

¹Centro de Investigación en Alimentación y Desarrollo, AC (CIAD, AC), Hermosillo, Sonora, México

²Instituto Tecnológico de Tepic, Tepic, Nayarit, México

Added dietary fiber affects antioxidant capacity and phenolic compounds content extracted from tropical fruit

A.E. Quirós-Sauceda¹, J.F. Ayala-Zavala¹, S.G. Sáyago-Ayerdi², R. Vélez-de la Rocha¹,
J.A. Sañudo-Barajas¹, G.A. González-Aguilar^{1*}

(Received January 15, 2014)

Summary

The effect of fruit dietary fiber (FDF) on the antioxidant capacity and phenolic compounds (PC) content from the same fruits was investigated. FDF was incubated (pH 2.5, room temperature, 2 h) with methanolic extracts (ME) containing PC. The phenolic compounds (total phenols and flavonoids) content and antioxidant capacity (DPPH and TEAC) were analyzed in the resulting supernatants. Results showed that the addition of FDF decreased the PC content (5-25 %) and their action as antioxidants (4-22 %), being mango fiber which affects in most proportion. Wheat dietary fiber (WDF) was used as control, and its addition to ME reduced in greatest proportion the phenolic content (16-38 %) and the antioxidant capacity (30-48 %) than the FDF. This suggests that apparently depending on the nature and type of dietary fiber present in the food, some physicochemical interactions between the polysaccharides that conforms the fiber and PC could occur and subsequently affect their quantification and mode of action at the level of antioxidant capacity.

Introduction

Several epidemiological studies have linked the consumption of tropical fruits to the prevention of chronic disease and different types of cancer (HOOPER and CASSIDY, 2006). The proposed beneficial effects of these foods have been attributed to their important source of numerous phytochemicals with potential biological activity, such as phenolic compounds (PC) (KRIS-ETHERTON et al., 2002; GONZALEZ-AGUILAR et al., 2010). The mechanisms by which PC confers health effects are mainly associated to the antioxidant protection, by scavenging free radicals, inducing antioxidant enzymes or inhibiting pro-oxidant enzymes. Mango (*Mangifera indica* L.), pineapple (*Ananas comosus* L.), papaya (*Carica papaya* L.) and guava (*Psidium guajava* L.) are popular tropical fruits that presented high antioxidant potential, due to the high concentration of PC presented in their pulp (SANCHO et al., 2010; FU et al., 2011; PALAFOX-CARLOS et al., 2012). However, to achieve any health beneficial effect, the PC must be first released from the food matrix in which is embedded (bioaccessibility) and consequently be bioavailable for absorption (MANACH et al., 2005; PALAFOX-CARLOS et al., 2011).

In plant cell, most PC are compartmentalized primarily within the vacuole, enclosed by tonoplast and cytoplasmic lipid membranes which are in turn compressed against the plant cell wall (PADAYACHEE et al., 2012b). Mechanical and biochemical break down of food matrix results in cell rupture allowing the PC release and therefore favoring the bioavailability during digestion (PADAYACHEE et al., 2012b). However, the exposition of PC with the cell wall polysaccharide-protein matrix, produces a potential physical and chemical binding that promotes interactions in one or other way. There is evidence that PC interact physicochemically with polysaccharides during fruit synthesis as part of cell growth and development (PALAFOX-CARLOS et al., 2011; PADAYACHEE et al., 2012b). These interactions may be

beneficial, non-beneficial, or even detrimental for the bioactive effects attributed to PC.

In previous reports, some authors have studied the interactions between different groups of PC and the primary components of the cell wall matrix, particularly polysaccharides of dietary fiber. RENARD et al. (2001) studied the interactions between apple cell walls and native polyphenols. They reported that hydroxycinnamic acids and epicatechin did not bind to cell walls. However, binding of procyanidins reached up to 0.6 g per g cell walls. MOTOMURA and YOSHIDA, (2002) reported that cell walls from a range of fruits have been found to protect ascorbic acid from oxidation. Subsequently, apple cell walls and their polysaccharides were shown to affect the antioxidant activity of quercetin and L-ascorbic acid (SUN-WATERHOUSE et al., 2007). SUN-WATERHOUSE et al. (2008) reported that fiber from onions have some type of favorable interaction with L-ascorbic acid, but not with quercetin. More recently, PADAYACHEE et al. (2012b; 2012a) studied the extent of anthocyanins and phenolic acids cell wall interaction, using cellulose and pectin as cell wall models. It was found that cellulose and pectin are each able to bind both compounds. Consequently the interactions between different PC and dietary fiber components may prevent the bioaccessibility or release of the PC, and thereby affect its antioxidant effect as well as could potentially impact the nutritional content and functional potential of diets (SUN-WATERHOUSE et al., 2008). However, a beneficial interaction could occur when a PC bound to fiber is not bioavailable for absorption in the small intestine; so it is transported to the large intestine where fermentation of fibrous material takes place and helps maintain good gut health (SAURA-CALIXTO, 2011; PADAYACHEE et al., 2012b).

The purpose of this study was to investigate the influence of the source of dietary fiber on the interaction with supplemented PC (total phenols and flavonoids), as well as the effect of the antioxidant capacity.

Materials and methods

Materials

Tropical fruits of mango cv. "Haden", pineapple cv. "Esmeralda", papaya cv. "Maradol" and guava cv. "Hawaiian white" were purchased from a local market in Hermosillo, Sonora, Mexico, and used in this study. Fruits free of defects and mechanical damage were selected and distributed into 2 lots, to obtain fiber and PC. Fruit used for PC extraction was freeze-dried and stored until use. In addition, wheat dietary fiber (WDF) was obtained from a local store and used as fiber control.

