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THE AMINO ACID COMPOSITION OF GUM EXUDATES FROM *PROSOPIS* SPECIES*

D. M. W. ANDERSON, J. F. HOWLETT† and C. G. A. MCNAB

Chemistry Department, The University, Edinburgh EH9 3JJ, U.K.; †Food Science Division, Ministry of Agriculture, Fisheries and Food, Horseferry Road, London, SW1P 2AE, U.K.

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Key Word Index—*Prosopis*; Leguminosae; gum exudates; amino acids.

Abstract—The amino acid compositions of the proteinaceous components of the gum exudates from *Prosopis alba*, *P. chilensis*, *P. glandulosa*, *P. laevigata*, *P. torreyana* and *P. velutina*, and for a sample of commercial gum mesquite, are presented. In agreement with data published previously for the polysaccharide components of their gums, only minor differences in composition are shown by these species. The amino acid compositions are characterized by very high proportions of hydroxyproline and by high proportions of proline and serine; these three amino acids account for 62.5% of those present in the gum from *Prosopis velutina*. The amino acid compositions of these *Prosopis* gums are remarkably similar to that established recently for the gum from *Acacia senegal* (gum arabic).

INTRODUCTION

The many advantages of *Prosopis* spp. emphasized in U.S. National Academy of Sciences publications [1,2] have stimulated economic [3–5], ecological [6,7] and taxonomic [8,9] interest. There is a strong case for the controlled introduction of carefully selected *Prosopis* spp. into under-developed arid zones, where their advantages as sources of rapidly growing timber and as sources of fodder for grazing animals, and their ability to enrich surrounding soils through nitrogen fixation are valuable attributes, particularly in the hottest and driest conditions. In Mexico and the South-Western arid regions of the U.S.A., the Indian natives locate their homes in the proximity of mesquite (*Prosopis*) trees, which provide valued shelter from the sun, prolific supplies of edible beans/pods, and a copious supply of a gum exudate that is collected for sale and also used as a foodstuff.

The botanical origin of commercial gum mesquite has frequently been attributed in chemical studies to *P. juliflora*, but this is one of the species known to have been identified incorrectly in the past [8,10]. A recent report [11] ascribes mesquite to *Prosopis laevigata* and comments on its foodstuff value. The chemical differences between the polysaccharide components of the gums from seven *Prosopis* spp. have been studied [12].

Mesquite gum is not included in the American GRAS list nor in any other list of permitted foodstuffs additives; there are no metabolic, teratological, mutagenicity nor toxicological studies available. Nevertheless, because mesquite gum is widely used as an ingredient of traditional native dishes in regions where it is produced prolifically, it was included in a Project 'The Characterisation of the Proteinaceous Components of Edible Gums' sponsored by The Ministry of Agriculture, Fisheries and Food within the period 1982–1984.

This paper presents the data obtained for the amino acid compositions of specimens of gum from seven *Prosopis* spp. and for a commercial sample of mesquite gum.

RESULTS

The amino acid data for the gum exudates from *Prosopis alba* Grisebach, *P. chilensis* (Mol.) Stuntz, *P. glandulosa* Torrey var. *torreyana* (Benson) Johnson ('*glandulosa A*'), *P. glandulosa* Torrey var. *glandulosa* ('*glandulosa B*'), *P. glandulosa* Torrey ('*glandulosa C*'), *P. laevigata* (Humb. and Bonpl. ex Willd.) M. C. Johnst., *P. velutina* Wooton, and a commercial sample of gum mesquite are given in Table 1. For comparison, the data for the gum from *Acacia senegal* Willd., obtained [13] as average values for eleven specimens, are also shown.

DISCUSSION

The data in Table 1 show that the *Prosopis* spp. studied give exudates that do not differ greatly in terms of their amino acid compositions. The largest variations occur in their proportions of alanine, glutamic acid, proline, and hydroxyproline; the latter varies from 201 (*P. chilensis*) to 425 (*P. velutina*) residues per 1000 residues. On the basis of the data available, *P. chilensis* shows more extreme values than any of the others. This may arise from the fact that the *P. chilensis* specimen was of African origin; the others originated in Mexico and the southern U.S.A. From the data available it appears that *P. glandulosa* '*C*' and *P. laevigata*, possibly admixed with minor amounts from *P. velutina*, were the most likely sources of the commercial sample of gum mesquite studied.

