



Review

# Pyrrolizidine Alkaloids –Pros and Cons for Pharmaceutical and Medicinal Applications

Kavindi Jayawickreme<sup>1,†</sup>, Dawid Świstak<sup>1,†</sup>, Ewa Ozimek<sup>2</sup>, Emilia Reszczyńska<sup>3,4</sup>, Anna Rysiak<sup>5</sup>, Anna Makuch-Kocka<sup>6</sup> and Agnieszka Hanaka<sup>4\*</sup>

<sup>1</sup> Student Scientific Club of Phytochemists, Institute of Biological Sciences, Faculty of Biology and Biotechnology, Maria Curie-Skłodowska University, Akademicka St. 19, 20-033 Lublin, Poland

<sup>2</sup> Department of Industrial and Environmental Microbiology, Institute of Biological Sciences, Faculty of Biology and Biotechnology, Maria Curie-Skłodowska University, Akademicka St. 19, 20-033 Lublin, Poland

<sup>3</sup> Department of Biochemistry and Molecular Biology, Medical University of Lublin, Chodźki St. 1, 20-093 Lublin, Poland

<sup>4</sup> Department of Plant Physiology and Biophysics, Institute of Biological Sciences, Faculty of Biology and Biotechnology, Maria Curie-Skłodowska University, Akademicka St. 19, 20-033 Lublin, Poland

<sup>5</sup> Department of Botany, Mycology, and Ecology, Institute of Biological Sciences, Faculty of Biology and Biotechnology, Maria Curie-Skłodowska University, Akademicka St. 19, 20-033 Lublin, Poland

<sup>6</sup> Department of Pharmacology, Medical University of Lublin, Radziwiłłowska St. 11, 20-080, Lublin, Poland

\* Correspondence: agnieszka.hanaka@mail.umcs.pl

† These authors contributed equally to this work.

**Table S1.** HPLC/UHPLC protocols of PAs detection. Abbreviations: HPLC/UHPLC - high-performance liquid chromatography/ ultra-high-performance liquid chromatography; PCX cartridge - a cation exchanger with mixed mode sorbent characteristics; GCB - graphitised carbon black; PSA - primary secondary amine; C18 - octadecylsilane.

Name of PAs	PAs-producing plants	Column	Extraction condition	Chromatography condition HPLC/UHPLC	Ref.
retrorsine; senecivernine; echimidine-N-oxide; retrorsine-N-oxide; sen-ecivernine-N-oxide; concentration: > 2 µg/ml	<i>Atractylodis Macrocephalae</i> <i>Rhizoma</i> ; <i>Paeoniae Radix Alba</i> ; <i>Angelicae Sinensis Radix</i> ; <i>Bupleuri Radix</i> ; <i>Glycyrrhizae Radix</i> Et <i>Rhizoma Praeparata Cum</i> Melle	Thermo Hypersil Gold C18 (100 × 2.1 mm, 1.9 µm)	1% formic acid; sonication: room temp., 20 min; supernatant applied to the PCX cartridge; pretreated: methanol + 0.05 M sulfuric acid solution (1:1, v/v); rinsed: methanol + 0.05 M sulfuric acid solution;  PAs: washed with ammonia + methanol (1:3, v/v) solution	<b>mobile phase A:</b> 0.05% formic acid solution (containing 2.5 mM/L ammonium formate); <b>mobile phase B:</b> methanol + 0.05% formic acid solution (containing 2.5 mM/L ammonium formate); <b>elution</b> <b>gradient:</b> 95% A, 0 min–0.5 min; changed with a linear gradient to: 90% A, 0.5 min–5 min; 85% A, 5 min–11 min; 80% A, 11 min–13 min; 35% A, 13 min–14 min; 5% A, 14 min–16.5 min; keeping isocratic elution for 2.5 min; 95% A, 16.5 min–16.6 min, 95% A, 16.6 min–18 min; <b>flow rate:</b> 0.4 mL/min, <b>column temperature:</b> 40°C, <b>injection volume:</b> 2 µL in 50% methanol	[1]
green tea, black tea, herbal infusion	<i>Asteraceae: Eupatorium can-</i> <i>nabinum, Petasites hybridus,</i> <i>Senecio vulgaris, Tussilago</i> <i>farfara</i> );	Omega Polar C18 (2.1 × 100 mm, 1.6 µm;	aqueous extracts aliquot of the aqueous solution (10 ml) brought to a concentration of 1	<b>mobile phase A:</b> H <sub>2</sub> O and <b>mobile phase B:</b> MeCN - both containing 0.1% of formic acid;	

