## **ABSTRACT**

The thesis is focused on a determination of heparin and chondroitin sulfate, using flow injection analysis with spectrofluorimetric detection. The determination is based on the interaction of negatively charged heparin, chondroitin sulfate resp., with a cationic dye (azure B or phenosafranine) which is manifested by the decrease in fluorescence intensity of the dye in its emission maximum. The optimal conditions for the determination in static mode were found, and calibration dependencies were measured. The conditions of FIA were optimized and following parameters were established: the volume of dispensed sample of 100 ml, the length of the reaction coil 60 cm, the flow rate 0.7 ml min<sup>-1</sup>, the concentration of azure B 1.6×10<sup>-5</sup> mol dm<sup>-3</sup>, the concentration of phenosafranine 3.5×10<sup>-5</sup> mol dm<sup>-3</sup>. For the determination of heparin using azure B it was found:  $LOD = 0.023 \text{ IU ml}^{-1}$ ,  $LOQ = 0.186 \text{ IU ml}^{-1}$ , and linear dynamic range 0.19–1.43 IU ml<sup>-1</sup>. For the determination of heparin using phenosafranine it was found:  $LOD = 0.102 \text{ IU ml}^{-1}$ ,  $LOQ = 0.192 \text{ IU ml}^{-1}$ , and linear dynamic range 0.19–1.79 IU ml<sup>-1</sup>. For the determination of chondroitin sulfate using azure B it was found:  $LOD = 0.58 \text{ mg dm}^{-3}$ ,  $LOQ = 2.37 \text{ mg dm}^{-3}$ , and linear dynamic range 2.37–8.32 mg dm<sup>-3</sup>. The developed determination was applied to the determination of heparin, resp.chondroitin sulfate, in pharmaceutical preparations.

## **Keywords**

Azure B

Chondroitin sulfate

Flow injection analysis (FIA)

Glycosaminoglycans

Heparin

Phenosafranine

Spektrofluorimetric detection