/day, 2.5 mL/h) was mixed with SIEM without the indigestible carbohydrates (pectin, starch, xylan, arabinogalactan) was fed in three different units of the system. Then the 72 h experimental period started and samples were collected at 0 h, 24 h, 48 h and 72 h and stored at −80 °C until analysis.

2.4. Extraction of phenolic compounds from mango peel, predigested mango peel and fermented samples

Phenolic compounds were extracted (Pérez-Jiménez et al., 2008) in 1 g of sample (mango peel or predigested mango peel). Each sample (n = 2) was extracted in aqueous methanol (50:50, v/v, with HCl 2 N, 1 h) by constant shaking, and centrifuged at 3000*g*. Supernatants were separated and the pellets washed with acetone/water (70:30, v/v) by constant shaking and centrifuged at 3000*g*. Supernatants from each extraction step were combined at 100 mL. An aliquot of 1 mL was concentrated under reduced pressure using a vacuum concentrator system (Speed-Vac, Thermo Fisher Scientific Inc., Waltham, MA, USA), and then resuspended in the same volume of 1% formic acid in deionized water (v/v), filtered (0.45 μM pore-size, cellulose-acetate membrane filters), dispensed in chromatographic vials and stored at −80 °C until analysis. The fermented samples, a 1 mL aliquot in an eppendorf was centrifuged for 30 min at 14,000*g* (4 °C), filtered (0.45 μm pore size cellulose-acetate membrane filter), dispensed in chromatographic vials and stored at −80 °C until analysis.

2.5. Characterization and quantification of the phenolic content of mango peel, predigested mango peel and fermented samples by LC-ESI-QToF analysis

Phenolic compounds from the mentioned samples were characterized by HPLC-ESI-QToF (Gómez-Juaristi, Martínez-López, Sarria, Bravo, & Mateos, 2018), in an Agilent 1200 series LC system coupled to an Agilent 6530A Accurate-Mass Quadrupole Time-of-Flight (Q-ToF) with ESI-Jet Stream Technology (Agilent Technologies). Compounds were separated on a reverse-phase Ascentis Express C18 (15 cm × 3 mm, 2.7 μm) column (Sigma-Aldrich Quimica, Madrid) preceded by a Supelco 55215-U guard column at 30 °C. Five μL of sample were injected and separated by using a mobile phase consisting of Milli-Q water (phase A) and acetonitrile (phase B), both containing 0.1% formic acid, at a flow rate of 0.3 mL/min. The mobile phase was initially programmed with 90% of solvent A and 10% of B. The elution program increased to 30% of solvent B in 10 min, 40% solvent B in 5 min, and 50% of solvent B in 5 min. Then, the initial conditions (10% solvent B) were recovered in 2 min and maintained for 8 min. The Q-ToF acquisition conditions were as follows: drying gas flow (nitrogen, purity >99.9%) and temperature were 10 L/min and 325 °C, respectively; sheath gas flow and temperature were 6 L/min and 250 °C, respectively; nebulizer pressure was 25 psi; cap voltage was 3500 V and nozzle voltage was 500 V. Mass range selected was from 100 up to 970 *m/z* in negative mode and fragmentor voltage of 150 V. Data were processed in a Mass Hunter Workstation Software. External calibration curves were prepared with the following standards (gallic acid, 3,4-dihydroxyphenylpropionic acid, 3-hydroxyphenylpropionic acid, 3,4-dihydrophenylacetic acid, 4-

hydroxyphenylacetic acid, protocatechuic acid, 4-hydroxybenzoic acid, coumaric acid, caffeic acid, ferulic acid, catechin, epicatechin, quercetin, mangiferin, maclurin and ellagic acid) at six different concentration levels from 0.001 to 20 μM. Limit of detection and quantification ranged from 0.001 to 0.005 μM and from 0.003 to 0.007 mM, respectively. The inter- and intra-day precision of the assay (as the coefficient of variation, ranging from 3.5 to 8.5%) were considered acceptable and allowed the quantification of phenolic compounds and their metabolites (quantified as equivalents of the respective parent molecules when were available or the most chemically related). Table 1 indicates the reference standard used to quantify each identified compound.

2.6. Statistical analysis

All analyses were performed by triplicate (n = 3), mean values and standard deviations from each value were calculated. Mango peel and predigested mango data was analysed using *t*-Student's test. Fermentation mango peel data was analysed by a two ways ANOVA/Fisher's least significant differences test for all samples ($p < 0.05$). All analyses were performed using STATISTICA software, version 10.0 (StatSoft. Inc. 1984–2007, Tulsa, OK, USA).

3. Results

3.1. Identification and quantification of the phenolic content of mango (Mangifera indica L) peel and predigested mango peel

The compound identification was performed on basis of their relative retention time and mass spectra obtained using QToF-MS together with commercial standards and/or information previously reported in the literature (Table 1). Eighty-eight compounds were determined in the peel, and predigested and fermented mango peel samples. Specifically, sixty-nine compounds were present in the peel, forty-one in the predigested peel, and forty-five in the fermented mango peel. Many of the identified compounds showed a chemical structure belonging to gallates and gallotannins such as isomers of mono-, di-, tri-, tetra- and pentagalloylglucose, methyl- and ethyl-gallate, methyl-digallate ester, ethyltrigallate, theogallin, gallic acid and ethyl-2,4-dihydroxy-3-(3,4,5-trihydroxybenzoyl) oxybenzoate. Most of the gallates were absent in the predigested mango peel (Table 1). Gallic acid was the main compound identified in the predigested mango peel, reaching 50.50% of the gallate group, followed by galloyl-glucose (33.30%). Isomers of tri-*O*-galloyl-glucose were identified for the first time in mango peel, based on their pseudomolecular ion of *m/z* 635.0890 and fragments ion at *m/z* 483 and 169 corresponding to digalloyl-glucose and gallic acid, respectively (Table 1). Gallates and gallotannins are important groups of polyphenols present in both mango peel and predigested mango peel, which accounted for 22.75 and 18.21%, respectively, of the total quantified polyphenols (Table 2). Among this group of compounds, isomers of methyl-digallate ester (isomer III) were the most abundant compounds in mango peel, followed by penta-*O*-galloyl-glucose, tetra-*O*-galloyl-glucose, and galloyl-glucose. Except for gallic acid, the compounds detected in predigested mango significantly decreased their amount in comparison to the mango peel ($p < 0.05$).

Mango peel is a rich source of ellagic acid and valoneic acid dilactone, after the gastrointestinal digestion only ellagic acid remained although significantly lower ($p < 0.05$) than in mango peel, which represented 24.92% of the total polyphenols (Table 2).

The xanthone group is the most characteristic phenolic compounds present in mango peel, amounting to 8.55 and 43.87% of the total phenols quantified in mango peel and predigested mango peel, respectively. Headed by mangiferin, both samples also contain *iso*mangiferin and their galloylated and hydroxybenzoyl derivatives, as well as dehydroxymangiferin and methyl-mangiferin (homomangiferin) and mango peel, therefore, norathyriol (deglycosilated form of mangiferin). Mangiferin and homomangiferin significantly increased their concentration in predigested sample showing a contrary tendency to the rest of compounds, that significantly decreased their content ($p < 0.05$).

Benzophenone derivatives composed by glycosylated derivatives of maclurin, iriflophenone, in addition to their galloylated forms, represented 6.78% and 1.58% of total polyphenols identified in mango peel and predigested mango peel, respectively. Glycosidic derivatives of maclurin (91.00 and 93.11% of the total benzophenones in mango peel and predigested mango peel, respectively) were substantially more abundant than their homologue derivatives of iriflophenone. The predigested sample significantly showed lower amount of this group of compounds than mango peel ($p < 0.05$).

Flavonoids, headed by quercetin and its glycosidic derivatives (glucoside, diglucoside, galactoside, xyloside, arabinofuranoside, and rhamnoside), were present in the mango samples, accounting for 37.21% and 10.18% of the total polyphenols identified in both mango peel and predigested mango peel, respectively (Table 2). Both free quercetin, and to a greater extent its glycosylated forms, constituted the main flavonoids identified in mango peel, while no free quercetin was present in predigested mango peel, which was exclusively glycosylated. Quercetin-3-*O*-diglucoside, quercetin-3-*O*-galactoside, quercetin-3-*O*-glucoside, quercetin-xyloside, quercetin-arabinopyranoside and quercetin-*O*-rhamnoside accounted for 92.49 and 94.79% of the total flavonoids identified in mango peel and predigested mango peel, respectively, in significantly lower amount in the predigested sample than in un-predigested one ($p < 0.05$). A minor amount of catechin as well as epigallocatechin-gallate and epicatechin-gallate were also detected, which barely reached 2.42 and 5.21% of the total flavonoids identified in mango peel and predigested mango peel, respectively, with similar significant tendency than the rest of flavonoids ($p < 0.05$).

