

Tocopherols, tocotrienols and a new radical scavenging substance in raw and processed rapeseed and rapeseed oil

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Abstract

The content of tocopherols (alpha, gamma, delta), alpha-tocotrienol and plastochromanol-8 was determined both in raw and processed rapeseed as well as in different rapeseed oils after saponification and hexane extraction by normal-phase liquid chromatography using fluorescence detection. The results show that gamma-tocopherol represented 54% and alpha-tocopherol 32% of the total tocopherols. The content of plastochromanol-8 accounted to 12% of the total tocopherols. The alpha-tocotrienol content was below 1%, which was separated with a high resolution. Applying a modified method of tocopherol analysis by decreasing the isopropanol fraction in the solvent a strong phenolic antioxidant was found in the crude rapeseed oil. The compound was identified as 4-vinylsyringol also referred to as canolol, a decarboxylation product of sinapinic acid. For the structural identification 4-vinylsyringol was isolated from crude rapeseed oil. 4-Vinylsyringol was chemically synthesized and compared with the natural compound. 4-Vinylsyringol was detected in cold pressed oil and in roasted rapeseed but not in fully refined oils. Interestingly, the 4-vinylsyringol content increased after roasting the rapeseed.

Key words: rapeseed oil, *Brassica napus*, tocopherols, tocotrienols, plastochromanol-8, 4-vinylsyringol, canolol

Introduction

Rapeseed and rapeseed oil are particularly rich in phenolic substances. The content of the substances is substantially higher than in soy bean, cotton, peanut, and sun flower oil (Kozlowka et al., 1990, Ciz et al., 2002). The concentration of the phenols is higher in raw rapeseed oil than in refined rapeseed oil. The loss of the phenols by the processing is also associated with a decrease of the antioxidative capacity and stability (Ciz et al., 2002). The majority of the phenols in rapeseed oil are non-polar substances like the tocopherols and tocotrienols. In the oil processing the most polar phenols remain in the rapeseed meal. In rapeseed oil 4-vinylsyringol is the predominating polar phenol. 4-Vinylsyringol is a stronger antioxidant than alpha-tocopherol, vitamin C, and beta-carotene as far as alkyl peroxy radicals are concerned (Wakamatsu et al., in 2005). In other investigations anti-mutagenic and anti-inflammatory properties were also proved (Wakamatsu et al., 2005, Vuorela et al., 2005).

Material and Methods

2 g of the rapeseed oil were dissolved in 10 mL *n*-hexane and a volume of 20 μ L was used for injection. Tocopherols, tocotrienols, plastochromanol-8, and 4-vinylcanolol were separated by a Shimadzu 10A series HPLC using a 250 mm x 4 mm, 5 μ m Nucleosil-100 NH₂ column (Macherey-Nagel, Dueren, Germany), by isocratic elution with *n*-hexane/2-propanol (96:1.2 v/v) at a flow rate of 1.2 mL/min. The tocopherols, tocotrienols, plastochromanol-8, and 4-vinylsyringol were detected using a RF 10AXL fluorescence detector (excitation wave length: 298 nm, emission wave length: 325 nm) and diode-array detector (275 nm), respectively. The rapeseed oil was analyzed before and after roasting in a microwave for 6 min (600 Watt). The rapeseed phenolics were extracted with *n*-hexan/ethylacetat (50:50 v/v) in a soxhlet extractor. The extract was evaporated to dryness in a rotavapor at 40°C. The residues were redissolved in *n*-hexan (10 mL) and 1 mL of this was diluted with the mobile phase to 10 mL. The parameters for the HPLC analysis were the same as for the oils.

Results

The results show that gamma-tocopherol represents 54% and alpha-tocopherol 32% of total tocopherols. The content of plastochromanol-8 accounted to 12% of the total tocopherols. The alpha-tocotrienol content was below 1%, which was separated with a high resolution.

The alpha-tocopherol content was not affected by the oil processing. However, the alpha-tocotrienol, gamma-tocopherol and plastochromanol-8 content of cold pressed and crude oil was higher than that of refined oil (Tab. 1). Applying a modified method of tocopherol analysis by decreasing the isopropanol fraction in the solvent a strong phenolic antioxidant was found in the crude rapeseed oil. The compound was identified as 4-vinylsyringol, a decarboxylation product of sinapinic acid. For the structural identification 4-vinylsyringol was isolated from crude rapeseed oil. 4-Vinylsyringol was synthesized and compared with the natural compound. 4-Vinylsyringol was detected in crude oil, cold pressed oil and in roasted rapeseed but not in fully refined oils and steam stripped oil (Fig 1). Because 4-vinylsyringol was found in canola oil it is also referred to as canolol. The level of canolol in rapeseed was determined before and after roasting. The canolol content increased after roasting the rapeseed by approximately 120 times (Fig. 1 and 2).

