

## **Abstract - The application of electrophoretic and chromatographic methods in clinical research**

RNDr. Jana Faulknerová Matějčková

Malondialdehyde (MDA) is considered to be the main biomarker for oxidative damage to biomembranes and an elevated level of this compound can act as an indicator for a number of diseases. I determined MDA using a HPLC method based on derivatization of blood plasma samples with 2,4-dinitrophenylhydrazine. I used HPLC to separate the MDA derivatives from the other components of the blood plasma and detected them at 307 nm. The MDA analysis time did not exceed 4.5 min. The sensitivity (detection limit  $0.27 \mu\text{mol l}^{-1}$ ) and repeatability of the determination of MDA are sufficient for monitoring the MDA level in real blood plasma samples.

My work also describes monitoring the level of the low-molecular secondary antioxidant of uric acid, whose concentration in the blood attains values of 140 to  $350 \mu\text{mol l}^{-1}$  and the secondary enzymatic antioxidant of superoxide dismutase (SOD). The uric acid level was analysed by the capillary electrophoresis and spectrophotometric methods, where the two methods yielded comparable results. SOD was analysed using a spectrophotometric kit.

The MDA levels were measured for vegans (who refuse to eat any food of animal origin, meat, eggs and also dairy products), with an average value of MDA  $5 \mu\text{mol l}^{-1}$  which is a statistically lower value than for a "non-vegan" population of comparable age. Further, the MDA level was monitored for patients with diabetes mellitus type I, where the average MDA level was  $9 \mu\text{mol l}^{-1}$ , compared with diabetes mellitus type II, with an average level of  $7 \mu\text{mol l}^{-1}$ , and healthy controls, with an average value of MDA  $6 \mu\text{mol l}^{-1}$ . The MDA level was also studied in the blood of patients with gynaecological tumours, where an average MDA concentration of  $11 \mu\text{mol l}^{-1}$  was found. These results were also statistically significant compared to health control subjects.

In the last part of the work, I demonstrated that surgical removal of the tumour leads to an increase in the plasmatic MDA concentration and that administration of a single dose of vitamin E is capable of substantially reducing the damage to the organism from oxidative stress. Administration of vitamin E did not lead to a reduction in the levels of either uric acid or SOD.