Finally, thirteen and eleven simple phenolic acids were also identified in both mango peel and the predigested sample, respectively. Particularly, glycosidic derivatives of dihydroxybenzoic, *p*-hydroxybenzoic, coumaric and ferulic acids were identified along with protocatechuic acid, 3- and 4-hydroxybenzoic acids, 3,4-dihydroxyphenylpropionic acid, 4-hydroxyphenylpropionic acid, 4-hydroxyphenylacetic acid, and hydroxycinnamic acids such as caffeic, ferulic and coumaric acids. Except *p*-hydroxybenzoic glucose and 4-hydroxyphenylpropionic acids which were absent in the digested mango peel, the rest of compounds were present in both samples with similar content, except for protocatechuic, 4-hydroxyphenylacetic and coumaric acids that were significantly more abundant in predigested mango ($p < 0.05$) (Table 2). While in mango peel, glycosidic derivatives represented 62.10% of the total phenolic acids identified, this amount decreased to 6.50% in in the predigested mango peel, where protocatechuic acid was the most abundant compound (65.02%) followed by 4-hydroxyphenylacetic acid (9.49%) and 3,4-dihydroxyphenylpropionic acid (5.95%). Phenolic content in pre-digested mango was less than 30% than compounds initially quantified in the peels prior to digestion (Table 2).

3.2. Identification and quantification of the phenolic compounds and metabolites of the fermented predigested mango peel

Polyphenol precursors and metabolites derived from *in vitro* microbial colonic fermentation of predigested mango peel are summarized in Table 3. Gallates and gallotannins detected in the predigested mango peel, only methyl- and ethyl-gallate were observed in the fermented samples, which significantly increased over fermentation time. Likewise, the content of gallic acid increased significantly over time, showing a direct relationship with incubation time ($p < 0.05$). No methyl- and ethyl-gallate, and very low amount of gallic acid were present at baseline before adding the predigested mango peel.

Ellagic acid was also identified in the fermented samples, with a discrete but statistically significant time-dependent relation (from 0.20 μmoles at 24 h up to 0.54 μmoles at 72 h). Related with this compound urolithins, specifically, urolithin A, *iso*urolithin A, urolithin C and *iso*-urolithin C, were detected. The amount of these compounds enhanced

Table 1
LC-QToF identification of phenolic compounds in mango peel, digested mango peel and fermented samples in a dynamic *in vitro* model of the human colon. Next to the identified compounds and between brackets the standard used for its quantification is indicated.

Name		RT (min)	Molecular formula	Molecular weight	$[M-H]^-$	MS^2 fragments	Location		
							Peel	Predigested sample	Fermented sample
GALLATES AND GALLOTANNINS									
Galloyl-glucose	(Gallic acid)	2.1	$C_{13}H_{16}O_{10}$	332.0743	331.0671	169	Yes	Yes	No
Galloyl-glucose	(Gallic acid)	2.3	$C_{13}H_{16}O_{10}$	332.0743	331.0671	169	Yes	Yes	No
Theogallin (3-galloylquinic acid)	(Gallic acid)	2.4	$C_{14}H_{16}O_{10}$	344.0743	343.0671	192; 91	Yes	Yes	No
Gallic acid	(Gallic acid)	2.6	$C_7H_6O_5$	170.0215	169.0142	125; 79; 69	Yes	Yes	Yes
Galloyl-glucose	(Gallic acid)	2.7	$C_{13}H_{16}O_{10}$	332.0743	331.0671	169	Yes	Yes	No
Digalloyl-glucose	(Gallic acid)	3.1	$C_{20}H_{20}O_{14}$	484.0853	483.0780	169; 125	Yes	No	No
Digalloylquinic acid	(Gallic acid)	3.8	$C_{21}H_{20}O_{14}$	496.0853	495.078	169	Yes	No	No
Digalloyl-glucose	(Gallic acid)	4.2	$C_{20}H_{20}O_{14}$	484.0853	483.0780	169; 125	Yes	No	No
Methyl-gallate	(Gallic acid)	6.3	$C_8H_8O_5$	184.0372	183.0299	124; 78	Yes	Yes	Yes
Trigalloyl-glucose	(Gallic acid)	7.6	$C_{27}H_{24}O_{18}$	636.0963	635.0890	483; 169	Yes	No	No
Trigalloyl-glucose	(Gallic acid)	7.9	$C_{27}H_{24}O_{18}$	636.0963	635.0890	483; 169	Yes	No	No
Trigalloyl-glucose	(Gallic acid)	8.2	$C_{27}H_{24}O_{18}$	636.0963	635.0890	483; 169	Yes	No	No
Trigalloyl-glucose	(Gallic acid)	8.4	$C_{27}H_{24}O_{18}$	636.0963	635.0890	483	Yes	No	No
Trigalloyl-glucose	(Gallic acid)	8.7	$C_{27}H_{24}O_{18}$	636.0963	635.0890	483	Yes	No	No
Tetragalloyl-glucose	(Gallic acid)	9.8	$C_{34}H_{28}O_{23}$	788.1012	787.0999	635; 617	Yes	No	No
Tetragalloyl-glucose	(Gallic acid)	10.1	$C_{34}H_{28}O_{23}$	788.1012	787.0999	635; 617	Yes	No	No
Ethyl-gallate	(Gallic acid)	10.2	$C_9H_{10}O_5$	198.0528	197.0455	169; 124	Yes	Yes	Yes
Tetragalloyl-glucose	(Gallic acid)	10.2	$C_{34}H_{28}O_{23}$	788.1012	787.0999	635; 617; 465	Yes	No	No
Tetragalloyl-glucose	(Gallic acid)	10.3	$C_{34}H_{28}O_{23}$	788.1012	787.0999	635; 617; 465	Yes	No	No
Methyl-digallate ester	(Gallic acid)	10.4	$C_{15}H_{12}O_9$	336.0481	335.0409	241; 183	Yes	No	No
Methyl-digallate ester	(Gallic acid)	10.9	$C_{15}H_{12}O_9$	336.0481	335.0409	183	Yes	No	No
Pentagalloyl-glucose	(Gallic acid)	11.3	$C_{41}H_{32}O_{26}$	940.1182	939.1109	787; 769; 617	Yes	No	No
Pentagalloyl-glucose	(Gallic acid)	11.5	$C_{41}H_{32}O_{26}$	940.1182	939.1109	787; 769; 617	Yes	No	No
Pentagalloyl-glucose	(Gallic acid)	11.6	$C_{41}H_{32}O_{26}$	940.1182	939.1109	787; 769; 617	Yes	No	No
Pentagalloyl-glucose	(Gallic acid)	11.8	$C_{41}H_{32}O_{26}$	940.1182	939.1109	787; 769	Yes	No	No
Methyl-digallate ester	(Gallic acid)	11.9	$C_{15}H_{12}O_9$	336.0481	335.0409	241; 183	Yes	No	No
Ethyl-2,4-dihydroxy-3-(3,4,5-trihydroxybenzoyl) oxybenzoate	(Gallic acid)	14.6	$C_{16}H_{14}O_9$	350.0638	349.0576	197; 169; 124	Yes	No	No
Ethyl-*p*-trigallate	(Gallic acid)	16.3	$C_{23}H_{18}O_{13}$	502.0747	501.0691	349; 197	Yes	No	No
ELLAGIC ACID AND RELATED COMPOUNDS									
Ellagic acid	(Ellagic acid)	10.6	$C_{14}H_6O_8$	302.0063	300.9990	300; 283; 145	Yes	Yes	Yes
Valoneic acid dilactone	(Ellagic acid)	11.3	$C_{21}H_{10}O_{13}$	470.0121	469.0524	301; 169	Yes	No	No
Urolithin C	(Urolithin C)	11.8	$C_{13}H_8O_5$	244.0372	243.0299		No	No	Yes
*Iso*urolithin C	(Urolithin C)	12.2	$C_{13}H_8O_5$	244.0372	243.0299		No	No	Yes
Urolithin A	(Urolithin A)	14.5	$C_{13}H_8O_4$	228.0423	227.0350		No	No	Yes
*Iso*urolithin A	(Urolithin A)	14.9	$C_{13}H_8O_4$	228.0423	227.0350		No	No	Yes
XANTHONES									
Mangiferin	(Mangiferin)	7.5	$C_{19}H_{18}O_{11}$	422.0849	421.0776	331; 301	Yes	Yes	Yes
*Iso*mangiferin	(Mangiferin)	8.0	$C_{19}H_{18}O_{11}$	422.0849	421.0776	331; 301	Yes	Yes	Yes
Homomangiferin	(Mangiferin)	8.6	$C_{20}H_{20}O_{11}$	436.1006	435.0933	345; 331; 315	Yes	Yes	Yes
6-Galloyl-mangiferin	(Mangiferin)	9.3	$C_{26}H_{22}O_{15}$	574.0959	573.0886	421; 403; 331; 301	Yes	Yes	Yes
*Iso*mangiferin gallate	(Mangiferin)	9.8	$C_{26}H_{22}O_{15}$	574.0959	573.0886	421; 403; 331; 301	Yes	Yes	Yes
Dehydroxymangiferin	(Mangiferin)	11.2	$C_{19}H_{18}O_{10}$	406.0900	405.0827		Yes	Yes	Yes
6-*O*-(*p*-Hydroxybenzoyl) mangiferin (Isomer I)	(Mangiferin)	11.5	$C_{26}H_{22}O_{13}$	542.1060	541.0988	403; 331; 301	Yes	Yes	Yes
6-*O*-(*p*-Hydroxybenzoyl) mangiferin (Isomer II)	(Mangiferin)	11.7	$C_{26}H_{22}O_{13}$	542.1060	541.0988	403; 331; 301	Yes	Yes	Yes
Norathyriol	(Mangiferin)	14.7	$C_{13}H_8O_6$	260.0321	259.0248		Yes	No	Yes
Euxanthone	(Mangiferin)	14.9	$C_{13}H_8O_4$	228.0423	227.0350		No	No	Yes
Methyl-norathyriol	(Mangiferin)	20.9	$C_{14}H_{10}O_6$	274.0477	273.0405		No	No	Yes
BENZOPHENONE DERIVATIVES AND RELATED COMPOUNDS									
Maclurin-3-C-β-D-glucoside	(Maclurin)	3.2	$C_{19}H_{20}O_{11}$	424.1006	423.0933	333; 303; 223; 193	Yes	Yes	Yes
Maclurin-3-C-(2-*O*-galloyl)-β-D-glucoside	(Maclurin)	4.8	$C_{26}H_{24}O_{15}$	576.1115	575.1042	557; 465; 333; 303	Yes	Yes	Yes
Iriflophenone-3-C-β-D-glucoside	(Maclurin)	5.2	$C_{19}H_{20}O_{10}$	408.1056	407.0984	317; 287	Yes	Yes	Yes
	(Maclurin)	7.6	$C_{26}H_{24}O_{14}$	560.1166	559.1093		Yes	Yes	Yes