lasiocarpine; retrosine; senecionine; monocrotaline; senkirikine; e.g., ~100 µg kg <sup>-1</sup>	<i>Boraginaceae: Anchusa officinalis; Borago officinalis, Echium italicum, Heliotropium europaeum, Lithospermum officinale, Symphytum officinale</i>	Phenomenex) column	M of MgSO <sub>4</sub> ·7H <sub>2</sub> O, 1.5 M Na <sub>2</sub> SO <sub>4</sub> , pH 9.6; centrifugated: 5 min (13,000 rpm); 2 mL of the aqueous solution extracted with 2 mL of MeCN by vortexing for 1 min; centrifuged: 5 min (13,000 rpm) to achieve the phase separation; upper organic phase (MeCN)-dry under a gentle nitrogen flow; re-dissolved in 200 µL of H <sub>2</sub> O/MeOH (7:3 v/v) for herbal infusions and tea	<b>elution gradient:</b> 2% B, 0–1 min; 2–8% B, 1–5.5 min; 8% B, 5.5–7.5 min; 8–12% B, 7.5–9.5 min; 12–18% B, 9.5–11 min; 18–20% B, 11–13 min; 20–40% B, 13–15 min; 40–60% B, 15–17 min; 60–80% B, 17–19 min; after each injection: washing (98% B, 4 min) and re-equilibration of the column (2% B, 5 min); <b>flow rate:</b> 0.4 mL/min, <b>column temperature:</b> 40 °C; <b>injection volume:</b> 5 µL in H <sub>2</sub> O/MeOH (7:3, v/v)	[2]
heliotrine; europine	<i>Heliotropium dasycarpum</i> Ledeb.	Luna Omega Polar C18 column (3 µm, 250 × 4.6 mm)	dried plant material (3 kg); extraction: distilled MeOH twice (5 L, room temp.); solvent: evaporated under reduced pressure to get 75 g mass; methanolic extract (75 g) suspended in distilled water (500 mL); extracted with hexane and ethyl acetate; achieved fractions: hexane (35 g), ethyl acetate (22 g), and residual water (16 g)	<b>mobile phase A:</b> 0.1% formic acid in H <sub>2</sub> O; <b>mobile phase B:</b> acetonitrile gradient: 10–100% B, 45 min; <b>flow rate:</b> 0.5 mL/min; <b>injection volume:</b> 3 µL; UV detector: 190–800 nm	[3]
heliotrine, heliotrine- <i>N</i> -oxide, retrorsine,	fresh tea leaves	HSS T3 column (2.1 × 100 mm,	leaves (1g); 10 mL of 0.1 M sulfuric acid solution; vortexed: 1 min;	<b>mobile phase A:</b> 0.1% formic acid + 1 mM ammonium formate in methanol'	

retrorsine- <i>N</i> -oxide, denecionine, senecionine- <i>N</i> - oxide, jacobine, jacobine- <i>N</i> -oxide, intermedine, intermedine- <i>N</i> - oxide, seneciphylline, seneciphylline <i>N</i> - oxide, europine, europine- <i>N</i> -oxide, senkirrine	1.8 µm, Waters)	ultrasonically extracted: 15 min; centrifuged: 10 min (9,390g); after repetition of extraction supernatant was combined; 2 mL aliquot of the supernatant placed with the adsorbents GCB:PSA:C18 (10:20:15 mg); vortexed: 1 min; centrifuged: 8 min (9,390g) at room temp.	<b>mobile phase B:</b> 0.1% formic acid + 1 mM ammonium formate in H <sub>2</sub> O; <b>elution gradient:</b> 10% A, 0–0.25 min; 10–30% A, 0.25–6.0 min; 30–40% A, 6.0–9.0 min; 40–98% A, 9.0–9.01 min; held for 1.9 min; 98–100% A 11.0–11.1 min; held for 2.9 min; <b>flow rate:</b> 0.25 mL/min; <b>column temperature:</b> 40 °C; <b>injection volume:</b> 3 µL;	[4]
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**Table S2.** Overview of PA-rich family *Boraginaceae*, where: life forms per – perennial plant, ann – annual, bienn – biennial. Geographical range and life forms based on the Plants of the World online [6] herbal raw materials according to Smith and Culvenor [7], and names of plant taxa according to the International Plant Names Index [8].

