Food Chemistry 128 (2011) 62–69

Contents lists available at ScienceDirect

Food Chemistry

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

Antioxidant properties of 4-vinyl derivatives of hydroxycinnamic acids

Petra Terpinc, Tomaž Polak, Nataša Šegatin, Andrej Hanzlowsky, Nataša Poklar Ulrih, Helena Abramovič *

Biotechnical Faculty, University of Ljubljana, SI-1111 Ljubljana, Slovenia

ARTICLE INFO

Article history:
Received 26 November 2010
Received in revised form 18 February 2011
Accepted 22 February 2011
Available online 24 February 2011

Keywords:
4-Vinylphenol
4-Vinylguaiacol
4-Vinylsyringol
4-Vinylcatechol
Hydroxycinnamic acids
Antioxidant activity

ABSTRACT

The compounds 4-vinylphenol (4-VP), 4-vinylguaiacol (4-VG), 4-vinylsyringol (4-VS) and 4-vinylcatechol (4-VC) were prepared by thermal decarboxylation of the corresponding hydroxycinnamic acids *p*-coumaric, ferulic, sinapic and caffeic acid, respectively. For confirmation of the synthesised products LC–MS followed by NMR analysis was used. To evaluate their antioxidant potential, their reducing power and efficiency in scavenging the alkylperoxyl radical generated in an emulsion system, the 2,2-diphenyl-1-picrylhydrazyl (DPPH·) radical and the superoxide anion radical ($O_2^{\cdot-}$) were determined. All tested 4-vinyl derivatives revealed weaker antioxidant activity in a homogeneous polar medium than the corresponding phenolic acids. In the emulsion system the activity for 4-vinyl derivatives was higher than was the activity of their corresponding phenolic acids, with 4-VG as the most active among the tested phenolic compounds.

1. Introduction

Hydroxycinnamic acids (*p*-coumaric, caffeic, ferulic and sinapic acid) are very common in fruits and vegetables in the form of esters and glycosides. Certain cinnamic acids behave as flavour precursors, since they break down and form new molecules with lower flavour thresholds (Rizzi & Boekley, 1992). These highly flavour-active volatile phenols, like 4-vinylphenol (4-VP) which originates from *p*-coumaric acid and 4-vinylguaiacol (4-VG), the decarboxylation product of ferulic acid, formed during thermal treatment and by enzyme activity, have been reported to influence the aroma and stabilise pigments of fruit juices (Fallico, Lanza, Maccarone, Asmundo, & Rapisarda, 1996; Rein, Ollilainen, Vahermo, Yli-Kauhaluoma, & Heinonen, 2005) and red wines (Morata, Gómez-Cordovés, Calderón, & Suárez, 2006).

Some of the 4-vinyl derivatives of hydroxycinnamic acids, 4-VG and 4-vinylsyringol (4-VS), the latter the decarboxylation product of sinapic acid, are inherent to the beer production process, where they are formed by specific enzymes during fermentation or by thermal decarboxylation (Callemien, Dasnoy, & Collin, 2006; Vanbeneden, Gils, Delvaux, & Delvaux, 2008). In addition, 4-vinyl derivatives are considered to contribute to the smokey aroma of cured meat products (Guillén & Ibargoitia, 1998). 4-VG and 4-VP are treated as food additives and are approved as flavouring agents by regulatory agencies (Joint Expert Committee on Food Additives (JECFA), 2001).