The amino acid profile for these *Prosopis* gums (%N *ca*

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Table 1. The amino acid composition of the gum exudates from some *Prosopis* species (residues per 1000 residues)

	Cml. gum mesquite	*Prosopis*							*Acacia**
		glandulosa							
		A	B	C	*laevigata*	*chilensis*	*velutina*	*alba*	*senegal* (av.)
Alanine	31	48	51	38	35	55	24	47	28
Arginine	14	16	11	13	16	18	15	14	5
Aspartic acid	50	74	73	56	50	76	49	71	50
Cystine	1	4	4	1	3	3	3	3	0
Glutamic acid	24	36	36	26	26	47	20	39	29
Glycine	34	48	43	34	33	56	25	43	41
Histidine	35	33	33	32	35	32	33	34	44
Hydroxyproline	381	292	298	363	376	201	425	289	328
Isoleucine	20	24	26	22	20	30	19	30	12
Leucine	20	45	37	29	35	51	24	43	67
Lysine	20	22	25	18	23	25	13	24	23
Methionine	2	2	3	1	2	0	2	2	1
Phenylalanine	14	17	18	14	12	22	11	22	22
Proline	101	86	78	106	79	134	82	92	88
Serine	109	108	109	112	113	100	118	101	136
Threonine	47	51	54	46	49	52	46	53	76
Tyrosine	38	38	35	35	32	35	35	35	10
Valine	53	59	64	54	61	63	56	60	36

*From ref. [13].

0.3–0.9% [12]) is remarkably similar to that observed [13] recently in studies of the gum from *Acacia senegal* (gum arabic). The data in Table 1 show that the proportions of arginine, isoleucine, tyrosine and valine are slightly less, and the proportions of histidine, leucine, serine and threonine slightly greater in *A. senegal* gum [13] than in the *Prosopis* spp. studied. The polysaccharide components of *Prosopis* gum exudates [12] are also similar to those of some *Acacia* gum exudates [14, 15].

EXPERIMENTAL

Origin of gum specimens. The origins of the gum specimens have been described [12], except for that from *P. chilensis* which was collected by one of the authors (D.M.W.A.) from trees growing in an experimental plot at the Forestry Research Institute, Soba, Republic of the Sudan, in January 1983. The sample of commercial mesquite gum was supplied by an American gum merchant; the author will be happy to provide the name and address of the supplier on request.

Amino acid analyses. Sufficient finely powdered sample to give 2 mg of nitrogen (12.5 mg of crude protein) was transferred to a 100 ml round-bottomed two-necked flask. After adding anti-bumping granules, 80 ml of *ca* 6 M HCl (AnalaR, sp. gr. 1.18, diluted with an equal vol. of distilled water) and 2 ml of 0.004 M *nor*-leucine in 0.1 M HCl (internal standard), the flask was fitted with an air-cooled condenser (800 mm) and the apparatus was purged with oxygen-free nitrogen. The flask contents were refluxed for exactly 20 hr under a continuous slow stream of oxygen-free nitrogen, then allowed to cool. The condenser was washed down with distilled water; the soln was filtered and taken to dryness at 42° (rotary evaporator). The residue was dissolved in 20 ml 0.01 M HCl, filtered (Millipore filter, 0.22 μ) and stored frozen in a glass vial pending instrumental analysis.

A suitable aliquot (normally 1 ml) of the hydrolysate was applied to a column of cationic exchange resin, the amino acid components were separated by elution with suitable buffers of increasing ionic strength of pH, detected by reaction with ninhydrin in a continuous flow analytical system (Rank-Hilger Chromaspek), and quantified by reference to standard solns of calibration standards. This technique determined all the amino acids commonly found in foodstuffs proteins with the exception of methionine, cystine and tryptophan.