Genus	Species	Geographical range		Life form	Raw material
		Native	Secondary		
<i>Alkanna</i>	<i>orientalis</i>	S Europe, W Asia		per	herba
	<i>tinctoria</i>	S Europe		per	herba
<i>Amblynotus</i>	<i>rupestris</i>	Asia C		subshrub	herba
<i>Amsinckia</i>	<i>carinata</i>	America N		ann	herba
	<i>douglasiana</i>	America N		ann	herba
	<i>eastwoodiae</i>	America N		ann	herba
	<i>grandiflora</i>	America N		ann	herba
	<i>caycina</i>	W, S America S		ann	herba
	<i>lunaris</i>	America N		ann	herba
	<i>lycopsoides</i>	America N		ann	herba
	<i>lycopsoides</i> × <i>menziesii</i>	America N	Europe, Asia	ann	herba
	<i>var. intermedia</i>				
	<i>menziesii</i>	America N		ann	herba
	<i>menziesii</i> var. <i>intermedia</i>	S America N		ann	herba
	<i>spectabilis</i> var. <i>microcarpa</i>	America N		ann	herba
	<i>specabtilis</i> var. <i>spectabilis</i>	America N		ann	herba
	<i>tessellata</i>	America N, S		ann	herba
	<i>tessellata</i> var. <i>gloriosa</i>	America N		ann	herba
	<i>tessellata</i> var. <i>tessellata</i>	America N, S		ann	herba
	<i>vernica</i> var. <i>furcata</i>	America N		ann	herba
<i>Anchusa</i>	<i>arvensis</i>	Europe	America N	ann	herba
	<i>hispida</i>	Africa, Asia		ann	herba
	<i>milleri</i>	Africa, Asia		ann	herba
	<i>officinalis</i>	Europe, Asia		per	herba
	<i>strigosa</i>	Europe, Asia		per	herba
<i>Arnebia</i>	<i>decumbens</i>	Europe, Asia		ann	herba
	<i>euchroma</i>	Asia		per	herba
	<i>hispidissima</i>	Africa, Asia		ann	herba
<i>Asperugo</i>	<i>procumbens</i>	Europe, Asia	America N	ann	herba
<i>Borago</i>	<i>officinalis</i>	Europe	America N	ann	herba, seeds
	<i>macranthera</i> var.	Asia		per	herba
<i>Caccinia</i>	<i>crassifolia</i>				
<i>Cerithe</i>	<i>glabra</i>	Asia		bienn	herba
	<i>minor</i>	Europe, Asia		ann	herba
<i>Cordia</i>	<i>myxa</i>	Asia		per/tree	herba
	<i>sinensis</i>	Africa, Asia		per/shrub/tree	herba
<i>Cryptantha</i>	<i>cana</i>	America N, S		ann	herba

<b>Cynoglossum</b>	<i>clevelandii</i>	America N, S		ann	herba
	<i>confertiflora</i>	America N, S		per	herba
	<i>crassipes</i>	America N, S		per	herba
	<i>fendleri</i>	America N, S		per	herba
	<i>flava</i>	America N, S		per	herba
	<i>inequata</i>	America N, S		per	herba
	<i>jamesii</i>	America N, S		per	herba
	<i>leiocarpa</i>	America N, S		per	herba
	<i>thyrsiflora</i>	America N, S		per	herba
	<i>utahensis</i>	America N, S		per	herba
	<i>virgata</i>	America N, S		per	herba
	<i>virginiensis</i>	America N, S		per	herba
	<i>amabile</i>	Asia	America N, S	ann/bienn	herba
	<i>australe</i>	Australia	America N, S	per	herba
	<i>clandestinum</i>	Europe	America N, S	bienn	herba
	<i>columnae</i>	Europe	America N, S	ann	herba
	<i>creticum</i>	Europe	America N, S	per	root, herba
	<i>furcatum</i>	Asia	America N, S	per	herba
	<i>germanicum</i>	Europe, Asia	America N, S	bienn	herba
	<i>glochidiatum</i>	Europe, Asia	America N, S	per	herba
	<i>lanceolatum</i>	Africa, Asia	America N, S	per	herba
	<i>latifolium</i>	Asia	America N, S	per	herba
	<i>macrostylum</i>	Europe	America N, S	per	herba
	<i>montanum</i>	Europe, Asia	America N, S	per	herba
	<i>nervosum</i>	Europe, Asia	America N, S	per	herba
	<i>officinale</i>	Europe	America N, S	per	root, herba
	<i>pictum</i>	Europe	America N, S	per	root, herba
	<i>viridiforum</i>	Asia	America N, S	per	root, herba
<b>Echium</b>	<i>amoenum</i>	Europe, Asia		bienn	herba
	<i>angustifolium</i>	Europe		per	herba
	<i>diffusum</i>	Europe		per	herba
	<i>glomeratum</i>	Europe, Asia		bienn	herba
	<i>horridum</i>	Africa		ann/per	herba
	<i>humile</i>	Africa, Europe		per	herba
	<i>hypertropicum</i>	Africa		shrub	herba
	<i>italicum</i>	Europe, Asia		bienn	herba
	<i>plantagineum</i>	Europe		ann/bienn	herba
	<i>pininana</i>	Europe		bienn/subshr ub	herba
	<i>rauwolfii</i>	Africa, Asia		ann	herba
	<i>angustifolium ssp.</i>	Africa, Asia		per	herba
	<i>sericeum</i>				

