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Chemical characterization of three *Thymus* species: *T. herba-barona*, *T. pseudolanuginosus* and *T. caespititius*

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The genus *Thymus*, belonging to *Lamiaceae* family, is rich in medicinal and aromatic species and well-known by several health promoting activities [1,2]. Despite this genus has been extensively studied, some species remain unexploited. In this study, *Thymus herba-barona*, *Thymus pseudolanuginosus* and *Thymus caespititius* decoctions were screened for their phenolic constituents by ultra-high performance liquid chromatography coupled to diode array detector and an electrospray mass spectrometer (UHPLC-DAD-ESI-MSn) operating in negative mode.

The three aqueous extracts were rich in caffeic acid derivatives, mainly rosmarinic acid (MW 359) and its structural isomers, that accounted for 55.8 ± 2.8 mg/g in T. herba-barona and 40.2 ± 0.9 and 43.2 ± 3.2 mg/g in *T. pseudolanuginosus* and *T. caespititius*, respectively. In turn, other depsides were differently distributed in the three Thyme extracts: while dihydro-salvianolic acid B (MW 716 Da) and caffeoyl rosmarinic acid were particularly representative in *T. herba-barona*, salvianolic acids K ([M-H]⁻ at *m/z* 555 493 359) and B ([M-H]⁻ at *m/z* 717 519 475) were found in moderate amounts in *T. caespititius* extract. On the other hand, *T. pseudolanuginosus* was clearly distinguished by its richness in the flavone luteolin-O-glucuronide ([M – H]⁻ at *m/z* 461→285).

Overall, this work is an important contribution for the phytochemical characterization of these three *Thymus* species, which are poorly explored.

Table 1. *Identification and quantification of main UHPLC eluting fractions by UHPLC-DAD-MSn of T. herba-barona, T. pseudolanuginosus, and T. caespititius aqueous extracts.*

	[M-H]⁻	T. h-b	T. p	T. c
Rosmarinic acid	359	55.8 ± 2.8	40.2 ± 0.9	43.2 ± 3.2
Luteolin-O-glcuronide	461	10.5 ± 0.2*	54.1 ± 0.6	17.3 ± 1.1
Dedihydro-Salvianolic Acid B	715	10.8 ± 0.1	-	-
Salvianolic Acid B	717	-	-	6.9 ± 0.5
Salvianolic Acid K	555	D	-	10.5 ± 0.1
Caffeoyl Rosmarinic acid	537	10.5 ± 0.06	D	D

**Structural isomer; D: detected*

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