(*continued on next page*)

Table 1 (*continued*)

Name		RT (min)	Molecular formula	Molecular weight	$[M-H]^-$	MS^2 fragments	Location		
							Peel	Predigested sample	Fermented sample
Iriflophenone-3-*C*-(2-*O*-galloyl)-β-*D*-glucoside						407; 389; 317; 287; 269; 245; 169			
Maclurin-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside	(Maclurin)	7.8	$C_{33}H_{28}O_{19}$	728.1225	727.1152	575; 333; 303	Yes	No	No
Iriflophenone-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside	(Maclurin)	9.7	$C_{33}H_{28}O_{18}$	712.1276	711.1203	559; 541; 317; 287; 271	Yes	Yes	No
Maclurin	(Maclurin)	8.1	$C_{13}H_{10}O_6$	262.0477	261.0405		No	No	Yes
Iriflophenone	(Maclurin)	14.0	$C_{13}H_{10}O_5$	246.0528	245.0455		No	No	Yes
FLAVONOIDS									
Catechin	(Catechin)	5.7	$C_{15}H_{14}O_6$	290.0790	289.0718	245	Yes	Yes	Yes
Epigallocatechin-gallate	(Epicatechin)	6.1	$C_{22}H_{18}O_{11}$	458.0849	457.0776	169	Yes	Yes	No
Epicatechin	(Epicatechin)	7.8	$C_{15}H_{14}O_6$	290.0790	289.0718	245; 137	No	No	Yes
Quercetin-3-*O*-diglucoside	(Quercetin)	9.6	$C_{26}H_{28}O_{16}$	596.1377	595.1305	301; 300	Yes	Yes	No
Quercetin-3-*O*-glucoside (*iso*quercitrin)	(Quercetin)	10.8	$C_{21}H_{20}O_{12}$	464.0955	463.0882	301; 300; 179; 151	Yes	yes	No
Epicatechin-gallate	(Epicatechin)	10.9	$C_{22}H_{18}O_{10}$	442.0900	441.0827	169	Yes	Yes	No
Quercetin-3-*O*-galactoside	(Quercetin)	11.1	$C_{21}H_{20}O_{12}$	464.0955	463.0882	301; 300	Yes	Yes	No
Quercetin-xyloside	(Quercetin)	11.6	$C_{20}H_{18}O_{11}$	434.0849	433.0776	301; 300	Yes	Yes	No
Quercetin-arabinopyranoside	(Quercetin)	11.8	$C_{20}H_{18}O_{11}$	434.0849	433.0776	301; 300	Yes	Yes	No
Quercetin-arabinofuranoside	(Quercetin)	12.1	$C_{20}H_{18}O_{11}$	434.0849	433.0776	301; 300	Yes	Yes	No
Quercetin-*O*-ramnoside	(Quercetin)	12.4	$C_{21}H_{20}O_{11}$	448.1006	447.0933	301; 300	Yes	Yes	No
Rhamnetin-hexoside	(Quercetin)	15.6	$C_{22}H_{22}O_{12}$	478.1111	477.1038	315; 300	Yes	No	No
Quercetin	(Quercetin)	16.1	$C_{15}H_{10}O_7$	302.0427	301.0354	151	Yes	No	No
OTHER PHENOLIC ACIDS AND RELATED COMPOUNDS									
Dihydroxybenzoic acid glucose	(Protocatechuic acid)	3.0	$C_{13}H_{16}O_9$	316.0794	315.0722	153; 109	Yes	Yes	No
Pyrogallol	(Pyrogallol)	3.2	$C_6H_6O_3$	126.0317	125.0244	51	No	No	Yes
p-Hydroxybenzoic acid glucose	(4-Hydrozybenzoic acid)	3.4	$C_{13}H_{16}O_8$	300.0845	299.0772	137	Yes	No	No
3,4-Dihydroxybenzoic acid (protocatechuic acid)	(Protocatechuic acid)	3.8	$C_7H_6O_4$	154.0266	153.0193	109	Yes	Yes	Yes
3-Methoxy-4-hydroxybenzoic acid (vanillic acid)	(Vanillic acid)	4.3	$C_8H_8O_4$	168.0423	167.0350	123	No	No	Yes
3,4-Dihydroxyphenylacetic acid	(3,4-Dihydroxyphenylacetic acid)	4.4	$C_8H_8O_4$	168.0423	167.0350	123; 108	No	No	Yes
4-Hydroxy-5-(3′,4′-dihydroxyphenyl)valeric acid	(3,4-Dihydroxyphenylpropionic acid)	5.1	$C_{11}H_{14}O_5$	226.0841	225.0768		No	No	Yes
3-Hydroxybenzoic acid	(3-Hydroxybenzoic acid)	5.8	$C_7H_6O_3$	138.0317	137.0244	93	Yes	Yes	Yes
4-Hydroxybenzoic acid	(4-Hydroxybenzoic acid)	6.0	$C_7H_6O_3$	138.0317	137.0244	93	Yes	Yes	Yes
Coumaric acid glucoside	(Coumaric acid)	6.1	$C_{15}H_{18}O_8$	326.1002	325.0929	163; 119	Yes	Yes	No
3,4-Dihydroxyphenylpropionic acid	(3,4-Dihydroxyphenylpropionic acid)	6.6	$C_9H_{10}O_4$	182.0579	181.0506	137	Yes	Yes	Yes
4-Hydroxyphenylacetic acid	(4-Hydroxyphenylacetic acid)	6.9	$C_8H_8O_3$	152.0473	151.0401	107	Yes	Yes	Yes
Caffeic acid	(Caffeic acid)	7.2	$C_9H_8O_4$	180.0423	179.0350		Yes	Yes	Yes
Ferulic acid hexoside	(Ferulic acid)	7.4	$C_{16}H_{20}O_9$	356.1107	355.1035	193; 149; 134	Yes	Yes	No
3-Methoxy-4-hydroxyphenylacetic acid	(3-Methoxy-4-hydroxyphenylacetic acid)	8.8	$C_9H_{10}O_4$	182.0579	181.0506	137	No	No	Yes
5-(3′,4′-Dihydroxyphenyl)-γ-valerolactone	(Epicatechin)	9.2	$C_{11}H_{12}O_4$	208.0736	207.0663	163	No	No	Yes
Coumaric acid	(Coumaric acid)	9.4	$C_9H_8O_3$	164.0473	163.0401	119	Yes	Yes	Yes
3-Hydroxyphenylpropionic acid	(3-Hydroxyphenylpropionic acid)	9.4	$C_9H_{10}O_3$	166.0630	165.0557	121	No	No	Yes
3-Hydroxyphenylacetic acid	(3-Hydroxyphenylacetic acid)	9.4	$C_8H_8O_3$	152.0473	151.0401	107; 121	No	No	Yes
Methoxy-hydroxyphenylpropionic acid	(Methoxy-hydroxyphenylpropionic acid)	10.5	$C_{10}H_{12}O_4$	196.0736	195.0663	136	No	No	Yes
4-Hydroxyphenylpropionic acid	(4-Hydroxyphenylpropionic acid)	10.7	$C_9H_{10}O_3$	166.0630	165.0557	121	Yes	No	Yes
Ferulic acid	(Ferulic acid)	11.2	$C_{10}H_{10}O_4$	194.0579	193.0506	149; 134	No	Yes	No

significantly over time, being particularly important the amount of *iso*-urolithin A (4.44 μmoles) and *iso*urolithin C (21.59 μmoles) after 3 days of colonic fermentation.

Regarding xanthones, all the compounds present in the predigested mango peel were also identified in the fermented samples, showing a concentration-dependent relationship with fermentation time ($p < 0.05$). Additionally, norathyriol, euxanthone and methyl-norathyriol were detected in the fermented samples, and these significantly ascended over time. Norathyriol, along with mangiferin and homo-mangiferin, were the most abundant of all the identified xanthones,

Table 2
Content of individual phenolic compounds present in mango peel and digested mango peel. Results represent the mean ± standard deviation (n = 2). N.D.: not detected; d.w.: dry weight. Means with the same letter are not significantly different from each other in the same line ($p < 0.05$).

