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Effect of an olive phenolic extract added to the oily phase of a tomato sauce, on the preservation of phenols and carotenoids during domestic cooking

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ABSTRACT

The protective effect of a phenolic extract from olive vegetation water on carotenoids and other phytonutrients has been evaluated during a home-cooking procedure to prepare tomato sauce. To specifically investigate on the complex mechanisms involved in the thermal degradation and solubilization of carotenoids, the olive phenols were added to a refined olive oil, chosen as control fat, in order to distinguish the effect of the simple oily matrix from that of the phenols occurring in virgin olive oils.

Three sauces were prepared using a commercial tomato "passata", carrots, celery and onion. Two sauces contained refined olive oil to which was added a phenolic extract at 40 or 60 mg/100 g of polyphenols, the third was the control with refined olive oil. The results revealed positive effect of the phenolic extract on the phytonutrient content of the tomato sauce. In the experimental sauces for the hydrophilic phenols, an improvement of approximately 100% was observed at both levels of phenolic extract addition in comparison to the control. There was a greater than 50% increase in *a*-tocopherol and a 43% increase in carotenoids with the lower level of phenols addition, and greater than 58% with the higher level of enrichment.

1. Introduction

The basis of the Mediterranean diet model is fresh and processed vegetables, providing a range of nutrients and different bioactive compounds, including phytochemicals, vitamins, minerals, and fibers (Liu, 2013). However, food-processing operations, particularly domestic and industrial thermal processing, strongly affect the stability of nutrients and other bioactive compounds, depending on the temperature and duration (Grajek & Olejnik, 2010; Ioannou, Hafsa, Hamdi, Charbonnel, & Ghoul, 2012; Tiwari & Cummins, 2013). Most heat processes lead to a degradation of phenolic acids, flavonoids, tocopherols and tocotrienols, due to their high likelihood of being involved in oxidative reactions. Carotenoids are also subjected to modifications that, depending on the processing conditions, may lead to their decrease or increase due to their ameliorated solubility following the degradation of vegetable tissues induced by heat, especially in the presence of a fat source. Recently, there has been an increasing need for consumer information about the healthy value of raw and processed foods, including those prepared under home-cooking conditions, together with an awareness that the thermal process may lower the phytonutrient content and a growing movement against conventional additives used as preserving agents. The phenolic substances occurring in virgin olive oils and their co-products possess strong antioxidant activities (Servili et al., 2004). Beyond the advantages offered by their presence in such food matrixes, recently a system to recover and purify them from the vegetation waters produced by the mechanical extraction process of virgin oil has been projected (Servili et al., 2011a), giving a valuable opportunity to gain added value from the depollution of these wastes, very abundant in the Mediterranean basin. Such kind of phenolic extract has been evaluated for its potential use as a functional food ingredient and as a food antioxidant of natural origin (Araújo, Pimentel, Alves, & Oliveira, 2015; Esposto et al., 2015; Servili et al., 2011a, 2011b). Of the most frequently consumed Mediterranean recipes, those based on tomatoes (raw or derivatives) are particularly precious for their richness in lipophilic and hydrophilic phytonutrients (flavonoids,

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tocopherols and carotenoids). For this reason, in this study, a traditional tomato sauce prepared with the addition of refined olive oil, with or without a purified olive phenolic extract, has been used as a model for evaluating the potential preservation activity of the extract in terms of the degradation of phytonutrients from the tomato sauce and from the "soffritto".

Soffritto, a combination of three key ingredients (celery, onion and carrot), is the essential base for Italian soups, stews and some sauces. Literally meaning under-fried, the technique can be considered a type of stir-frying or a sautéing, where the food is fried very quickly at a very high temperature in a thin layer of fat, during which time it is stirred continuously to prevent the food from burning. The subsequent step in domestic preparations of tomato sauce is a cooking step at approximately 100 °C for several minutes. Using this process, the experiments described here have taken into account the modifications of the hydrophilic and lipophilic phenolic compounds of the dish, and of the carotenoid fraction affected by the presence and the concentration of phenols from the olive extract.

2. Materials and methods

2.1. Materials and chemicals

2.1.1. Reference compounds

The (p-hydroxyphenyl)ethanol (p-HPEA) was purchased from Fluka (Milan, Italy), while the 3,4-(dihydroxyphenyl)ethanol (3,4-DHPEA), was obtained from Cabru s.a.s. (Arcore, Milan, Italy). The α-tocopherol was provided by Sigma Aldrich (Milano, Italy), while (*E*)-lycopene, β-carotene, *p*-cumaric acid, caffeic acid, ferulic acid, quercetin-3,4′-*O*-diglucoside, quercetin-3-*O*-rutinoside, quercetin-4-*O*-glucoside, quercetin, apigenin luteolin, kaempferol and lutein were obtained from Extrasynthese (Genay, France). The verbascoside was extracted from the olive fruit and the dialdehyde form of elenolic acid linked to 3,4-DHPEA (3,4-DHPEA-EDA) was extracted from Virgin Olive Oil (VOO), according to the procedures reported in previous papers, followed also for assessing the purity and the chemical structures of those substances (Montedoro et al., 1993; Servili, Baldioli, Selvaggini, Macchioni, & Montedoro, 1999).

