Botanica Pacifica. A journal of plant science and conservation. 2026.
DOI: 10.17581/bp.2026.15211

RESEARCH PAPER

Comparative phytochemical profiling of *Crinum latifolium* leaf extracts obtained by supercritical CO_2 extraction and ethanol maceration using HPLC-ESI-MS/MS

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Manuscript received: 06.05.2026
Review completed: 25.08.2026
Accepted for publication: 06.09.2026
Published online: 09.09.2026

ABSTRACT

Crinum latifolium L. is an Amaryllidaceae medicinal plant known for its alkaloid constituents, but its detectable phytochemical profile may vary with extraction conditions. We compared leaf extracts obtained by supercritical CO_2 (SC-CO_2) extraction and ethanol maceration using HPLC-ESI-MS/MS. We tentatively annotated 98 compounds in 12 chemical classes, with flavonoids and flavonoid glycosides forming the largest class, followed by alkaloids and phenolic acids. We detected 68 compounds in the SC-CO_2 extract and 42 in the ethanol extract, with 12 occurring in both, giving a Jaccard similarity index of 0.122. Based on the literature examined, 66 compounds represent potential first records for *C. latifolium*. The two extraction procedures produced distinct but complementary detectable profiles, and the broader chemical coverage observed for SC-CO_2 under these conditions does not imply higher compound concentrations or extraction yields.

Keywords: *Crinum latifolium*, Amaryllidaceae alkaloids, supercritical CO_2 extraction, ethanol maceration, comparative phytochemical profiling, HPLC-ESI-MS/MS, flavonoids

РЕЗЮМЕ

Чан В.К., Разгонова М.П., Голохваст К.С. Сравнительный фитохимический профиль экстрактов листьев *Crinum latifolium*, полученных методом сверхкритической CO_2-экстракции и этанольной мацерации, с использованием ВЭЖХ-ЭСИ-МС/МС. *Crinum latifolium* L. – лекарственное растение семейства Amaryllidaceae, известное как источник алкалоидов; при этом его выявляемый фитохимический профиль может зависеть от условий экстракции. Мы сравнили экстракты листьев, полученные методом сверхкритической CO_2-экстракции (SC-CO_2) и мацерации этанолом, с использованием ВЭЖХ-ЭСИ-МС/МС. В общей сложности мы предварительно аннотировали 98 соединений, относящихся к 12 химическим классам; наиболее представленной группой были флавоноиды и флавоноидные гликозиды, за которыми следовали алкалоиды и фенольные кислоты. В SC-CO_2-экстракте было обнаружено 68 соединений, а в этанольном экстракте 42, причем 12 соединений были общими для обоих экстрактов; индекс сходства Жаккара составил 0,122. На основании изученной литературы 66 соединений можно рассматривать как предполагаемые первые находки для *C. latifolium*. Два метода экстракции дали различающиеся, но взаимодополняющие фитохимические профили. Более широкий химический охват, наблюдаемый для SC-CO_2-экстракта в использованных условиях, не свидетельствует о более высоких концентрациях отдельных соединений или большем выходе экстракта.

Ключевые слова: *Crinum latifolium*, алкалоиды амариллисовых, экстракция сверхкритическим CO_2, мацерация этанолом, сравнительное фитохимическое профилирование, ВЭЖХ-ЭСИ-МС/МС, флавоноиды

Crinum latifolium L. is a perennial herb of the family Amaryllidaceae, characterized by a bulbous habit, strap-shaped basal leaves, and white to pale-pink flowers (Dolai & Nandi 2021, Gasca-Silva et al. 2022) (Fig. 1). The species is distributed in tropical and subtropical Asia and is used as a medicinal plant in Vietnam (Tram & Khanh 2012).

Previous investigations of *C. latifolium* have addressed its traditional use, isolated constituents, and biological properties of extracts or purified compounds (Tram & Khanh 2012, Chen et al. 2018, Gasca-Silva et al. 2022, Thongphichai et al. 2022). These studies provide useful background; however, biological activity cannot be inferred solely from the detection of a compound in an extract.

Phytochemical studies have established Amaryllidaceae alkaloids as characteristic constituents of *C. latifolium*, particularly compounds of the lycorine, crinine, haemanthamine, and tazettine types (Gasca-Silva et al. 2022). Leaves and bulbs have yielded compounds such as 6-methoxyundulatine, 6-methoxycrinamidine, undulatine N-oxide, crinalatifolines, and 6-epihydroxypowelline (Chen et al. 2018, Hanh et al. 2018, Tian et al. 2021, Chaichompoo et al. 2024, Trang et al. 2024). However, most previous work has

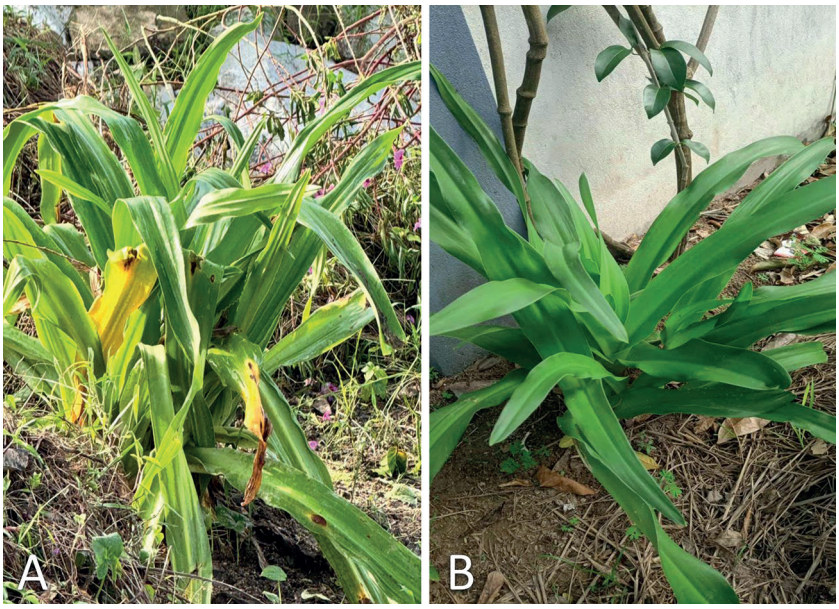

Figure 1 *Crinum latifolium* L. (A, B). Photo by V.C. Tran (June 2024)

focused on individual alkaloids or selected fractions rather than on broad comparative profiling of extracts obtained by different methods.

