## THE COMPOSITION AND PROPERTIES OF GUM EXUDATES FROM SUBSPECIES OF ACACIA TORTILIS\*

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Abstract—Gum specimens from Acacia tortilis ssp. spirocarpa, ssp. raddiana var. pubescens (two specimens) and ssp. heteracantha (three specimens) have been analysed. The results are of chemotaxonomic interest. Although the gum from ssp. raddiana var. pubescens appears to be intermediate between those from ssp. spirocarpa and ssp. heteracantha in terms of some of the analytical parameters, the overall impression is that ssp. raddiana is more similar chemically to ssp. spirocarpa than to ssp. heterocantha. The latter yields a viscous, proteinaceous polysaccharide that differs from those from both ssp. spirocarpa and ssp. raddiana by having a much higher ratio of arabinose to galactose, higher nitrogen and methoxyl contents, and much higher intrinsic viscosity and molecular weight; preliminary experiments have shown this gum to consist of two components.

## INTRODUCTION

BENTHAM<sup>2</sup> classified Acacia heteracantha Burch., A. spirocarpa Hochst., and A. tortilis Hayne as closely related species (Numbers 315–317 respectively) within his Series 4, Gummiferae, and he listed A. perrottetii Steud., A. fasciculata Guill. et Perr. non Kunth non R. Br., and A. raddiana Savi as synonyms of A. tortilis. Later, two varieties, var. raddiana and var. pubescens A. Chev., were recognized in A. raddiana.

A. tortilis is a widespread species, complicated genetically, and apparently divisible into a number of more or less distinct geographical races. In Brenan's opinion, the following four subspecies can be distinguished: A. tortilis (Forsk.) Hayne ssp. tortilis; A. tortilis (Forsk.) Hayne ssp. spirocarpa (Hochst. ex A. Rich.) Brenan; A. tortilis (Forsk.) Hayne ssp. heteracantha (Burch.) Brenan; and A. tortilis (Forsk.) Hayne ssp. raddiana (Savi) Brenan, which exists as two varieties: var. raddiana (young branchlets glabrous or subglabrous and the pods glabrous) and var. pubescens A. Chev. (young branchlets and rhachides of the leaves more or less pubescent, as are the pods). We are most grateful to Mr. J. P. M. Brenan for the information that, of these ssp., raddiana is the most distinctive and still regarded by some workers as a separate species; ssp. spirocarpa and ssp. heteracantha are closely related morphologically, but typical ssp. tortilis is rather isolated from the rest, both morphologically and geographically, being the only one found east of the Red Sea.

When good gum specimens from A. tortilis ssp. heteracantha, ssp. spirocarpa, and ssp. raddiana var. pubescens were obtained, their chemical composition was investigated to ascertain whether significant differences exist at the subspecies level.

<sup>\*</sup> Part XLV of the Series "Studies of Uronic Acid Materials". For Part XLIV see Ref. 1.

<sup>&</sup>lt;sup>1</sup> Anderson, D. M. W. and Bell, P. C. Phytochemistry 13, 1871.

<sup>&</sup>lt;sup>2</sup> Bentham, G. (1875) Trans. Linn. Soc (Lond.) 30, 444.

<sup>&</sup>lt;sup>3</sup> Brenan, J. P. M. private communication.

## RESULTS AND DISCUSSION

The analytical data obtained for the six specimens studied are shown in Table 1. When all the analytical parameters are taken into account, there is little doubt that the three subspecies examined each give a gum exudate that is analytically distinguishable from the others, and from all other *Acacia* exudates examined to date.

The overall impression is that ssp. *raddiana* var. *pubescens* is intermediate between ssp. *spirocarpa* and ssp. *heteracantha*, but, on the basis of their closely similar viscosities, molecular weight, methoxyl and rhamnose contents, and arabinose to galactose ratio, ssp. *raddiana* appears to be closer chemically to ssp. *spirocarpa* than to ssp. *heteracantha*, in spite of its morphological relationship with the latter subspecies. Ssp. *raddiana* is certainly not an entirely distinct species.

TABLE 1. ANALYTICAL DATA FOR PURIFIED GUM POLYSACCHARIDES	FROM Acacia tortilis subspecies	
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	Cookers	Acacia Subsp. raddiana var, pubescens		Tortilis Subsp. heteracantha		
	Subsp. spirocarpa	l	II	1	II	III
Moisture (%)	9.9	10.3	9-6	9.9	11-8	10.7
Ash (%)*	1.6	1.9	1.3	1.5	2.0	1.7
Nitrogen (%)*	0.46	0.96	1-22	1.51	1.49	1.51
Hence protein $\binom{9}{6}$ $(N \times 6.25)^*$	2.9	6.0	7.6	9.4	9-3	9.4
Methoxyl (%)†	0.58	0.61	0.66	0.96	0.97	1.06
$[\alpha]_D$ , In H <sub>2</sub> O, deg	+ 74	+88	+87	+97	+79	$\pm 87$
$[\alpha]_D$ , In 7 M urea, deg	+ 78	+93	+92	+99	+82	+91
Intrinsic viscosity (mlg -1)	9.8	9-3	11-2	22-5	16.9	19:4
Mol wt (MW $\times$ 10 <sup>4</sup> )	25	50	51	210	220	140
Equivalent wt†	1590	2040	1940	2440	1730	2100
Hence uronic anhydride (%)†‡	11.1	8.6	9-1	7.2	10.1	8.7
Sugar composition after hydrolysis:						
4-O-Methylglucuronic acid	3.5	3.7	4·()	5.8	5.8	6.4
Glucuronic acid	7.6	4.9	5.1	1.4	4.3	2.3
Galactose	39	36	37	21	24	24
Arabinose	43	49	46	68	62	64
Rhamnose	7	6	8	4	3	3

<sup>\*</sup> Corrected for moisture content.