Isolation of dietary fiber

Dietary fiber of tropical fruits were obtained as alcohol insoluble solids according to the methodology reported by SANUDO-BARAJAS et al. (2009). A sample of 100 g of cubed flesh from the pooled tissue was homogenized (IKA[®] Works, Model T25, Willmington, NC) with 300 mL of 95 % ethanol to dissolve low molecular weight sugars and

* Corresponding author

organic acids and the slurry was boiled with continuous stirring for 45 min to ensure enzyme inactivation and prevention of autocatalytic breakdown of polysaccharides (ROSE et al., 1998). The insoluble material was filtered through glass fiber filter, recovered and sequentially washed with 250 mL of ethanol, 250 mL of chloroform:methanol (1:1, v/v), and 250 mL of acetone. Insoluble, acetone-washed material was oven-dried at 37 °C, weighed and stored in a desiccator until use. This crude extract was named fruit dietary fiber (FDF).

Dietary fiber characterization

FDF and WDF were analyzed for total, soluble and insoluble dietary fiber contents by using the Megazyme total dietary fiber analysis kit (Megazyme International Ireland Ltd Wicklow, Ireland). In addition, the uronic acids, total sugars and neutral sugar and starch content were determined. The uronic acids was assayed in two mg of sample hydrolyzed in H₂SO₄ and calculated from a standard curve prepared with galacturonic acid (GalA) (Sigma) as described by AHMED and LABAVITCH (1977). For total sugars, 3 mg of sample were stirred with 3 mL of 67 % H₂SO₄ for 4 h and aliquots were assayed by the anthrone reaction according to YEMM and WILLIS (1954), using glucose (Sigma) for the standard curve. The neutral sugars composition was obtained from two mg of sample residue hydrolyzed with one mol L⁻¹ trifluoroacetic acid at 121 °C for 1 h. After hydrolysis the samples were centrifuged and the residue was digested with 67 % H₂SO₄ and used for cellulose assay using the anthrone reagent (crystalline cellulose Sigma was used for a standard curve). The trifluoroacetic acid-soluble supernatants were dried and converted to alditol acetates (BLAKENEY et al., 1983). For analysis, they were injected into a gas chromatograph (Model 3800, Varian Inc.) with a 30 m x 0.25 mm i.d. capillary column (model DB-23, J & W Scientific, Folsom, CA) as described in CARRINGTON et al., (1993). Results were calculated using standards of rhamnose, fucose, arabinose, xylose, mannose, galactose, glucose and myo-inositol as internal standard (Sigma). Starch was determined in 250 mg of sample adding 300 U thermostable α -amylase and 3 ml MOPS buffer (50 mM, pH 7.0) and hydrolyzed during 45 min in a boiling water bath. Thereafter, 100 U amyloglucosidase and 4 mL sodium acetate (200 mM, pH 4.5) were added; mixture was incubated for 45 min at 50 °C, then cooled and centrifuged until the sedimentation of insoluble solids. Aliquots (0.1 mL) in triplicate were taken from the supernatant and mixed with the glucose oxidase/peroxidase reagent; the mixture was incubated for 10 min at 50 °C; absorbance was recorded at 510 nm using a Cary 1E-UV spectrophotometer. By means of the glucose standard the concentration was obtained.

Phenolic compounds extraction

Freeze-dried samples (1 g) of tropical fruits were homogenized in 10 mL solution of 80 % methanol (IKA® Works, Model T25, Wilmington, NC) at room temperature. The homogenate was sonicated for 30 min (Bransonic Ultrasonic Co., Model 2210, Danbury, CT, USA) and then centrifuged at 14000 rpm for 15 min at 4 °C. The supernatants were collected and the pellet was resuspended again with 10 mL of 80 % methanol, under the conditions previously described. The two supernatants were mixed, filtered (Whatman No. 1, Springfield Mill, Maidstone Kent, UK) and made up to a final volume of 30 mL. The final concentration of the methanolic extracts (ME) was 0.033 g mL⁻¹ which was stored at -25 °C to be used for later evaluations.

Phenolic compounds content and antioxidant capacity

Total phenols

Total phenols were determined according to SINGLETON and ROSSI (1965), with some modifications. Briefly, 30 μ L of fruit ME were

added to 150 μ L Folin-Ciocalteu reagent (dilution 1:10), after reacting for 5 min, 120 μ L of 7.5 % Na₂CO₃ solution were added. The phenols were incubated for 2 h in the dark, and absorbance at 765 nm was measured using a microplate reader (BMG Labtech Inc., Model Omega, USA). The results were reported as mg of gallic acid equivalents (GAE)/100 g of fresh weight.

Total flavonoids

The total flavonoids content was measured using a colorimetric assay in accordance with the method of (ZHISHEN et al., 1999). Flavonoids were extracted 5 % NaNO₂ 10 % AlCl₃ and 1 mol L⁻¹ NaOH and measured spectrophotometrically at 510 nm using quercetin as standard. The results were expressed as mg of quercetin equivalents (QE)/100 g of fresh weight.

DPPH

Antioxidant capacity was evaluated by radical-scavenging activity using the DPPH (2, 2-diphenyl-1-picryl-hydrazyl-hydrate) as reported by PALAFOX-CARLOS et al. (2012). The stock solution was prepared by mixing 2.5 mg of DPPH radical with 100 mL of pure methanol. The solution was adjusted at an absorbance of 1.0 \pm 0.02 at 515 nm. Samples of 20 μ L of the ME were placed in a microplate and 280 μ L of DPPH radical were added. The mixture was kept in the dark for 30 min. The absorbance was read using a microplate (BMG Labtech Inc., Model Omega, USA) reader device, at a wavelength of 515 nm. The capacity of the antioxidants to reduce the absorbance of the radical after incubation time was calculated and the results were expressed as μ mol of trolox equivalents (TE)/100 g of fresh weight

TEAC

ABTS^{•+} cation was generated through the interaction of 19.2 mg of ABTS (2'-azino-bis(3-ethylbenzotriazoline-6-sulfonic acid)), dissolved in 5 mL of HPLC-grade water and 88 μ L of potassium persulfate (K₂S₂O₈) (0.0378 g mL⁻¹). It was incubated in the dark at room temperature for 16 h; then 1 mL of ABTS activated radical was taken and 88 mL of pure ethanol was added. The radical was adjusted at an absorbance of 0.7 \pm 0.02 at 734 nm. The reaction was initiated adding 245 μ L of ABTS^{•+} and 5 μ L of ME of tropical fruits. Absorbance was monitored at 734 nm at 1 and 6 min. The percentage of inhibition was calculated and the results were expressed as μ mol of TE/100g of fresh weight.

