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New sources of lycopsamine-type pyrrolizidine alkaloids and their distribution in Apocynaceae

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1. Subject and source

Tissues were sampled from 19 species from 14 genera of six tribes of Apocynaceae; 17 species were tested for pyrrolizidine alkaloids for the first time (Table 1). Sampling focused on Echiteae (4 genera, 7 species) and Apocynae (5 genera, 8 species) because these tribes include species that have been previously reported as containing pyrrolizidine alkaloids (Fig. 1, Table 2). In addition, 1 species each was sampled from tribes Asclepiadeae, Melodineae, Mesechiteae, and Vincae (Table 1). Vouchers were identified by Dr. Tatyana Livshultz, Dr. Mary Endress (University of Zurich), Dr. Justin K. Williams (Sam Houston State University), and Dr. David Middleton (Singapore Botanical Garden) and voucher specimens deposited in publically accessible herbaria (Table 1). Lycopsamine-type pyrrolizidine alkaloids were identified by LC-MS using leaf samples of *Parsonsia alboflavescens* as an authentic reference standard (Abe et al., 1991a,b; Abe and Yamauchi, 1987).

Abbreviations: GC-MS, gas chromatography mass spectroscopy; LC-MS, liquid chromatography mass spectroscopy; MRM, multiple reaction monitoring scan; NMR, nuclear magnetic resonance; PA, pyrrolizidine alkaloid; PIS, precursor ion scan; RT, room temperature.

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Table 1

Samples tested for lycopsamine type pyrrolizidine alkaloids (PAs), voucher specimens [herbarium abbreviations from *Index Herbariorum* (Thiers, 2012)], and results. Presence of PAs detected with LC-MS Q1 scan (Q1), precursor ion scan (PIS), and/or multiple reaction monitoring scan (MRM). Compounds identified with MRM except as noted. All identified compounds belong to the parsonsine (C4) subgroup of lycopsamine PAs. The compounds and their (retention time in minutes, m/z [M + H]⁺) are as follows: 1) 17-methyl paronsianidine (10.47, 470.2); 2) 17-methyl paronsianidine *N*-oxide (10.67, 486.2); 3) heterophylline (11.97, 454.3); 4) heterophylline *N*-oxide (12.12, 470.2); 5) ideamine B (10.32, 426.2); 6) ideamine B *N*-oxide (10.52, 442.2); 7) paronsianidine (9.37, 456.1); 8) paronsianidine *N*-oxide (9.58, 472.0); 9) paronsianine (9.37, 442.2); 10) paronsianine *N*-oxide (9.58, 458.3); 11) paronsine or isomer 1 (11.17, 440.3); 12) paronsine or isomer 2 (11.27, 440.3); 13) paronsine or isomer 3 (11.32, 440.2); 14) spiraline (9.82, 456.3); 15) spiraline *N*-oxide (10.12, 472.3); 16) Unknown A (10.82, 458.3); 17) Unknown B (12.82, 468.2); 18) Unknown C (11.47, 472.3). It has been previously noted by Wuilloud et al. (2004) and others that the *N*-oxide analogs of PAs elute at a later retention time than the corresponding free amines under acidic LC conditions.

