TOCOPHEROLS OF THE GRAPE

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The amounts of the various forms of tocopherols in the grape have not been studied. There is only information in the presence of vitamin E in a lipid extract of grape seed [1].

We have investigated the qualitative and quantitative composition of various forms of tocopherols in the component parts of the grape (flesh, skin, seeds) of the cultivated grape *Vitis vinifera* L., of the variety Pinot gris.

The tocopherols were isolated as part of the total lipid fraction separately from the flesh, seeds, and skin by a modification of Bligh and Dyer's method [2]. All the operations up to the isolation and separation of the tocopherols were carried out in an inert gas atmosphere. The tocopherols were freed from contamination with other lipids by hydrolysis in a 12% ethanolic solution of KOH in the presence of pyrogallol (80°C, 5 min). The unsaponifiable fraction was extracted with diethyl ether and washed free from impurities. The various forms of tocopherols were separated in a thin layer of Woelm silica gel in the hexane–diethyl ether–sulfuric acid–acetic acid (70:27:0.4:1) solvent system [3]. The spots of the tocopherols were detected by spraying with a 2% ethanolic solution of p-dimethylphenylenediamine. Three spots were detected, with R_f 0.52, 0.42, and 0.34, which were assigned, respectively, to the α-, (β + γ)-, and δ-tocopherols. It must be mentioned that β- and γ-tocopherols, which are position isomers, form a "critical pair" which is not separated under the given conditions [3]. As markers in the TLC separation we used synthetic α-tocopherol and γ- and δ-tocopherols which we had isolated from linseed and soybean oils, respectively.

For the quantitative determination of the forms of tocopherols, they were eluted from the unsprayed part of the plate with ethanol and their amounts were determined by the iron–bipyridyl method [4]. Calibration graphs were plotted for the individual α-, γ-, and δ-tocopherols.

The highest concentration of tocopherols was found in the grape seeds — 110.4 mg/kg (α, 39.3%; β + γ, 36.2%; δ, 24.5%), the amount ot tocopherols in the flesh being 4.6 mg/kg (α, 59.4%; β + γ, 26.9%; δ, 13.7%), and the amount in the skin being 21.2 mg/kg (α, 49.2%; β + γ, 31.0%; δ, 19.8%).

Since Pinot gris grapes are a technical variety used predominantly in wine making, the results obtained indicate that the main structural component of the grape responsible for the passage of tocopherols into wine is represented by the seeds. In actual fact, the wine material obtained by the juice expressed from the flesh contained only 0.1 mg/liter of α-tocopherol, while the wine material obtained under identical conditions but using the whole grape contained 1.2 mg/liter of tocopherol (α, 81.9%; β + γ, 18.1%). The absence of δ-tocopherols from the finished product is due, to all appearances, to the fact that this form of tocopherol possesses the highest antioxidant activity and consequently is most susceptible to oxidative transformation [5].

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