Incubation system

The incubation system was carried out to enhance interaction between the addition of fibers and PC contained in the ME of tropical fruits. 300 mg of FDF were added in 6 mL of ME pH 2.5 for 2 h. Samples were mixed and shaken constantly in the dark at room temperature. In order to compare the different type of fibers, a WDF was used as a positive control in the same study. Control solutions of the ME of tropical fruits free of fiber were also incubated under the same conditions. After 2 h, supernatant was collected and evaluated for phenolic content and antioxidant capacity. Moreover, a dynamic interaction was conducted to study the effect of incubation of both molecules with respect to the time. Aliquots were taken at 0, 10, 30, 60, 90 and 120 min and antioxidant capacity was evaluated.

Statistical analysis

Results were expressed as means \pm SD. Data were statistically analyzed by one-way ANOVA procedure, and the Tukey-Kramer multiple comparison tests were used at 95 confidence level. Number Cruncher Statistical System version 6.0 software (NCSS, LLC) was used. Four replicates were used for each experiment.

Results and discussion

Dietary fiber characterization

The dietary fiber total composition of samples is summarized in Tab. 1. Among the FDF, the lowest content of total fiber was observed in mango and the highest in guava. Insoluble fiber content ranged from 10.1 to 72.1 g/100 g in mango and guava, respectively. The soluble fiber content ranged from 10.7 in pineapple to 36.1 g/100 g in papaya. Interestingly, mango and papaya fibers had the highest proportion of soluble fiber; whereas pineapple and guava fibers showed highest proportion of insoluble fiber. This relationship agrees with that reported by RAMULU et al. (2003), indicating that mango and papaya were rich in soluble fiber whereas pineapple and guava contained mainly insoluble fiber. The total sugars range in samples from 47.5 to 91.4 g/100g in papaya and mango, respectively. Papaya fiber had the highest content of uronic acids represented by pectin and soluble dietary fiber; whereas the cellulose concentration (representing insoluble dietary fiber) was higher in guava and pineapple fiber. In addition, starch was detected only in mango fiber. While, by definition, the starch content is not necessarily part of the fiber; however, it was determined and included in the FDF due to it has been demonstrated that starch molecules (amylose and amylopectin) interacts effectively with phenolic (BARROS et al., 2012). As for the WDF (used as control), this presented a high content of total dietary fiber compared to the FDF, being insoluble fiber the highest proportion. Besides, WDF showed presence of starch. In terms of health benefits, both type of fibers are complementary and derived from individual polymeric composition, each fraction could play different physiological role depending on their fermentability and the reactivity of the sugars found in fiber. Insoluble fiber relates to both water and other compounds absorption and intestinal regulation, whereas soluble fiber associates with cholesterol in blood and it has been observed that it influenced the intestinal absorption (ANDERSON et al. 2009). In general, both types of dietary fibers are associated with the entrapment and interaction with different molecules with biological activity presented in foods.

Tab. 2 shows the neutral sugars composition of the fiber samples. Glucose and galactose were the main neutral sugars in mango and papaya fibers, while xylose and arabinose were the major monosaccharides in pineapple and guava fibers. This contrasts the high proportion of soluble dietary fiber in mango and papaya, and insoluble dietary fiber in pineapple and guava; as these sugars are major for those types of fibers respectively. The rest of monosaccharides were maintained at relatively low concentrations. This is in line with results reported by others authors (MEDLICOTT and THOMPSON, 1985; LARRAURI et al., 1997; GOMEZ et al., 2006) for neutral sugars in same fruits. Indeed, the high concentration of specific neutral sugars could

Tab. 1: Total, soluble and insoluble dietary fiber, uronic acids, total sugars, starch and cellulose content of fibers (g/100 g).

Fiber	TDF	IDF	SDF	TS	UA	Cellulose	Starch
Pineapple	76.1 ^b	65.3 ^c	10.7 ^b	50.5 ^{ab}	21.6 ^b	43.3 ^d	ND
Mango	28.3 ^a	10.1 ^a	18.1 ^c	91.4 ^d	19.4 ^b	10.6 ^a	48.6 ^b
Papaya	68.9 ^b	32.8 ^b	36.1 ^d	47.5 ^a	46.1 ^c	30.1 ^b	ND
Guava	85.2 ^c	72.1 ^d	13.1 ^b	56.4 ^b	17.9 ^b	43.6 ^d	ND
Wheat (control)	93.5 ^d	89.0 ^e	4.50 ^a	72.5 ^c	5.20 ^a	35.1 ^c	15.9 ^a

ND: not detected. TDF: total dietary fiber. IDF: insoluble dietary fiber. SDF: soluble dietary fiber. TS: total sugars. UA: uronic acids. Means values in each column followed by a different letter are significantly different ($p < 0.05$).

Tab. 2: Sum of the neutral sugars composition of fibers (g/100 g).

