

Article

Phytochemical Analysis, Mineral Composition and GC-MS Profiling of Three *Iris* Species from the Southeastern Part of Kazakhstan

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Abstract

This study examined the phytochemical profiles and bioactive potential of *Iris alberti*, *I. pallasii*, and *I. sogdiana* collected from southeastern Kazakhstan. GC-MS analysis identified fatty acids, sterols, long-chain alcohols, and hydrocarbons in each species. The key phytochemicals β -sitosterol, stigmasterol, and campesterol were present in all three. *I. alberti* showed the highest sterol content of γ -sitosterol (28.49%) and campesterol (6.51%). *I. pallasii* contained significant amounts of hexacosanol (19.74%) and γ -sitosterol (13.24%), while *I. sogdiana* was notable for octacosanol (22.87%) and γ -sitosterol (12.18%). Fatty acid composition varied: *I. sogdiana* was rich in α -linolenic acid (4.77%) and palmitic acid (2.77%), *I. pallasii* had 3.96% and (3.48%). *I. alberti* contained dodecanoic acid (3.83%) and branched-chain fatty acids. Tannin content was highest in *I. alberti* (1.88%), while alkaloid levels were moderate across species (0.63–0.77%). Mineral analysis showed *I. pallasii* had the highest Ca (586.62 mg/100 g), Mg and K, whereas *I. sogdiana* had the highest potassium (538.36 mg/100 g). HPLC-DAD analysis revealed distinct phenolic profiles. Water–alcohol extracts from rhizomes of *I. alberti* had an IC_{50} of 16.90 ± 6.04 μ g/mL in DPPH and were inactive in ABTS. *I. pallasii* exhibited IC_{50} values of 28.77 ± 2.93 μ g/mL (DPPH) and 10.93 ± 7.29 μ g/mL (ABTS), while *I. sogdiana* showed negligible activity in both assays. Ethyl acetate extracts displayed higher IC_{50} values, consistent with lower phenolic content.

Keywords: *Iris* species; phytochemical profile; GC-MS analysis; phenolic profile; antioxidant activity



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1. Introduction

The genus *Iris* Tourn. ex L. (*Iridaceae* Juss.) encompasses over 300 species, many of which remain poorly explored chemically, particularly in Central Asia [1–4]. These species are cosmopolitan and have significant horticultural, medicinal (homoeopathy), cosmetic

(perfumery), food, and ecological importance, and served as templates for hydrophobic surfaces [5–11]. Floristic studies indicate that Kazakhstan has 19–22 *Iris* species distributed across all natural zones and ecosystems [2–4,12–15]. Three rare species—*Iris alberti* Regel, *I. ludwigii* Maxim., and *I. tigridia* Bunge are listed in the Red Book of Kazakhstan (2014) [16], and only eight have been used in folk and experimental medicine [17].

Among Kazakhstan's *Iris* flora, *Iris alberti* Regel, *I. pallasii* Fisch. ex Trevir., and *I. sogdiana* Bunge occur in southeastern Kazakhstan and represent ecologically distinct taxa adapted to different environmental conditions [18]. Long-term phytocenotic introduction of wild-growing iris species in this region began in 2018 and is ongoing [19–21], although comprehensive comparative chemical analysis of these species is limited. According to the herbarium collections of the Institute of Botany and Phytointroduction (IBPh), *I. sogdiana* is found across various floristic regions, whereas *I. pallasii* and *I. alberti* exhibit a more restricted yet ecologically distinct distribution.

Taxonomic revisions within the genus *Iris* complicate comparisons of previously reported chemical data. *Iris alberti* is consistently assigned to the subgenus and section *Iris* [22–26]. Central Asian *Iris*, based on both morphological and molecular evidence, places *I. sogdiana* and *I. pallasii* in the subgenus *Limniris* [26]. Consequently, chemical and pharmacological data are dispersed across different species names, limiting direct comparability.