RT (min)	Proposed compound	Mango peel (mg/100 g d.w.)	Predigested mango peel (mg/100 g d.w.)
GALLATES AND GALLOTANNINS			
2.1	Galloyl glucose	2.96 ± 0.04^{a}	47.28 ± 3.90^{b}
2.3	Galloyl glucose	35.33 ± 0.94^{b}	2.06 ± 0.49^{a}
2.4	Theogallin	3.82 ± 0.04^{b}	7.04 ± 0.23^{a}
2.6	Gallic acid	13.83 ± 0.04^{a}	71.72 ± 0.80^{b}
2.7	Galloyl glucose	11.31 ± 0.43^{b}	3.06 ± 0.29^{a}
3.1	Digalloyl glucose	13.72 ± 1.55	N.D.
3.8	Digalloylquinic acid	5.74 ± 0.14	N.D.
4.2	Digalloyl glucose	5.78 ± 0.52	N.D.
6.3	Methyl-gallate	18.79 ± 0.17^{b}	7.08 ± 0.69^{a}
7.6	Trigalloyl glucose (Isomer I)	10.86 ± 0.62	N.D.
7.9	Trigalloyl glucose (Isomer II)	3.73 ± 0.01	N.D.
8.2	Trigalloyl glucose (Isomer III)	5.24 ± 0.41	N.D.
8.4	Trigalloyl glucose (Isomer IV)	3.69 ± 0.09	N.D.
8.7	Trigalloyl glucose (Isomer V)	2.41 ± 0.09	N.D.
9.8	Tetra-*O*-galloyl glucose (Isomer I)	21.94 ± 0.19	N.D.
10.1	Tetra-*O*-galloyl glucose (Isomer II)	9.10 ± 0.09	N.D.
10.2	Ethyl-gallate	1.09 ± 0.05^{a}	3.76 ± 0.15^{b}
10.2	Tetra-*O*-galloyl glucose (Isomer III)	61.10 ± 1.09	N.D.
10.3	Tetra-*O*-galloyl glucose (Isomer IV)	23.61 ± 0.84	N.D.
10.4	Methyl-digallate ester (Isomer I)	9.34 ± 0.01	N.D.
10.9	Methyl-digallate ester (Isomer II)	28.93 ± 0.62	N.D.
11.3	Pentagalloyl glucose	127.64 ± 1.89	N.D.
11.5	Pentagalloyl glucose	9.57 ± 1.29	N.D.
11.6	Pentagalloyl glucose	29.31 ± 0.04	N.D.
11.8	Pentagalloyl glucose	12.18 ± 0.15	N.D.
11.9	Methyl-digallate ester (Isomer III)	149.14 ± 2.48	N.D.
14.6	Ethyl-2,4-dihydroxy-3-(3,4,5-trihydroxybenzoyl)oxybenzoate	2.05 ± 0.14	N.D.
16.3	Ethyl-*p*-trigallate	1.38 ± 0.01	N.D.
TOTAL (mg/100 g) (%)		*623.59 ± 13.98^{b} (22.75 %)*	*142.01 ± 6.55^{a} (18.21 %)*
ELLAGIC ACID AND RELATED COMPOUNDS			
10.6	Ellagic acid	130.83 ± 3.63^{a}	194.36 ± 4.49^{b}
11.3	Valoneic acid dilactone	508.60 ± 9.08	N.D.
TOTAL (mg/100 g) (%)		*639.43 ± 12.71^{b} (23.33%)*	*194.36 ± 4.49^{a} (24.92%)*
XANTHONES			
7.5	Mangiferin	147.60 ± 1.71^{a}	259.54 ± 0.75^{b}
8.0	*Iso*mangiferin	26.60 ± 1.13^{b}	10.80 ± 0.19^{a}
8.6	Homomangiferin	33.92 ± 0.36^{a}	61.29 ± 0.03^{b}
9.3	6-Galloyl-mangiferin	8.56 ± 0.19^{b}	0.16 ± 0.01^{a}
9.8	*Iso*mangiferin gallate	13.91 ± 0.14^{b}	0.36 ± 0.02^{a}
11.2	Dehydroxymangiferin	0.15 ± 0.01^{b}	0.07 ± 0.01^{a}
11.5	6-*O*-(*p*-Hydroxybenzoyl)mangiferin	3.08 ± 0.01^{a}	7.85 ± 0.12^{b}
11.7	6-*O*-(*p*-Hydroxybenzoyl)mangiferin	1.02 ± 0.04^{a}	2.00 ± 0.14^{b}
14.7	Norathyriol	0.10 ± 0.01	N.D.
TOTAL (mg/100 g) (%)		*234.36 ± 3.60^{a} (8.55%)*	*342.06 ± 1.27^{b} (43.87%)*
BENZOPHENONE DERIVATIVES			
3.2	Maclurin-3-*C*-β-*D*-glucoside	14.78 ± 0.06^{b}	1.33 ± 0.33^{a}
4.8	Maclurin-3-C-(2-*O*-galloyl)-β-D-glucoside	75.75 ± 1.24^{b}	10.09 ± 0.09^{a}
5.2	Iriflophenone-3-C-β-D-glucose	0.24 ± 0.01^{a}	0.27 ± 0.01^{b}
7.6	Iriflophenone-3-*C*-(2-*O*-galloyl)-β-*D*-glucoside	8.79 ± 0.53^{b}	0.33 ± 0.09^{a}
7.8	Maclurin-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside	78.61 ± 0.70	N.D.
9.7	Iriflophenone-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside	7.68 ± 0.11^{b}	0.25 ± 0.01^{a}
TOTAL (mg/100 g) (%)		*185.86 ± 2.65^{b} (6.78%)*	*12.26 ± 0.53^{a} (1.57%)*
FLAVONOIDS			
5.7	Catechin	0.83 ± 0.04^{a}	0.68 ± 0.17^{a}
6.1	Epigallocatechin-gallate	0.85 ± 0.01	N.D.
9.6	Quercetin-3-*O*-diglucoside	42.19 ± 1.18^{b}	4.97 ± 0.98^{a}
10.8	Quercetin-3-*O*-glucoside (isoquercitrin)	312.15 ± 5.01^{b}	30.22 ± 3.35^{a}
10.9	Epicatechin-gallate	23.03 ± 0.21^{b}	3.46 ± 0.28^{a}
11.1	Quercetin-3-*O*-galactoside	325.09 ± 8.60^{b}	36.66 ± 19.47^{a}
11.6	Quercetin-xyloside	136.47 ± 1.89^{b}	0.57 ± 0.05^{a}
11.8	Quercetin-arabinopyranoside	78.14 ± 3.77^{b}	0.83 ± 0.35^{a}
12.1	Quercetin-arabinofuranoside	28.58 ± 0.02^{b}	0.21 ± 0.04^{a}
12.4	Quercetin-*O*-ramnoside (quercitrin)	20.67 ± 0.06^{b}	1.80 ± 0.03^{a}
15.6	Rhamnetin-hexoside	3.33 ± 0.11	N.D.
16.1	Quercetin	48.56 ± 2.11	N.D.
TOTAL (mg/100 g) (%)		*1019.90 ± 23.01^{b} (37.21%)*	*79.38 ± 24.71^{a} (10.18)*
PHENOLIC ACIDS			
3.0	Dihydroxybenzoic acid glucose	10.71 ± 0.45^{b}	0.20 ± 0.14^{a}
3.4	*p*-Hydroxybenzoic acid glucose	3.88 ± 0.13	N.D.

(*continued on next page*)

Table 2 (*continued*)

RT (min)	Proposed compound	Mango peel (mg/100 g d.w.)	Predigested mango peel (mg/100 g d.w.)
3.8	3,4-Dihydroxybenzoic acid (protocatechuic acid)	0.64 ± 0.05^{a}	6.32 ± 0.59^{b}
5.8	3-Hydroxybenzoic acid	0.25 ± 0.03^{a}	0.28 ± 0.09^{a}
6.0	4-Hydroxybenzoic acid	0.19 ± 0.01^{a}	0.32 ± 0.05^{a}
6.1	Coumaric acid glucoside	8.28 ± 0.05^{b}	0.39 ± 0.01^{a}
6.6	3,4-Dihydroxyphenylpropionic acid	0.68 ± 0.03^{b}	0.58 ± 0.08^{a}
6.9	4-Hydroxyphenylacetic acid	0.57 ± 0.01^{a}	0.92 ± 0.01^{b}
7.2	Caffeic acid	0.20 ± 0.01^{a}	0.29 ± 0.01^{b}
7.4	Ferulic acid hexoside	0.57 ± 0.01^{b}	0.04 ± 0.01^{a}
9.4	Coumaric acid	0.07 ± 0.01^{a}	0.25 ± 0.01^{b}
10.7	4-Hydroxyphenylpropionic acid	11.72 ± 0.30	N.D.
11.2	Ferulic acid	N.D.	0.14 ± 0.01
TOTAL (mg/100 g) (%)		*37.75 ± 1.08^{b} (1.38%)*	*9.72 ± 1.03^{a} (1.25%)*
TOTAL	**PHENOLIC COMPOUNDS (mg/100 g) (%)**	**2740.90 ± 57.01^{b} (100%)**	**779.80 ± 38.58^{a} (100%)**

reaching values of 19.60, 17.72 and 13.46, μmoles, respectively, after 72 h of colonic fermentation.

Benzophenone derivatives were another important group of phenolic compounds that resisted colonic fermentation. Except maclurin-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside and iriflophenone-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside, which were absent in fermented samples, the rest of the compounds identified in the predigested mango peel were identified after 72 h of fermentation, showing a significantly direct relationship between their concentration and fermentation time. Glycosylated derivatives of maclurin (0.20–0.60 μmoles) were more abundant than the equivalent forms of iriflophenone (0.005–0.035 μmoles) at 72 h. Apart from these compounds, maclurin and iriflophenone, which were absent in the predigested mango peel, were identified and quantified in the fermented samples, reaching final concentrations up to 0.50 μmoles and 0.96 μmoles at 72 h, respectively.

