

Quantitative analysis of anthocyanins in natural fruit Juices and evidence-based rationale for their inclusion as bioactive agents in individuals with increased physical activity

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INTRODUCTION & AIM

Anthocyanins, as a class of water-soluble flavonoid pigments are natural antioxidants with proven biological activity. Recently, **anthocyanin supplementation** has been recognized as one of the approaches for faster recovery after intense physical activity, reduction of inflammation, and modulation of vascular function. The aim of this study was to analyze the concentration of anthocyanins in several natural fruit juices obtained from various plants: pomegranate (*Punica granatum*), blueberry (*Vaccinium myrtillus*), grape (*Vitis vinifera*), elderberry (*Sambucus nigra*), and chokeberry (*Aronia melanocarpa*) in order to justify their nutritional and functional potential.

METHODS

pH

The pH value was measured using a pH meter, calibrated with standard buffers pH 4.00 and 7.00, and the measurement was performed directly in the sample after stabilization of the electrode (~30 seconds).

Total acids

The total acids were determined by titration with 0.1 M NaOH to an endpoint of pH 7.0, using an automatic titrator, and the results were expressed as g tartaric acid/L and mmol H⁺/L (equivalents of H₂SO₄).

Brix

The soluble solid content was determined with a digital refractometer, calibrated with distilled water (0.0 °Brix), and the results were presented as % soluble solids (°Brix).

Total anthocyanins – Diatron P500

The total concentration of anthocyanins was determined using a Diatron P500 clinical analyzer with the Oenolab Diagnostics reagent kit, where the absorbance was measured at 505 nm, and the concentration (mg/L) was calculated according to the formula: $\text{mg/L} = A_{505} \times 850$.

Total anthocyanins Cary Scan 50 VIS

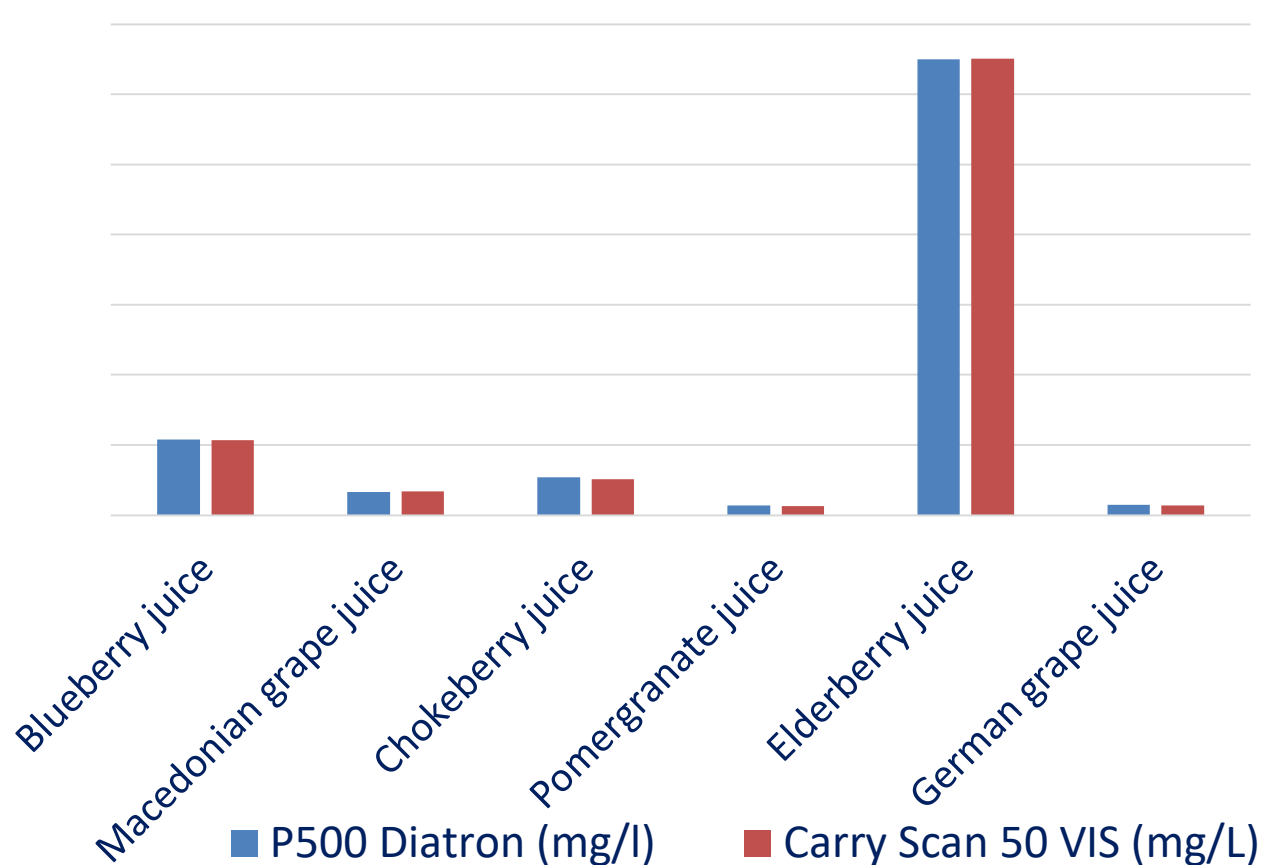
The anthocyanins were also determined with a UV-VIS spectrophotometer, after extraction with 2% HCl in ethanol, where the absorbance was measured at 540 nm, and the concentration (mg/L) was calculated according to the formula: $\text{mg/L} = A_{540} \times 26 \times 15$.