The detectable composition of plant extracts depends strongly on solvent properties and extraction conditions (de Araújo et al. 2022). Supercritical carbon dioxide (SC-CO_2) extraction offers tunable selectivity through control of pressure, temperature, and co-solvent conditions and facilitates solvent removal (Arumugham et al. 2021, de Araújo et al. 2022). Its application to *C. latifolium* remains limited, making a direct comparison with conventional ethanol maceration useful for evaluating extraction-dependent chemical profiles (Pilařová et al. 2025).

In this study, SC-CO_2 extraction and ethanol maceration were applied to C. latifolium leaves to compare their detectable phytochemical profiles by HPLC-ESI-MS/MS. The study was designed as a qualitative comparison of extraction-dependent chemical profiles.

MATERIAL AND METHODS

Material and processing

Leaves of *Crinum latifolium* were collected from the Hong Linh Mountain area, Ha Tinh Province, North Central Vietnam (18°33'29"N 105°47'20"E), during 15–17 June 2024. All samples were morphologically authenticated in accordance with the current standards of the State Pharmacopoeia of the Russian Federation (Pharmacopoeia of the Eurasian Economic Union 2020).

All reagents used in this study were of analytical grade. HPLC-grade acetonitrile was purchased from Fisher Scientific (Kent, UK), while MS-grade formic acid and ethanol (EtOH) were obtained from Sigma-Aldrich (Steinheim, Germany). Ultrapure water was supplied by Siemens Water Technologies (Munich, Germany).

The leaves of *C. latifolium* were dried at 50 °C for approximately 6 h until they reached a constant weight. A moderate drying temperature was used to remove moisture while limiting prolonged thermal exposure of the plant material.

Dried leaves of *C. latifolium* (11.2 g) were soaked in 50 mL of ethanol for 7 days. After extraction, the mixture was filtered, and the crude filtrate was directly subjected to HPLC-ESI-MS/MS analysis without further purification.

Supercritical carbon dioxide extraction

SC-CO_2 extraction was performed using an SFE-500 system (Thar SCF Waters, Milford, CT, USA). The system included a Thar Waters P-50 high-pressure pump for ethanol delivery as a co-solvent, a Siemens CO_2 flow meter (Munich, Germany) for monitoring the CO_2 supply, and multiple extraction vessels for processing different sample sizes. The liquid CO_2 and ethanol flow rates were set at 20 mL/min and 1.00 mL/min, respectively, and the extraction was conducted at 300 bar and 50°C. For each extraction run, 59 g of dried *C. latifolium* leaves were loaded into the extraction vessel. The extraction was maintained for 60 min after the system reached the target pressure and a stable flow rate.

Liquid chromatography

Chromatographic analysis was performed using a Shimadzu LC-20 Prominence HPLC system (Shimadzu, Kyoto, Japan) equipped with an SPD-20A UV-Vis detector and a Shim-pack Velox C18 reversed-phase column (4.6 × 150 mm, 2.7 µm; Shimadzu, Kyoto, Japan). Deionised water was used as mobile phase A, and acetonitrile containing 0.1 % (v/v) formic acid was used as mobile phase B. The gradient program started at 0 % B for 0–2 min, increased linearly from 0 to 100 % B over 2–50 min, and remained at 100 % B from 50 to 60 min. The eluate was monitored at 230 nm, and the column temperature, flow rate, and injection volume were maintained at 50°C, 0.25 mL/min, and 10 µL, respectively. The same chromatographic conditions were used for HPLC-ESI-ion trap-MS/MS analysis. Five injections of the SC-CO_2 extract and four injections of the ethanol extract were performed. These technical replicate injections were used to assess the reproducibility of compound detection rather than biological variability.

Mass spectrometry

Mass spectrometric analysis was performed using an amaZon SL ion trap mass spectrometer (Bruker Daltonics, Bremen, Germany) equipped with an electrospray ionisation source operated in both positive and negative modes. The ion source temperature was set to 70°C, the drying gas flow rate to 4 L/min, the nebuliser pressure to 7.3 psi, the capillary voltage to 4500 V, the end plate offset to 1500 V, the fragmentor voltage to 280 V, and the collision energy to 60 eV. Full-scan MS and MS/MS spectra were acquired over an m/z range of 100–1700 at scan rates of 1 spectrum/s

for MS and 2 spectra/s for MS/MS. Selected precursor ions were further examined by multistage fragmentation up to MS^4. Instrument control, data acquisition, and processing were performed using Bruker Daltonics software.

Comparative analysis of phytochemical profiles

Phytochemical profiles of the SC-CO_2 and ethanol extracts were compared using manually curated detection/non-detection data for tentatively annotated compounds. The overlap between the two extracts was summarised using a Venn diagram, and the Jaccard similarity index was calculated as $J = a/(a + b + c)$, where a represents the number of compounds detected in both extracts, b the number detected only in the SC-CO_2 extract, and c the number detected only in the ethanol extract. Analytical reproducibility was assessed by calculating the detection frequency of each compound across replicate injections and classified as low (+; detected in ≤25 % of runs), moderate (++; >25 % but <75 %), or high (+++; ≥75 %). These categories reflect only the reproducibility of compound detection across analytical runs and do not represent relative or absolute compound abundance.

Tentative compound annotation

Compounds were tentatively annotated by comparing chromatographic retention, ionisation behaviour, precursor ions, and MS/MS or multistage MS^n fragmentation patterns with previously published data. Because authentic reference standards were not used for structural confirmation, all compound assignments were considered tentative. Only reproducibly detected peaks with interpretable fragmentation spectra were included in the final compound list. This analytical workflow was used for qualitative phytochemical profiling rather than validated quantitative determination.