Regarding flavonoids, glycosilated derivatives of quercetin were not detected in the fermented samples. Only catechin and epicatechin were identified in the fermented samples although their concentrations were very low and did not show changes over time.

Finally, phenolic acids and related compounds were detected in the fermented samples, showing a significantly direct relation between the amount formed and fermentation time ($p < 0.05$). 5-(3′,4′-dihydroxyphenyl)-γ-valerolactone (DHPVL) and 4-hydroxy-5-(3′,4′-dihydroxyphenyl)valeric acid (DHPVA) have been identified in the fermented samples, with a significantly direct relationship between their concentration and incubation time (from 2.60 μmoles up to 6.32 μmoles and from 0.41 μmoles up to 1.14 μmoles for DHPVL and DHPVA, respectively). Moreover, derivatives of hydroxyphenylpropionic, hydroxyphenylacetic, and hydroxybenzoic were also identified at 72 h, being 4-hydroxyphenylpropionic acid (236.67 μmoles) the most abundant of all the metabolites, followed by 3-hydroxyphenylpropionic acid (99.87 μmoles), 3,4-dihydrophenylacetic acid (20.58 μmoles), 3-hydroxyphenylacetic acid (19.83 μmoles) and 4-hydroxyphenylacetic acid (17.47 μmoles) among others (Table 3). Finally, pyrogallol, absent in the predigested mango peel (Tables 1 and 2), was formed after three days colonic fermentation, reaching 32.34 μmoles.

In summary, the metabolites identified amounted up to 33.14%, 40.48% and 47.52% of the colonic fermented mango peel polyphenols after 24 h, 48 h and 72 h, respectively.

4. Discussion

Mango byproducts are a relevant source of phenolic compounds which have already shown to display a multitude of health beneficial properties (Sáyago-Ayerdi et al, 2019; Dorta et al, 2014). However, it should not be disregarded that the biological activity of a compound is partly determined by its chemical stability in the gastrointestinal tract. Therefore, evaluating digestive stability is the first step in estimating the amount of a phytochemical that might be available for absorption in the intestine and/or later bio-transformation in the colon.

In vitro digestion models represent a valid approach to understand potential interactions in the food matrix (dietary fiber and bioactive compounds) (Bohn, 2014). In order to evaluate the stability of the main polyphenols in mango peel during simulated digestion, a double incubation with pepsin and pancreatin, the main enzymes involved in human gastric and intestinal digestions, respectively, was performed. Gallotannins and gallates were present in relatively large amounts in mango peel (22.75% of the total polyphenols quantified), mainly gallotannins such as isomers of mono-, di-, tri-, tetra- and pentagalloylglucose, theogallin (3-galloylquinic acid) and digalloylquinic acid, in addition to gallates such as methyl- and ethyl-gallate, methyl-digallate ester, ethyl-trigallate and ethyl-2,4-dihydroxy-3-(3,4,5-trihydroxybenzoyl) oxybenzoate. Minor amounts of free gallic acid were also present in mango peel (Table 2). Most of gallotannins and gallates were hydrolyzed by digestive enzymes during mango peel digestion, leaving only galloyl glucose, theogallin, methyl- and ethyl-gallate, in addition to gallic acid, which reached up to 50.50% of the phenolic compounds of this group, compared to 2.2% of gallic acid present in mango peel. As far, this is the first time isomers of tri-pentagalloylglucose have been identified in mango peel based on their quasi-molecular ion at *m/z* 635.0890 and fragment ions at *m/z* 483 and 169 corresponding to digalloyl-glucose and gallic acid, respectively. However, Sáyago-Ayerdi et al. (2013) were able to identify penta- to trideca-*O*-galloylglucoside in mango peel by MALDI-TOF MS.

Mango peel is a rich source of ellagic acid and valoneic acid dilactone, both adding up to 23.33% of the total polyphenols identified, of which only ellagic acid was detected after gastrointestinal digestion in the predigested mango peel (24.92%). Hydrolysis of both ellagic acid and valoneic acid dilactone contributed to the increased gallic acid content, and their oxidation to quinone derivatives might also justify the loss of these compounds after gastrointestinal digestion.

Mango contains a characteristic polyphenol, mangiferin, which represented the 63.0% and 75.9% of the total xanthones quantified in mango peel and predigested mango peel, respectively. The higher rate of mangiferin in the predigested mango peel compared to mango peel is a consequence of the partial hydrolysis of galloyl- and *p*-hydroxybenzoyl derivatives during gastrointestinal digestion. Xanthones were the most stable compounds of all the identified phenolics, and the percentage of this group in the digested mango peel increased from 8.55% up to 43.87%, at the expense of other polyphenols such as benzophenones and flavonoids, as detailed below.

The presence of several galloylated and galloyl-glycosylated derivatives of benzophenone has already been reported in mango peel (López-Cobo et al., 2017, among others). Maclurin-3-*C*-(2,3-di-*O*-galloyl)-β-*D*-glucoside, maclurin-3-*C*-(2-*O*-galloyl)-β-*D*-glucoside and maclurin 3-*C*-β-*D*–glucoside summed up to more than 90% of benzophenones in both samples. This group of compounds resulted to be unstable since the percentage of their content decreased in the digested mango peel down to 1.57% of the total polyphenols, compared to 6.78% in the original mango peel.

Table 3
Content of individual phenolic compounds present in the fermented mango peel in a dynamic *in vitro* model of the human colon from 0 to 72 h. Results represent the mean ± standard deviation (n = 3). N.D.: not detected. Means with the same letter are not significantly different from each other in the same line ($p < 0.05$).