Phytochemical studies of the genus *Iris* indicate that its representatives are rich sources of structurally diverse secondary metabolites, including isoflavonoids, xanthones, stilbenoids, quinones, phenolic acids, and triterpenoids, many of which exhibit pronounced biological activity [27,28]. However, research has predominantly focused on species such as *I. lactea*, *I. ensata*, *I. pseudacorus*, and *I. tenuifolia*, while data on *I. alberti*, *I. pallasii*, and *I. sogdiana* remain fragmentary or indirect [29–34].

Despite limited data, *Iris alberti* has a phytochemical profile primarily characterised by vitamin C and phenolic acids (sinapic and ferulic) in the leaves, and its roots and leaves are rich in tannins (2.99 and 3.52% [29]) and xanthones (mangiferin and isomangiferin) [28]. Traditionally, the roots are used for digestive issues, and the flowers serve as a source of natural green dye [17,29]. In contrast, *I. pallasii* (*I. lactea* s.l. or *I. pallasii* var. *chinensis*) exhibits a diverse secondary metabolite profile that includes benzoquinones, C-glycosylflavones, xanthones, stilbenoids, phenolic acids, and a significant proportion of unsaturated fatty acids in its seeds [30] (Appendix A, Table A1). This chemical diversity suggests adaptability for harsh environments. The presence of anticancer benzoquinones, such as irisquinone and pallasone, within the *Iridaceae* family likely functions as potent defence agents against biotic stressors. The pronounced antiviral activity of oligostilbenes, including vitisin B and C, further emphasises the pharmacological significance of this species [27–29]. *Iris sogdiana* accumulates carbohydrates and polysaccharides in roots (0.62–0.63%) and leaves (1.8–2.4%), with seeds containing glucomannans [30]. *Iris sogdiana* is considered a synonym of *I. spuria* [25], which is characterised by glycosylated iridals, and iso-iridal type of triterpenes, and isoflavonoids (peltogynoids and rotenoid). Likewise, *I. pallasii* is frequently treated as *I. lactea* Pall. or *I. pallasii* var. *chinensis*; thus, all phytochemical and pharmacological findings published under these names are integrated to provide a consistent comparative framework [27,32–34].

Considering the ecological heterogeneity of southeastern Kazakhstan and the sensitivity of plant secondary metabolism to environmental factors, a targeted comparative phytochemical study of these species is justified. In this context, the present comparison of *I. alberti*, *I. pallasii* and *I. sogdiana* provides important insights into their phytochemical profiles.

Therefore, the objective of this study was to undertake a comparative phytochemical and biological assessment of *Iris alberti*, *I. pallasii*, and *I. sogdiana* collected from southeastern

Kazakhstan, thereby contributing novel chemical data for underexplored taxa. The species nomenclature is provided in accordance with the floristic summary [2–4].

2. Results

2.1. Collection Sites

The studied plants were collected during expedition trips in April–May 2023 in different phases of vegetation in the valleys of the desert rivers Ili (*Iris pallasii*, *I. sogdiana*) and Karatal (*I. sogdiana*) and in the downhill of the gorges Kasquelen, Shamalgan ridge of Zayliskiy Alatau (*I. alberti*) (Figure 1). All collected species specimens were submitted to the herbarium of AA belonging to the Institute of Botany and Phytointroduction, under numbers AA0042423, AA0042424, AA0042425. The species was identified using key signs [2–4,12–14] by Ramzanova M.S. and Prof. Gemejiyeva N.G.

