

## HPLC Separation of Some Unsaturated and Saturated Fatty Acids

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HPLC procedure for the determination of saturated fatty acids (from C<sub>8</sub> to C<sub>22</sub>) and some unsaturated fatty acids containing eighteen carbon atoms is described. Milk and intestinal digesta samples were hydrolyzed with 2 mol L<sup>-1</sup> NaOH. The hydrolysates were acidified to pH ~ 2 and free fatty acids were extracted with dichloromethane. Fatty acids were then assayed after derivatization with 2,4'-dibromoacetophenone in the presence of triethylamine. The separation of derivatized fatty acids was achieved using two C<sub>18</sub> columns and UV detection at 256 nm. Derivatized conjugated linoleic acid isomers (CLA) in the effluent were monitored using UV detection at 235 and 256 nm (method I). Gradient elution with UV detection at 234.5 nm (method II) allowed the determination of CLA without derivatization. The low detection limits (from  $2.3 \times 10^{-4}$  to 5.1 pg) for assayed fatty acids, good „purity” of fatty acid peaks (~100%) and very simple procedure for preparation of free fatty acid extracts point to the satisfactory sensitivity and reliability of the presented methods.