Fiber	Rha	Fuc	Ara	Xyl	Man	Gal	Glc	Total
Pineapple	0.79 ^b	0.38 ^b	9.63 ^c	19.5 ^c	1.23 ^c	5.77 ^c	2.91 ^a	40.2 ^b
Mango	0.64 ^b	0.39 ^b	5.09 ^b	2.01 ^a	0.45 ^a	5.12 ^c	75.9 ^c	89.6 ^d
Papaya	1.03 ^c	0.35 ^b	0.90 ^a	2.74 ^a	0.98 ^b	2.82 ^b	3.05 ^a	11.8 ^a
Guava	0.89 ^{bc}	0.31 ^b	5.39 ^b	21.6 ^c	0.43 ^a	2.69 ^b	2.45 ^a	34.1 ^b
Wheat (control)	0.13 ^a	0.04 ^a	10.8 ^c	12.7 ^b	0.40 ^a	1.18 ^a	24.7 ^b	49.9 ^c

Rha: rhamnose. Fuc: fucose. Ara: arabinose. Xyl: xylose. Man: mannose. Gal: galactose. Glu: glucose. Means values in each column followed by a different letter are significantly different ($p < 0.05$).

be associated to the result of the transformation of some other sugars, by hydrolysis or bond. Xylose and arabinose indicate the presence of arabinoxylyans. Galactose can be converted to galactan by hydrolysis. Binding of arabinose and galactose generates arabinogalactans and xyloglucans have a backbone composed by xylose, galactose and fucose. Although, these molecules may also bind to other compounds such as phenolic. In addition, the highest neutral sugars present in the control fiber (WDF) were glucose, arabinose and xylose. Thus agrees with that reported in the literature, indicating that arabinoxylyans are the major constituents in wheat bran (BATAILLON et al., 1998).

Phenolic compounds content and antioxidant activity

Determination of total phenols and flavonoids content as well as antioxidant activity of crude ME are shown in Tab. 3. Total phenols varied in all fruits, papaya had lowest content but not different for mango, followed by pineapple and guava. Moreover, the flavonoids content measured was lower than the content of total phenols; however, it showed the same trend. Contrasting these results with others reported in the literature, we observed comparable values for the same fruits (ROCHA RIBEIRO et al., 2007; CORRAL-AGUAYO et al., 2008; FU et al., 2011; ROSAS-DOMINGUEZ, 2011). The variation found in fruits and previous studies, may be attributed to differences in varieties, climate and ripeness, among others.

The antioxidant capacity evaluated by the DPPH assay showed a range of radical scavenging capacity, 7.19 to 14.30 $\mu\text{mol TE}/100\text{ g}$ of fresh weigh. The highest activity was found in guava and the lowest in pineapple. With the TEAC assay the extracts showed higher activity than DPPH assay but the same trend, showing the pineapple

Tab. 3: Phenolic compounds content and antioxidant activity of methanolic extracts of tropical fruits.

Fruit	Phenolic compounds		Antioxidant capacity	
	Total phenols (mg GAE/100g FW)	Total flavonoids (mg QE/100g FW)	DPPH ($\mu\text{mol TE}/100\text{ g FW}$)	TEAC ($\mu\text{mol TE}/100\text{ g FW}$)
Pineapple	77.59 ^b	63.30 ^c	7.196 ^a	189.5 ^a
Mango	55.85 ^a	49.39 ^b	10.80 ^c	224.4 ^c
Papaya	51.01 ^a	34.71 ^a	9.679 ^b	325.7 ^b
Guava	221.8 ^c	108.2 ^d	14.30 ^d	959.1 ^d

FW: fresh weight. GAE: gallic acid equivalents. QE: quercetin equivalents. TE: trolox equivalents. Means values in each column followed by a different letter are significantly different ($p < 0.05$).

the lower activity, followed by mango, papaya and guava. The fact that both reactions have same mechanism explains the behavior between their results. Moreover, the antioxidant activity did not follow the same tendency that PC present in the ME; as reported by TABART et al. (2009). This can be explained largely because the antioxidant activity of PC depends on the structure, in particular the number and positions of the hydroxyl groups and the nature of substitutions on the aromatic rings (BALASUNDRAM et al., 2006). In this sense, it appears that the differences in the antioxidant capacity of ME are attributed to the specific structure and concentration of each PC present in them.

Effect of dietary fiber on phenolic compounds content and antioxidant capacity

Fig. 1 shows the total phenols content of ME after 2 h of contact with fibers. Total phenols decreased significantly ($p < 0.05$) in all ME of tropical fruits when both types of fibers were added. Mango FDF mostly decreased total phenols content (23.72 %), followed by papaya (13.06 %), pineapple (10.82 %) and guava (5.86 %). Nevertheless, addition of WDF increased 15 % the depletion of the phenols content, following the same trend than FDF, mango (38 %), papaya (31.30 %), pineapple (28.77 %) and guava (16.17 %). Interestingly, the addition of FDF and WDF decreased at a higher ratio the phenols content in mango. By contrast, flavonoids content was affected in lowest proportion; however, similarly, the compounds were mostly decreased when WDF and mango fiber were present (Fig. 2).

The effect on the antioxidant capacity of ME with the addition of FDF and WDF evaluated by DPPH assay are shown in Fig. 3. When FDF was added, ME of tropical fruits shows a significantly decrease ($p < 0.05$) in the DPPH scavenging capacity. Mango fiber decreased the antioxidant capacity in greater extent (22.13 %), followed by papaya (8.83 %), pineapple (5.76 %), and guava (4.69 %). Moreover, addition of WDF decreased the antioxidant capacity of ME in greater proportion, being the pineapple the highest reduction (48.27 %), followed by mango (41.58 %), papaya (33.20 %) and guava (30.35 %). Similar results and the same tendency was showed in the effect on the antioxidant capacity assessed by TEAC (Fig. 4). It was observed that the decrease of PC content as well as antioxidant capacity is not directly related to the concentration of FDF, it could be more related to the type of fiber present as well as by the type of sugars that conform it. For example, guava showed a higher content of fiber but less decreased of PC content and antioxidant capacity. However, this behavior could be attributed to previous reports that indicated that