The gum from ssp. heteracantha gives colourless or pale yellow, clear solutions, and has a combination of properties that makes it of considerable potential commercial interest. It has the highest ratio of arabinose to galactose recorded so far (cf. A. nubica<sup>4</sup>); a high methoxyl content, surpassed only by A. nilotica, A. parramattensis, and A. giraffae; a nitrogen content as high as that previously reported (cf. A. parramattensis); a molecular weight that is almost as high as that recorded previously (cf. A. arabica) and an intrinsic viscosity, which, for specimen III, equals the highest values previously recorded (cf. A.

<sup>†</sup> Corrected for moisture and protein content.

<sup>‡</sup> If all acidity arises from uronic acids.

<sup>§</sup> If all methoxyl groups located in this acid.

<sup>&</sup>lt;sup>4</sup> Anderson, D. M. W. and Cree, G. M. (1968) Carbohyd. Res. 6, 385.

<sup>&</sup>lt;sup>5</sup> Anderson, D. M. W., Cree, G. M., Herbich, M. A., Karamalla, K. A. and Stoddart, J. F. (1964) *Talanta* 11, 1559.

<sup>&</sup>lt;sup>6</sup> ANDERSON, D. M. W., BELL, P. C., and McNab C. G. A. (1971) Carbohyd. Res. 20, 269.

<sup>&</sup>lt;sup>7</sup> Anderson, D. M. W. and Dea, I. C. M. (1969) Carbohyd. Res. 10, 161.

laeta, 8 A. parramattensis<sup>6</sup>). It is clearly desirable to study further specimens to ascertain whether the extent of the differences between the three specimens studied here are typical of a genetically variable species; it is customary for the analytical parameters for different specimens of a particular species to vary by not more than  $\pm 10\%$ . Attempts are also being made to secure gum specimens from ssp. tortilis and ssp. raddiana var. raddiana, but the location of these has proved to be difficult.

With a view to proceeding to a detailed structural study of the gum from A. tortilis ssp. heteracantha, the homogeneity of the available specimens has been examined. Zone electrophoresis of dyed samples<sup>9</sup> on Phoroslides in acetate and ammonium carbonate buffers gave two distinct bands for each specimen. Molecular-sieve chromatography (Bio-Gel Al50) of a dyed<sup>9</sup> sample of specimen I showed the presence of two elution peaks (one at the void volume). Examination in the Spinco ultracentrifuge at 44000 rpm showed the presence of 2 components in each specimen. A. tortilis ssp. heteracantha therefore shows the clearest evidence obtained to date in this laboratory for the presence of two components in an Acacia exudate. In contrast, only 1 band was given by ssp. spirocarpa, and by specimen I of ssp. raddiana var. pubescens, although specimen II of this ssp. gave 2 bands. Clearly, careful attempts to achieve fractionation or the elimination of some impurity will be an essential pre-requisite to structural studies.

With such a high protein content, and the presence of more than one component, it appeared that contributions to the optical rotation in water from tertiary structures were possible. Rotations in 7 M urea were therefore determined, but the values were only slightly different and always more positive than in water, as was found<sup>10</sup> in a previous study.

## **EXPERIMENTAL**

Origins of gum specimens. Gum from Acacia tortilis (Forsk.) Hayne ssp. spirocarpa (Hochst. ex A. Rich.) Brenan was collected by Mr. A. G. Seif-el-Din, Gum Research Officer to the Republic of the Sudan in March 1964; this ssp. is abundant on hard soils in the region 13° 45' N, 30° 20' E. Gum from A. tortilis (Forsk.) Hayne ssp. raddiana (Savi) Brenan var. pubescens A. Chev. was collected by Mr Seif-el-Din as follows: specimen 1 was obtained in April 1973 in the region 13° 45′ N, 30° 20′ E, where this ssp. is abundant on loose sandy soils; specimen II was obtained in March 1970 from the main trunk (normally it is the upper branches that are tapped) of a tree at Um Badir, N. Kordofan. (Botanical Vouchers for specimens of gum from A. tortilis ssp. spirocarpa and A. tortilis ssp. raddiana var. pubescens have been authenticated by Mr. J. P. M. Brenan). Gum from A. tortilis (Forsk.) Hayne subsp. heteracantha (Burch.) Brenan was collected as follows; specimen I was collected at Salisbury, Rhodesia on 28th February 1971 (botanical voucher Kelly 479 in SRGH); specimens II and III (botanical vouchers G. Pope 426 and 427 in SRGH) were collected in May 1971 at Botswana, near Rakops, Rhodesia (24° 30' E, 21° 4' S) and sent by Mr Th. Müller, Curator, Botanical Gardens, Salisbury. Preparation of samples for analysis. All of the specimens dissolved in cold water overnight to give clear, colourless solutions (ssp. spirocarpa, ssp. heteracantha specimens I and II) or clear, pale yellow solutions (ssp. raddiana var. pubescens specimens I and II, ssp. heteracantha specimen III). The three specimens of ssp. heteracantha all gave a very slight jelly-type residue which was readily dispersed with alkaline borohydride. 11 The gum solns were treated as described in the previous paper.1

Analytical methods. These have been described.1

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<sup>&</sup>lt;sup>8</sup> Anderson, D. M. W., Dea, I. C. M. and Smith, R. N. (1968) Carbohyd. Res. 7, 320.

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<sup>&</sup>lt;sup>11</sup> Anderson, D. M. W., Bell, P. C. and King, H. A. R. (1972) Carbohyd. Res. 22, 453.