	<i>simplex</i>	Europe		bienn/subshr ub	herba
	<i>stenosiphon</i> ssp.			subshrub/shr ub	herba
	<i>stenosiphon</i>	Africa			
	<i>tuberculatum</i>	Africa, Spain		bienn	herba
	<i>vulgare</i>	Europe, Asia		ann/bienn	herba
	<i>wildpretti</i>	Europe		bienn/subshr ub	herba
<b>Ehretia</b>	<i>aspera</i>	Asia		shrub/tree	herba
<b>Euploca</b>	<i>bracteatum</i>	Australia		ann	herba
	<i>bursiferum</i>	America C		ann	herba
	<i>ovalifolium</i>	N Africa, Asia		ann/subshrub	herba
	<i>parviantrum</i>	C, E Australia		ann	herba
	<i>procumbens</i>	S America N		ann	herba
	<i>queretaroanum</i>	S America N		ann	herba
	<i>racemosum</i>	S America N		subshrub	herba
	<i>marifolia</i> ssp. <i>marifolia</i>	S Asia		ann/subshrub	herba
	<i>sessei</i>	S America N		ann	herba
	<i>humilis</i>	S America N		subshrub	herba
	<i>wigginsii</i>	S America N		ann	herba
<b>Hackelia</b>	<i>californica</i>	America N		per	herba
	<i>uncinata</i>	C Asia		per	herba
	<i>floribunda</i>	America N		bienn/per	herba
	<i>velutina</i>	America N		per	herba
<b>Heliotropium</b>	<i>acutiflorum</i>	Asia		subshrub	herba
	<i>amplexicaule</i>	America S		subshrub	herba
	<i>angiospermum</i>	America S, C, N		ann/subshrub	herba
	<i>arbainense</i>	Africa, Asia		subshrub	herba
	<i>arborescens</i>	America S		subshrub/shr ub	herba
	<i>arguzioides</i>	Europe, Asia		subshrub	herba
	<i>bacciferum</i>	Africa		subshrub	herba
	<i>bovei</i>	Europe		ann	herba
	<i>arboreum</i>	E Africa, W Australia		shrub	herba
	<i>sibiricum</i>	SE Europe, Asia		per	herba
	<i>circinatum</i>	Asia		ann	herba
	<i>confertifolium</i>	Asia		ann	herba
	<i>crassifolium</i>	Asia		ann	herba
	<i>curassavicum</i>	Australia, America N, S	Africa N	ann/subshrub	herba, seeds
	<i>curassavicum</i> var. <i>argentinum</i>	America S, Australia	Africa N	ann/subshrub	herba

