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Nectar Sugar Composition in the Subfamily Alooideae (Asphodelaceae)

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Key Word Index—*Aloe*; *Astroloba*; *Chortolirion*; *Gasteria*; *Haworthia*; *Poellnitzia*; Asphodelaceae; Alooideae; nectar sugars; chemotaxonomy.

Abstract—HPLC analyses of nectar samples from 82 species of the genera *Aloe, Astroloba, Chortolirion, Gasteria, Haworthia, Kniphofia, Lomatophyllum* and *Poellnitzia* have shown that the sugar composition of nectar is remarkably invariable within each of the genera. In the Alooideae, distinct suprageneric groups can be distinguished, based only on the relative proportions of sucrose, glucose and fructose. Three nectar types are present in the subfamily: (1) an alooid type (in *Aloe, Kniphofia, Lomatophyllum* and *Poellnitzia*) with less than 5% sucrose and equal proportions of glucose and fructose; (2) a gasterioid type (in *Gasteria* only) with sucrose dominant and equal proportions of glucose and fructose; (3) a haworthioid type (in *Astroloba, Chortolirion* and *Haworthia*) with sucrose dominant but with much more glucose than fructose.

Introduction

A hypothesis of generic relationships amongst *Aloe* and related genera (Asphodelaceae, subfamily Alooideae) was recently published (Smith and Van Wyk, 1991), in which nectar sugars were used as phylogenetic characters in a cladistic study. The data on which the character polarities were based is presented in this paper. Nectar sugar compositions of one species of *Gasteria*, two species of *Haworthia* and one species of *Kniphofia* have been determined semi-quantitatively by Percival (1961) but her results were subjective and do not allow meaningful comparisons. Since all the genera of the subfamily are here rigorously compared, our study is the first one in which the taxonomic value of nectar sugars can be evaluated.

Materials and Methods

Nectar samples were taken from cultivated plants in various botanical gardens (see Table 1) and only a few from plants in their natural habitat. Collecting voucher specimens was therefore not only unpractical, but also unnecessary (most of the co-authors of this paper are taxonomic experts on the various genera of the subfamily and furthermore, differences in sugar composition between species proved to be unimportant). A total of 98 samples were analysed, including 82 species representing all seven genera of the subfamily Alooideae and also *Kniphofia* of the Asphodeloideae.

Procedures. Nectar was sampled as spots (5–15 mm diameter) on filter paper (Whatman no. 1). After air-drying, the papers were stored at −18°C awaiting analysis. Nectar was recovered from the papers by repeated rinsing (3×) with 50 to 100 μl distilled water, followed by centrifugation. Samples were analysed by isocratic HPLC operating at 2.5 ml min^{-1}, with a "Waters Sugarpack" column and acetonitrile–water (87:13) as eluent. For detection we used a refractive index detector. The presence of fructose, glucose and sucrose was determined as percentages of total sugars, using peak height and 8 mg ml^{-1} of each sugar as external standard.

Results

A comprehensive list of the samples studied and the composition of nectar sugars is given in Table 1. On the basis of the percentage sucrose, the genera can be divided into

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TABLE 1. NECTAR SUGAR COMPOSITION IN 98 NECTAR SAMPLES FROM 82 SPECIES AND 8 GENERA OF THE FAMILY ASPHODELACEAE