No	Name	RT (min)	0 h (μmoles)	24 h (μmoles)	48 h (μmoles)	72 h (μmoles)
GALLATES AND GALLOTANINS						
1	Gallic acid	2.6	0.010 ± 0.001^{a}	5.148 ± 0.600^{b}	7.338 ± 0.505^{c}	10.214 ± 0.476^{d}
2	Methyl-gallate	6.3	N.D.	0.208 ± 0.030^{a}	0.343 ± 0.052^{b}	0.323 ± 0.064^{b}
3	Ethyl-gallate	10.2	N.D.	0.006 ± 0.001^{a}	0.038 ± 0.024^{b}	0.112 ± 0.078^{c}
ELLAGIC ACID AND RELATED COMPOUNDS						
4	Ellagic acid	10.6	N.D.	0.196 ± 0.023^{a}	0.299 ± 0.031^{b}	0.543 ± 0.005^{c}
5	Urolithin C	11.8	N.D.	0.003 ± 0.001^{a}	0.121 ± 0.013^{b}	0.338 ± 0.098^{c}
6	*Iso*urolithin C	12.2	N.D.	1.816 ± 0.283^{a}	8.028 ± 1.796^{b}	21.596 ± 1.957^{c}
7	Urolithin A	14.5	N.D.	0.014 ± 0.002^{b}	0.096 ± 0.006^{a}	0.166 ± 0.127^{a}
8	*Iso*urolithin A	14.9	N.D.	0.990 ± 0.183^{a}	3.215 ± 0.685^{b}	5.476 ± 1.055^{c}
XANTHONES						
9	Mangiferin	7.5	N.D.	11.199 ± 1.143^{a}	15.726 ± 0.103^{b}	17.721 ± 0.476^{c}
10	*Iso*mangiferin	8.0	N.D.	0.411 ± 0.028^{a}	0.601 ± 0.025^{b}	0.727 ± 0.043^{c}
11	Homomangiferin	8.6	N.D.	2.497 ± 0.330^{a}	7.483 ± 0.621^{b}	13.459 ± 1.143^{c}
12	6-Galloyl-mangiferin	9.3	N.D.	0.042 ± 0.005^{a}	0.099 ± 0.005^{b}	0.138 ± 0.011^{c}
13	*Iso*mangiferin gallate	9.8	N.D.	0.021 ± 0.005^{a}	0.057 ± 0.011^{b}	0.096 ± 0.014^{c}
14	Dehydroxymangiferin	11.2	N.D.	0.005 ± 0.001^{a}	0.014 ± 0.003^{b}	0.025 ± 0.004^{c}
15	6-*O*-(p-Hydroxybenzoyl)mangiferin (Isomer I)	11.5	N.D.	0.016 ± 0.006^{b}	0.034 ± 0.010^{a}	0.048 ± 0.012^{a}
16	6-*O*-(p-Hydroxybenzoyl)mangiferin (Isomer II)	11.7	N.D.	0.021 ± 0.004^{a}	0.074 ± 0.016^{b}	0.145 ± 0.038^{c}
17	Norathyriol	14.7	N.D.	1.995 ± 0.551^{a}	11.999 ± 1.042^{b}	19.598 ± 1.999^{c}
18	Euxanthone	14.9	N.D.	0.027 ± 0.001^{a}	0.101 ± 0.014^{b}	0.175 ± 0.018^{c}
19	Methyl-norathyriol	20.9	N.D.	0.023 ± 0.002^{a}	0.057 ± 0.004^{b}	0.117 ± 0.016^{c}
BENZOPHENONE DERIVATIVES						
20	Maclurin 3-C-β-D-glucoside	3.2	N.D.	0.085 ± 0.020^{a}	0.135 ± 0.029^{a}	0.201 ± 0.029^{b}
21	Maclurin-3-C-(2-*O*-galloyl)-β-D-glucoside	4.8	N.D.	0.323 ± 0.111^{a}	0.466 ± 0.111^{ab}	0.597 ± 0.121^{b}
22	Iriflophenone-3-C-β-D-glucoside (Isomer II)	5.2	N.D.	0.006 ± 0.001^{a}	0.009 ± 0.001^{a}	0.009 ± 0.005^{a}
23	Iriflophenone-3-*C*-(2-*O*-galloyl)-β-*D*-glucoside	7.6	N.D.	0.020 ± 0.006^{a}	0.027 ± 0.005^{ab}	0.035 ± 0.005^{b}
24	Maclurin	8.1	N.D.	0.064 ± 0.011^{a}	0.293 ± 0.017^{b}	0.503 ± 0.017^{c}
25	Iriflophenone	14.0	N.D.	0.041 ± 0.019^{a}	0.335 ± 0.117^{b}	0.962 ± 0.208^{c}
FLAVONOIDS						
26	Catechin	5.7	N.D.	0.011 ± 0.004^{a}	0.011 ± 0.002^{a}	0.010 ± 0.001^{a}
27	Epicatechin	7.8	N.D.	0.033 ± 0.009^{b}	0.055 ± 0.008^{a}	0.059 ± 0.004^{a}
PHENOLIC ACIDS AND RELATED COMPOUND						
28	Pyrogallol	3.2	0.050 ± 0.001^{a}	16.593 ± 3.494^{c}	27.922 ± 5.044^{b}	32.343 ± 4.389^{b}
29	3,4-Dihydroxybenzoic acid (protocatechuic acid)	3.8	N.D.	0.366 ± 0.016^{a}	0.704 ± 0.050^{b}	1.300 ± 0.141^{c}
30	3-Methoxy-4-hydroxybenzoic acid (vanillic acid)	4.3	N.D.	0.673 ± 0.108^{a}	1.702 ± 0.200^{b}	2.658 ± 0.293^{c}
31	3,4-Dihydroxyphenylacetic acid	4.4	N.D.	5.810 ± 0.590^{a}	13.308 ± 1.046^{b}	20.577 ± 1.650^{c}
32	4-Hydroxy-5-(3′,4′-dihydroxyphenyl)valeric acid	5.1	0.050 ± 0.001^{a}	0.409 ± 0.101^{a}	0.762 ± 0.193^{ab}	1.144 ± 0.199^{b}
33	3-Hydroxybenzoic acid	5.8	N.D.	0.023 ± 0.002^{a}	0.057 ± 0.004^{b}	0.117 ± 0.016^{c}
34	4-Hydroxybenzoic acid	6.0	N.D.	3.796 ± 0.195^{a}	7.910 ± 0.166^{b}	11.566 ± 0.297^{a}
35	3,4-Dihydroxyphenylpropionic acid	6.6	0.097 ± 0.005^{a}	0.133 ± 0.039^{b}	0.456 ± 0.028^{c}	0.604 ± 0.009^{d}
36	4-Hydroxyphenylacetic acid	6.9	N.D.	4.300 ± 1.330^{a}	10.656 ± 2.388^{b}	17.469 ± 1.042^{c}
37	Caffeic acid	7.2	0.029 ± 0.002	Traces	Traces	Traces
38	3-Methoxy-4-hydroxyphenylacetic acid	8.8	N.D.	0.452 ± 0.193^{b}	1.913 ± 0.701^{a}	2.471 ± 0.903^{a}
39	5-(3′,4′-Dihydroxyphenyl)-γ-valerolactone	9.2	0.347 ± 0.001^{a}	2.595 ± 0.455^{b}	4.691 ± 0.880^{c}	6.322 ± 0.911^{d}
40	3-Hydroxyphenylacetic acid	9.4	0.122 ± 0.030^{a}	4.918 ± 1.303^{b}	13.823 ± 2.135^{c}	19.827 ± 1.084^{d}
41	3-Hydroxyphenylpropionic	9.4	N.D.	8.413 ± 0.773^{a}	40.845 ± 3.527^{b}	99.868 ± 14.412^{c}
42	Coumaric acid	9.4	N.D.	0.009 ± 0.001^{b}	0.018 ± 0.001^{a}	0.017 ± 0.003^{a}
43	3-Methoxy-4-hydroxyphenylpropionic acid	10.5	1.684 ± 0.210^{a}	3.495 ± 0.120^{b}	7.168 ± 0.253^{c}	10.735 ± 0.322^{d}
44	4-Hydroxyphenylpropionic acid	10.7	14.95 4 ± 0.120^{a}	52.252 ± 6.675^{b}	127.357 ± 4.693^{c}	236.676 ± 18.326^{d}
45	Ferulic acid	11.2	N.D.	Traces	Traces	Traces
TOTAL (μmoles)			**17.333 ± 0.370^{a}**	**129.252 ± 19.295^{b}**	**315.751 ± 27.187^{c}**	**556.043 ± 53.232^{d}**

Flavonoids is another important group of phenolic compounds present in mango peel, mainly as quercetin glycosides, which accounted for 37.21% of the total polyphenols. Most mango flavonoids disappeared after gastrointestinal digestion, leaving only 10.18% of the total polyphenols in the predigested mango peel. Interestingly, quercetin was not detected in the digested sample, so either the quercetin glycosides had been oxidized to quinones due to the alkaline media during gastrointestinal digestion, or they had been hydrolyzed to generate quercetin and subsequently oxidized to form quinone derivatives.

Finally, the least abundant phenolic group identified were simple phenolic acids, which represented 1.38 and 1.25% of all the polyphenols quantified in mango peel and predigested mango peel, respectively. It is important to mention that most of the glycosidic derivatives of phenolic acids, such as dihydroxybenzoic acid glucose, *p*-hydroxybenzoic acid glucose, coumaric acid glucose and ferulic acid hexoside, almost disappeared during gastrointestinal digestion in favor of free simple phenols, such as protocatechuic acid or ferulic acid, among others.

In the present work, mango peel contained 2741 mg of phenolic compounds in 100 g dry matter. This result was in agreement with data reported by Barreto et al., (2008), who detailed profiles and amounts for the cultivars 'Van Dyke' and 'Embrapa-141-Roxa' ranging from 2424 to 5907 mg/100 g dry matter in mango peels. Recently, López-Cobo et al. (2017) showed that phenolic compounds in mango peel of three cultivars ('Sensación', 'Osteen', and 'Keitt') were in the range of 1811.43–2476.08 mg/100 g d.w. in 'Keitt' and 'Sensación', respectively). Barreto et al. (2008) described that penta-*O*-galloyl-glucoside was the most abundant compound in mango peel, followed by methyl-digallate ester and mangiferin. López-Cobo et al. (2017) also reported

that gallotannins were the most abundant group followed by flavonoids, in agreement with the results presented in this study. Regarding phenolic compounds present in mango peel, only 28.5% was available for the fermentation process, highlighting xanthones as the most abundant group (43.87% of the total polyphenols).

A novel aspect of this study is the evaluation of the effects of human microbiota on polyphenols in mango peel, in order to further understand the microbial-derived metabolites responsible for the beneficial effects of mango consumption, or at least in part. The generation of phenolic catabolites and/or metabolites was monitored at different incubation times: 24, 48, and 72 h. It is important to remark that TIM-2 was continuously fed with the pre-digested mango peel over 72 h at a rate of 2.5 mL/min to mimic a physiological situation. Regarding gallates and gallotannins, which accounted for almost 20% of the total phenols in the digested mango peel, only methyl- and ethyl-gallate at very low concentration were detected in the fermented samples. This result indicates an extensive hydrolysis of gallates to form gallic acid (Fig. 1), as confirmed its increasing concentration over time, from 5.15 μmoles at 24 h to 10.21 μmoles at 72 h (Table 3). In addition, gallic acid might also derive from degalloylation of mangiferin, maclurin, iriflophenone and epigallocatechin gallates. Afterwards, gallic acid might also be dehydroxylated, yielding 3,4-dihydroxybenzoic acid and subsequently 3- and 4-hydroxybenzoic acid, as suggested by the increase of both compounds over time (Table 3). Another plausible mechanism in which gallic acid could be involved, is linked with the formation of pyrogallol by decarboxylation, in accordance with the high amount observed after 72 h (32.34 μmoles) (Table 3, Fig. 1). These biotransformation routes associated with gallic acid are in agreement with results described by Pereira-Caro et al., (2017), where colonic catabolism of black tea theaflavins was evaluated. Gallic acid has protective actions against cardiovascular diseases through increasing antioxidant enzymes capacity, inhibition of lipid peroxidation and decreasing serum levels of cardiac marker enzymes, modulation of hemodynamic parameters, recovery of electrocardiogram aberrations, and preservation of histopathological changes, as reported in a recent review carried out by Akbari (2020).

Mango peel is a rich source of ellagic acid, which was also present in the fermented samples. However, the highest concentration observed for ellagic acid after 72 h of fermentation (0.50 μmoles) was low, likely as a result of its biotransformation into urolithins, particularly A and C (Fig. 1), which increased significantly over time, reaching values of 4.61 and 21.93 μmoles after 72 h of colonic fermentation, respectively, in agreement with other studies that investigated the microbial conversion of ellagic acid and ellagitannins (García-Villalba, Beltrán, Espín, Selma, & Tomás-Barberán, 2013) (Table 3). A recent review has addressed the studies supporting the potential of urolithins in treatment interventions against gastrointestinal ailments (Kujawska & Jodynis-Liebert, 2020).