Determination of cystine and methionine. A sample containing *ca* 10 mg N was weighed ($\pm$0.10 mg) into a 250 ml round-bottomed flask and cooled in iced water. Ice-cooled performic acid (2.5 ml) was added, care being taken to ensure that the sample was wetted thoroughly. The flask was transferred to an oven at 50° for 30 min, then allowed to cool; 0.75 M HBr was added, with swirling, in an ice-bath. After the addition of 200 ml 6 M HCl, hydrolysis and chromatography were carried out as described above, the cystine and methionine being determined as cysteic acid and methionine sulphone respectively [16].

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ZANTHOMAMIDE: AN AROMATIC AMIDE FROM *ZANTHOXYLUM THOMENSE*

JOËL SIMERAY, JEAN-PIERRE CHAUMONT, FRANÇOISE BEVALOT* and JACQUELINE VAQUETTE*†

Laboratoire de Botanique et Cryptogamie, UFR de Médecine et Pharmacie, Place St Jacques, F 25030 Besancon Cédex, France; *Laboratoire de Pharmacognosie, UFR de Médecine et Pharmacie, Place Saint-Jacques, F 25030 Besancon Cédex, France

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Key Word Index—*Zanthoxylum thomense*; Rutaceae; aromatic amide; zanthomamide; benzophenanthridines.

Abstract—From the stem bark of *Zanthoxylum thomense* a new amide, zanthomamide has been isolated and characterized as *N*-methyl, *N*-cinnamyl-(3′,4′-methylenedioxy)-phenylethylamine. Other constituents identified are the known benzophenanthridines, decarine, norchelerythrine and angoline.

INTRODUCTION

The genus *Zanthoxylum* (including *Fagara*) is known to contain various components such as terpenes, lignans, amides and alkaloids [1]. We report here the examination of the stem bark of *Zanthoxylum thomense* (Engl.) A. Chev. *ex* Waterm. [2], a woody rutaceous plant indigenous to West Equatorial Africa.

RESULTS AND DISCUSSION

After preliminary defatting with petrol (bp 60–80°), the ground stem bark was extracted with chloroform, then with chloroform in an alkaline medium and finally with methanol. A total yield of 0.15% crude alkaloids was obtained.

From the concentrated petrol extract, the common triterpene lupeol crystallized, and the new amide, zanthomamide (**1**) was obtained by CC of the supernatant over silica gel, then by CC of eluates ($CHCl_3$) from the silica gel column over alumina.

From the neutral chloroform extract, three known benzophenanthridine alkaloids, decarine (**2**), norchelerythrine (**3**) and angoline (**4**) were successively separated by CC over silica gel then alumina. They were identified by direct comparison of their spectra (UV, MS, ^{1}H NMR) with those of authentic samples. Decarine (**2**) was the main alkaloid (55% of crude alkaloids).

The new amide, zanthomamide (**1**), was obtained only in an amorphous form. The EI mass spectrum showed a weak $[M]^+$ at *m/z* 309 ($C_{19}H_{19}O_3N$). In the UV spectrum (methanol) there is one maximum at 276 nm (log ε5.92); it is not modified by alkali. The IR spectrum exhibited an absorption band at 1650 cm^{-1}, characteristic of a tertiary amide [3], while the absence of absorption bands between 3200 and 3500 cm^{-1} confirmed the tertiary nature of the amide [3]. EI mass spectral fragmentation of the amide gave major ions at *m/z* 148 $[M-131-30]^+$, 131 (100%), 103 and 77. The latter three ions suggested that the acidic part of the amide was cinnamic acid. This argument was supported by the occurrence in the ^{1}H NMR (60 MHz, $CDCl_3$) spectrum of a broad signal for five aromatic protons at δ7.37 and, in the ^{1}H NMR (250 MHz, $CDCl_3$) spectrum, of an AB signal for two protons at δ7.73 and 6.88 (J = 15 Hz), characteristic of *trans*-cinnamic acid protons [4]. Alkaline hydrolysis [5] of **1** gave cinnamic acid, identified by direct comparison (TLC, mmp) with an authentic sample.

Further examination of the ^{1}H NMR spectrum of the amide showed that the amino part was a phenylethylamine substituted by an *N*-methyl group (3H, *s*, δ3.04) and by a methylenedioxy group on the aromatic nucleus

†To whom correspondence should be addressed.