RESULTS AND DISCUSSION

Phytochemical profiling of *Crinum latifolium* led to the tentative identification of 98 compounds, which were classified into 12 chemical groups. Table 1 summarises all tentatively identified constituents, their chemical classes, and their distribution across the extracts. Among them, flavonoids and flavonoid glycosides represented the largest chemical class, with 40 tentatively annotated compounds, followed by alkaloids (30 compounds), phenolic acids and their derivatives (8 compounds), fatty acids (6 compounds), phytosterols (5 compounds), lignans (2 compounds), and quinones (2 compounds). The remaining constituents were distributed among several minor groups, including phenolic glycosides, phenolic amides, coumarin glycosides, oxidised fatty acids, and sugar acids. These findings indicate that *C. latifolium* possesses a chemically diverse metabolite profile, characterised not only by the presence of Amaryllidaceae-type alkaloids but also by a substantial number of flavonoids and phenolic constituents.

Representative HPLC-ESI-MS chromatograms of the ethanol and SC-CO_2 extracts are shown in Figure 2. Both extracts exhibited multiple chromatographic signals across the analytical run, with differences in the distribution of detectable signals. These chromatograms provide a qualitative overview of the chemical profiles and are not intended for quantitative comparison of compound abundance.

Figure 3 shows representative collision-induced dissociation (CID) spectra of selected metabolites tentatively identified in *C. latifolium* extracts by HPLC-ESI-MS/MS.

The MS^n spectrum of crinamine (Fig. 3A) showed a protonated molecular ion $[M+H]^+$ at m/z 302.29, which produced major fragment ions at m/z 259.21 and 199.14. Further fragmentation of the m/z 199.14 ion generated a product ion at m/z 141.10, followed by an ion at m/z 129.25. This fragmentation pathway agrees with previous reports for crinamine in *C. latifolium* (Tram et al. 2002, Chen et al. 2018, Tian et al. 2021) and supports its tentative annotation in the present study.

Lycorine (Fig. 3B) exhibited a $[M+H]^+$ ion at m/z 287.25, with a product ion at m/z 151.11. Fragmentation of the m/z 151.11 ion subsequently produced an ion at m/z 121.10. The observed fragmentation pattern is consistent with published data for lycorine in *C. latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022, Thongphichai et al. 2022, Trang et al. 2024).

Similarly, ambelline (Fig. 3C) gave a protonated molecular ion at m/z 332.31 and major product ions at m/z 316.30 and 272.22. Successive fragmentation of the m/z 272.22 ion yielded ions at m/z 243.20 and 213.19. These

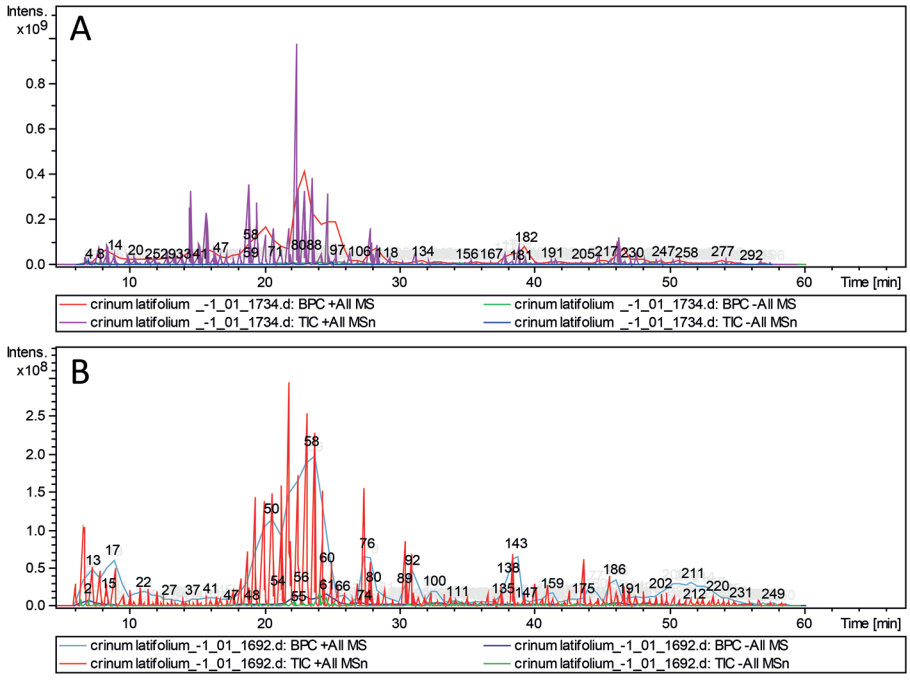

Figure 2 Representative HPLC-ESI-MS chromatograms of *Crinum latifolium* leaf extracts obtained by (A) ethanol maceration and (B) supercritical CO_2 extraction

