

# Comparative content of some phytochemicals in Spanish apples, peaches and pears

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**Abstract:** Dietary fibre, total polyphenols and phenolic acids in Spanish apples, peaches and pears were analysed and compared with their total radical-trapping antioxidative potential (TRAP). There were no significant differences in the content of dietary fibre among the studied fruits. The content of total polyphenols was  $2.4 \pm 0.4$ ,  $2.1 \pm 0.3$  and  $6.9 \pm 0.7 \text{ g kg}^{-1}$  in peeled fruits and  $4.7 \pm 0.4$ ,  $4.5 \pm 0.4$  and  $11.1 \pm 1.2 \text{ g kg}^{-1}$  in their peels for peaches, pears and apples respectively. The contents of dietary fibre, total polyphenols, caffeic, *p*-coumaric and ferulic acids and the TRAP values were significantly ( $P < 0.05$ ) higher in peels than in peeled fruits. The contents of all studied compounds and the TRAP values were significantly higher in peeled apples and their peel than in peaches and pears. We observed a strong correlation between the contents of total polyphenols and phenolic acids and the total radical-trapping antioxidative potential in all three fruits. The relatively high content of dietary fibre, the highest contents of total polyphenols, caffeic, *p*-coumaric and ferulic acids and the highest value of TRAP make apples preferable among the studied fruits for dietary prevention of atherosclerosis and other diseases.

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**Keywords:** dietary fibre; polyphenols; phenolic acids; total radical-trapping antioxidative potential

## INTRODUCTION

It is an established fact that phenolic compounds, particularly flavonoids of vegetables and fruits, possess antioxidant properties which are anticarcinogenic, antimicrobial and cardioprotective.<sup>1</sup> Epidemiological and clinical investigations demonstrate significant decreases in morbidity and mortality from cardiovascular and other diseases among fruit and vegetable consumers.<sup>1,2</sup> The positive influence of these natural products is attributed also to their dietary fibre.<sup>2,3</sup> It was shown that high-dietary-fibre diets are associated with the prevention of the dangerous coronary atherosclerosis and other diseases.<sup>2–4</sup> Therefore high-dietary-fibre formulated food products are currently being developed.<sup>5–8</sup>

The objectives of this investigation were (a) to determine the contents of dietary fibre, total polyphenols and phenolic acids in peeled apples, pears and peaches and their peels and compare these data with the values of total radical-trapping antioxidative

potential (TRAP) and (b) to find the most suitable fruit for a disease-preventing diet.

## MATERIALS AND METHODS

All reagents were of analytical grade and were purchased from Sigma Chemical Co, St Louis, MO, USA. Solvents were verified to have negligible TRAP.

We have studied peaches (*Prunus persica* L Batsch var Catherina), pears (*Pyrus communis* L var Blanquilla) and apples (*Malus domestica* Borkh var Golden Delicious) grown in Lleida (Segrià, Spain). Harvest time was 2 weeks around the following dates for each fruit: peaches, 1–5 August; pears, 20–25 August; apples, 5–10 September. These harvest times are the best for obtaining the respective fruits at optimal maturity in Lleida. At harvest time the values of sugars (Brix, %) were 12.7, 13.8 and 14.7 and acidity ( $\text{g l}^{-1}$ ) 7.03, 3.15 and 1.80 for peaches, pears and apples respectively.

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## Samples

Samples (12 peaches, 12 pears and 12 apples) were obtained from 36 randomly selected fruits for determination of all studied compounds. Peeled fruits and peels were studied separately. Six replicates were performed for each part of the studied fruits.

## Total radical-trapping antioxidative potential (TRAP) measurement

Extracts were prepared by mixing portions of 10 g of dry powdered peels and pulps from peaches, pears and apples with 50 ml of methanol each for 23 h at room temperature in stoppered flasks (Mallinckrodt, Baker Inc, Phillipsburg, NJ, USA). The extracts were filtered and concentrated under reduced pressure at 43 °C in a Savant Speed Vac Plus SC210A concentrator (Farmingdale, NY, USA).

## Chemiluminescence (CL) assay

The TRAP measurement was performed as previously described.<sup>9</sup> Peroxyl radicals, produced at a constant rate by thermal decomposition of 2,2-azo-bis-2-amidinopropane hydrochloride (ABAP; Polyscience, Warrington, PA, USA), were monitored by luminol-enhanced CL. The reaction was initiated by mixing 475 µl of phosphate-buffered saline, 50 µl of 10 mM luminol in 100 mM borate buffer (pH 10.0) and 50 µl of ABAP. This mixture was incubated (37 °C) in the temperature-controlled sample carousel of a BioOrbit 1251 luminometer (BioOrbit, Turku, Finland) for 15 min. During this period of time a steady state of the CL signal was reached. Then 20 µl of methanolic extract was added directly to the cuvette, and the sample was measured for a further period of time ( $\tau$ ) until 50% recovery of the original steady state CL signal was achieved. Trolox 8.0 nM; (Aldrich Chemical Co, Milwaukee, WI, USA), a water-soluble analogue of tocopherol, was used as a reference inhibitor instead of the sample. The stoichiometric factor of trolox (the number of peroxyl radicals trapped per added molecule of antioxidant) is 2.0. The TRAP value for each sample measured was obtained from the equation:

