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Optimization of the high-performance liquid chromatographic separation of a mixture of natural and synthetic corticosteroids

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Abstract-

Systematic optimization of the HPLC separation of a mixture of natural and synthetic corticosteroids was carried out for screening purposes. The method involves binary, ternary or quaternary mixtures containing water, methanol, acetonitrile and tetrahydrofuran. It was possible to separate thirteen out of fourteen corticosteroids contained in a sample in about 26 min, with a 5-?m Hypersil-C18 (250 mm × 4.6 mm I.D.) column thermostated at 30°C, using a mobile phase composed of water-tetrahydrofuran (72:28, v/v). This separation was not improved using other C8 or C18 columns. The effect of temperature on the separation of these compounds was also studied. Calibration graphs were established for each corticosteroid up to 8 ?g/ml using indapamide as internal standard. The detection limits were in the range 0.02-0.14 ng. The optimized method was applied to urine samples spiked with corticosteroids and showed potential for future applications.

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