<i>Euploca</i>	<i>curassavicum</i> var.	Australia, America N,	Africa N	ann/subshrub	herba
	<i>curassavicum</i>	S			
	<i>dasycarpum</i>	Asia		subshrub	root, herba
	<i>digynum</i>	Africa N, Asia		subshrub	herba
	<i>disciforme</i>	Asia		per	herba
	<i>dissitiflorum</i>	Asia		per	herba
	<i>elipticum</i>	Asia		per	herba, seeds
	<i>esfandarii</i>	Asia		per	herba
	<i>europaeum</i>	Europe, Asia		per	herba
	<i>floridum</i>	America S		subshrub	herba
	<i>foliosissimum</i>	America N		subshrub	herba
	<i>fruticosum</i>	America N, S		per	herba
	<i>hirsutissimum</i>	Europe		per	herba
			America C,		
	<i>indicum</i>	America S	Africa C, Asia SE	per	herba
	<i>keralense</i>	Asia		per	herba
	<i>lasiocarpum</i>	Africa, Asia		per	herba
	<i>marifolium</i>	Asia		per	herba
	<i>maris mortui</i>	Asia		per	herba
	<i>megalanthum</i>	America S		subshrub/shrub	herba
	<i>molle</i>	S America N		ann	herba
	<i>olgae</i>	C Asia		per	root, herba
	<i>popovii</i> ssp. <i>gillianum</i>	Asia		per	seed
	<i>ramosissimum</i>	NE Africa, W Asia		subshrub	herba
	<i>rotundifolium</i>	N Africa		subshrub	herba
	<i>curassavicum</i> var.	N America N		per	herba
	<i>obovatum</i>				
	<i>steudneri</i>		S, N Africa	subshrub/shrub	herba
	<i>ophiglosum</i>		NE Africa, W Asia	subshrub	herba
	<i>suaveolens</i>		SE, E Europe, W Asia	ann	herba
	<i>zeylandicum</i>		S Africa, S Asia	per	herba
	<i>sarmentosum</i>		E Asia, Australia	shrub	herba
	<i>supinum</i>		E, S, C Europe, Africa, W Asia	ann	leaf, herba
		S America N, S			root,
	<i>humilis</i>	America S		subshrub	herba
	S America N, S		subshrub/shrub	herba	
<i>transalpinum</i>	America S		ub		



	<i>transalpinum</i> var.			subshrub/shrub	herba
	<i>transalpinum</i>	America N, C		ub	
	<i>dasycarpum</i> ssp.				
	<i>transoxanum</i>	C Asia		subshrub	herba
	<i>intermedia</i>	C, E Asia		ann	herba
	<i>squarrosa</i>	Eurasia	America N	bienn	herba
	<i>spinocarpos</i>	N Africa, W Asia		ann	herba
<b>Lindelofia</b>	<i>anchusoides</i>	C Asia		per	herba
	<i>stylosa</i> ssp. <i>stylosa</i>	C Asia		per	herba
	<i>longiflora</i>	C Asia		per	herba
	<i>olgae</i>	C Asia		per	herba
	<i>stylosa</i> ssp. <i>pterocarpa</i>	C Asia		per	herba
	<i>longiflora</i> var. <i>longiflora</i>	C Asia		per	herba
	<i>stylosa</i>	C Asia		per	seed
	<i>tschimganica</i>	C Asia		per	herba
<b>Lithospermum</b>	<i>canesens</i>	C America N		per	herba
	<i>erythrorhizon</i>	E Asia		per	herba
	<i>officinale</i>	Eurasia	E America N	per	herba
<b>Lithodora</b>	<i>fruticosa</i>	N Africa, France, Spain		subshrub	herba
<b>Moltikiopsis</b>	<i>ciliata</i>	N Africa, W Asia		subshrub	herba
<b>Myosotis</b>	<i>scorpioides</i>	Eurasia	America N	per	herba
	<i>sylvatica</i>	Eurasia	America N	per	herba
<b>Neatostema</b>	<i>apulium</i>	S Europe, N Africa, W Asia		ann	herba
<b>Nonea</b>	<i>lutea</i>	W Asia, C Europe		ann	herba
	<i>setosa</i>	C Asia		per	herba
	<i>echioides</i>	S Europe, W Asia		ann	herba
<b>Omphalodes</b>	<i>verna</i>	S Europe	C, N Europe, E America N	per	herba
<b>Onosma</b>	<i>alborosea</i>	W Asia		subshrub	herba
	<i>arenaria</i>	C, SE Europe		bienn/per	herba
	<i>arenaria</i> ssp. <i>pennina</i>	S Europe		per	herba
	<i>erecta</i>	S Europe		per/subshrub	herba
	<i>hetrophyllum</i>	S Europe		per/subshrub	herba
	<i>leptantha</i>	S Europe		per	herba
	<i>stellulata</i>	S Europe		per	herba
<b>Paracaryum</b>	<i>himalayense</i>	C Asia		ann	herba
<b>Microparacaryum</b>	<i>intermedium</i>	ES Europe, W Asia		ann	herba
	<i>regulosum</i>	ES Europe, W Asia		bienn/per	herba
<b>Cynoglossum</b>	<i>zeylandicum</i>	S Asia		bienn	root, herba
<b>Pulmonaria</b>	<i>obscura</i>	Europe to W. Siberia		per	herba