$$\text{TRAP} = \frac{2.0[\text{trolox}]\tau_{\text{sample}}}{f\tau_{\text{trolox}}}$$

where  $f$  is the dilution of the sample measured. The obtained results are expressed as nmol of peroxyl radicals trapped by 1 ml of sample.

## Determination of dietary fibre, total polyphenols and phenolic acids

Total polyphenols were extracted with ethanol as well as with methanol and ethyl acetate. Portions of 10 g of peeled peaches, pears and apples and of their peels were separately homogenised with 125 ml of 95% ethanol for 1 min and then gently boiled. After this procedure the fruit samples were cooled and filtered under vacuum using Whatman No 1 paper. The filtrates were reduced by evaporation under vacuum at 60 °C to a volume of 10 ml and then made up to a volume of 100 ml with distilled water.

Total polyphenols were determined according to the Folin–Ciocalteu method and measured at 675 nm.<sup>10</sup>

Determination of phenolic acids was done according to the method of García-Sánchez *et al.*,<sup>11</sup> with our modifications and changes in the extraction procedure of samples, using a combination of methanol, petroleum ether and ethyl acetate.<sup>12</sup> Fluorescence emission was measured with a Jasco model FP-770, spectrofluorometer (Japan Spectroscopic Co, Ltd, Hachioji City, Japan) at excitation ( $\lambda_{\text{ex}}$ ) and emission ( $\lambda_{\text{em}}$ ) wavelengths appropriate for each phenolic acid determined.

Total, soluble and insoluble dietary fibre contents were determined according to Prosky *et al.*<sup>13</sup>

## Statistical analyses

The results of this investigation are given as the mean  $\pm$  SD of six replicates for each fruit sample. When appropriate, differences between groups were tested by two-way ANOVA. In the assessment of TRAP the Spearman correlation coefficient ( $R$ ) was used.  $P$  values  $<0.05$  were considered significant.

## RESULTS

The ranges of total, insoluble and soluble dietary fibre content were 26.1–33.2 and 19.1–27.1, 15.2–21.1 and 11.1–16.3, and 9.2–13.3 and 7.2–11.1 g kg<sup>-1</sup> fresh fruit for peels and peeled fruits respectively. There were no significant differences in total, insoluble and soluble dietary fibre content in peeled peaches, pears and apples and their peels (Table 1). The content of dietary fibre in peels was significantly higher than in peeled fruits ( $P < 0.05$ ).

The content of total polyphenols was 2.4  $\pm$  0.4, 2.1  $\pm$  0.3 and 6.9  $\pm$  0.7 g kg<sup>-1</sup> in peeled fruits and 4.7  $\pm$  0.4, 4.5  $\pm$  0.4 and 11.1  $\pm$  1.2 g kg<sup>-1</sup> in their peels

Dietary fibre	Peeled fruits			Peels		
	Peaches	Pears	Apples	Peaches	Pears	Apples
Total	20.9 $\pm$ 2.1a	22.8 $\pm$ 2.2a	21.9 $\pm$ 2.1a	27.9 $\pm$ 3.1a	29.7 $\pm$ 3.0a	28.7 $\pm$ 0.1a
Soluble	8.1 $\pm$ 0.6a	8.9 $\pm$ 0.7a	8.9 $\pm$ 0.7a	10.1 $\pm$ 1.1a	11.8 $\pm$ 1.1a	10.8 $\pm$ 1.1a
Insoluble	12.6 $\pm$ 1.2a	13.9 $\pm$ 1.3a	13.0 $\pm$ 1.2a	17.8 $\pm$ 1.6a	17.9 $\pm$ 1.6a	17.9 $\pm$ 1.6a

**Table 1.** Dietary fibre contents (g kg<sup>-1</sup>) in fresh peaches, pears and apples

Values are mean  $\pm$  SD of six measurements. Means in a row without a letter in common differ significantly ( $P < 0.05$ ).