2.1.2. Ingredients

Commercial tomato "passata" and fresh carrots, celery and onions were bought in a supermarket. The refined olive oil (ROO) was kindly provided by Monini SpA (Perugia, Italy).

2.1.3. Phenolic extract

A crude phenolic concentrated from fresh olive vegetation water arising from a three-phase decanter oil separation system was obtained by membrane treatments and purified following the procedures described in a previous paper (Servili et al., 2011a) to obtain a phenolic extract (PE) whit a total concentration in phenolic compounds of 730 mg/g.

2.1.4. ROO samples enriched with phenolic extract (ROOPE)

PE was mixed until complete solubilization into two samples of ROO (400 g each) to reach a final concentration of phenolic compounds of 400 and 600 mg/kg, expressed as the sum of p-HPEA, 3,4-DHPEA, 3,4-DHPEA-EDA and verbascoside. Those two levels of addition were chosen in a range of concentrations comparable to those occurring in extra virgin olive oils of good quality (Taticchi, Esposto, & Servili, 2014). The two new samples were named ROOPE (1) and ROOPE (2). Table 1 gives their phenolic composition.

2.1.5. Manufacture of the experimental tomato sauces

The experimental sauces were prepared following the traditional Italian recipe for a simple tomato sauce, in which the test variable was the oil used for making the soffritto. The ingredients for the soffritto were: 20 g of fresh celery, 20 g of fresh onion and 20 g of fresh carrot chopped and mixed, 100 g of ROO, 100 g of ROOPE (1) or 100 g of ROOPE (2). All the soffritto ingredients were sautéed for 10 min, reaching temperatures that were taken with a temperature probe and reported in Table 2. Next, a sample of oil (20 g) and of vegetable mix (2 g) were taken from each trial for analytical evaluation, and replaced with 20 g of the respective oil [ROO, ROOPE (1) or ROOPE (2)], preheated to 80 °C. To each sautéed experimental soffritto was added 800 g of tomato passata and the mixes were boiled for 20 min, reaching temperatures that were taken with a temperature probe and reported in Table 2. These samples were the tomato sauce samples. An aliquot of tomato sauce was centrifuged at 9327 g for 3 min (with a centrifuge Universal 32 Hettich, Germany) to recover the supernatant oily fraction for analytical evaluation. The decrease in water content, calculated in each sample after the soffritto sautéing and the tomato sauce cooking, corresponded to an average of 2.2 g/100 g.

2.2. Analyses

2.2.1. Extraction and HPLC analysis of the phenolic compounds

PE. The phenolic composition of the PE obtained was characterized by HPLC following the procedure reported by Selvaggini et al. (2006). The reversed-phase HPLC analyses of the phenolic extracts were conducted with an Agilent Technologies system (Santa Clara, CA, USA) Mod. 1100 composed of a vacuum degasser, a quaternary pump, an autosampler, a thermostatted column compartment, a DAD, and a FLD. A Spherisorb column ODS-1250 × 4.6 mm with a particle size of 5 μm (Phase Separation Ltd., Deeside, U.K.) was used to fractionate the samples, using as solvents methanol, water and acetic acid.

Oils. Phenols were extracted from the oils used for the soffritto preparation [ROO, ROOPE (1) and ROOPE (2)] and the oily fractions separated from the three corresponding experimental soffrittos and tomato sauces. For the hydrophilic phenols the method reported by Montedoro, Servili, Baldioli, and Miniati (1992) was applied; the HPLC analyses of the phenolic extracts were conducted with the same equipment as described above; the operating conditions of the chromatographic analysis were those reported by Selvaggini et al. (2006), using as solvents methanol, water and acetic acid. For the α-tocopherol the analysis was carried out according to Esposto et al. (2015), the solvents used were hexane and isopropyl alcohol.

Soffritto and tomato sauce. The phenolic fraction of the vegetables in the soffritto and tomato sauce were extracted when uncooked and after the thermal process, separated from the oily phase. For the hydrophilic phenols, the extraction procedure was adapted from Motilva et al. (2014). Briefly, 5 g of soffritto and tomato sauce was homogenized by Ultraturrax T25 for 1 min with 10 mL of a mixture of acetone with 500 mL/L of ethanol. The homogenate was centrifuged at 9327 g for 10 min and the supernatant recovered; the operation was repeated. The two recovered supernatant fractions were then combined and the organic solvent was evaporated under vacuum. The fraction was dissolved in 1 mL of methanol, filtered by a syringe filter (0.45 μm CA, Whatman, Clifton, NJ, USA), injected in the HPLC (the same equipment as described above) and analyzed following the procedure described by Selvaggini et al. (2006), using as solvents methanol, water and acetic acid. Flavonoids and phenolic acids were detected operating a DAD set at wavelength 324 nm for caffeic acid, *p*-coumaric acid, and ferulic; 360 nm for quercetin, quercetin-3,4′-*O*-diglucoside, quercetin-3-*O*-rutinoside, quercetin-4-*O*-glucoside, and