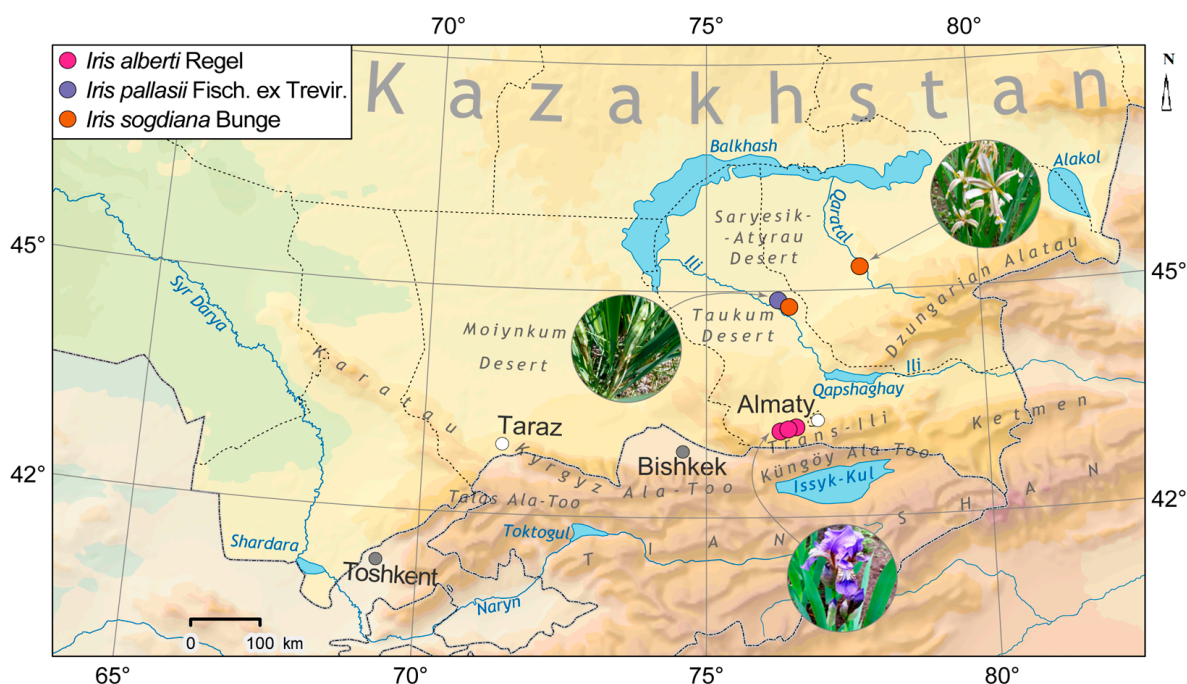


Figure 1. Locations of Field Collection for the Investigated *Iris* species: in the valleys of the desert rivers Ili (*Iris pallasii*, *I. sogdiana*) and Karatal (*I. sogdiana*) and in the downhill of the gorges Kasquelen, Shamalgan ridge of Zayliskiy Alatau (*I. alberti*). (Map prepared by Rybakov I.A.).

2.2. Qualitative and Quantitative Analysis

Phytochemical screening provides a preliminary basis for identifying bioactive natural compounds and assessing their potential. The leaves and rhizomes of the three *Iris* species examined were screened for flavonoids, alkaloids, terpenoids, and tannins, and all these bioactive classes were confirmed. However, qualitative analysis was performed on rhizomes, as these parts are most significant for therapeutic value and are widely used in folk medicine. Additionally, moisture and ash content were calculated (Table 1).

Quantitative phytochemical analysis of three *Iris* samples revealed significant variation in the measured parameters. Flavonoid content was relatively uniform across samples, ranging from 0.29% to 0.33%. Tannin level illustrates considerable variability, with the highest concentration in *I. alberti* (1.88%) and the lowest in *I. sogdiana* (1.06%). Alkaloid content remained moderate and comparable across the three samples (0.63–0.77%).

Table 1. Qualitative and quantitative analysis of *Iris* species from the south-eastern part of Kazakhstan.

