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CHEMICAL CONSTITUENTS AND CYTOTOXIC ACTIVITY OF *Smallanthus maculatus*

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Smallanthus maculatus (Cav) H. Rob. (Asteraceae, nomenclatural synonym: *Polymnia maculata* Cav. Icon. [1, 2]) is used by the Highlands Mayas of Chiapas, Mexico, for the treatment of gastrointestinal diseases [3]. To our knowledge, no phytochemical investigation has been reported on *S. maculatus*; however, from *P. maculata* has been isolated *ent*-kaur-16-en-19-oic acid, kauradiene-9,16-dioic acid, polymatin A, polymatin B, polymatin C [4], maculatin [5], melampodin D, and 9-desacetoxymelcanthin F [6]; consequently, these are chemical constituents of *S. maculatus* in reality. In our hands, the acetone extract from the aerial parts of *S. maculatus* was cytotoxically active (ED_{50} = 17.0 μg/mL against HCT-15 COLADCAR and ED_{50} = 18.4 μg/mL against OVCAR-5). Their bioguided analysis yield two active fractions: F-4 (ED_{50} = 17.4 μg/mL against HCT-15 COLADCAR, ED_{50} = 15.5 μg/mL against UISO-SQC-1 and ED_{50} = 19.5 μg/mL against OVCAR-5) and F-5 (ED_{50} = 7.2 μg/mL against HCT-15 COLADCAR, ED_{50} = 16.2 μg/mL against KB, ED_{50} = 4.0 μg/mL against UISO-SQC-1 and ED_{50} = 3.0 μg/mL against OVCAR-5), both composed of the mixture of *ent*-kaur-16-en-19-ioc acid and *ent*-kaur-15-en-19- ioc acid [7], 15α-angeloyloxy-*ent*-kaur-16-en-19-oic acid [7], 12α-hydroxy-*ent*-kaur-16-en-19-oic acid [8], and ursolic acid (**1**, [9, 10]) in different proportions. The cytotoxic analysis of these natural products showed that **1** is a unique cytotoxic compound from this plant (ED_{50} = 3.7 μg/mL against HCT-15 COLADCAR, ED_{50} = 3.4 μg/mL against UISO-SQC-1, and ED_{50} = 3.6 μg/mL against OVCAR-5). Compound **1** has been previously isolated from several species and its antitumor activity has been documented in mouse and human cell lines [10] as well as *in vivo* induced mice skin cancer [11].

The aerial parts of *S. maculatus* were collected in Rancho Merced Bason, Huixtan, Chiapas, Mexico and were identified by M. C. Abigail Aguilar. A voucher specimen (number 12700) was deposited at the Instituto Mexicano del Seguro Social Herbarium (IMSSH, Mexico City). The air-dried aerial parts (1.3 kg) were powdered and extracted by maceration at room temperature with acetone (10 L × 3) and concentrated to dryness *in vacuum* to obtain 52.6 g of a syrup residue, which was adsorbed on 53 g of silica gel and chromatographed on open CC over silica gel 60 (400 g), using a gradient of *n*-hexane:acetone as eluent. The composition of the fractions obtained (400 mL each) was monitored by TLC, visualizing the compounds by spraying with a 1% solution of $CeSO_4 \cdot NH_3$ in H_2SO_4 2N. The chromatographically identical fractions were combined, yielding six mixtures of compounds: F-1 [3.2 g, *n*-hexane 100%], F-2 [6.1 g, *n*-hexane-acetone (95:05)], F-3 [1.4 g, *n*-hexane-acetone (95:05)], F-4 [4.8 g, *n*-hexane-acetone (90:10)], F-5 [3.2 g, *n*-hexane-acetone (80:20)], and F-6 [2.0 g, *n*-hexane-acetone (60:40)]. The natural products were isolated by means of successive chromatographic process, F-1: caryophyllene β-oxide, F-2: triacontanol and stigmasterol, F-3: stigmasterol, *ent*-kaur-16-en-19-oic acid and *ent*-kaur-15-en-19-oic acid, F-4: *ent*-kaur-16-en-19-oic acid, *ent*-kaur-15-en-19-oic acid, 15α-angeloyloxy-*ent*-kaur-16-en-19-oic acid, 12α-hydroxy-*ent*-kauren-19-oic acid, and ursolic acid (**1**), F-5: 12α-hydroxy-*ent*-kauren-19-oic acid and **1** and F-6: β-sitosteryl β-D-glucopyranoside, stigmasteryl β-D-glucopyranoside and sucrose. All these compounds were identified by comparison of their IR, ^{1}H, and ^{13}C NMR data with those previously reported. The cytotoxic activity of the extract, fractions, and pure compounds was accomplished according to the previously described procedure [12].

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REFERENCES

1. H. Robinson, *Smithsonian Contrib. Bot.*, **51**, 38 (1981).
2. E. Berlin and B. Berlin, *Ethnobiology of the Highland Maya of Chiapas, Mexico. The Gastrointestinal Diseases*, Princeton University Press, Princeton, New Jersey, 1996.
3. N. LeVan and N. H. Fischer, *Phytochemistry*, **18**, 851 (1979).
4. W. Herz and S. V. Bhat, *Phytochemistry*, **12**, 1737 (1973).
5. A. J. Malcolm and N. H. Fischer, *J. Nat. Prod.*, **50**, 167 (1987).
6. N. Ohno, T. J. Mabry, V. Zabel, and W. H. Watson, *Phytochemistry*, **18**, 1687 (1979).
7. J. Jakupovic, A. Schuster, F. Bohlmann, U. Ganzer, R. M. King, and H. Robinson, *Phytochemistry*, **28**, 543 (1989).
8. P. J. Houghton and L. M. Lian, *Phytochemistry*, **25**, 1939 (1986).
9. A. Numata, P. Yang, C. Takahashi, R. Fujiki, M. Nabae, and E. Fujita, *Chem. Pharm. Bull*., **37**, 648 (1989).
10. K. S. Hoon, A. Byung-Zun, and R. S. Yong, *Phytotherapy Res.*, **12**, 553 (1998).
11. H. M. Tuan, H. C. Tang, W. Z. Yuan, F. Thomas, L. Y. Rong, S. Kathe, M. Wei, G. Constantino, L. Jeffrey, and C. Allan, *Cancer Res.*, **54**, 701 (1994).
12. J. Popoca, A. Aguilar, D. Alonso, and M. L. Villarreal, *J. Etnopharmacol.*, **59**, 173 (1998).