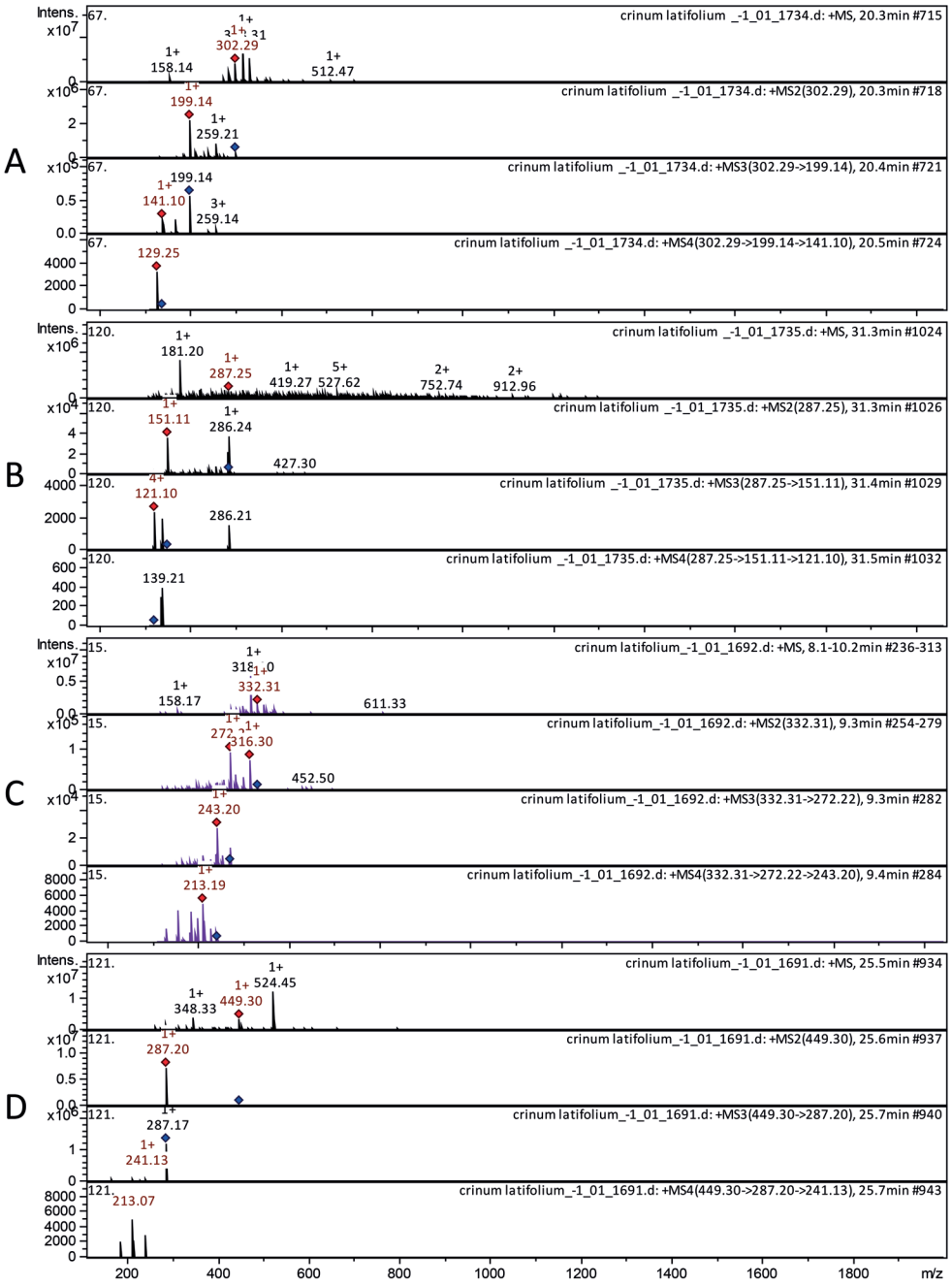

Figure 3 Representative CID spectra of selected compounds tentatively identified in *Crinum latifolium* extracts: A – crinamine, precursor ion at m/z 302.29; B – lycorine, precursor ion at m/z 287.25; C – ambelline, precursor ion at m/z 332.31; and D – astragalin, precursor ion at m/z 449.30

spectral features are consistent with data reported for ambelline in *C. latifolium* (Tram et al. 2002, Hanh et al. 2018, Gasca-Silva et al. 2022) and with reports on other Amaryllidaceae taxa (Feu et al. 2026).

Astragalin (Fig. 3D) showed a $[M+H]^+$ ion at m/z 449.30, which generated a major product ion at m/z 287.20. Further fragmentation produced an ion at m/z 241.13, with the latter yielding an ion at m/z 213.07. This fragmentation behaviour agrees with previous reports of astragalin in *Moringa oleifera* (Ganjayi et al. 2022) and *Trifolium resupinatum* (Marzouk et al. 2024), supporting its tentative annotation.

The ethanol maceration and SC-CO_2 extracts showed distinct but complementary phytochemical profiles. A total of 98 compounds were tentatively annotated, of which 68 occurred in the SC-CO_2 extract and 42 in the ethanol extract. Twelve compounds were common to both extracts, while 56 occurred only in the SC-CO_2 extract and 30 only in the ethanol extract. The Jaccard similarity index was 0.122, indicating low compositional overlap between the two profiles. Although the SC-CO_2 extract contained a larger number of detectable compounds, this finding reflects broader chemical coverage under the conditions used and does not indicate higher compound concentrations or greater extraction yields.

The compositional differences between the two extracts are consistent with the different physicochemical properties of the extraction systems. Ethanol is a polar protic solvent, whereas SC-CO_2 is relatively non-polar. In SC-CO_2 extraction, pressure and temperature affect solvent density and consequently influence extraction yield and selectivity (Arumugham et al. 2021). The addition of ethanol as a co-solvent can further modify the extraction characteristics of the system. Different solvents and extraction approaches have been reported for *Crinum* species (de Araújo et al. 2022), while recent studies have demonstrated the selective extraction of Amaryllidaceae alkaloids using SC-CO_2 with polar modifiers (Pilařová et al. 2025). Under the conditions used in the present study, the two extraction procedures therefore provided complementary chemical coverage rather than directly comparable extraction efficiencies.

Table 1 summarises the detection frequency across replicate HPLC-ESI-MS/MS analyses. Among the 98 compounds tentatively annotated in this study, no previous reports were found for 66 compounds in *C. latifolium* after reviewing the available literature (Tram et al. 2002, Tram & Khanh 2012, Chen et al. 2018, Hanh et al. 2018, Thong-

Table 1. Tentatively identified compounds from the CO_2 and ethanol extracts of *C. latifolium* in positive and negative ionization modes and detection frequency of tentatively identified compounds in ethanol maceration and supercritical CO_2 extracts using HPLC-ESI-ion trap-MS/MS

