Abstract

This bachelor thesis solves the question of sulphated metabolites of silybin by their interactions with sulfatases. Silybin is flavonolignan prepared from the seeds of milk thistle (*Silybum marianum)*. Sulfate of silybin (metabolite) has not been isolated from organism so far thus its exact structure has not been determined.

In this work synthesis of silybin-7,23-disulfate from natural silybin is presented. Silybin disulfate was isolated and spectrally characterized (NMR, MS). In the next step the kinetics of the reaction *p*NPS with sulfatase *from Helix pomatia* was measured: K_m = 0.0494 mmol/l a V_{max} = 0.0325 mmol/dm^3/min. Conditions for sulfatase reaction from *Helix pomatia* were selected according to literature.

One of the main goal was to determine, whether silybin-7,23-disulfate is a substrate and/or an inhibitor of sulfatase from *Helix pomatia.* Activity measurements of the sulfatase from *Helix pomatia* in the presence of various concentrations of silybin-7,23-disulfate enabled us to determine that silybin disulfate at concentration higher than 0.2 mM is a strong inhibitor of the sulfatase tested.

Series of reactions with sulfatase from *Helix pomatia* were performed to determine whether silybin-7,23-disulfate is a substrate. Reactions were monitored by HPLC and it was demonstrated that the sulfatases from *Helix pomatia* decomposes silybin-7,23-disulfate (to the concentration 0.2 mM). From measuring, we suppose, that the incurred product decomposed from silybin-7,23-disulfate is silybin.