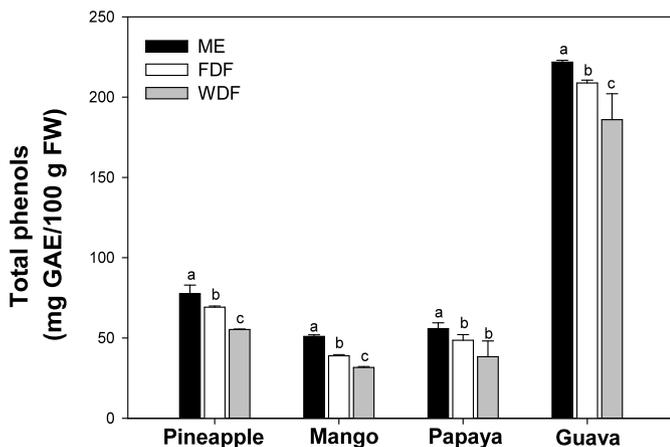


Fig. 1: Total phenols content of methanolic extracts of tropical fruits and reduction by adding fruit dietary fiber (FDF) and wheat dietary fiber (WDF). Different letter in bars indicates significant difference ($p < 0.05$) among fiber types by fruit.

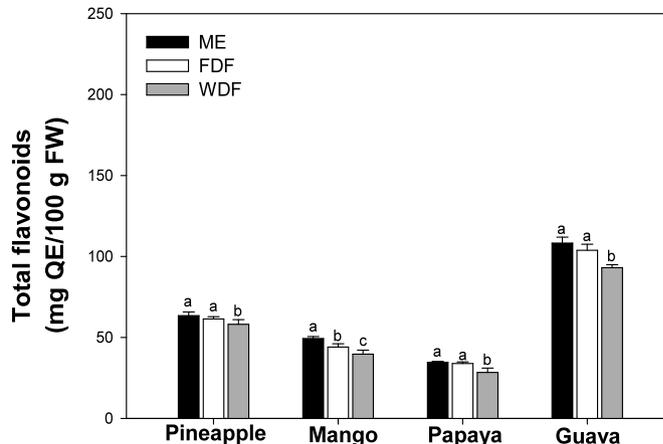


Fig. 2: Total flavonoids content of methanolic extracts of tropical fruits and reduction by adding fruit dietary fiber (FDF) and wheat dietary fiber (WDF). Different letter in bars indicates significant difference ($p < 0.05$) among fiber types by fruit.

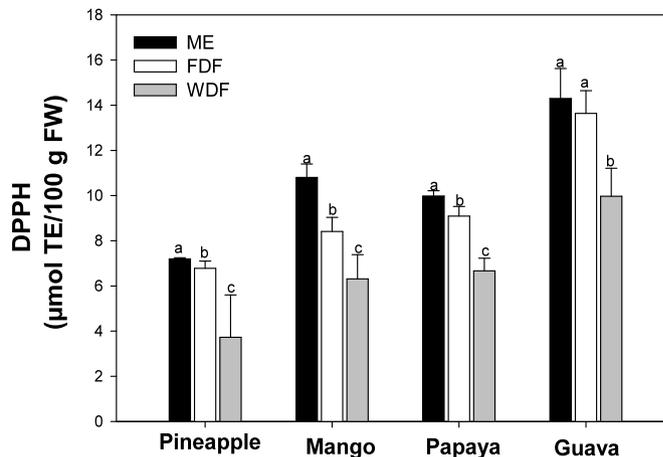


Fig. 3: Radical scavenging activity assessed by the DPPH assay, of methanolic extracts (ME) of tropical fruits by adding fruit dietary fiber (FDF) and wheat dietary fiber (WDF). Different letter in the same fruit indicates significant difference ($p < 0.05$) between fiber types.

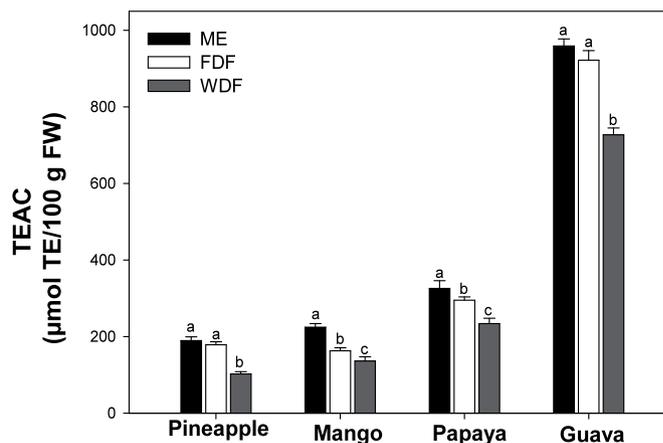


Fig. 4: Radical scavenging activity assessed by the TEAC assay, of methanolic extracts (ME) of tropical fruits by adding fruit dietary fiber (FDF) and wheat dietary fiber (WDF). Different letter in the same fruit indicates significant difference ($p < 0.05$) between fiber types.

guava is an excellent source of antioxidant dietary fiber; this indicates that fiber has associated phenols, which confer antioxidant activity to it (JIMENEZ-ESCRIG et al., 2001). In this sense, due to that guava fiber has in its structure associated PC, there could be no space to interact with other phenols. Furthermore, it was observed that in all assays, the highest depletion of PC and antioxidant capacity occurred with the addition of WDF and mango fiber, which can be associated to the presence of starch; these fibers being the only ones that had starch in their structure. It is evident that the addition of both types of fibers affects adversely the PC content and consequently the antioxidant capacity of ME. This could be attributed to physicochemical interactions that may occur between PC and the components of dietary fiber, causing PC physical trapping, consequently affecting their quantification and their action as antioxidants (PALAFOX-CARLOS et al., 2011). Next, will be described some possible explanations that could be causing the observed results.

