

A highly selective two-way purification method using liquid chromatography for isolating α S2-casein from goat milk of five different breeds

ABSTRACT

The main challenges in the purification of α S2-casein are due to the low quantity in milk and high homology with other casein subunits, i.e., α S1-casein, β -casein, and κ -casein. To overcome these challenges, the aim of this study was to develop a two-step purification to isolate native α S2-casein in goat milk from five different breeds; British Alpine, Jamnapari, Saanen, Shami, and Toggenburg. The first step of the purification was executed by anion-exchange chromatography under optimal elution conditions followed by size exclusion chromatography. Tryptic peptides from in-gel digestion of purified α S2-casein were sequenced and analyzed by LC-ESI-MS/MS. From 1.05 g of whole casein, the highest yield of α S2-casein (6.7 mg/mL) was obtained from Jamnapari and the lowest yield (2.2 mg/mL) was from Saanen. A single band of pure α S2-casein was observed on SDS-PAGE for all breeds. The α S2-casein showed coverage percentage of amino acid sequence from 76.68 to 92.83%. The two-step purification process developed herein was successfully applied for isolating native α S2-casein from goat milk with high purity, which will allow for future in vitro studies to be conducted on this protein.

Keyword: Goat α S2-casein; Protein identification; Goat breed; Ion-exchange chromatography; Size-exclusion chromatography; LC-ESI-MS/MS