

Major Constituents and Cytotoxic Effects of *Ajuga chamaecistus* ssp. *tomentella*

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The *n*-butanolic fraction of a methanolic extract (80%) from aerial parts of *Ajuga chamaecistus* ssp. *tomentella* was analysed using different chromatographic methods. Column (CC) and high-performance liquid chromatography (HPLC) were used for isolation and purification. ¹³C, ¹H NMR, H-H COSY, HSQC, HMBC, and ESI-MS were employed for identification of the compounds isolated from this fraction. The structures of the compounds were determined to be *cis*-melilotoside (**1**), *trans*-melilotoside (**2**), lavandulifolioside (**3**), 20-hydroxyecdysone (**4**), leonoside B (**5**), martynoside (**6**), ajugalactone (**7**), makisterone A (**8**), and 24-dehydroprecysterone (**9**). This is the first report on the presence of *cis*- and *trans*-melilotoside in *Ajuga* species. Cytotoxic evaluation of the *n*-butanolic fraction, *cis*- and *trans*-melilotoside against cancer (T47D, HT-29, and Caco-2) and normal (NIH 3T3) cell lines by the mitochondrial tetrazolium test (MTT) showed no cytotoxic effects up to 400 µg/mL. The results of this study suggest that melilotoside, phenylethyl glycosides, and phytoecdysteroids are the main constituents of the *n*-butanolic fraction of *Ajuga chamaecistus* ssp. *tomentella*.

Key words: *Ajuga chamaecistus* ssp. *tomentella*, Melilotoside, Cytotoxic Effect