**Table 2.** Contents of some major phenolics ( $\text{mg kg}^{-1}$ ) and TRAP values ( $\text{mmol ml}^{-1}$ ) in fresh pears, peaches and apples

Phenolic compound	Peeled fruits			Peels		
	Pears	Peaches	Apples	Pears	Peaches	Apples
Ferulic acid	11.2±1.1a	41.2±3.5b	112.2±9.1c	14.5±1.2a	47.8±4.6b	134.8±1.3c
<i>p</i> -Coumaric acid	38.7±3.1a	112.1±10.1b	369.2±32.1c	51.7±4.5a	151.3±11.3b	523.7±43.7c
Caffeic acid	189.1±17a	702.3±67.1b	1994.1±99.2c	245.2±19.3a	878.3±9.5b	2599.1±173c
TRAP	1.54±0.2a	1.61±0.2a	3.23±0.3b	2.7±0.2a	2.9±0.2a	6.9±0.6b

Values are mean±SD of six measurements. Means of acids in a column and means of TRAP in a row without a letter in common differ significantly ( $P < 0.05$ ).

for peaches, pears and apples respectively. Total polyphenols were significantly higher in peels than in peeled fruits.

The highest content of all studied phenolic acids was in peeled apples and their peel and the lowest was in peeled pears and their peel (Table 2). Phenolic acids in peels of peaches, pears and apples were significantly higher than in peeled fruits.

The highest TRAP values were in peeled apples and their peel and the lowest were in peeled pears and their peel. The TRAP values in peels of peaches, pears and apples were significantly ( $P < 0.005$ ) higher than in peeled fruits (Table 2).

When the TRAP values were correlated with the content of total polyphenols, a very good correlation was observed (Fig 1,  $R^2 = 0.9505$ ). When the TRAP values were correlated with the content of individual phenolic acids, the best correlations were found between *p*-coumaric acid and TRAP (Fig 2,  $R^2 = 0.7849$ ) and between TRAP and caffeic acid (Fig 2,  $R^2 = 0.7259$ ), while a weaker correlation was found between TRAP and ferulic acid (Fig 2,  $R^2 = 0.6747$ ).

In contrast, a poor correlation was observed when the TRAP values were correlated with total dietary fibre contents (Fig 3,  $R^2 = 0.2969$ ).

## DISCUSSION

The aim of our investigation was to compare the content of some phytochemicals in Spanish apples, peaches and pears and to find among them the best for dietary prevention of atherosclerosis and other diseases.

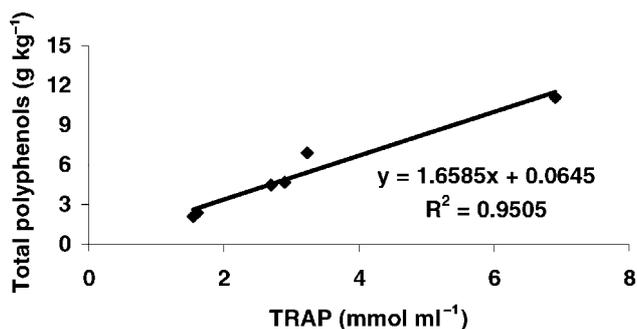


Figure 1. Correlation of total polyphenol content with TRAP value.

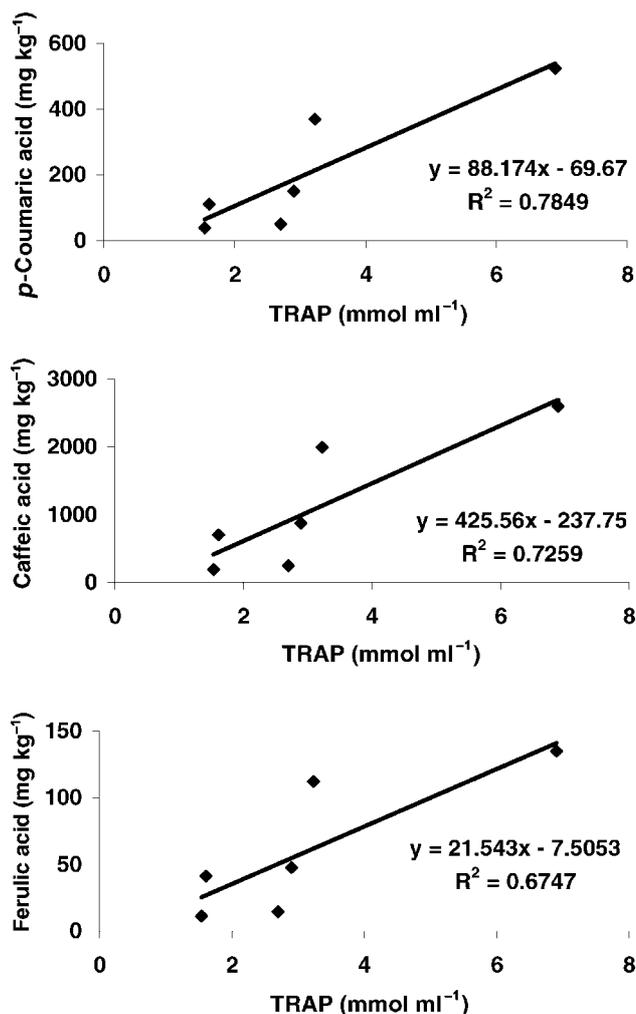


Figure 2. Correlation of phenolic acid content with TRAP value.