Tentatively identified compounds	Formula	Mass	Molecular Ion		MS/MS fragment ions			EtOH Maceration	CO_2 Extraction	References
			[M−H]−	[M+H]+	2	3	4			
Column number	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**
Alkaloids										
Crinamine [Crinamin; (+)-Crinamine]	$C_{17}H_{19}NO_4$	301.3371		302	283; 258; 202; 199	254; 199; 156; 141	224; 129	+	+++	*Crinum latifolium* (Tram et al. 2002, Chen et al. 2018, Tian et al. 2021)
Powelline	$C_{17}H_{19}NO_4$	301.3371		302	283; 211	254	224	+	+	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022); Amaryllidaceae (Zhang et al. 2009, Lin et al. 2025)
Filifoline	$C_{24}H_{24}N_2O_6$	436.4572		437	282	252	221	+	n/d	*Crinum latifolium* (Hanh et al. 2018, Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
Epoxy-3,7-dimethoxy-crinane-11-one	$C_{18}H_{19}NO_6$	345.3466		346	316; 275; 173	286; 239; 144		+	+	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022)
1,2beta-Epoxyambelline	$C_{18}H_{21}NO_6$	347.3624		347	319; 275; 246; 173	273; 239; 218; 144	172	+	+++	*Crinum latifolium* (Ghosal et al. 1985, Tram et al. 2002, Gasca-Silva et al. 2022)
Trisphaeridine*	$C_{14}H_9NO_2$	223.2268		224	222	158		n/d	+	*Zephyranthes candida* (Ogbole et al. 2015)
11,12-Dehydroanhydro-lycorine	$C_{16}H_{11}NO_2$	249.2640		250	248; 126	230; 204; 172		n/d	+++	*Crinum erubescens* (Guerrieri et al. 2016); Amaryllidaceae (Soto-Vásquez et al. 2022)
Hippeastrine	$C_{17}H_{17}NO_5$	315.3206		316	297; 296; 279; 230; 224	211; 194	166	n/d	++	*Crinum latifolium* (Gasca-Silva et al. 2022)
Ambelline	$C_{18}H_{21}NO_5$	331.3630		332	316; 272	275; 243	213	n/d	+++	*Crinum latifolium* (Tram et al. 2002, Hanh et al. 2018, Gasca-Silva et al. 2022); Amaryllidaceae (Feu et al. 2026)
Aulicine	$C_{18}H_{25}NO_4$	319.3954		319	218; 273	172	142; 124	n/d	+++	*Hippeastrum aulicum* (de Andrade et al. 2014); Amaryllidaceae (Lin et al. 2025)
8-O-Demethylmaritidine	$C_{16}H_{19}NO_3$	273.3270		274	256; 244; 182; 174	238; 174	175	n/d	+	*Hippeastrum aulicum* (de Andrade et al. 2014); Amaryllidaceae (Lin et al. 2025)
Sternbergine	$C_{18}H_{21}NO_5$	331.3630		332	300; 272; 178	256; 241	199; 184	n/d	+	*Pyrolirion albicans* (Huaylla et al. 2021); *Crinum latifolium* (Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
6α-Hydroxyundulatine	$C_{18}H_{21}NO_6$	347.3624		347	319; 316; 246	286; 273; 218	257; 172	n/d	+++	*Crinum latifolium* (Tram et al. 2002, Hanh et al. 2018, Gasca-Silva et al. 2022)
Lycorine [Amarylline; Galanthidine; Narcissine]	$C_{16}H_{17}NO_4$	287.3105		287	286; 242; 151	121; 119		n/d	+	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022, Thongphichai et al. 2022, Trang et al. 2024)
11-Hydroxyvittatine	$C_{16}H_{17}NO_4$	287.3105		288	242; 188	119		n/d	+	*Hippeastrum aulicum* (de Andrade et al. 2014); Amaryllidaceae (Lin et al. 2025)
Flexinine	$C_{16}H_{17}NO_4$	287.3105		288	242; 188	119		n/d	+	*Crinum latifolium* (Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
Tazettine	$C_{18}H_{21}NO_5$	331.3630		331	303; 184	256; 202	126	n/d	+	*Crinum latifolium* (Gasca-Silva et al. 2022); Amaryllidaceae (Zhang et al. 2009, Lin et al. 2025)
Delagoensine	$C_{18}H_{21}NO_5$	331.3630		332	300; 272; 184	256; 241	232; 184	n/d	+	*Crinum delagoense* (Nair et al. 1998); *Crinum latifolium* (Gasca-Silva et al. 2022)
6α-Hydroxybuphanidrine	$C_{18}H_{21}NO_5$	331.3630		332	300; 272; 184	256; 241	232; 184	n/d	+	*Crinum latifolium* (Hanh et al. 2018, Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
Undulatine [Undulatin]	$C_{18}H_{21}NO_5$	331.3630		332	300; 272; 184	256; 241	232; 184	n/d	+	*Crinum latifolium* (Tram et al. 2002, Hanh et al. 2018, Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
6-Hydroxypowelline	$C_{17}H_{19}NO_5$	317.3795		318	300; 176	282; 253	252; 180	n/d	++	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022)

Table 1. Continued.