The group of complex polysaccharides that conform dietary fiber has the ability to act forming physicochemical interactions with the PC, thereby preventing its action as antioxidants (SERRANO et al., 2009). These interactions may occur by hydrogen and ester bonds (with ferulic and cinnamic acids), hydrophobic interactions, and covalent bonds or simply by a physical entrapment (PALAFOX-CARLOS et al., 2011). However, the composition, functional group substitution and physical properties of the fibers are key factors for this interaction. In addition, another key factor is the type of PC present in the extract. PC that have more hydroxyl groups and also contain more hydrophobic domains provide hydrogen bonding and would promote stronger interactions. Comparing between fiber types, WDF declined proportionally the phenolic compounds (total phenols and flavonoids) and antioxidant capacity of ME of tropical fruits. WDF is an insoluble fiber type, consisting mainly of cellulose. According to its architecture, the cellulose has the ability to trap and bind phenolic. PC may be interacting through non-covalent interactions with neutral sugars (xyloglucans) present in cellulose chain (PADAYACHEE et al., 2012a). Furthermore, when the cellulose are in contact with water or aqueous solutions (as study was conducted) can form multiple compact fibers by hydrogen bonds between the hydroxyl groups on different chains of glucose (LE-BOURVELLEC et al., 2004). Thus, some PC can be caught in its structure, thereby reducing its bioaccessibility or release, in consequence affecting their bioactive action. Also, molecular size and structure of PC are key factors that could influence the possible interactions between both molecules. Therefore, architecture of the cellulose may be more conducive to PC penetration through and potentially binding to the surface than pectin composites (LE-BOURVELLEC et al., 2004). Another possible mechanism of interaction is due to the presence of starch. It was reported that presence of starch can significantly decrease the phenols content, due to strong interactions with their constituents, particularly with the amylose. The amylose possess a linear nature that affords a more optimum configuration for stronger bond formation between starch and PC in solution, particularly hydrophobic interactions. However, also the amylopectin double helix structure might provide a physically trapped of PC within the bulky amylopectin matrix, without necessarily chemically interacting with the starch (BARROS et al., 2012).

As for FDF, mango and papaya fibers were those that decreased in largest proportion the PC and antioxidant capacity of ME. These fruits fibers are characterized by showing highest uronic acid composition that is associated with the content of pectin, the fibers also showed presence of cellulose. PADAYACHEE et al. (2012a) reported that PC presented in various fruits and vegetables had the ability to interact with both cellulose and pectin. As mentioned before, the architecture of the cellulose may be more conducive to PC penetrating through and potentially binding to the surface of the cellulose, than pectin composites. However, pectin components also play

a role in binding PC, especially on prolonged exposure. Pectin contains hydrophobic cavities that could potentially encapsulate PC, as LE BOURVELLEC et al. (2004) concluded. Also, the portion of pectin polygalacturonic acid may be complex with some types of antioxidants through Ca^+ ions, which are the main cations present in the cell walls. Furthermore, in less proportion to the cellulose, the porosity of the pectin composites might also be a contributing factor to PC entrapment. Nevertheless, the high presence of starch in mango fiber could be associated with the greatest depletion of PC, as mentioned before. Moreover, the major neutral sugars identified in the insoluble fibers (xylose and arabinose) and soluble fibers (glucose and galactose) may be directly involved in the interaction with the PC, facilitating their physics interactions (SUN-WATERHOUSE et al., 2008; PADAYACHEE et al., 2012a; SIVAM et al., 2013).

Dynamic interaction

Fig. 5 shows a dynamic interaction of the addition of FDF and WDF on the antioxidant capacity of the ME of tropical fruits with respect to time. The addition of FDF shows a slow steady decline in the antioxidant capacity of ME of tropical fruits; however, the addition of WDF shows a faster decrease. With the addition of both types of fibers, mango fruit was decreased antioxidant capacity in less time. This indicates that the physical interaction or entrapment between these compounds was carried out more easily and more quickly, in comparison with the rest of the tropical fruits. That could be attributed to the type of PC presents in mango ME.

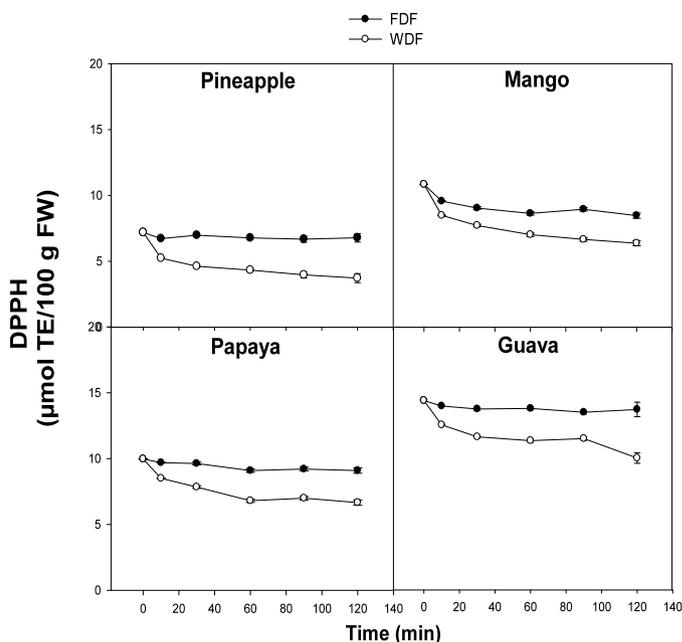


Fig. 5: Antioxidant capacity depletion of methanolic extract (ME) by adding fruit dietary fiber (FDF) and wheat dietary fiber (WDF) respect to time.

Conclusion

This study has shown that addition of FDF to ME containing PC decreased their phenolic content, as well as their antioxidant capacity. This can be attributed to the result of different physicochemical interactions between the two molecules, which affects the determination and antioxidant action of PC, as reported by other authors. However, it is shown that the type of fiber (soluble or insoluble) and the specific components, such as starch, are a key factors in this

process. However, is necessary further work to evaluate the type of the physicochemical interactions that can be carried out between fiber and phenols. Spectroscopy studies (IR, NMR, UV/Vis) are tools that can provide more information to understand this phenomenon. Furthermore, it is important to know if this interaction affects the absorption and bioavailability of PC during digestion process.