Rindera	austroechinata	C Asia	per	leaf, root, herba, seed	
	tetraspis	E Europe, W Asia	per	herba	
	echinata	C Asia	per	herba	
	oblongifolia	C Asia	per	herba	
	umbellata	SE Europe	per	herba	
Solenanthus	circinnatus	W, C Asia	per	herba, seed, root	
	coronatus	S Europe	per	herba	
	karateginus	C Asia	per	herba	
	turkestanicus	W, C Asia	per	herba	
Symphytum	aintabicum	W Asia	per	root, herba	
	asperum	W Asia	America N, N Europe	per	root, herba
	bohemium	E. Europe to Caucasus.	S, W Europe	per	root, herba
	caucasium	Caucasus	S, W Europe	per	root, herba
	consolidum	Europe		per	root, herba
	grandiflorum	Europe	N, C America S; C Asia	per	root, herba
	ibericum	W Asia	W Europe	per	root, herba
	officinale	Eurasia	America N, C, S	per	root, herba
	orientale	SE, E. Europe, W Asia	C Europe	per	root, herba
	officinale ssp. officinale	Eurasia	America N	per	root, herba
	sylvaticum ssp. sepulcare var. sepulcare	W Asia		per	root, herba
	tanaicense	C, E Europe		per	root, herba
	tuberosum	C, W Europe		per	root, herba
	× uplandicum	C Asia	Europe	per	root, herba
Trahlenthus	hissaricus	Asia		per	herba
	korolkovii	Asia		per	herba
Trichodesma	africanum	Africa		per	herba
	ehrenbergii	Africa, Asia		ann	herba
	incanum	Asia		per	herba, seed, root
	zeylanicum	Africa, Asia		ann	herba
Ulugbekia	tschimganica	Asia		per	herba

**Table S3.** Dynamics of PAs occurrence in species of the family *Boraginaceae*. Group I – rare, exclusive compounds, found in 1 to 3 species; Group II – moderately frequent compounds, recognized in 4 to 9 species; Group III – frequent compounds, found in 10 to 15 species; Group IV – very frequent compounds, recognized in 16 to 20 species; Group V – common compounds, found in more than 20 species. In each group metabolites are ranked alphabetically by increasing occurrence in plants. List of the compounds was prepared according to El-Shazly and Wink [5]. Metabolites from each group ranked alphabetically by increasing occurrence in plants.

Number and Name of PA							
Group I							
1	3'-Acetylcanesine	45	Dihydroechinatine	89	(7S,8S) Petranine	133	Erythro-2'',3''-chloro-2''-hydroxyechiumine
2	3'-Acetylcanescenine	46	Echiupine	90	Pictumine	134	Heliospathuline
3	7-Acetyl-9-curassavoylheliotridine	47	Echivulgarine	91	Platynecine	135	Heliotridine
4	7-Acetyl-9-(2,3-dihydroxybutryl) retronecine	48	Ehretinine	92	Platynecine N-oxide 2S-hydroxy-2S (1S-hydroxyethyl)-4-methyl-pentanosyl ester	136	9-(3'-Isovaleryl)viridiflory retronecine
5	7-Acetyl-9-(2-dimethylbutryl) retronecine	49	7-Epi-echimiplate	93	Punctanecine	137	Longitubine
6	7-Acetylechinatine	50	1 $\alpha$ -2 $\alpha$ -Epoxy-1 $\beta$ -hydroxymethyl-8 $\alpha$ -pyrrolizidine	94	Pycnanthine	138	7-(2-Methylbutyryl)retronecine
7	3'- Acetylechihumiline	51	Floridanine	95	Retronecine-7:9- dibenzoate	139	7-(2-Methylbutyryl)-9-(2,3-dihydroxybutyryl)retronecine
8	3'-Acetylechiumine	52	Floridimine	96	Retronecine 2S-hydroxy-2S(1S-hydroxyethyl)-4-methyl-pentanosyl ester	140	Monocrotaline
9	3'-Acetylechimidine	53	Floridine	97	Retronecine 2S-hydroxy-2S(1R-hydroxyethyl)-4-methyl-pentanosyl ester	141	Neolatifoline
10	5'-Acetylechimidine	54	Floridinine	98	Scorpioidine	142	7-Senecioylhelotridine
11	5'-Acetyleuropine	55	Hackelidine	99	7-Senecioylrinderine	143	9-Senecioylretronecine
12	7-Acetyleuropine	56	Helibracteatine	100	7-Senecioyllycopsamine	144	7-Tigloyl-9-(2,3-dihydroxypropanoyl)retronecine
13	7-Acetyl-9-(2-hydroxy-3-methylbutryl) retronecine	57	Helibractinecine	101	Sincamidine	145	7-Tigloyllycopsamine
14	5'-Acetylasiocarpine	58	Helibracteatinecine	102	Senkirkine	146	Trachelanthine
15	7-Acetyl-9-latifolylretronecine	59	Helibracteatinine	103	Strigosine	147	Triangularine
16	3'-Acetylolithosenine	60	Heliofoline	104	Supinidine N-oxide 2S-hydroxy-2S(1S-hydroxyethyl)-4-methyl-pentanoyl ester	148	Triangularicine
17	7-Acetyl-9-(2-methylbutyryl) retronecine	61	Helindicine	105	Thesinine	149	Viridantine
18	7-Acetylretronecine	62	Heliocoromandaline	106	Thesinine-4'-O- $\beta$ -D-glucoside	150	3'-Acetylheliosupine
19	7-Acetyl-9-sarracinoyl retronecine	63	Heliocurassavine	107	7-Tigloyl-9-(2-deoxy-2-methyl) echimidinyheliotridin	151	9-Acetytessellatine
20	7-Acetylscorpioidine	64	Heliocurassavicine	108	7-Tigloylheliotridine	152	7-Angeloylheliotridine
21	3'-Acetylsupinine	65	Heliocurassavinine	109	7-Trachelanthyl-laburnine	153	Asperumine