Sample number (see also Fig. 1)	Genera and species	Locality*	Nectar composition (%) Fructose	(%) Glucose	(%) Sucrose
1.	*Aloe abyssinica* Lavranos	PRE	47	49	4
2.	*A. aculeata* Pole Evans	PRE	45	55	—
3.	*A. affinis* Berger	PRE	46	53	1
4a.	*A. arborescens* Mill. sample 1	PRE	45	55	—
4b.	sample 2	PRE	46	53	1
5.	*A. asperifolia* Berger	PRE	46	51	3
6.	*A. bellatula* Reynolds	PRE	49	51	—
7.	*A. bowiea* Schult. & J. H. Schut.				
7a.	sample 1	NBG	51	49	—
7b.	sample 2	POTCH	49	51	—
8.	*A. branddraaiensis* Groenewald	PRE	40	60	—
9.	*A. capitata* Bak. var. *gneissicola* H. Perr.	PRE	47	53	—
10.	*A. castanea* Schonl.	PRE	46	52	2
11.	*A. ciliaris* Haw.	PRE	49	49	2
12.	*A. citrina* Lavranos	PRE	49	47	4
13.	*A. dewinteri* Geiss	PRE	48	49	3
14.	*A. divaricata* Berger	PRE	46	54	—
15.	*A. erinacea* Hardy	PRE	46	54	—
16.	*A. fourei* Glen & Hardy	PRE	47	50	3
17.	*A. gariepensis* Pillans	PRE	49	51	—
18.	*A. greatheadii* Schonl.				
18a.	var. *greatheadii*	PRE	48	50	2
18b.	var. *davyana* (Schonl.) Glen & Hardy	PRE	49	49	2
19.	*A. hardyi* Glen	PRE	46	54	—
20.	*A. hereroensis* Engl. var. *hereroensis*	PRE	47	52	1
21.	*A. humilis* (L.) Mill.	POTCH	47	53	—
22.	*A. littoralis* Bak.	PRE	46	50	4
23.	*A. lutescens* Groenewald	PRE	47	52	1
24.	*A. massawana* Reynolds	POTCH	46	53	1
25.	*A. melanacantha* Berger	PRE	47	52	1
26.	*A. meyeri* Van Jaarsveld	PRE	49	50	1
27.	*A. microstigma* Salm-Dyck	PRE	50	50	—
28.	*A. monotropa* Verdoorn	NBG	47	52	1
29.	*A. mutabilis* Pillans	PRE	44	56	—
30.	*A. nubigena* Groenewald	PRE	48	51	1
31.	*A. pachygaster* Dinter	PRE	49	51	—
32.	*A. parvibracteata* Schonl.	POTCH	45	55	—
33a.	*A. pearsonii* Schonl. (red form)	PRE	47	51	2
33b.	(yellow form)	PRE	50	50	—
34.	*A. perfoliata* L.	PRE	48	49	3
35.	*A. petricola* Pole Evans	PRE	48	52	—
36.	*A. pictifolia* Hardy	POTCH	48	52	—
37.	*A. ramosissima* Pillans	PRE	55	45	—
38.	*A. sinkata* Reynolds	POTCH	42	57	1
39.	*A. speciosa* Bak.	PRE	48	52	—
40.	*A. suprafoliata* Pole Evans	PRE	49	50	1
41.	*A. thompsoniae* Groenewald	PRE	48	49	3
42a.	*A. tricosantha* Berger sample 1	PRE	46	53	1
42b.	sample 2	PRE	47	52	1
42c.	sample 3	POTCH	46	53	1
43.	*A. vanbalenii* Pillans	PRE	45	55	—
44.	*A. vaombe* Dec. & Poisson	PRE	45	55	—
45.	*A. variegata* L.	PRE	45	55	—
46.	*A. vera* (L.) Burm.f.	PRE	46	54	—
47.	*A. verecunda* Pole Evans	NBG	48	52	—