Column number	1	2	3	4	5	6	7	8	9	10
Crinamidine	$C_{17}H_{19}NO_5$	317.3795		318	300; 299; 218	282; 253; 172	252; 124	n/d	+++	*Crinum latifolium* (Tram et al. 2002, Hanh et al. 2018, Gasca-Silva et al. 2022); Amaryllidaceae (Zhang et al. 2009, Lin et al. 2025)
1-Epideacetylbowdensine	$C_{17}H_{21}NO_5$	319.3523		319	218	172		n/d	+	*Crinum latifolium* (Trang et al. 2024); Amaryllidaceae (Lin et al. 2025)
Sanguinine [O-Desmethyl Galanthamine]	$C_{16}H_{19}NO_3$	273.3270		274	256; 182	238		n/d	+	Amaryllidaceae (Lin et al. 2025, Feu et al. 2026)
6-Hydroxycrinamine	$C_{17}H_{19}NO_5$	317.3795		318	300; 176	298		n/d	+	Amaryllidaceae (Lin et al. 2025)
Haemanthamine	$C_{17}H_{19}NO_4$	301.3371		302	283; 211	254	224	n/d	+	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022); Amaryllidaceae (Zhang et al. 2009, Lin et al. 2025)
Augustine	$C_{17}H_{19}NO_4$	301.3371		302	283; 211	254	224	n/d	+	Amaryllidaceae (Lin et al. 2025)
Pratorimine	$C_{16}H_{11}NO_3$	265.2634		266	248	220; 118		n/d	+	*Crinum latifolium* (Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
Ismine	$C_{15}H_{15}NO_3$	257.2845		257	238; 196	169		n/d	+	*Crinum erubescens* (Guerrieri et al. 2016); Amaryllidaceae (Lin et al. 2025)
6α-Hydroxycrinamidine	$C_{17}H_{19}NO_6$	333.3359		333	316; 133	289; 120	180	n/d	+	*Crinum latifolium* (Tram et al. 2002, Gasca-Silva et al. 2022); Amaryllidaceae (Lin et al. 2025)
Flavonoids and flavonoid glycosides										
Kaempferol-C-hexoside*	$C_{21}H_{20}O_{11}$	448.3769	447		401	269; 161	161	++	n/d	Tomato-based products (Vallverdú-Queralt et al. 2011)
Kaempferol-3-O-α-L-rhamnoside*	$C_{21}H_{20}O_{10}$	432.3775	431		385; 223	223	151	+	n/d	*Copaifera oblongifolia* (Ramos et al. 2025)
Quercetin-3-O-hexoside*	$C_{21}H_{20}O_{12}$	464.3763		465	447; 303	303; 257; 229	229; 201	++	n/d	*Corema album* (León-González et al. 2013); *Tibouchina urvilleana* (Solarte et al. 2022)
Delphinidin 3-O-hexoside*	$C_{21}H_{21}O_{12}^+$	465.3905		465	303	303; 257; 229	229; 201	++	n/d	*Corema album* (León-González et al. 2013)
Dihydrokaempferol-O-hexoside*	$C_{21}H_{22}O_{11}$	450.3928	449		431; 269	208	165	+	n/d	*Vigna radiata* (Yang et al. 2020)
Eriodictyol-7-O-glucoside* [Pyracantho-side; Miscanthoside]	$C_{21}H_{22}O_{11}$	450.3928	449		431; 269	208	165	+	n/d	*Viscum articulatum* (Li et al. 2008)
Quercitrin* [Quercetin 3-O-rhamnoside; Quercetrin]	$C_{21}H_{20}O_{11}$	448.3769		449	287	287; 241	213	+	n/d	*Copaifera oblongifolia* (Ramos et al. 2025)
Astragalin* [Astragaline]	$C_{21}H_{20}O_{11}$	448.3769		449	287	287; 241	213	+	n/d	*Moringa oleifera* (Ganjayi et al. 2022); *Trifolium resupinatum* (Marzouk et al. 2024)
Kaempferol-3-O-galactoside* [Trifolin]	$C_{21}H_{20}O_{11}$	448.3769		449	287	287; 241	213	+	n/d	*Amygdalus persica* (Zhang et al. 2023)
Cyanidin-3,5-O-dihexoside*	$C_{27}H_{31}O_{16}$	611.5254		611	449; 287; 181	287; 214		+	n/d	*Aronia melanocarpa* (Lin et al. 2022)
Kaempferol-3,7-O-di-hexoside*	$C_{27}H_{30}O_{16}$	610.5175		611	449; 287; 181	287; 214		+	n/d	*Allium macrostemon* (Nakane & Iwashina 2015)
Cyanidin-3-O-hexoside*	$C_{21}H_{21}O_{11}^+$	449.3849		449	431; 287	287; 257; 213	157	+	n/d	*Aronia melanocarpa* (Lin et al. 2022)
Luteolin-7-O-glucoside* [Cynaroside; Luteoloside]	$C_{21}H_{20}O_{11}$	448.3769	447		285	255	255	+	n/d	*Dendranthema morifolium* (Lin et al. 2015)
Apigenin-O-rhamnoside*	$C_{22}H_{22}O_{11}$	462.4035	461		415; 311; 301	293; 149	131	+	n/d	*Echium humile* (Aouadi et al. 2021)

Table 1. Continued.