Xanthones constitute a very stable group of phenolic compounds, which remained after the colonic fermentation. Thus, mangiferin, *iso*-mangiferin and homomangiferin showed a direct relationship between concentration and fermentation time as a consequence of the hydrolysis of galloylated and *p*-hydroxybenzoylated derivatives of mangiferin and *iso*mangiferin (Table 3, Fig. 1). This is an interesting result considering that mangiferin is widely recognized for its anti-inflammatory, neuroprotective and immunomodulatory effects (García, Leiro, Delgado, Sanmartín, & Ubeira, 2003). In addition, mangiferin can influence apoptosis by suppressing the activation of nuclear factor kappa B-inducing kinase (Takeda et al., 2016). Dehydroxymangiferin was also identified in the fermented samples, although its low concentration revealed that it was not the main biotransformation pathway compared with the amount described for norathyriol after deglycosylation of mangiferin. Similarly, methyl-norathyriol was formed as a result of the sugar release from homomangiferin. Finally, euxanthone was identified as consequence of the dehydroxylation of norathyriol (Fig. 1), which tended to further accumulate after 72 h (Table 3). Recently, Fernández-Ochoa et al. (2020) evaluated the metabolic changes in liver and serum of streptozotocin-induced diabetic rats who received a mango dietary supplementation, using LC-MS untargeted metabolomic strategy. The mango supplemented group revealed an increased hepatic bioaccumulation of euxanthone, which shows the importance of the colon microbiota in the biotransformation of phenolic compounds. This result is noteworthy because euxanthone has shown protective effect against ox-LDL-induced endothelial cell injury via Nrf2 (Li et al., 2019), and suppression of tumour growth and metastasis in colorectal cancer (Wang et al., 2018).

Benzophenone derivatives showed lower stability than xanthones, although a discrete amount of maclurin and iriflophenone were detected in the fermented samples, at the expense of the hydrolysis of their

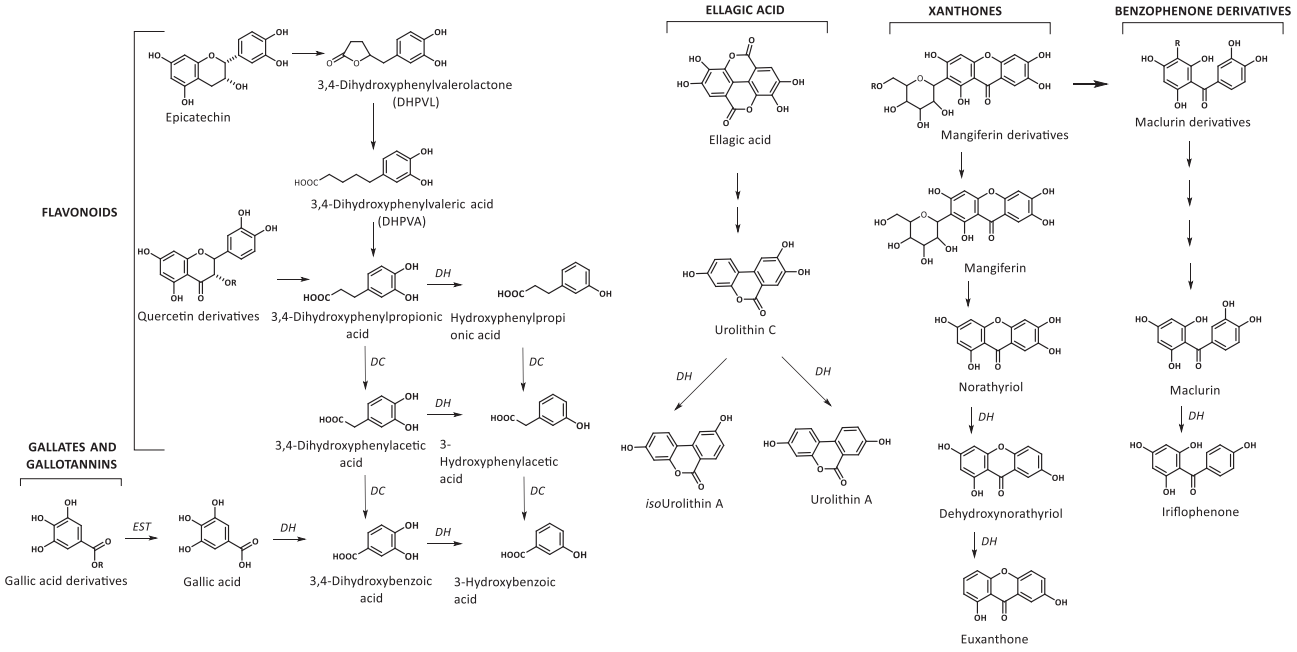

Fig. 1. Biotransformation pathways by the gut microbiota of the main phenolic compounds contained in predigested mango peels. DC: decarboxylases; DH: dehydrogenases; EST: esterases.

corresponding galloylated and glycosylated derivatives. Oxidation of benzophenones into xanthones cannot be ruled out (Fig. 1).

Regarding the flavonoid group, glycosylated derivatives of quercetin were completely absent during fermentation of samples with human feces. Quercetin fermentation is related to the formation of 3,4-dihydroxyphenylpropionic acid that subsequently can evolve to form phenylacetic acid derivatives, and then into benzoic acids by decarboxylation (Fig. 1, Table 3).

Within the flavonoid group, catechin and epicatechin were identified in the fermented samples although at low concentrations. These flavanols undergo microbiota-mediated conversion yielding 5C-ring fission metabolites, 5-(3′,4′-dihydroxypheynyl)-γ-valerolactones (DHPVL) and 4-hydroxy-5-(3′,4′-dihydrophenyl) valeric acid (DHPVA) (Gómez-Juaristi et al., 2018), which were detected in the fermented samples, showing the tendency to further accumulate over time (Table 3). Therefore, DHPVA might evolve to 3,4-dihydroxyphenylpropionic acid and its monohydroxylated derivatives (3- and 4-dihydroxyphenylpropionic acids) and, subsequently, to phenylacetic and benzoic derivatives, contributing to the formation of common phenolic acids (Fig. 1).

Regarding hydroxycinnamic acids present in the predigested mango peel (caffeic, ferulic and coumaric acids, as well as ferulic acid hexoside and coumaric acid glucose), only traces of caffeic and ferulic acids were detected in the fermented samples along with low amount of coumaric acid, which also might derive from the dehydroxylation of caffeic acid (Fig. 1). However, formation of reduced forms of hydroxycinnamic acids have been widely described in studies carried out with coffee, a rich source of hydroxycinnamics (Gómez-Juaristi et al., 2018). Thus, dihydrocaffeic, dihydroferullic and dihydrocoumaric acids were identified and quantified in the fermented samples (3,4-dihydroxyphenylpropionic, 3-methoxy-4-hydroxyphenylpropionic and 4-hydroxyphenylpropionic acids, respectively) showing concentration and incubation time a direct relationship (Table 3). It is important to mention that the amount of 4- and 3-hydroxyphenylpropionic acids increased up to 236.68 μmoles and 99.87 μmoles at 72 h, respectively, considering that both compounds have shown interesting bioactivity against cardiovascular diseases, among other effects (Álvarez-Cilleros, Ramos, Goya, & Martín, 2018).

Some phenylpropionic and phenylacetic acid derivatives were present in baseline samples because they are also involved in biotransformation pathways of other compounds, but their concentration was significantly enhanced after the colonic fermentation of predigested mango peel, confirming their role in the biotransformation of the phenolic compounds contained in this by-product.

To end, it is important to mention that the quantified phenolic metabolites in the colonic fermented samples revealed a recovery of about 33%, 40% and 48% after 24 h, 48 h and 72 h, respectively, with a tendency to further accumulate. This justifies that polyphenol metabolites remain longer in the body and, therefore, may be responsible for an extended bioactivity.

5. Conclusion

The present study revealed a wide spectrum of microbial-derived metabolites formed after the fermentation of mango peel with human fecal microbiota in TIM-2, a validated colon model. Besides simple phenolic acid derivatives of phenylpropionic, phenylacetic and benzoic acids, common in the biotransformation of flavonoids, a phenolic group characteristic of mango appeared, xanthones. Interestingly, these bioactive phenols showed high stability after colonic fermentation, thus expanding the horizons of the potential bioactivity and health effects of mango polyphenols. Therefore, most of the biological activity associated with the intake of mango peel might be associated to microbial-derived metabolites considering the low bioavailability at small intestinal level. More studies with mango polyphenol microbial metabolites are necessary to better understand the biological properties linked to this fruit and its sub-products.

CRediT authorship contribution statement

Sonia G. Sáyago-Ayerdi: Conceptualization, Formal analysis, Visualization, Writing - original draft. **Koen Venema:** Funding acquisition, Conceptualization, Methodology. **Maria Tabernero:** Formal analysis, Data curation, Investigation. **Beatriz Sarriá:** Writing - review & editing. **L. Laura Bravo:** Project administration, Writing - review & editing, Resources. **Raquel Mateos:** Supervision, Funding acquisition, Formal analysis, Writing - original draft, Writing - review & editing.