TABLE 1—*CONTINUED*

Sample number (see also Fig. 1)	Genera and species	Locality*	Nectar composition (%) Fructose	(%) Glucose	(%) Sucrose
1.	*Astroloba congesta* Salm-Dyck	NBG	4	32	64
2.	*A. foliolosa* (Haw.) Uitew. ssp. *foliolosa*	POTCH	4	16	80
3.	*A. spiralis* Salm-Dyck	KBG	2	13	85
1.	*Chortolirion angolensis* (Bak.) Berger				
1a.	sample 1	POTCH	8	21	71
1b.	sample 2	POTCH	8	19	73
1c.	sample 3	POTCH	7	20	73
1.	*Gasteria acinacifolia* (Jacq.) Haw.	NBG	10	14	76
2.	*G. baylissiana* Rauh	NBG	2	4	94
3.	*G. brachyphylla* (Salm-Dyck) Van Jaarsveld	NBG	1	1	98
4.	*G. carinata* (Mill.) Haw.	NBG	5	8	87
5.	*G. croucheri* (Hook.f.) Bak.	NBG	2	2	96
6.	*G. disticha* (L.) Haw.	NBG	2	4	94
7.	*G. excelsa* Bak.	NBG	7	9	84
8.	*G. maculata* (Thunb.) Haw				
8a.	var. *maculata*	NBG	2	2	96
8b.	var. *liliputana* (V. Poelln.) sample 1	NBG	7	8	85
	sample 2	PRE	6	7	87
9.	*G. pillansii* Kensit	NBG	2	3	95
10.	*G. pulchra* (Ait.) Haw.	NBG	6	8	86
11.	*G. rawlinsonii* Oberm.	NBG	12	13	75
12.	*G. vlokii* Van Jaarsveld	NBG	5	9	86
13.	*Gasteria maculata* X *Astroloba* sp.	POTCH	7	10	83
14.	*Gasteria sp.* X *Haworthia longiana*	NBG	16	18	66
1.	*Haworthia arachnoidea* (L.) Duval	NBG	13	51	36
2.	*H. bolusii* Bak.	PRE	6	39	55
3.	*H. comptoniana* G. G. Smith	POTCH	4	54	42
4.	*H. cooperi* Bak.	NBG	5	39	56
5.	*H. glauca* Bak.	NBG	1	19	80
6a.	*H. granulata* Marl. sample 1	PRE	5	25	70
6b.	sample 2	PRE	4	24	72
7.	*H. herbacea* (Mill.) Stearn *sensu* Bayer	POTCH	19	46	35
8.	*H. koelmaniorum* Oberm. & Hardy	PRE	5	23	72
9a.	*H. limifolia* Marl. sample 1	NBG	4	41	55
9b.	sample 2	PRE	3	24	73
10.	*H. longiana* v. Poelln.	NBG	3	20	77
11.	*H. tessellata* (Salm-Dyck) Bak. sample 1	NBG	1	29	70
11b.	sample 2	PRE	2	24	74
12.	*H. viscosa* (L.) Haw.	NBG	2	32	66
1a.	*Kniphofia porphyrantha* Bak. sample 1	GGNP	47	53	—
1b.	sample 2	GGNP	47	53	—
2a.	*K. uvaria* (L.) Oken sample 1	PRE	49	49	2
2b.	sample 2	NBG	50	47	3
1.	*Lomatophyllum* cf. *purpureum* (Lam.) Th. Dur.				
1a.	sample 1	JHB	51	49	—
1b.	sample 2	JHB	50	50	—
1.	*Poellnitzia rubriflora* (L. Bol.)				
1a.	sample 1	KBG	48	52	—
1b.	sample 2	KBG	47	53	—

*Locality abbreviations: GGNP, Golden Gate National Park, Orange Free State; JHB, Johannesburg Botanic Garden; KBG, Karoo Botanic Garden, Worcester; NBG, Kirstenbosch Botanic Garden, Cape Town; POTCH, Potchefstroom University Botanical Garden; PRE, Pretoria Botanic Garden.

two mutually exclusive groups: those with low (less than 5%) sucrose (*Aloe, Kniphofia, Lomatophyllum* and *Poellnitzia*) and those with high (more than 50%) sucrose (*Astroloba, Chortolirion, Gasteria* and *Haworthia*). The latter can be further subdivided into two groups: those with more or less equal amounts of fructose and glucose (*Gasteria* only) and those with much more glucose than fructose (*Astroloba, Chortolirion* and *Haworthia*). These distinct generic and suprageneric discontinuities, as well as the remarkable uniformity within each of the genera, are clearly visible in Fig. 1. The lack of variation in *Aloe* is particularly noteworthy, since the nectar samples