Table 1. Tocopherol and tocotrienol content in the tested rapeseed oils [mg/100g]

Rapeseed oil	alpha-tocopherol	alpha-tocotrienol	gamma-tocopherol	delta-tocopherol	plastochoymanol-8	total tocopherols
Cold pressed	29.7	1.2	45.1	1.8	10.1	87.9
Refined	28.4	< 0.2	40.0	1.6	7.0	77.0
Kernel oil	20.2	1.0	43.3	2.1	10.8	77.4
Washed	30.4	< 0.2	40.6	1.3	10.5	82.8
Crude	35.0	< 0.2	48.4	2.2	10.1	95.7

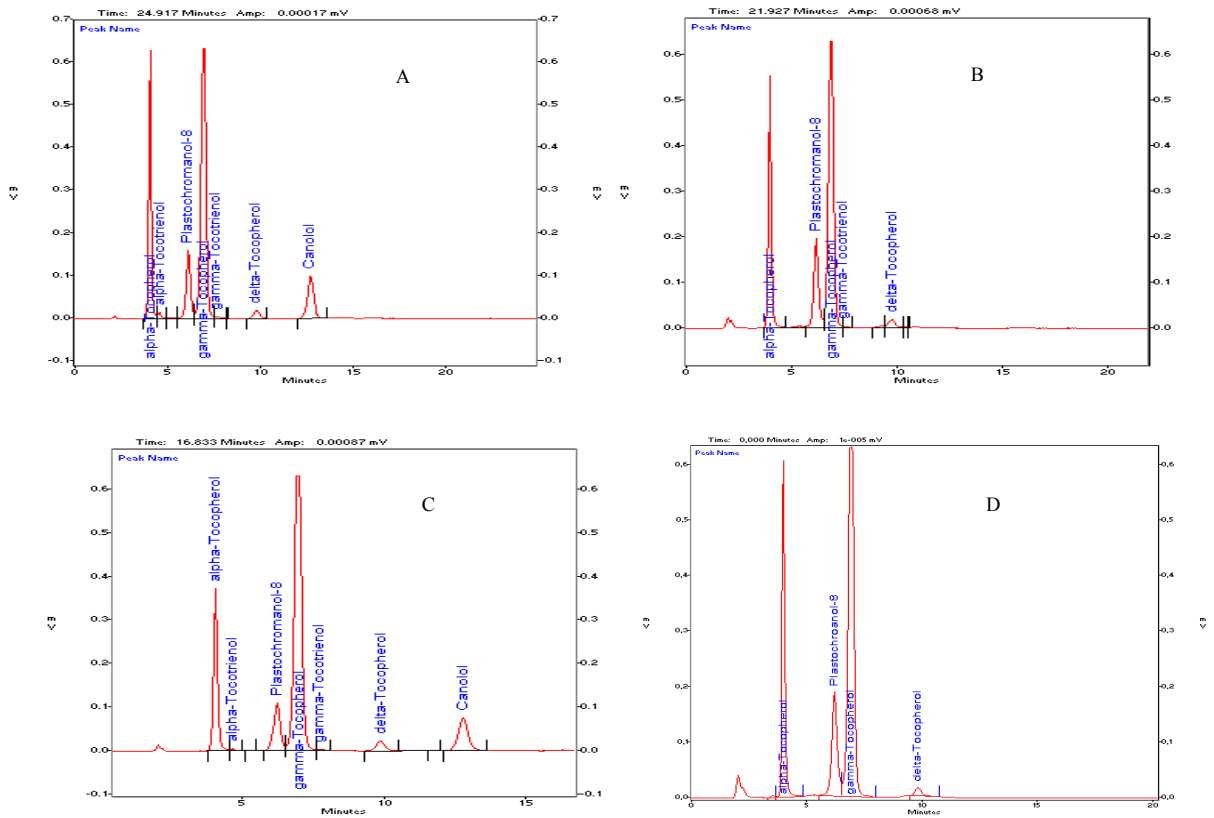


Fig. 1. HPLC chromatogram of different rapeseed oils samples; A: cold pressed rapeseed oil; B: refined rapeseed oil; C: kernel oil; D: washed rapeseed oil

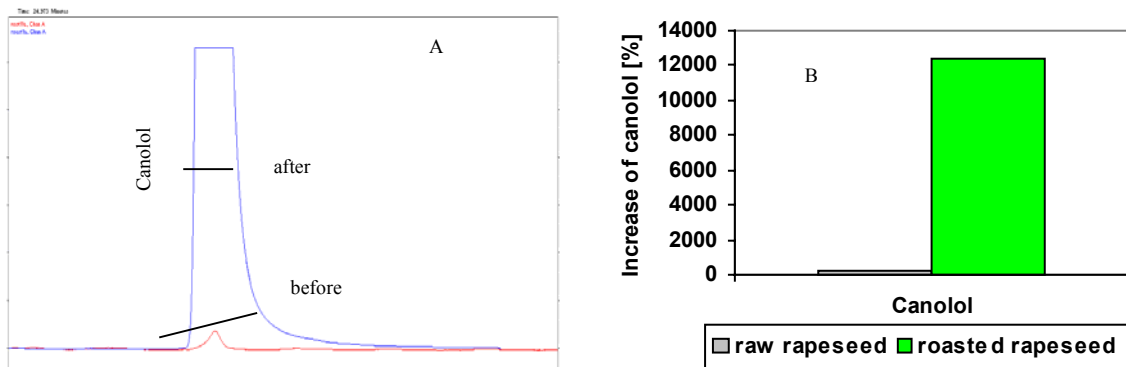


Fig. 2. A: HPLC chromatogram of rapeseed sample before and after roasting; B: Comparison between the increase of canolol before and after roasting

Conclusion

In conclusion, crude rapeseed oil comprises of a high content of antioxidants like tocopherols, tocotrienols, plastochoymanol-8, and canolol. The present results suggest that the amount of the antioxidants particularly the content of canolol is reduced or completely removed after refining. Notable is the abundant increase of canolol after roasting rapeseed.

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