Column number	1	2	3	4	5	6	7	8	9	10
Quercetin-O-dihexoside*	$C_{27}H_{30}O_{17}$	626.5169	625		578; 463; 301	418; 301		+	n/d	*Pseudoconyza viscosa* (Zahid et al. 2025)
Eriodictyol* [3',4',5,7-tetrahydroxy-flavanone]	$C_{15}H_{12}O_6$	288.2522		289	271; 188	211; 142		+	+	*Corynebacterium glutamicum* (Wu et al. 2022); *Lycium barbarum* (Yang et al. 2022)
3',4',7,8-Tetrahydroxy-flavanone*	$C_{15}H_{12}O_6$	288.2522	287		269; 127	269; 169		+	n/d	*Acacia confusa* (Chang & Chang 2018)
(epi)-Catechin*	$C_{15}H_{14}O_6$	290.2681		291	273; 261; 172; 156	265; 255; 172	222; 142	+	+++	*Schotia latifolia* (Masika et al. 2004); *Litchi chinensis* (Izu et al. 2024)
Pelargonidin-3-O-glucoside* [Callistephin]	$C_{21}H_{21}O_{10}$	433.3854	431		385; 223	223	151	+	n/d	*Aristotelia chilensis* (Pineda et al. 2022)
Isorhamnetin 3-O-hexoside*	$C_{22}H_{22}O_{12}$	478.4029		479	461; 317	302; 139	274; 153	+	n/d	*Anthyllis cytisoides* (Bouammali et al. 2026)
Stellarin-2*	$C_{28}H_{32}O_{16}$	624.5441		625	607; 502; 295; 177	572; 312; 210	463	+	n/d	*Citrus sinensis* (Barreca et al. 2014)
Lucenin-2,4'-methyl ether* [Diosmetin-6,8-Di-C-hexoside]	$C_{28}H_{32}O_{16}$	624.5441		625	607; 502; 295; 177	572; 312; 210	463	+	n/d	*Citrus sinensis* (Pepe et al. 2017)
di-C,C-hexosyl-chryso-eriol*	$C_{28}H_{32}O_{16}$	624.5441		625	607; 502; 295; 177	572; 312; 210	463	+	n/d	*Citrullus* spp. fruits (Yuan et al. 2021)
Luteolin* [Tetrahydroxy-flavone]	$C_{15}H_{10}O_6$	286.2363		287	287; 240	213	157	+	n/d	Propolis (Belmehdi et al. 2021)
Apigenin* [5,7-Dixydroxy-2-(4 Hydroxyphenyl)-4H-Chromen-4-One]	$C_{15}H_{10}O_5$	270.2369		271	225	224		+	n/d	Propolis (Belmehdi et al. 2021)
Quercetin	$C_{15}H_{10}O_7$	302.2357		303	275; 202	156	126	n/d	+++	*Crinum latifolium* (Viet et al. 2011); *Cucumis sativus* (Anjani et al. 2023)
Herbacetin*	$C_{15}H_{10}O_7$	302.2357		303	275; 256; 202	156		n/d	+++	*Rhodiola rosea* (Olennikov & Chirikova 2022)
Catechin*	$C_{15}H_{14}O_6$	290.2681		291	261; 156	242; 172	142	n/d	+++	*Schotia latifolia* (Masika et al. 2004); *Arbutus unedo* (Mrabti et al. 2018)
(epi)-Afzelechin*	$C_{15}H_{14}O_5$	274.2687		275	256; 244; 174	174; 156		n/d	+++	*Astilbe rivularis* (Hori et al. 2018); Fermented tea leaf (Wangkarn et al. 2021)
Epiafzelechin derivative*	$C_{18}H_{16}O_{10}$	392.3136		393	274	244	156	n/d	++	*Mascagnia pubiflora* (Garcez et al. 2009)
Dihydroxy-dimethoxy-isoflavan*	$C_{16}H_{16}O_5$	288.2952		289	271; 188	211; 142		n/d	+	*Astragalus membranaceus* (Li et al. 2014)
6α-Hydroxymaackiain*	$C_{16}H_{12}O_6$	300.2629		301	255	199		n/d	+	*Ventilago denticulata* (Azizah et al. 2020)
Jaceosidin* [5,7,4'-trihydroxy-6',3'-dimethoxyflavone]	$C_{17}H_{14}O_7$	330.2889		331	303; 184	275; 256; 202	184; 126	n/d	+++	Jaceosidin (Woo & Kwon 2016; Nageen et al. 2021)
Cirsiliol*	$C_{17}H_{14}O_7$	330.2889		331	315; 303; 196	275	244	n/d	++	*Juglans mandshurica* (Huo et al. 2018); *Inula viscosa* (Kheyar-Kraouche et al. 2018)
Dihydroquercetin* [Taxifolin; Taxifoliol]	$C_{15}H_{12}O_7$	304.2516		305	287	259; 162	232	n/d	+	*Abies nephrolepis* (Wei et al. 2020)
Afzelechin*	$C_{15}H_{14}O_5$	274.2687		275	244	214; 156	156	n/d	+	*Astilbe rivularis* (Hori et al. 2018)
Tricin* [5,7,4'-trihydroxy-3',5'-dimetoxyflavone]	$C_{17}H_{14}O_7$	330.2889		331	315	238		n/d	+	Tricin (Zheng et al. 2021)

Table 1. Continued.

Column number	1	2	3	4	5	6	7	8	9	10
Phloretin* [Dihydro-naringenin; Phloretol]	$C_{15}H_{14}O_5$	274.2687		275	258; 256; 182	238; 210; 196	164	+	+	Phloretin (Tuli et al. 2022)
3,5-Dihydroxy-6,7,3',4'-tetramethoxyflavone*	$C_{19}H_{18}O_8$	374.3414		375	345	319; 258	228	n/d	+	*Artemisia annua* (Han et al. 2008)
Dihydroxy-tetramethoxy-flavone*	$C_{19}H_{18}O_8$	374.3414		375	345	319; 258	228	n/d	+	Propolis (Belmehdi et al. 2021)
Phenolic acids and derivatives										
p-Coumaric acid-O-hexoside*	$C_{15}H_{18}O_8$	326.2986		327	309	281; 190		+	n/d	*Polypodium hastatum* (Yao et al. 2016)
Caffeoyl shikimic acid*	$C_{16}H_{16}O_8$	336.2934		337	318; 180	138		+	n/d	*Vitis vinifera* (Šuković et al. 2020)
Caffeoyl-hexose-deoxyhexoside*	$C_{24}H_{24}O_{11}$	488.4408	487		341; 281; 161	251; 179		+	n/d	*Alhagi maurorum* (El-Zahar et al. 2022)
Gallic acid hexoside* [Monogalloyl hexoside]	$C_{13}H_{16}O_{10}$	332.2601		333	315	297; 188		n/d	+	*Canavalia gladiata* (Gan et al. 2018)
Caffeic acid*	$C_9H_8O_4$	180.1574		180	160; 134	144; 134; 118		n/d	++	*Adina rubella* (Yin et al. 2016)
Caftaric acid* [Cis-Caftaric acid; 2-Caffeoyl-L-Tartaric acid]	$C_{13}H_{12}O_9$	312.2290	311		293	275	232	n/d	+	*Echinacea purpurea* (Lekar' et al. 2013)
Salvianolic acid G*	$C_{18}H_{12}O_7$	340.2837		341	323; 273; 208; 158	305; 174		n/d	+	*Salvia miltiorrhiza* (Xia et al. 2023)
Rosmarinic acid*	$C_{18}H_{16}O_8$	360.3148		361	343; 180	134		n/d	+	*Rosmarinus officinalis* (Sharma et al. 2020)
Phenolic glycosides										
Arbutin*	$C_{12}H_{16}O_7$	272.2512		273	163	131		+	n/d	*Arbutus pavarii* (Buzgaia et al. 2021)
Phenolic amides										
N-feruloyloctopamine*	$C_{18}H_{19}NO_5$	329.3472		330	312; 123	294	265; 123	+	n/d	*Salsola* L. (Selim et al. 2023)
Lignans										
Syringaresinol*	$C_{22}H_{26}O_8$	418.4436		419	401; 326; 273	382; 381; 298	253	++	+	*Gymnema sylvestre* (Pham et al. 2022)
Arctigenin*	$C_{21}H_{24}O_6$	372.4117		373	337	319; 209	180	n/d	+	*Lavatera cashmeriana* (Sofi et al. 2022)
Coumarin glycosides										
Xanthotoxol glucoside*	$C_{17}H_{16}O_9$	364.3035		365	259	228; 142	126	n/d	+	*Clausena lansium* (Xu et al. 2014)
Oxidized fatty acids										
13-Trihydroxy-Octadecenoic acid* [THODE]	$C_{18}H_{34}O_5$	330.4596	329		311; 229	211		n/d	+	*Meloidogyne javanica* (Fitoussi et al. 2021)
Fatty acids										
Linoleic acid [Linolic acid; Telfairic acid]	$C_{18}H_{32}O_2$	280.4455	279		261	243; 217		+	n/d	*Crinum latifolium* (Shukla et al. 2018, Gasca-Silva et al. 2022)
Stearidonic acid*	$C_{18}H_{28}O_2$	276.4137		277	257; 231	213	211	+	+++	*Buglossoides* spp. seeds (Chelh et al. 2022); *Maackia amurensis* (Razgonova et al. 2023)
Linolenic acid* [Alpha-Linolenic acid; Linolenate]	$C_{18}H_{30}O_2$	278.4296		279	262; 204; 149	121		+	++	*Linum usitatissimum* (Kumar et al. 2023)