Despite numerous studies on the antioxidant behaviour of hydroxycinnamic acids, only a few works have focused on the antioxidant activity of their 4-vinyl derivatives. 4-VG and 4-VS are reported to be potent antioxidants (Fujioka & Shibamoto, 2006; Koski, Pekkarinen, Hopia, Wähälä, & Heinonen, 2003; Vuorela, Meyer, & Heinonen, 2004). According to Fujioka and Shibamoto (2006), 4-VG, an important phenolic compound found in coffee, exhibits antioxidant activity comparable to that of α-tocopherol. The decarboxylation product of caffeic acid, 4-vinylcatechol (4-VC), acts as an inhibitor for phenylalanine hydrolase and some monooxygenases. 4-VS is an effective scavenger of the DPPH· radical (Koski et al., 2003; Vuorela, Kreander, et al., 2005) and also a strong antioxidant against protein and lipid oxidation (Vuorela, Salminen, et al., 2005). Further, 4-VS is known for its scavenging capacity against the endogenous mutagen peroxynitrite (Kuwahara et al., 2004) and showed peroxyl radical-scavenging activity (Wakamatsu et al., 2005). 4-VS, also referred to as canolol (named after its isolation from crude canola oil) can be produced by decarboxylation of sinapic acid during the pressing process or roasting of seeds (Koski et al., 2003). According to Spielmeyer, Wagner, and Jahreis (2009), heating of rapeseed before pressing can lead to a higher canolol content in the oil. On the other hand, Andueza, Manzocco, de Peña, Cid, and Nicoli (2009) reported the pro-oxidant properties of 4-VC. Although decarboxylation products of hydroxycinnamic acids have been recognised as antioxidants, reports on systematic evaluation of the antioxidant properties of these derivatives determined under different conditions are lacking.

Our aim was to prepare the decarboxylation products 4-VP, 4-VG, 4-VS and 4-VC (Fig. 1) of four hydroxycinnamic acids, namely *p*-coumaric, ferulic, sinapic and caffeic acid, respectively, and to

* Corresponding author.
E-mail address: helena.abramovic@bf.uni-lj.si (H. Abramovič).

0308-8146/$ - see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.foodchem.2011.02.077

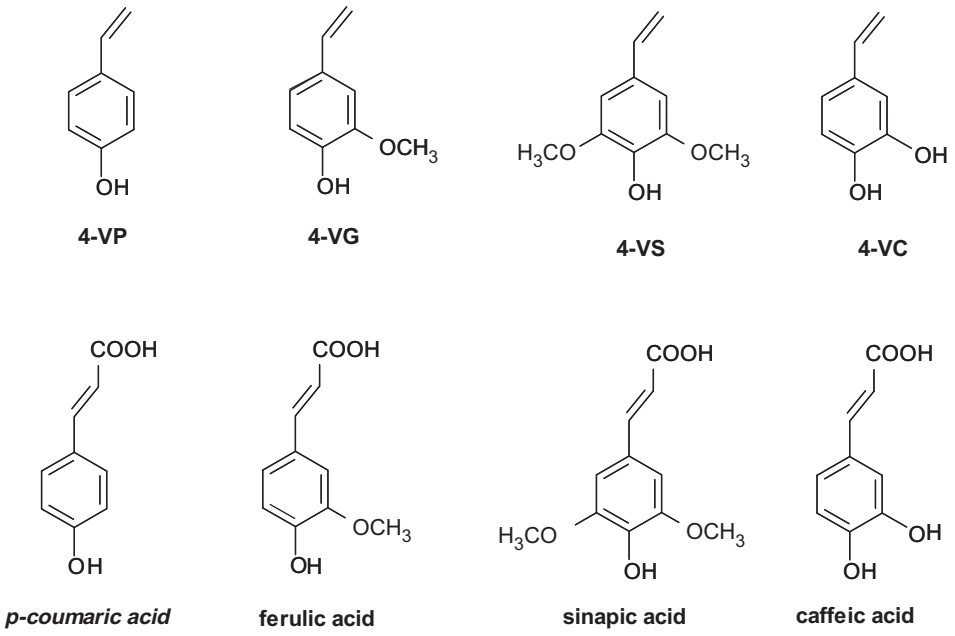

Fig. 1. Structural formula of 4-vinylphenol (4-VP), 4-vinylguaiacol (4-VG), 4-vinylsyringol (4-VS), 4-vinylcatechol (4-VC) and their corresponding hydroxycinnamic acids.

evaluate their antioxidant potential. The synthesised products were first subjected to a purification step by solid phase extraction (SPE). For confirmation of the purity and identity of the synthesised products, LC–DAD, LC–MS and NMR analysis were used. Antioxidant activity determinations based on different approaches were carried out. For this purpose the reducing power, the efficiency in scavenging the DPPH· radical, the superoxide anion radical and the efficiency in scavenging the alkylperoxyl radical generated in the linoleic acid emulsion system were determined. The results obtained were ranked and discussed with regard to the structure of each compound, in particular the presence of substituents on the phenyl ring. Further, the results obtained for the antioxidant activity of the 4-vinyl derivatives investigated were compared to those of the corresponding hydroxycinnamic acids.