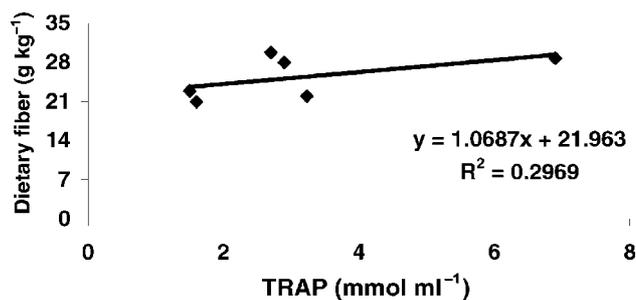


Figure 3. Correlation of dietary fibre content with TRAP value.

The results of this investigation showed that the content of dietary fibre was high in all three fruits, without significant differences among them. This is an advantage, as high-dietary-fibre diets are associated with the prevention and treatment of some diverse diseases such as diverticular and coronary heart disease.<sup>3</sup>

It is known that dietary fibre has different physiological effects. Soluble dietary fibre becomes viscous when mixed with water. This viscosity is associated with delayed gastric emptying, altered mixing of the intestinal contents and slower transit of digesta along the small intestine and thus with a slower rate of glucose, lipid or sterol absorption along a greater length of the small intestine.<sup>14</sup> The insoluble part is related to both water absorption and intestinal regulation, whereas the soluble fraction may influence lipid metabolism by decreasing the levels of LDL-C and is associated with the reduction of cholesterol in blood.<sup>3</sup> These observations support the opinion that dietary fibre is a very important component of a healthy diet. Thus health organisations recommend the ingestion of 30–45 g of dietary fibre per day.<sup>15</sup> The results of this investigation showed that the content of dietary fibre was high in all three fruits, without significant differences among them. Therefore, from the point of view of the content and composition of dietary fibre, the three studied fruits are comparable.

The content of the studied compounds is influenced by various conditions, including region, climate and ripeness.<sup>16</sup> Thus some of our results differ from others, but most were in accordance with the results of others investigators.<sup>17–19</sup>

Some authors claim that dietary fibre possesses antioxidant properties.<sup>20,21</sup> In our previous investigations *in vitro* and *in vivo* we did not find that dietary fibre provides an antioxidant effect.<sup>22,23</sup> Similarly, in this investigation we found a very poor correlation between dietary fibre and TRAP ( $R^2=0.2969$ ). Therefore this investigation, like our previous ones, did not support the claim that dietary fibre possesses antioxidant properties.

The total polyphenol content in peeled apples and their peel was significantly higher than in peeled peaches and pears ( $P < 0.05$  in all cases). The contents of caffeic, *p*-coumaric and ferulic acids in peeled apples and their peel were significantly higher than in peaches and pears ( $P < 0.05$  in all cases). The contents of these compounds in peels were significantly higher than in peeled fruits. These results are in accordance with the data of other investigators.<sup>24</sup>

The total polyphenol content of the studied fruits was compared with TRAP. The highest TRAP values were found in apple peel and peeled apples. Therefore this investigation confirms that the higher the total polyphenol content, the greater is the antioxidant capacity.<sup>25</sup>

The phenolic acid contents in the investigated fruits were in the following order: caffeic > *p*-coumaric > ferulic. It is known that antioxidant activity of phenolic

acids is generally governed by their chemical structures. Their activity increases with the number of hydroxyl groups. Therefore our results are of particular interest regarding the amount of caffeic acid found in apples and are in agreement with others.<sup>26</sup>

## CONCLUSIONS

The antioxidant properties of the studied fruits can be attributed to their content of phenolic compounds. The degree of antioxidant potential depends upon the level of total polyphenols and some phenolic acids.

Total polyphenol and phenolic acid contents and TRAP values in peeled apples and their peel are significantly higher than in peeled peaches and pears and their peels. All these indices are higher in peels than in peeled fruits.

There are no significant differences in the high content of dietary fibre in all studied fruits. The content of dietary fibre in peels is significantly higher than in peeled fruits. The antioxidant potential of dietary fibre in the studied fruits is questionable.

The high content of dietary fibre, the highest total polyphenol and phenolic acid contents and the highest TRAP values make apples preferable for dietary prevention of cardiovascular and other diseases.

Peels of apples, peaches and pears are rich in dietary fibre and phenolics and can be used in industrial processing for extraction of these constituents.

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