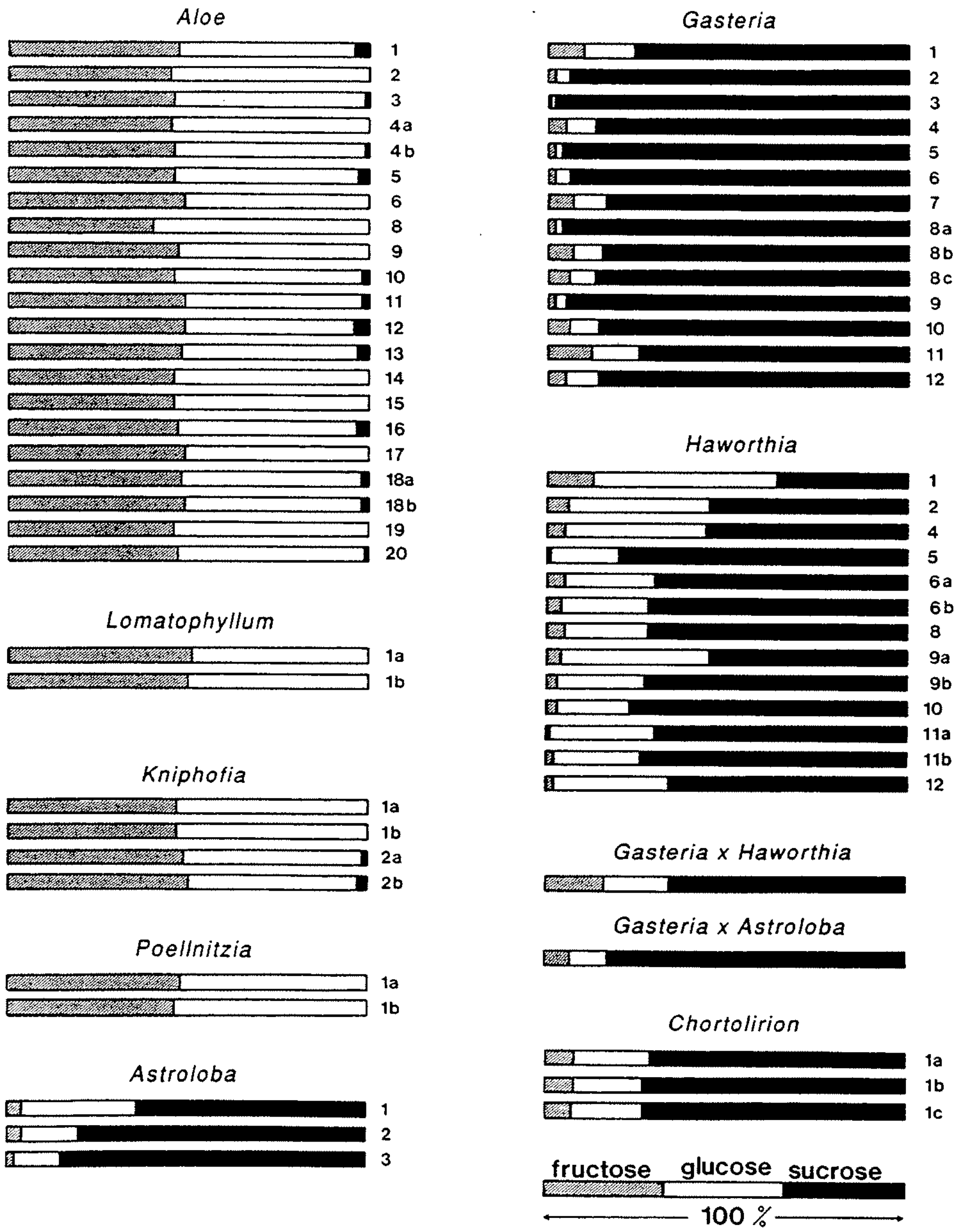

FIG. 1. NECTAR SUGAR COMPOSITIONS IN SAMPLES FROM ALL THE GENERA OF THE SUBFAMILY ALOOIDEAE AND *KNIPHOFIA* OF THE SUBFAMILY ASPHODELOIDEAE. (Samples and species are numbered as in Table 1; only a small selection of samples from *Aloe* is shown.)

came from many different infrageneric groups, representing a wide range of flower morphologies.