Tribe	Species	Voucher collector and collector number, date (herbarium)	Provenance	Organ/tissue	Age when tested	Storage conditions	PAs present			Compounds identified
							Q1	PIS	MRM	
Apocynaceae	<i>Aganosma cymosa</i> (Roxb.) G. Don	Middleton 1101, August 16, 2002 (A)	Thailand	Leaf	12 Years	Silica dried/RT	No	No		
Apocynaceae	<i>Anodendron affine</i> (Hook. & Arn.) Druce	Livshultz 2014-3, May 23, 2014 (PH)	Taiwan	Leaf	1 Month	Silica dried/RT			No	
Apocynaceae	<i>Anodendron oblongifolium</i> Hemsl.	Takeuchi 14513, June 27, 2000 (A)	Papua New Guinea	Leaf	14 Years	Dry/RT			No	
Apocynaceae	<i>Apocynum cannabinum</i> L.	Livshultz 03-28a (BH)	New York, USA	Leaf	10 Years	Silica dried/RT	No	No		
Apocynaceae	<i>Epigynum auritum</i> (C.K. Schneid.) Tsiang & P.T. Li	Middleton 1457, August 29, 2002 (A)		Leaf	11 Years	Silica dried/RT	No	No		
Apocynaceae	<i>Urceola lucida</i> (Wall. ex Don) Kurz	Middleton 2058, April 9, 2003 (A)	Thailand	Leaf	10 Years	Silica dried/RT	No	No		
Apocynaceae	<i>Urceola rosea</i> (Hook. & Arn.) D.J. Middleton	Livshultz 2014-1, May 17, 2014 (PH)	Taiwan	Leaf	1 month	Silica dried/RT			No	
Apocynaceae	<i>Urceola rosea</i> (Hook. & Arn.) D.J. Middleton	Livshultz 2014-1, May 17, 2014 (PH)	Taiwan	Stems & shoots	1 Month	Silica dried/RT			No	
Asclepiadeae	<i>Tylophora ovata</i> (Lindl.) Hook. ex Steud.	Livshultz 2011-2, July 2, 2011 (PH)	Taiwan	Leaf	3 Years	Silica dried/RT	No	No		
Echiteae	<i>Echites panduratus</i> A. DC.	Gomez s.n. (Z)	Costa Rica	Leaf	—	Silica dried/RT	No	No		
Echiteae	<i>Echites turriiger</i> Woodson	Spencer & Williams 2 (SHST)	Mexico	Leaf	8 Years	Silica dried/RT	No	No		
Echiteae	<i>Echites umbellatus</i> Jacq.	Vincent 11468, 15 January, 2004 (GH)	Bahamas	Leaf	10 Years	Silica dried/RT		Yes	Yes 1, 3, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18	
Echiteae	<i>Echites umbellatus</i> Jacq.	Livshultz s.n., December 20, 2011 (PH)	Bahamas	Leaf	2 Years	Silica dried/RT		Yes	Yes 1, 3, 4, 5, 6, 8, 9, 10, 11, 13, 15, 16, 17, 18	
Echiteae	<i>Echites umbellatus</i> Jacq.	Livshultz s.n., December 20, 2011 (PH)	Bahamas	Nectar	2 Years	Dried on filter paper/RT	Yes	No ^a	11, 15, 16 Q1 scan	
Echiteae	<i>Echites umbellatus</i> Jacq.	Livshultz s.n., December 20, 2011 (PH)	Bahamas	Nectar	2 Years	Dried on filter paper/RT		Yes	1, 3, 4, 5, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 18	
Echiteae	<i>Echites umbellatus</i> Jacq.	Livshultz s.n., December 20, 2011 (PH)	Bahamas	Sap	2 Years	Dried on filter paper/RT	Yes	Yes	Yes 1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 13, 15, 16, 17, 18	
Echiteae	<i>Laubertia contorta</i> (M. Martens & Galeotti) Woodson	Williams 2005-1 (SHST)	Mexico	Leaf	9 Years	Silica dried/RT	No	No		
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz 2011-14, July 9, 2011 (PH)	Taiwan	Leaf	3 Years	Silica dried/RT		Yes	1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 13, 14, 15, 16, 17, 18	
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz 2012-23, August 19, 2012 (PH)	Taiwan	Leaf	1 Year	Silica dried/RT	Yes	Yes	Yes 1, 3, 4, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18	
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz 2014-4, May 26, 2014 (PH)	Taiwan	Nectar	1 Month	Dried on filter paper/RT		Yes	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17	
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz 2014-4, May 26, 2014 (PH)	Taiwan	Nectar	1 Month	Dried on filter paper/RT		Yes	1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17	
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz 2012-23, August 19, 2012 (PH)	Taiwan	Nectar	1 Year	Dried on filter paper/RT	Yes	Yes	Yes 1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18	
Echiteae	<i>Parsonsia alboflavescens</i> (Dennst.) Mabb.	Livshultz s.n., August 18, 2013 (PH)	Taiwan	Sap	1 Year	Dried on filter paper/RT		Yes	Yes 1, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18	

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