Monomeric anthocyanins – pH differential method

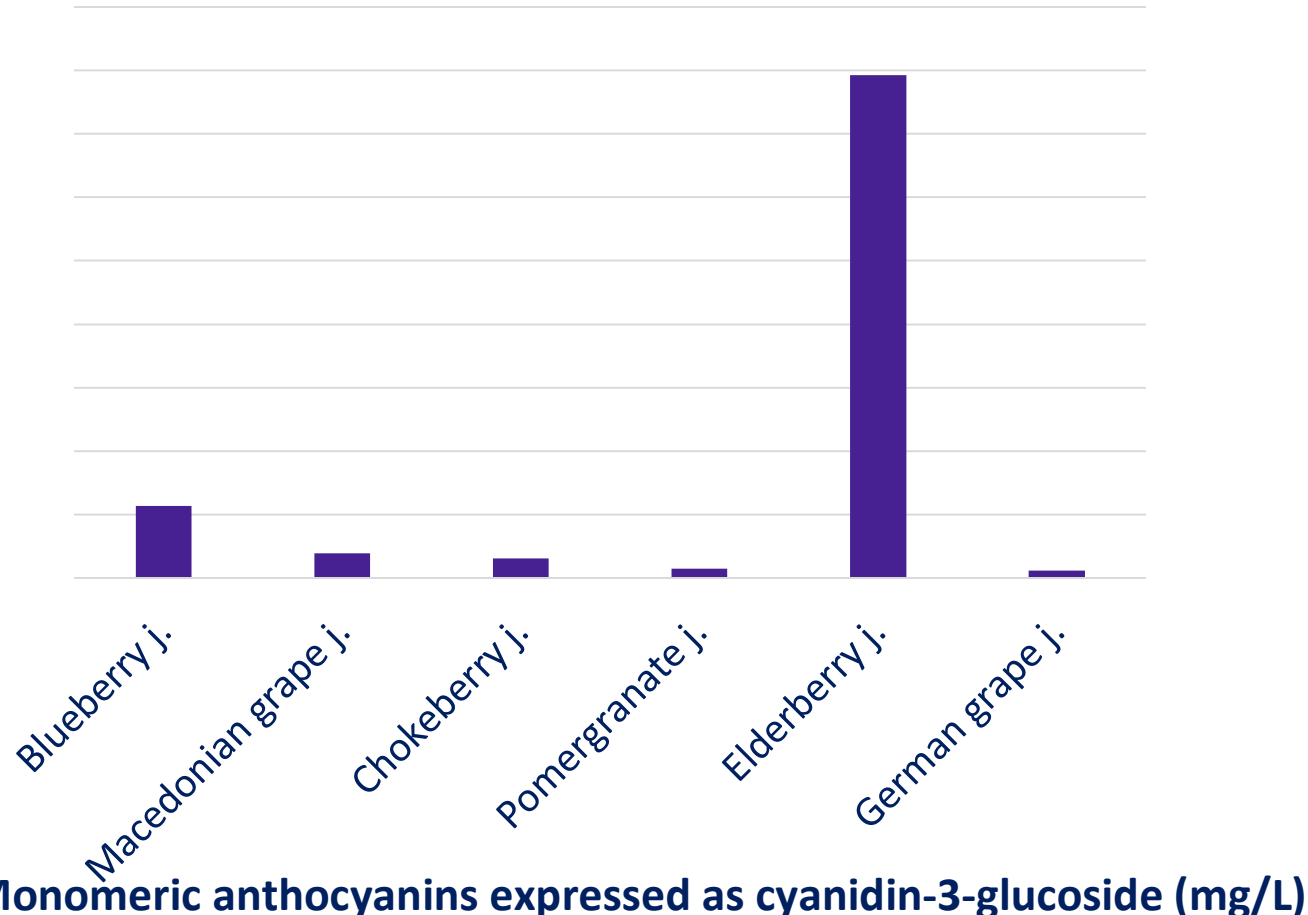
The monomeric anthocyanins were determined with the pH differential method, where the samples were prepared in two buffers: KCl–HCl (pH 1.0) and Na-acetate–HCl (pH 4.5). The absorbance was measured at 520 and 700 nm with a UV-VIS spectrophotometer, and the concentration was expressed as cyanidin-3-glucoside or malvidin-3-glucoside equivalent.

RESULTS

Graph 1 Comparison between the precision of the two methods for determining the total anthocyanins in the analyzed samples



Graph 2. Concentration of monomeric anthocyanins in the analyzed samples



DISCUSSION

The obtained results showed significant differences among the samples. Pomegranate and blueberry juices had the lowest pH values (**3.16 and 3.29**, respectively), which increases the stability of their anthocyanins. Pomegranate and elderberry showed the highest acidity, while chokeberry juice had the highest Brix value (**26.2 °Brix**), indicating a high concentration of soluble solids. The highest concentration of total anthocyanins was measured in elderberry juice (**>3,200 mg/L**), and elderberry juice also dominated in monomeric anthocyanins (**over 1,500 mg/L**). This phytochemical characterization is of particular importance for the pharmaceutical industry, as the obtained results are consistent with the literature and confirm the value of these juices as natural sources of bioactive components with potential application in functional and sports pharmacy.

CONCLUSION

This data confirms the potential for using precisely dosed fruit juices as an alternative to commercial encapsulated supplements, with the added benefit of natural sugars, organic acids, and polyphenols present in the matrix. Consequently, the integration of fruit juices into routine supplementation represents an accessible and effective approach for supporting health and sports performance, with potential for further standardization and application in functional foods.