Discussion

Baker and Baker (1982; 1983a) developed a terminology to describe four classes of nectar based on sucrose/hexose ratios, but the importance of fructose/glucose ratios may have been underestimated. It is clear from Fig. 1. that three distinct nectar types are present in the subfamily: (1) an alooid type (in *Aloe, Kniphofia, Lomatophyllum* and *Poellnitzia*) with less than 5% sucrose and equal proportions of glucose and fructose; (2) a gasterioid type (in *Gasteria* only) with sucrose dominant and equal proportions of glucose and fructose; (3) a haworthioid type (in *Astroloba, Chortolirion* and *Haworthia*) with sucrose dominant but with much more glucose than fructose. Our results for two intergeneric hybrids (*Gasteria*×*Astroloba* and *Gasteria*×*Haworthia*) suggest that the fructose/glucose ratio is inherited from the pod parent. Baker and Baker (1979; 1982; 1983a) demonstrated quantitative intermediacy in a hybrid species of *Erythrina* but greater resemblance to one of the two parents in *Penstemon* and *Campsis*. The pattern of nectar sugars in the Alooideae agrees with current ideas on taxonomic affinities [see Smith and Van Wyk (1991) for a detailed discussion].

Nectar sugar composition, along with nectar quantity and sugar concentrations, are considered to be important variables in pollination studies. Baker and Baker (1979; 1982; 1983a; 1983b) found that nectar sugar composition (and particularly the ratio of sucrose: fructose+glucose) is broadly correlated with various types of pollinators. As a result, differences between taxa have mostly been discussed from an ecological point of view (Baker and Baker, 1975; 1982; 1983a; 1983b; Freeman and Lammers, 1986; Freeman and Worthington, 1985; Freeman *et al.*, 1983; 1984; 1985). Our results show that nectar sugar composition in the subfamily Alooideae is a conservative character and that the overall pattern reflects taxonomic affinities rather than pollinator types. Similar discontinuities amongst closely related genera are likely to be found in other plant families if nectar sugar composition is studied from a taxonomic perspective.

References

Baker, H. G. and Baker, I. (1975) Studies of nectar-constitution and pollinator–plant coevolution. In *Coevolution of Animals and Plants* (Gilbert, L. E. and Raven, P. H., eds), pp. 100–140. University of Texas Press, Austin.

Baker, H. G. and Baker, I. (1982) Chemical constituents of nectar in relation to pollination mechanisms and phylogeny. In *Biochemical Aspects of Evolutionary Biology* (Nitecki, M. H., ed.), pp. 131–171. University of Chicago Press, Chicago.

Baker, H. G. and Baker, I. (1983a) Floral nectar sugar constituents in relation to pollinator type. In *Handbook of Experimental Pollination Biology* (Jones, C. E. and Little, R. J., eds), pp. 117–141. Van Nostrand Reinhold, New York.

Baker, H. G. and Baker, I. (1983b) A brief historical review of the chemistry of floral nectar. In *The Biology of Nectaries* (Bentley, B. and Elias, T., eds), pp. 126–152. Columbia University Press, New York.

Baker, I. and Baker, H. G. (1979) Chemical constituents of the nectars of two *Erythrina* species and their hybrid. *Ann. Missouri Bot. Gard.* **66**, 446–450.

Freeman, C. E., Reid, W. H. and Becvar, J. E. (1983) Nectar sugar composition in some species of *Agave* (Agavaceae). *Madrono* **30**, 153–158.

Freeman, C. E., Reid, W. H., Becvar, J. E. and Scogin, R. (1985) Similarity and apparent convergence in the nectar–sugar composition of some hummingbird-pollinated flowers. *Bot. Gaz.* **145**, 132–135.

Freeman, C. E. and Worthington, R. D. (1985) Some floral nectar–sugar compositions of species from south-eastern Arizona and southwestern New Mexico. *Madrono* **32**, 78–86.

Freeman, C. E., Worthington, R. D. and Corral, R. D. (1984) Some floral nectar–sugar compositions from Durango and Sinaloa, Mexico. *Biotropica* **17**, 309–313.

Lammers, T. G. and Freeman, C. E. (1986) Ornithophily among the Hawaiian Lobeliaceae (Campanulaceae): evidence from floral nectar sugar compositions. *Am. J. Bot.* **73**, 1613–1619.

Percival, M. S. (1961) Types of nectar in angiosperms. *New Phytol.* **60**, 235–281.

Smith, G. F. and Van Wyk, B.-E. (1991) Generic relationships in the Alooideae (Asphodelaceae). *Taxon* **40**, 557–581.