Table 1. Continued.

Column number	1	2	3	4	5	6	7	8	9	10
Eicosatetraenedioic acid*	$C_{20}H_{30}O_4$	334.4498		335	289; 172	261; 175	204	n/d	+	Eicosanoids (Tsikas & Zoerner 2014)
Myristoleic acid*	$C_{14}H_{26}O_2$	226.3550		227	209; 206	193; 189; 139		+	+	*Maackia amurensis* (Razgonova et al. 2023)
Arachidic acid* [Eicosanoic acid]	$C_{20}H_{40}O_2$	312.5304		313	277	247		n/d	+	*Cyperus laevigatus* (Ayoub et al. 2022)
Phytosterols										
Stigmastanol* [Sitostanol; Fucostanol; Spinastanol]	$C_{29}H_{52}O$	416.7226		417	399; 398	380; 363		+	+	Phytosterols (Oliva et al. 2024)
Beta-Sitosterin [Beta-Sitosterol]	$C_{29}H_{50}O$	414.7067		415	397	379		n/d	+	*Crinum latifolium* (Tram & Khanh 2012, Nhung & Hue 2020)
Gamma-Sitosterol* [Clionasterol]	$C_{29}H_{50}O$	414.7067		415	397; 297	379		n/d	+	*Synedrella nodiflora* (Yakubu et al. 2014)
Avenasterol* [Delta7-Avenasterol; 7-Dehydroavenasterol]	$C_{29}H_{48}O$	412.6908		413	377	323; 220	319; 172	n/d	+	Vegetable oils (Schlag et al. 2022)
Gramisterol*	$C_{29}H_{48}O$	412.6908		413	377	323; 220	319; 172	n/d	+	Vegetable oils (Schlag et al. 2022)
Quinones										
8,8'-Dihydroxy-2,2'-binaphthalene-1,1';4,4'-tetrone*	$C_{20}H_{10}O_6$	346.2898		347	319; 255	217	172	n/d	+	*Juglans mandshurica* (Huo et al. 2018)
6-Methyl-rhein*	$C_{16}H_{10}O_6$	298.2470		299	281	263; 121		n/d	+	*Rheum emodi* (Singh et al. 2005)
Sugar acids										
Gluconic acid* [Gluconate]	$C_6H_{12}O_7$	196.1553		197	178	160; 158	132	n/d	++	*Colchicum micranthum* (Gulsoy-Toplan et al. 2018); Perilla leaves (Chen et al. 2022)

Note: "*", putative first records for *Crinum latifolium* based on the literature examined.

phichai et al. 2022, Gasca-Silva et al. 2022, Chaichompoo et al. 2024, Trang et al. 2024). These compounds were therefore considered putative first records for *C. latifolium* based on the literature examined. Additional phytochemical and analytical studies on *Crinum* species and other Amaryllidaceae taxa were also consulted when evaluating the compound annotations (Ghosal et al. 1985, Nair et al. 1998, Zhang et al. 2009, de Andrade et al. 2014, Guerrieri et al. 2016, Huaylla et al. 2021, Soto-Vásquez et al. 2022, Lin et al. 2025, Feu et al. 2026).

Alkaloids remain of particular interest because they are characteristic metabolites of *Crinum*. However, the detection of flavonoids, phenolic acids, lignans, fatty acids, phytosterols, quinones, and sugar acids broadens the known phytochemical profile of *C. latifolium* beyond its established alkaloid composition. Overall, these findings expand the phytochemical information available for this species and provide a basis for further targeted chemical investigation.

CONCLUSION

Our HPLC-ESI-MS/MS analysis tentatively annotated 98 compounds in *Crinum latifolium* leaf extracts obtained by SC-CO_2 extraction and ethanol maceration. The two extraction procedures produced distinct but complementary detectable phytochemical profiles, with 68 compounds detected in the SC-CO_2 extract, 42 in the ethanol extract, and 12 shared between them. Based on the literature examined, 66 compounds represent putative first records for *C. latifolium*. These findings expand the known phytochemical profile of the species and show that extraction conditions influence detectable chemical coverage. All compound annotations remain tentative and require confirmation using authentic reference standards or complementary structural methods.

ACKNOWLEDGEMENTS

This work was supported by Program "Prioritet-2030" of the Tomsk State University.

AUTHOR CONTRIBUTIONS

Conceptualization – V.C.T.; methodology – V.C.T. and M.P.R.; formal analysis – V.C.T. and M.P.R.; investigation – V.C.T.; data curation – V.C.T.; writing original draft – V.C.T.; writing review and editing – M.P.R.; visualization – V.C.T.; supervision – K.S.G. All authors have read and agreed to the published version of the manuscript.

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