22	3'-Acetyltessellatine	66	Heliospathine	110	7-Trachelanthylretronecine	154	Carategine
23	3'-Acetyltrachelanthamine	67	Heliotridine 2S-hydroxy-2S (1S-hydroxyethyl)-4-methyl-pentanoyl ester	111	Transalpinecine	155	Curassavine
24	9-Acetyltrachelanthamine	68	Heliovicine	112	Uluganine	156	Cynaustine
25	3'-Acetylviridiflorine	69	Heliscabine	113	Vulgarine	157	3',7-Diacetylintermediate
26	7-Acetylvulgarine	70	Hydroxymyoscorpine	114	3'-Acetylfurcatine	158	thero-2'',3''-Dihydroxyechiumine
27	7 $\alpha$ -Angeloyl-1-chloromethyl-1,2-dihydropyrrolizidine	71	Ilamine	115	3'-Acetylindicine	159	Echimiplate
28	7-Angeloyl-9-(2,3-dihydroxybutyryl) heliotridine	72	Indicine	116	3'-Acetylmyscorpine	160	Echiuplatine
29	7-Angeloyl-1-formyl-6,7-dihydro-5H-pyrrolizidine	73	Isoechinatinine	117	3'-Acetylinderine	161	Furcatine
30	7-Angeloylechinatine	74	Isolycopsamine	118	9-(3'-Acetyl)viridiflory retronecine	162	Incanine
31	7-Angeloylinderine	75	Isoretronocanol (or its isomer)	119	9-(3'-Acetyl)viridifloryl turnifordine	163	7-Seneciylretronecine
32	9-Angeloyltrachelanthamidine	76	Lactodine	120	Anadoline	164	Symlandine
33	7-Angeloyl-9-(+)-trachelanthylheliotridine	77	9-Latifolylretronecine	121	7-Angeloyl-9-(2,3-dihydroxypropanoyl)retronecine	165	Symviridine
34	9-(Butyryl-2-ene) supinidine	78	Lindelofamine	122	7-Angeloyl-9-(hydroxypropenoyl) retronecine	166	Trichodesmine
35	Canescine	79	Lithosenine	123	7-Angeloyllycopsamine	167	Turkestanine
36	Canescenine	80	Macrophylline	124	7-Angeloyl-9-(2-methylbutyryl)heliotridine	168	Uplandicine
37	Cryptanthine	81	Macrotamine	125	7-Angeloyl-9-(2-methylbutyryl)retronecine		
38	Curassanecine	82	Megalanthonine	126	Coromandaline		
39	9-Curassavorylheliotridine	83	Methyechiuplatine	127	Coromandalinine		
40	Cynoglossamine	84	1-Methylene-8 $\alpha$ -pyrrolizidine	128	Curassavinine		
41	Dehydroheliotrine	85	9-(2-Methylbutyryl) retronecine	129	3',7-Diacetyllycopsamine		
42	5-Deoxylasiocarpine	86	Neocoromandaline	130	Dihydroxytriangularine		
43	3',9-Diacetyltessellatine	87	Onosmerectine	131	Dihydroxytriangularicine		
44	5,6-Dihydro-7,9-dimethoxy-7H-pyrrolizidine	88	(7S,8R) Petranine	132	2'',3''-Epoxyechiumine		

