## **Annotation:**

This work is concerned with the development and application of high performance separation methods suitable for determination of clinically considerable fragments of collagen and elastin as the most important proteins of connective tissues. For their analyses, various chromatographic and electrophoretic methods have been used, including high performance liquid chromatography (HPLC), capillary zone electrophoresis (CZE) and micellar electrokinetic capillary chromatography (MEKC).

This work has aimed at optimization of the developed and modified analytical separation procedures and methods and their utilization for determination of clinically important markers of connective tissue degradation. The key validation parameters of the analytical methods used in this work were evaluated too, which enabled their comparison with other approaches. One of the most important results of this work is thus high sensitivity, precision, repeatability and robustness of the methods which was found in a number of biological sample types. Successful was also very good HPLC separation of the analyzed cross-links in both collagen and elastin tissues.

High performance methods are described which have been suggested and optimized for separation and quantitative determination of pentosidine, pyridinolines and desmosines as the most important collagen and elastin cross-links. The developed and tested methods (based on more than 6 000 measurements) have already been successfully applied in analysis of more than 1 000 real patient samples and the results obtained utilized in clinical practice.

## **Keywords:**

HPLC; Capillary Electrophoresis; Collagen; Elastin; Pentosidine; Pyridinolines; Desmosines; Connective Tissue; Cross-links