LINOLEIC ACID AS A PLANT GROWTH INHIBITOR FROM SEEDS OF SESBANIA PUNICEA

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As part of a program involving a search for new naturally occurring plant growth regulants, application of an ethanolic extract of the seeds of *Sesbania punicea* (Fabaceae) resulted in the inhibition of meristematic growth of several test plant species. Linoleic acid was identified as the major growth inhibitory compound present in the seeds. Studies on the isolation of cytotoxic compounds from the seeds of the related *Sesbania drummondii* have not been completed (1).

EXPERIMENTAL¹

PLANT MATERIAL.—The seeds of Sesbania punicea (Cav.) Benth. (PR 48527) were collected in Florida for the Economic Botany Laboratory (USDA-ARS), Beltsville, MD.

EXTRACTION AND ISOLATION OF ACTIVE COMPOUND.—Coarsely-ground seeds (5 g) were extracted with acetonitrile-water (9:1). After concentration under nitrogen, the extract was fractionated by preparative tlc (silica gel, dichloromethane-methanol 95:5) using multiple development. The active compound was located by observing inhibition of root growth using a lettuce-seed germination assay (2) and inhibition of meristematic growth in the bean second internode assay (3). The active compound (ca. 50 μ g by glc) had ir, ¹H-nmr and ms identical to those of linoleic acid. Linoleic acid caused similar growth responses when tested in the above-mentioned bioassay systems.

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FLAVONOIDS OF RETAMA RAETAM

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Retama raetam (Forssk.) Webb. (=Lygos raetam (Forssk.) Heywood) [Retama=conserved name (1)] is an erect desert shrub belonging to the Leguminosae. The only flavonoids reported in Retama are the C-glycosides in two Lygos species (2) as well as daidzein, genistein, 5-methylgenistein and C-glycosides reported in Retama monosperma (L.) Boiss. (=Lygos monosperma (L.) Heywood (3). We present here the investigation of the flavonoids of R. raetam for the first time. The 7-glucosides of apigenin, luteolin, and chrysoeriol were identified, along with orientin its 4'-glucoside and apigenin 6,8-di-Cglucoside. Also identified were daidzein and its 7,4'-dimethyl ether. Small amounts of

¹Full details of the isolation and identification of the compound are available on request to the author.

two uncommon flavone aglycones were detected; however, their identity was not possible due to the small amounts available.

EXPERIMENTAL¹

GENERAL EXPERIMENTAL PROCEDURES.—Spectra were recorded with uv Beckman model 26. Adsorbent for tlc was from Fluka while Whatman No. 1, and 3 MM paper was used for pc.

PLANT MATERIAL.—The plant material was collected from plants on the Cairo-Suez road, 50 km from Cairo. Identification was carried out by Professor Dr. M.N. El-Hadidi, Department of Botany, Cairo University. Voucher specimens are deposited in the herbarium of Cairo University.

EXTRACTION AND ISOLATION.—Air-dried leaves and stems of *R. raetam* (120 g) were extracted with 70% aqueous ethanol three times. The combined extracts were concentrated under reduced pressure until only a concentrated water extract remained. The extract was applied to a polyamide column (10 x 90 cm). The column was eluted first with water and then by increasing concentrations of ethanol. Similar flavonoid fractions were combined and further purified through elution techniques on paper chromatography (Whatman 3 MM).

IDENTIFICATION OF FLAVONOIDS.—All structural assignments employed both β -glucosidase and acid hydrolysis, uv spectra (MeOH; NaOMe; AlCl₃; AlCl₃-HCl; NaOAc; NaOAc-H₃BO₃), and cochromatography with authentic samples on paper chromatography (H₂O; 15% HOAc; BAW; PhOH). Standard methods of identification were applied (4,5).

APIGENIN 7-GLUCOSIDE, LUTEOLIN 7-GLUCOSIDE, CHRYSOERIOL 7-GLUCOSIDE, ORIENTIN.— Each of the compounds yielded the expected aglycones and glucose when hydrolyzed with both enzyme and acid (the latter giving orientin and isoorientin). The uv spectral data, colors under uv before and after hydrolysis, and direct comparison with authentic samples established their structures.

ORIENTIN 4'-GLUCOSIDE.—This glycoside gave orientin, traces of isorientin, and glucose on acid hydrolysis. The uv data were: MeOH, 270, 290 sh, 333; NaOMe, 270, 276, 305 sh, 377; AlCl₃, 278, 295, 346, 383; AlCl₃HCl, 278, 295, 342, 383; NaOAc, 276, 320 sh, 372; NaOAc-H₃BO₃, 270, 334. That glycosylation is in position 4' is shown by the lack of increase in intensity with NaOMe, as well as by its chromatographic colors under uv with and without ammonia before and after acid hydrolysis (dark→dark before acid hydrolysis and dark→yellow after acid hydrolysis). Rf values (orientin in parenthesis) H₂O: 11 (3); 15% HOAc: 36 (18); BAW: 8 (19); PhOH: 37 (40).

APIGENIN 6.8-DI-C-GLUCOSIDE.—This compound was unchanged on acid hydrolysis, and cochromatographed wih an authentic sample from *Salvia triloba* (6). The uv data were: MeOH, 274, 311 sh, 333; NaOMe, 283, 331, 398; AlCl₃, 265 sh, 281, 306, 352, 390; AlCl₃-HCl, 262 sh, 280, 304, 346, 386; NaOAc, 283, 308 sh, 334 sh, 395; NaOAc-H₃BO₃, 276 sh, 284, 323, 351, 414 sh.

DAIDZEIN AND DAIDZEIN 7,4'-DIMETHYL ETHER.—Daidzein was identified through cochromatography with an authentic sample as well as uv spectroscopy. Only small amounts of the second isoflavone were available. It gave daidzein on demethylation with pyridenium hydrochloride. Rf value on silica gel in chloroform-methanol (9:1)=92. The uv data were: MeOH, 255 sh, 266, 310 sh, unchanged on addition of NaOAc or NaOMe.

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¹Full details of the isolation and identification of the compounds are available on request to the senior author.