Acknowledgements

We thank CIAD and CONACYT-México for financial support. This work is part of the project "Nutrigenómica e interacciones moleculares de fenoles y fibra dietaria del mango "Ataulfo" (*Mangifera indica*, L.) en un sistema Murino" Project 179574CB-2012-01.

References

- AHMED, A.E.L.R., LABAVITCH, J.M., 1977: A simplified method for accurate determination of cell wall uronide content. *J. Food Biochem.* 1(4), 361-365.
- ANDERSON, J.W., BAIRD, P., DAVIS Jr., R.H., FERRERI, S., KNUDTSON, M., KORAYM, A., WATERS, V., WILLIAMS, C.L., 2009: Health benefits of dietary fiber. *Nutr. Res.* 67(4), 188-205.
- BALASUNDRAM, N., SUNDRAM, K., SAMMAN, S., 2006: Phenolic compounds in plants and agri-industrial by-products: Antioxidant activity, occurrence, and potential uses. *Food Chem.* 99(1), 191-203.
- BARROS, F., AWIKA, J.M., ROONEY, L.W., 2012: Interaction of tannins and other sorghum phenolic compounds with starch and effects on in vitro starch digestibility. *J. Agr. Food Chem.* 60(46), 11609-11617.
- BATAILLON, M., MATHALY, P., NUNES CARDINALI, A.-P., DUCHIRON, F., 1998: Extraction and purification of arabinoxylan from destarched wheat bran in a pilot scale. *Ind. Crop Prod.* 8(1), 37-43.
- BLAKENEY, A.B., HARRIS, P.J., HENRY, R.J., STONE, B.A., 1983: A simple and rapid preparation of alditol acetates for monosaccharide analysis. *Carbohydr. Res.* 113(2), 291-299.
- CARRINGTON, C.M.S., GREVE, L.C., LABAVITCH, J.M., 1993: Cell wall metabolism in ripening fruit. Effect of the antisense polygalacturonase gene on cell wall changes accompanying ripening in transgenic tomatoes. *Plant Physiol.* 103(2), 429-434.
- CORRAL-AGUAYO, R.D., YAHIA, E.M., CARRILLO-LOPEZ, A., GONZALEZ-AGUILAR, G., 2008: Correlation between some nutritional components and the total antioxidant capacity measured with six different assays in eight horticultural crops. *J. Agr. Food Chem.* 56(22), 10498-10504.
- FU, L., XU, B.T., XU, X.R., GAN, R.Y., ZHANG, Y., XIA, E.Q., LI, H.B., 2011: Antioxidant capacities and total phenolic contents of 62 fruits. *Food Chem.* 129(2), 345-350.
- GOMEZ, M., LAJOLO, F., CORDENUNSI, B., 2006: Evolution of soluble sugars during ripening of papaya fruit and its relation to sweet taste. *J. Food Sci.* 67(1), 442-447.
- GONZALEZ-AGUILAR, G.A., VILLA-RODRIGUEZ, J.A., AYALA-ZAVALA, J.F., YAHIA, E.M., 2010: Improvement of the antioxidant status of tropical fruits as a secondary response to some postharvest treatments. *Trends Food Sci. Tech.* 21(10), 475-482.
- HOOPER, L., CASSIDY, A., 2006: A review of the health care potential of bioactive compounds. *J. Sci. Food Agric.* 86(12), 1805-1813.
- JIMENEZ-ESCRIG, A., RINCON, M., PULIDO, R., SAURA-CALIXTO, F., 2001: Guava fruit (*Psidium guajava* L.) as a new source of antioxidant dietary fiber. *J. Agr. Food Chem.* 49(11), 5489-5493.
- KRIS-ETHERTON, P.M., HECKER, K.D., BONANOME, A., COVAL, S.M., BINKOSKI, A.E., HILPERT, K.F., GRIEL, A.E., ETHERTON, T.D., 2002: Bioactive compounds in foods: their role in the prevention of cardiovascular disease and cancer. *Am. J. Med. Sci.* 113.
- LARRAURI, J.A., RUPEREZ, P., SAURA-CALIXTO, F., 1997: Pineapple shell as a source of dietary fiber with associated polyphenols. *J. Agr. Food Chem.* 45(10), 4028-4031.
- LE-BOURVELLEC, C., GUYOT, S., RENARD, C., 2004: Non-covalent interaction between procyanidins and apple cell wall material: Part I. Effect of some environmental parameters. *Biochim. Biophys. Acta, Gen. Subj.* 1672(3), 192-202.
- MANACH, C., WILLIAMSON, G., MORAND, C., SCALBERT, A., REMESY, C., 2005: Bioavailability and bioefficacy of polyphenols in humans. I. Review of 97 bioavailability studies. *Am. J. Clin. Nutr.* 81(1), 230-242.
- MEDLICOTT, A., THOMPSON, A., 1985: Analysis of sugars and organic acids in ripening mango fruits (*Mangifera indica* L var Keitt) by performance liquid chromatography. *J. Sci. Food Agric.* 36(7), 561-566.
- MOTOMURA, Y., YOSHIDA, Y., 2002: Antioxidative ability of cell wall components in fruits against ascorbic acid oxidation. XXVI International Horticultural Congress: Issues and Advances in Postharvest Horticulture 628.
- PADAYACHEE, A., NETZEL, G., NETZEL, M., DAY, L., ZABARAS, D., MIK-KELSEN, D., GIDLEY, M., 2012a: Binding of polyphenols to plant cell wall analogues – Part 2: Phenolic acids. *Food Chem.* 135(4).
- PADAYACHEE, A., NETZEL, G., NETZEL, M., DAY, L., ZABARAS, D., MIK-KELSEN, D., GIDLEY, M., 2012b: Binding of polyphenols to plant cell wall analogues – Part 1: Anthocyanins. *Food Chem.* 134(1), 155-161.
- PALAFIX-CARLOS, H., AYALA-ZAVALA, J.F., GONZALEZ-AGUILAR, G.A., 2011: The role of dietary fiber in the bioaccessibility and bioavailability of fruit and vegetable antioxidants. *J. Food Sci.* 76(1), 6-15.
- PALAFIX-CARLOS, H., YAHIA, E., ISLAS-OSUNA, M., GUTIERREZ-MARTINEZ, P., ROBLES-SANCHEZ, M., GONZALEZ-AGUILAR, G., 2012: Effect of ripeness stage of mango fruit (*Mangifera indica* L., cv. Ataulfo) on physiological parameters and antioxidant activity. *Sci. Hort.* 135, 7-13.
- RAMULU, P., UDAYASEKHARA RAO, P., 2003: Total, insoluble and soluble dietary fiber contents of Indian fruits. *J. Food Comp. Anal.* 16(6), 677-685.
- RENARD, C.M., BARON, A., GUYOT, S., DRILLEAU, J.-F., 2001: Interactions between apple cell walls and native apple polyphenols: quantification and some consequences. *Int. J. Biol. Macromol.* 29(2), 115-125.
- ROCHA RIBEIRO, S.M., QUEIROZ, J.H., LOPES RINEIRO DE QUEIROZ, M.E., CAMPOS, F.M., PINHEIRO SANT'ANA, H.M., 2007: Antioxidant in mango (*Mangifera indica* L.) pulp. *Plant Foods for Human Nutrition (Formerly Qualitas Plantarum)* 62(1), 13-17.
- ROSAS-DOMINGUEZ, C., 2011: Contenido de compuestos bioactivos y su contribución a la capacidad antioxidante durante la maduración de piña cv. "Esmeralda". Coordinación de Tecnología de Alimentos de Origen Vegetal, Centro de Investigación en Alimentación y Desarrollo, A.C.
- ROSE, J.K.C., HADFIELD, K.A., LABAVITCH, J.M., BENNETT, A.B., 1998: Temporal sequence of cell wall disassembly in rapidly ripening melon fruit. *Plant Physiol.* 117(2), 345-361.
- SANCHO, L.E.G.G., YAHIA, E.M., MARTINEZ-TELLEZ, M.A., GONZALEZ-AGUILAR, G.A., 2010: Effect of maturity stage of papaya maradol on physiological and biochemical parameters. *Am. J. Agr. Biol. Sci.* 5(2), 194-203.
- SAÑUDO-BARAJAS, J.A., LABAVITCH, J., GREVE, C., OSUNA-ENCISO, T., MUY-RANGEL, D., SILLER-CEPEDA, J., 2009: Cell wall disassembly during papaya softening: Role of ethylene in changes in composition, pectin-derived oligomers (PDOs) production and wall hydrolases. *Postharvest Biol. Tec.* 51(2), 158-167.
- SAURA-CALIXTO, F., 2011: Dietary fiber as a carrier of dietary antioxidants: an essential physiological function. *J. Agr. Food Chem.* 59(1), 43-49.
- SERRANO, J., PUUPPONEN-PIMIA, R., DAUER, A., AURA, A.M., SAURA-CALIXTO, F., 2009: Tannins: current knowledge of food sources, intake, bioavailability and biological effects. *Mol. Nutr. Food Res.* 53(2), 310-329.
- SINGLETON, V., ROSSI, J.A., 1965: Colorimetry of total phenolics with phosphomolybdic-phosphotungstic acid reagents. *Am. J. Enol. Viticult.* 16(3), 144-158.
- SIVAM, A., SUN-WATERHOUSE, D., PERERA, C., WATERHOUSE, G., 2013: Application of FT-IR and Raman spectroscopy for the study of biopolymers in breads fortified with fibre and polyphenols. *Food Res. Int.* 50(2), 574-585.