Class of Compounds	Content (%)		
	<i>Iris alberti</i>	<i>Iris pallasii</i>	<i>Iris sogdiana</i>
Flavonoids	0.33 ± 0.09	0.29 ± 0.06	0.31 ± 0.06
Tannins	1.88 ± 0.5	1.61 ± 0.2	1.06 ± 0.4
Alkaloids	0.63 ± 0.04	0.77 ± 0.05	0.73 ± 0.08
Moisture content	5.1 ± 1.3	6.9 ± 0.9	5.9 ± 1.1
Ash content	9.7 ± 3.2	11.6 ± 2.8	10.9 ± 1.8

2.2.1. Extract Yield (%)

The extract yield was isolated from creeping rhizomes of three *Iris* species by solvent extraction. Water–ethanol mixture (50–70%) was applied as a solvent in the extraction process. In all cases, 50% ethanol solution yielded higher extractive values than a 70% ethanol solution, indicating better solubility of the major constituents in the polar solvent system. For *I. pallasii* the extractive value decreased from 18.9% with 50% water–ethanol to 15.5% (70% w–ethanol). A similar trend was observed for *I. alberti* (36.7% to 30.3%) and *I. sogdiana* (31.2% to 29.1%). Among the studied species, the abundance of ethanol-soluble constituents follows the order *I. alberti* > *I. sogdiana* > *I. pallasii*.

2.2.2. Mineral Composition (mg/100 g)

Quantitative analysis of minerals in rhizomes of the three *Iris* species revealed inter-specific differences (Table 2). All samples contained higher levels of macronutrients (Ca, Mg, Na, K) compared to trace elements (Fe, Mn), indicating a consistent trend of decreasing content from major to minor elements across species. However, notable species-specific differences were observed.

Table 2. Elemental composition of rhizomes (roots) of selected *Iris* species from the south-eastern part of Kazakhstan.

Elements	Content (mg/100 g)		
	<i>Iris alberti</i>	<i>Iris pallasii</i>	<i>Iris sogdiana</i>
Calcium (Ca)	295.64	586.62	476.26
Magnesium (Mg)	33.84	167.99	76.55
Potassium (K)	200.76	385.63	538.36
Sodium (Na)	46.85	267.97	155.75
Iron (Fe)	22.11	19.57	17.55
Manganese (Mn)	1.09	1.16	1.157

Across species, a tendency toward accumulation of Ca, K, and Na was observed. *Iris pallasii* accumulates the highest levels of Ca (586.62 mg/100 g), Mg (167.99 mg/100 g), and Na (267.97 mg/100 g), considerably surpassing both *I. alberti* and *I. sogdiana*. In contrast, potassium content is highest in *I. sogdiana* (538.36 mg/100 g), followed by *I. pallasii* (385.63 mg/100 g) and *I. alberti* (200.76 mg/100 g). Among trace elements, iron concentration is highest in *I. alberti* at 22.11 mg/100 g. Manganese levels are relatively low and consistent across all species, with *I. pallasii* showing a slight increase at 1.16 mg/100 g.

2.3. GC-MS Analysis of the Native Nonpolar Extracts from the Leaves

GC–MS analysis of native CH₂Cl₂ leaf extracts from *Iris alberti*, *I. pallasii*, and *I. sogdiana* showed complex yet similar metabolite profiles dominated by lipophilic compounds. Compounds were identified using retention time, linear retention indices (LRIs), and mass

spectral matching. Relative abundances were reported as percentage peak areas. Figure 2 compares compounds identified in native and TMS-derivatised extracts. Derivatisation significantly influenced the number of detected metabolites in a species-specific manner, reflecting differences in derivatisable functional groups. In *I. alberti*, TMS derivatisation increased detectable compounds from 54 to 77, indicating a higher proportion of hydroxylated or carboxyl-containing metabolites. In contrast, *I. pallasii* showed a decrease (74 to 48), while *I. sogdiana* had a slight increase (70 to 74), highlighting species-specific differences in metabolite composition.