2. Materials and methods

2.1. Reagents and solvents

Chloroform, ethanol (96%), methanol (99.9% and HPLC grade), diethyl ether, trichloroacetic acid, sodium carbonate, formic acid, acetic acid, ethyl acetate, dimethyl sulfoxide, potassium hexacyanoferrate(III), *p*-coumaric acid (98%) and ferulic acid (98%) were obtained from Merck (Darmstadt, Germany). β-Carotene, DPPH· reagent, linoleic acid (95%), Tween 20, nitroblue tetrazolium (NBT), β-nicotinamide adenine dinucleotide (NADH), phenazine methosulphate (PMS), Folin–Ciocalteu reagent, caffeic acid (98%) and sinapic acid (97%) were purchased from Sigma–Aldrich GmbH (Steinheim, Germany). Magnesium sulphate was obtained from Fluka Chemika (Buchs, Switzerland). Potassium dihydrogen phosphate, di-sodium hydrogen phosphate, sodium hydrogen carbonate and sodium acetate were obtained from Kemika (Zagreb, Croatia). N,N-Dimethyl formamide (DMF) and acetonitrile were obtained from Riedel-de-Haën (Seelze, Germany). Iron(III) chloride was purchased from Carlo Erba (Milano, Italy). Silicone oil was obtained from Julabo Labortechnik GmbH (Seelbach, Germany). For preparation of solutions ultrapure water (Milli-Q, Millipore, Billerica, MA) was used.

2.2. Synthesis of 4-vinyl derivatives from hydroxycinnamic acids

Ferulic, *p*-coumaric, caffeic or sinapic acid (500 mg) were dissolved in 5 mL of DMF. Sodium acetate (100 mg) was added as a catalyst for decarboxylation and the reaction mixture was heated on an oil bath at 130 °C (110 °C for caffeic acid) in a round-bottomed flask equipped with a condenser. Every 15 min, the reactions were monitored by thin layer chromatography, which was carried out using silica gel 60 F_{254} 20 × 20 cm plates on aluminium sheet (Merck) and 1% acetic acid in ethyl acetate (v/v) as solvent. The process of decarboxylation was completed within 1 h. Then 50 mL of Milli-Q water were added and the product was extracted three times with 10 mL of diethyl ether. The organic phase was collected and further extracted with 5 mL of freshly prepared carbonate buffer at pH 7. After that it was dried with anhydrous magnesium sulphate. The organic solvent was removed under reduced pressure. The crude synthesised products were subjected to purification by SPE.

2.3. Solid phase extraction

Two different procedures were used for purification, SPEa for 4-VP, 4-VS, 4-VG and SPEb for 4-VC. The products were passed through a 500-mg Strata-X cartridge (Phenomenex, Torrance, CA) previously conditioned with 8 mL of methanol (SPEa, SPEb), followed by 8 mL of 50% methanol (v/v) (SPEa) or by 8 mL of 30% methanol (v/v) (SPEb). 4-VG, 4-VS, 4-VP were eluted with 8 mL of 50%, 60% and 70% methanol (v/v) (SPEa) in contrast to the more polar 4-VC, which was eluted with 8 mL of 30%, 40% and 50% methanol (v/v) (SPEb). Those less polar compounds remaining were removed with 100% methanol. The purity of individual fractions was determined by LC–DAD. In the next step, eluates containing more than 95% of 4-vinyl derivatives were combined, adequately diluted and loaded onto a 60-mg Strata-X cartridge previously conditioned with 2 mL of methanol, followed by 2 mL of 25% methanol (v/v). Elution was performed with methanol. Eluates were filtered through a 0.20 μm filter and evaporated to dryness under vacuum at 30 °C. The products were weighed and redissolved in methanol. The final purity of synthesised products was determined by