- SUN-WATERHOUSE, D., MELTON, L.D., O'CONNOR, C.J., KILMARTIN, P.A., SMITH, B.G., 2007: Effect of apple cell walls and their extracts on the activity of dietary antioxidants. *J. Agr. Food. Chem.* 56(1), 289-295.
- SUN-WATERHOUSE, D., SMITH, B.G., O'CONNOR, C.J., MELTON, L.D., 2008: Effect of raw and cooked onion dietary fibre on the antioxidant activity of ascorbic acid and quercetin. *Food Chem.* 111(3), 580-585.
- TABART, J., KEVERS, C., PINCEMAIL, J., DEFRAIGNE, J.-O., DOMMES, J., 2009: Comparative antioxidant capacities of phenolic compounds measured by various tests. *Food Chem.* 113(4), 1226-1233.
- YEMM, E., WILLIS, A., 1954: The estimation of carbohydrates in plant extracts by anthrone. *Biochem. J.* 57(3), 508.
- ZHISHEN, J., MENGCHENG, T., JIANMING, W., 1999: The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chem.* 64(4), 555-559.

Address of the authors:

A.E. Quirós-Sauceda, J.F. Ayala-Zavala, R. Vélez-de la Rocha, J.A. Sañudo-Barajas, G.A. González-Aguilar*, Centro de Investigación en Alimentación y Desarrollo, AC (CIAD, AC), Carretera a la Victoria Km 0.6, La Victoria, Hermosillo, Sonora 83000, México

E-mail corresponding author: gustavo@ciad.mx

S.G. Sáyago-Ayerdi, Laboratorio Integral de investigación en Alimentos, División de Estudios de Posgrado, Instituto Tecnológico de Tepic, Av. Tecnológico 2595, CP 63175, Tepic, Nayarit, México