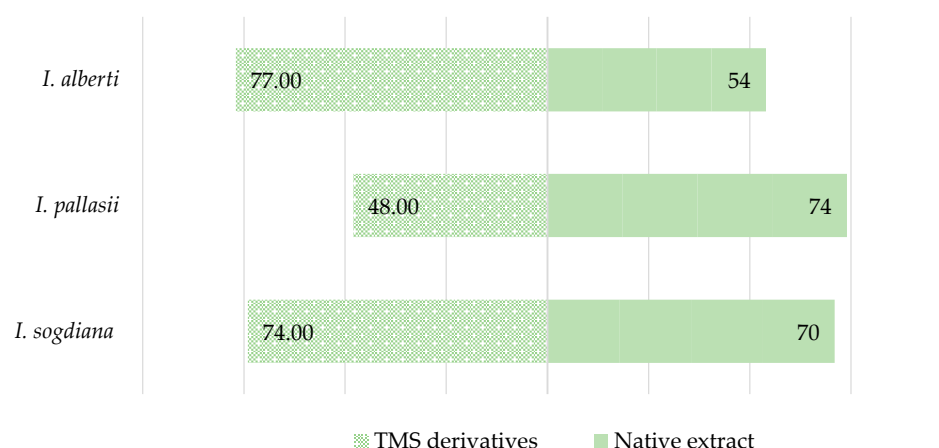


Figure 2. Comparison of the number of compounds identified by GC–MS in CH₂Cl₂ extracts and TMS-derivatised leaf extracts of the investigated *Iris* species.

Sterols, triterpenoids, fatty acids, and fatty acid esters were the dominant chemical classes in all three species. Quantities of identified compounds varied; many compounds had consistent retention times across species, demonstrating analytical reliability and a conserved core lipophilic metabolome in *Iris*. For instance, dodecanoic acid and squalene were present in all samples with nearly identical retention times, confirming accurate compound identification.

Identified compounds in CH₂Cl₂ leaf extracts from three *Iris* species were classified according to the chemical classes (Figure 3). Fatty acid esters and fatty acids were the most abundant chemical classes in all three species. *Iris sogdiana* had the highest number of fatty acid esters, while *I. pallasii* showed a similar but slightly lower diversity in this class. Sterols and terpenes/terpenoids were especially prominent in *I. pallasii*, which contained the highest number of these compounds among the species analysed. Aliphatic hydrocarbons were another major class, with *I. sogdiana* containing more identified compounds than *I. alberti*. In contrast, polyphenols and flavonoids, aromatic heterocycles (benzofuran, 2,3-dihydro-; 1,1,5-trimethyl-1,2-dihydronaphthalene; naphthalene, 1,2,3,4-tetrahydro-1,1,6-trimethyl-), alcohols, aldehydes, and ketones were present in limited quantity across all species, with only minor interspecies differences observed and grouped in the other category. In the other category, different classes of compounds were listed, including isoprenoid γ -lactone (4,8,12,16-Tetramethylheptadecan-4-olide), simple ethers, and unidentified compounds.

The following list comprises the 26 compounds that were found in all three samples (Table 3), and full peak assignments are provided in Supplementary Tables S1, S3 and S5.

