

9. SUMMARY

Family Asteraceae and Cichoriaceae are a broadly spread taxons comprising a great number of species. Many plants with important pharmacological and toxicological properties belong to this family. Within the systematic research, 30 taxons growing in the Middle Europe have been assayed for their influence on the platelet aggregation process.

The antiplatelet activity of herbal extracts was assayed *in vitro* on the model of human platelets-rich plasma (PRP; 250×10^9 platelets/L) at a concentration of 500 $\mu\text{g/mL}$ PRP. Arachidonic acid (AA; final concentration in cuvette was 0.5 mM), adenosine diphosphate (ADP; final conc. 10 μM), collagen (COL; final conc. 2 $\mu\text{g/mL}$) and thrombin (TR; final conc. 1 U/mL) were used as agonists of platelet aggregation.

From the tested plants, only extracts from flowering tops of *Cnicus benedictus* and *Galinsoga ciliata* were found to have antiplatelet activity. *Cnicus benedictus* inhibited the aggregation induced by AA ($15 \pm 5\%$), ADP ($12 \pm 2\%$) and COL ($42 \pm 6\%$). *Galinsoga ciliata* decreased platelet aggregation triggered by ADP ($34 \pm 6\%$). Extracts exhibited no influence on TR induced aggregation.

Cnicus benedictus was chosen as a suitable plant for further isolation. 3.34 kg of dry cut flowering tops were percolated with ethanol and in the next step, petrolether, chloroform, ethyl-acetate and butan-1-ol portion was prepared from this extract. All these extracts were tested on antiplatelet activity; chloroform extract was the most active one and it was subsequently chromatographically separated.

The individual fractions were then bioassayed and fraction 47–59 was selected for isolation of active substances (average activity in the case of three agonists: ADP

31 ± 4 %, AA 47 ± 6 %, collagen 38.5 ± 5 %). After crystalization, it yielded 7.24 g of a crystalline substance named CB-01. It was re-crystallized and a white and fine compound was obtained. The isolated compound was identified as a germacranolide sesquiterpenic lactone cnicin. Measured values of EC₅₀ for cnicin were: 552.3 ± 29 μM for AA, 1377 ± 68 μM for ADP and 215.9 ± 21 μM for COL. None activity was observed for TR induced aggregation. The antiplatelet activity of cnicin was lower than for acetylsalicylic acid (16.1 ± 1.2 μM for AA, 84.8 ± 11 μM for COL) and dipyridamole (122.5 ± 13 μM for ADP).