ABSTRACT

Charles University, Faculty of Pharmacy in Hradec Králové
Department of Pharmacognosy and Pharmaceutical Botany
Candidate: Viriyanata Wijaya, M.Sc.
Supervisor: Prof. RNDr. Lubomír Opletal, CSc.
Title of Doctoral Thesis: Alkaloids of *Dicranostigma franchetianum* (Prain) Fedde and their selected biological activity

Dicranostigma franchetianum (Prain) Fedde (Papaveraceae) is one of the representatives of the small genus *Dicranostigma* Hook. f. & Thomson. *D. franchetianum* (Prain) Fedde has been selected for the phytochemical investigation according to the screening study. In the primary screening of the alkaloid extract for cholinesterases inhibition, the inhibitory value was high (*h*AChE/*h*BChE, IC_{50} μg/ml; 1.67 ± 0.11 and 3.85 ± 0.31) and together at least 15 alkaloids were found in the extract. The primary ethanol extract was prepared from 11.8 kg of dry herb (Garden of Medicinal Plants, Faculty of Pharmacy in Hradec Kralove). Using common chromatographic methods, 21 isoquinoline alkaloids of various structural types were isolated. All compounds have been identified using various spectrometric techniques (GC-MS, HPLC-MS, and 1D- and 2D-NMR, optical rotatory. The alkaloids obtained in sufficient amounts were determined for human acetylcholinesterase (*h*AChE), human butyrylcholinesterase (*h*BChE), and prolyloligopeptidase (POP). In the *h*AChE assay, the quaternary alkaloids berberine (**DF-18**) and palmatine (**DF-19**) demonstrated the strongest inhibition potency, with values of IC_{50} 0.71 ± 0.10 μM and 1.1 ± 0.1 μM, respectively. Whereas in the *h*BuChE assay, benzophenanthridine alkaloids 6-ethoxydihydrochelerythrine (**DF-03**) and sanguinarine (**DF-07**) showed the strongest inhibition potency, with values of IC_{50} 3.2 ± 0.2 μM and 3.8 ± 0.6 μM, respectively. Furthermore, sanguinarine (**DF-07**) was the most effective in inhibiting POP with IC_{50} values of 1.5 ± 0.1 μM higher than baicalein as standard (IC_{50} = 14 ± 1 μM). However, CNS availability was calculated by applying the blood-brain barrier (BBB) score. Based on the score, compound **DF-02, -03, -05, -07, -11, 18-20** was assumed that compounds can pass through the BBB.

Furthermore, the compounds isolated in sufficient amounts have been tested for their antimycobacterial potential against five *Mycobacterium* or *Mycolicibacterium* strains. Alkaloids 6-ethoxydihydrochelerythrine and bis-[6-(5,6-dihydro-chelerythrinyl)]ether, which

contain a benzophenanthridine skeleton in their structure, showed moderate activity against all the tested mycobacterial strains (MICs 15.625–31.25 μg/ml).

Keywords: *Dicranostigma franchetianum* (Prain) Fedde, Papaveraceae, alkaloids, biological activity, acetylcholinesterase, butyrylcholinesterase, prolyloligopeptidase, antimycobacterial activity