Declaration of Competing Interest

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

Acknowledgments

We are grateful to the Analysis Service Unit (USTA) facilities of ICTAN for the analysis of Chromatography and Mass Spectrometry and the volunteers who provided the fecal samples. RMB and SGSA acknowledge to Science and Technology for Development Program (CYTED) through the Ibero-American Network of Underutilized Autonomous Foods (ALSUB-CYTED, 118RT0543). SGSA acknowledge to CONACYT-Mexico for the sabbatical grant 260935. The Spanish Ministry of Science, Innovation and Universities (Project AGL2015-69986-R) is also acknowledged.

References

Akbari, G. (2020). Molecular mechanisms underlying gallic acid effects against cardiovascular diseases: An update review. *Avicenna Journal of Phytomedicine, 10*(1), 11–23. https://pubmed.ncbi.nlm.nih.gov/31921604.

Álvarez-Cilleros, D., Ramos, S., Goya, L., & Martín, M.Á. (2018). Colonic metabolites from flavanols stimulate nitric oxide production in human endothelial cells and protect against oxidative stress-induced toxicity and endothelial dysfunction. *Food and Chemical Toxicology, 115*, 88–97. https://doi.org/10.1016/j.fct.2018.03.006.

Barreto, J. C., Trevisan, M. T. S., Hull, W. E., Erben, G., De, E. S., Pfundstein, B., … Owen, R. W. (2008). Characterization and Quantitation of Polyphenolic Compounds in Bark, Kernel, Leaves, and Peel of Mango (Mangifera indica L.). *Journal of Agricultural and Food Chemistry, 56*, 5599–5610. https://doi.org/10.1021/jf800738r.

Blancas-Benitez, F. J., Mercado-Mercado, G., Quiros-Sauceda, A. E., Montalvo-Gonzalez, E., Gonzalez-Aguilar, G. A., & Sayago-Ayerdi, S. G. (2015). Bioaccessibility of polyphenols associated with dietary fiber and in vitro kinetics release of polyphenols in Mexican "Ataulfo" mango (Mangifera indica L.) by-products. *Food & Function, 6*(3), 859–868. https://doi.org/10.1039/C4FO00982G.

Bohn, T. (2014). Dietary factors affecting polyphenol bioavailability. *Nutrition Reviews, 72*(7), 429–452. https://doi.org/10.1111/nure.12114.

Crozier, A., Jaganath, I. B., & Clifford, M. N. (2009). Dietary phenolics: Chemistry, bioavailability and effects on health. *Natural Product Reports, 26*(8), 1001. https://doi.org/10.1039/b802662a.

Dorta, E., González, M., Lobo, M. G., Sánchez-Moreno, C., & de Ancos, B. (2014). Screening of phenolic compounds in by-product extracts from mangoes (Mangifera indica L.) by HPLC-ESI-QTOF-MS and multivariate analysis for use as a food ingredient. *Food Research International, 57*, 51–60. https://doi.org/10.1016/j.foodres.2014.01.012.

Espín, J. C., González-Sarrías, A., & Tomás-Barberán, F. A. (2017). The gut microbiota: A key factor in the therapeutic effects of (poly)phenols. *Biochemical Pharmacology, 139*, 82–93. https://doi.org/10.1016/j.bcp.2017.04.033.

FAOSTAT (2016). FAO Statistical Databases-Agriculture. Retrieved December 1, 2018, from http://www.fao.org/corp/statistics/en/.

Fernández-Ochoa, Á., Cázares-Camacho, R., Borrás-Linares, I., Domínguez-Avila, J. A., Segura-Carretero, A., & González-Aguilar, G. A. (2020). Evaluation of metabolic changes in liver and serum of streptozotocin-induced diabetic rats after Mango diet supplementation. *Journal of Functional Foods, 64*, 103695. https://doi.org/10.1016/j.jff.2019.103695.

García-Villalba, R., Beltrán, D., Espín, J. C., Selma, M. V., & Tomás-Barberán, F. A. (2013). Time course production of urolithins from ellagic acid by human gut microbiota. *Journal of Agricultural and Food Chemistry, 61*(37), 8797–8806. https://doi.org/10.1021/jf402498b.

García, D., Leiro, J., Delgado, R., Sanmartín, M. L., & Ubeira, F. M. (2003). Mangifera indica L. extract (Vimang) and mangiferin modulate mouse humoral immune responses. *Phytotherapy Research, 17*(10), 1182–1187. https://doi.org/10.1002/ptr.1338.

Gómez-Juaristi, M., Martínez-López, S., Sarria, B., Bravo, L., & Mateos, R. (2018). Bioavailability of hydroxycinnamates in an instant green/roasted coffee blend in humans. Identification of novel colonic metabolites. *Food & Function, 9*(1), 331–343. https://doi.org/10.1039/c7fo01553d.

González-Aguilar, G. A., Blancas-Benitez, F. J., & Sáyago-Ayerdi, S. G. (2017). Polyphenols associated with dietary fibers in plant foods: Molecular interactions and bioaccessibility. *Current Opinion in Food Science, 13*, 84–88. https://doi.org/10.1016/j.cofs.2017.03.004.

Koenen, M. E., Rubio, J. M. C., Mueller, M., & Venema, K. (2016). The effect of agave fructan products on the activity and composition of the microbiota determined in a dynamic in vitro model of the human proximal large intestine. *Journal of Functional Foods, 22*, 201–210. https://doi.org/10.1016/j.jff.2016.01.018.

Kujawska, M., & Jodynis-Liebert, J. (2020). Potential of the ellagic acid-derived gut microbiota metabolite – Urolithin A in gastrointestinal protection. *World Journal of Gastroenterology, 26*, 23. https://doi.org/10.3748/wjg.v26.i23.3170.

Li, S., Sun, Y., Han, Z., Bu, X., Yu, W., & Wang, J. (2019). Cytoprotective effects of euxanthone against ox-LDL-induced endothelial cell injury is mediated via Nrf2. *Life Sciences, 223*(December 2018), 174–184. https://doi.org/10.1016/j.lfs.2019.03.032.

López-Cobo, A., Verardo, V., Diaz-de-Cerio, E., Segura-Carretero, A., Fernández-Gutiérrez, A., & Gómez-Caravaca, A. M. (2017). Use of HPLC- and GC-QTOF to determine hydrophilic and lipophilic phenols in mango fruit (*Mangifera indica* L.) and its by-products. *Food Research International, 100*, 423–434. https://doi.org/10.1016/j.foodres.2017.02.008.

Maathuis, A., Hoffman, A., Evans, A., Sanders, L., & Venema, K. (2009). The effect of the undigested fraction of maize products on the activity and composition of the microbiota determined in a dynamic in vitro model of the human proximal large intestine. *Journal of the American College of Nutrition, 28*(6), 657–666. https://doi.org/10.1080/07315724.2009.10719798.

Pereira-Caro, G., Moreno-Rojas, J. M., Brindani, N., Del Rio, D., Lean, M. E. J., Hara, Y., & Crozier, A. (2017). Bioavailability of black tea theaflavins: Absorption, metabolism, and colonic catabolism. *Journal of Agricultural and Food Chemistry, 65*(26), 5365–5374. https://doi.org/10.1021/acs.jafc.7b01707.

Pérez-Jiménez, J., Arranz, S., Tabernero, M., Díaz- Rubio, M. E., Serrano, J., Goñi, I., & Saura-Calixto, F. (2008). Updated methodology to determine antioxidant capacity in plant foods, oils and beverages: Extraction, measurement and expression of results. *Food Research International, 41*(3), 274–285. https://doi.org/10.1016/j.foodres.2007.12.004.

Santos, L. E. O. (2002). *Proyecto Estudio Generación de Residuos Vegetales en el Valle del Cauca*. Sede Palmira: Universidad Nacional de Colombia.

Sáyago-Ayerdi, Sonia G., Zamora-Gasga, V. M., & Venema, K. (2019). Prebiotic effect of predigested mango peel on gut microbiota assessed in a dynamic in vitro model of the human colon (TIM-2). *Food Research International, 118*(October 2017), 89–95. https://doi.org/10.1016/j.foodres.2017.12.024.

Sáyago-Ayerdi, S. G., Moreno-Hernández, C. L., Montalvo-González, E., García-Magaña, M. L., de Oca, M.-M.-M., Torres, J. L., & Pérez-Jiménez, J. (2013). Mexican 'Ataulfo' mango (Mangifera indica L) as a source of hydrolyzable tannins. Analysis by MALDI-TOF/TOF MS. *Food Research International, 51*(1), 188–194. https://doi.org/10.1016/j.foodres.2012.11.034.

Takeda, T., Tsubaki, M., Kino, T., Yamagishi, M., Iida, M., Itoh, T., … Nishida, S. (2016). Mangiferin induces apoptosis in multiple myeloma cell lines by suppressing the activation of nuclear factor kappa B-inducing kinase. *Chemico-Biological Interactions, 251*, 26–33. https://doi.org/10.1016/j.cbi.2016.03.018.

Venema, K., van Nuenen, M., Smeets-Peeters, M., Minekus, M., & Havenaar, R. (2000). TNO's in vitro large intestinal model: An excellent screening tool for functional food and pharmaceutical research. *Ernährung, 24*(12), 558–564.

Venema, K. (2015). The TNO in vitro model of the colon (TIM-2). In *The impact of food bioactives on health* (pp. 293–304). Cham: Springer.

Wang, N., Zhou, F., Guo, J., Zhu, H., Luo, S., & Cao, J. (2018). Euxanthone suppresses tumor growth and metastasis in colorectal cancer via targeting CIP2A/PP2A pathway. *Life Sciences, 209*(88), 498–506. https://doi.org/10.1016/j.lfs.2018.08.052.