

**Development and validation of an RP-HPLC method  
for the determination of caffeine and preservatives (sodium benzoate  
and potassium sorbate) in sports and energy drinks**

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**Abstract**

Preservatives in sports and energy drinks play a crucial role, especially in maintaining their quality and safety, while caffeine is included mainly in energy drinks as a stimulant to enhance alertness, focus, and physical performance. In this study, a rapid, simple and sensitive RP-HPLC method for simultaneous determination of caffeine, sodium benzoate (SB) and potassium sorbate (PS) in sports and energy drinks was developed. Separation of analytes was achieved using Poroshell 120 EC - C18 (3.0× 50 mm, 2.7 μm) analytical column. The best results were obtained by isocratic elution with mobile phase consisting of acetonitrile and phosphate buffer (KH<sub>2</sub>PO<sub>4</sub>/H<sub>3</sub>PO<sub>4</sub>) with pH = 3.8 in volume ratio 85/15 (V/V), flow rate of 1.0 mL/min, and a constant column temperature of (25 °C). The detection was performed at 230, 260 and 275 nm. The developed RP-HPLC method was validated regarding the selectivity/specificity, linearity ( $R^2 \geq 0.996$ ), accuracy (recoveries between 95.99 and 101.5%), precision (intra-day RSD < 1%), limit of detection (LOD) and limit of quantification (LOQ). All the validation parameters were within the acceptance range. The linearity of the RP-HPLC method was confirmed with  $R^2$  values higher than 0.996 in the concentration range of 49.95 - 449.55 mg/L for caffeine, 29.97 - 269.73 mg/L for SB, and 60.53 - 544.85 mg/L for PS. Intra-day repeatability data demonstrated that proposed method was reliable, and it is characterized by high precision (RSD ≤ 0.49%). The recovery data ranged between 95.99 - 101.50%, indicating excellent recoveries for SB, PS and caffeine by the proposed RP-HPLC method. LODs were 0.015 mg/L for caffeine, 0.025 mg/L for SB, and 0.018 mg/L for PS, while LOQs were 0.083 mg/L for SB, 0.059 mg/L for PS and 0.049 mg/L for caffeine. The proposed method was successfully applied for determination of caffeine, SB and PS in sports and energy drinks.

Keywords: Caffeine · Potassium sorbate · RP-HPLC · Sodium benzoate · Sports and energy drinks.