#### Number and Name of PAs

Group II	Group III	Group IV	Group V
1 3'-Acetylechinatine	1 7-Acetyllycopsamine	1 3'-Acetylintermediate	1 Retronecine

2	7-Angeloyl-9-(2,3-dihydroxybutyryl)ret- ronecine	2	Echiumine	2	Lasiocarpine	2	7-Acetyllycopsamine
3	Echiumine	3	Trachelanthamine	3	7-Angeloylretronecine	3	Heliotrine
4	7-(2-Methylbutyryl)-9- echimidinyl retronecine	4	Symphytine	4	7-Acetylintermediate	4	Echimidine
5	7-Tigloyl-9-(2,3-dihydroxybutyryl) ret- ronecine	5	3'-Acetyllycopsamine	5	Amabiline	5	Supinine
6	7-Tigloyl-9-(2-methylbutyryl)ret- ronecine	6	9-Angeloylretronecine			6	Echinatine
7	9-Angeloyl-7- viridiflorylretronecine	7	Europine			7	Intermedine
8	Cynaustaline	8	Heliosupine			8	Lycopsamine
9	Cynaustaline	9	Rinderine				
10	Echimidine isomer (tig- loyl)	10	Supinidine				
11	Leptanthine	11	Viridiflorine				
15	Subulacine	12	Lindelofidine				
13	7-Tigloylretronecine	13	Tessellatine				
14	7-Viridiflorylret- ronecine	14	Trachelanthamidine				
15	Heleurine						
16	Indicine						
17	Latifoline						
18	Lindelofine						
19	Myoscorpine						
20	9-Tigloylretronecine						
21	7-Angeloylheliotridine						

## References

- Cheng, S.; Sun, W.; Zhao, X.; Wang, P.; Zhang, W.; Zhang, S.; Chang, X.; Ye, Z. Simultaneous determination of 32 pyrrolizidine alkaloids in two traditional Chinese medicine preparations by UPLC-MS/MS. *J. Anal. Methods Chem.* **2022**, 2022, doi:10.1155/2022/7611501.
- Rizzo, S.; Celano, R.; Piccinelli, A.L.; Serio, S.; Russo, M.; Rastrelli, L. An analytical platform for the screening and identification of pyrrolizidine alkaloids in food matrices with high risk of contamination. *Food Chem.* **2023**, 406, doi:10.1016/j.foodchem.2022.135058.
- Mukhtar, M.; Saleem, M.; Nazir, M.; Riaz, N.; Shafiq, N.; Saleem, H.; Tauseef, S.; Khan, S.; Ehsan Mazhar, M.; Bakhsh Tareen, R.; et al. Identification of pyrrolizidine alkaloids and flavonoid glycosides through HR-LCMS/MS analysis, biological screening, DFT and molecular docking studies on *Heliotropium dasycarpum* Ledeb. *Arab. J. Chem.* **2023**, 16, doi:10.1016/j.arabjc.2023.104655.

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4. Jiao, W.; Zhu, L.; Shen, T.; Wang, L.; Li, Q.X.; Wang, C.; Wu, X.; Chen, H.; Hua, R. Simultaneous determination of 15 pyrrolizidine alkaloids and their N-oxides in weeds, soil, fresh tea leaves, and tea: exploring the pollution source of pyrrolizidine alkaloids in tea. *Food Chem.* **2024**, *434*, 137305, doi:10.1016/j.foodchem.2023.137305.
  5. El-Shazly, A.; Wink, M. Diversity of pyrrolizidine alkaloids in the Boraginaceae structures, distribution, and biological properties. *Diversity* **2014**, *6*, 188–282, doi:10.3390/d6020188.
  6. Plants of the World online; Board of Trustees of the Royal Botanic Gardens, Kew Available online: <https://powo.science.kew.org>.
  7. Culvenor, C.C.J. Tumor-Inhibitory activity of pyrrolizidine alkaloids. *J. Pharm. Sci* **1968**, *57*, 1112–1117.
  8. IPNI – The International Plant Names Index Available online: <https://www.ipni.org>.