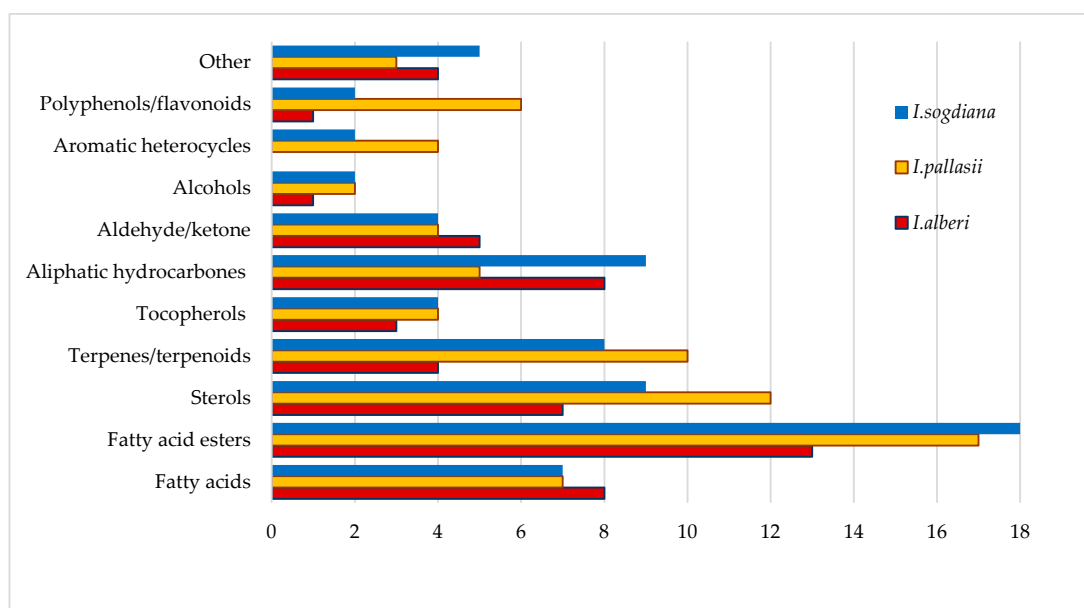


Figure 3. Identified compounds by major chemical class in CH_2Cl_2 leaf extracts of three *Iris* species.

Table 3. Recurring compounds identified in the CH_2Cl_2 extracts of leaf samples from the studied *Iris* species (retention time, LRI, relative peak area, %).

Compound	RT, min	LRI	<i>I. sogdiana</i>	<i>I. pallasii</i>	<i>I. alberti</i>
Dodecanoic acid	16.50	1570	0.74	0.06	3.83
Dodecanoic acid, propyl ester	17.99	1695	0.06	0.04	0.21
Tetradecanoic acid	18.77	1769	1.24	0.32	0.77
Tetradecanoic acid, ethyl ester	19.13	1799	0.17	0.04	0.08
Neophytadiene	19.62	1947	1.85	1.53	1.82
n-Hexadecanoic acid	20.85	1969	2.29	1.68	2.10
Phytol	22.31	2115	0.56	0.30	0.65
9,12-Octadecadienoic acid (Z,Z)-	22.54	2144	2.77	0.92	0.26
9,12,15-Octadecatrienoic acid (Z,Z,Z)-	22.60	2250	4.77	1.89	2.15
Linoleic acid ethyl ester	22.75	2169	2.00	0.88	0.50
9,12,15-Octadecatrienoic acid ethyl ester (Z,Z,Z)-	22.82	2177	2.32	0.85	1.04
Octadecanoic acid, ethyl ester	23.00	2199	0.58	0.27	0.12
Pentacosane	25.53	2500	0.37	0.24	0.45
Heptacosane	27.04	2600	1.53	1.16	0.95
Squalene	28.03	2838	1.08	1.22	1.76
Nonacosane	28.47	2900	4.48	6.23	5.00
Phytol decanoate	29.01	2971	1.12	1.29	1.08
Octacosanal	29.64	3047	1.59	2.85	0.90
γ -Tocopherol	29.93	3081	0.80	0.41	0.33
Triacontanal	31.74	3236	1.38	2.03	0.53
Campesterol	32.07	3265	1.48	3.28	6.51
Stigmasterol	32.48	3302	3.84	6.49	3.67
γ -Sitosterol	33.31	3366	12.18	13.24	28.49
Stigmastanol	33.44	3376	0.76	0.78	1.15
Stigmast-4-en-3-one	35.44	3526	1.17	1.86	1.70
Phytol palmitate	36.09	3570	2.78	2.15	4.26

Sterols and terpenoids were the most prominent compound classes identified across all analysed species. Among these, γ -sitosterol was the dominant constituent, accounting for 12.18% in *I. sogdiana*, 13.24% in *I. pallasii*, and 28.49% in *I. alberti*. Saturated and unsaturated fatty acids were found in the analysed species. Among the saturated fatty

acids, n-hexadecanoic acid was particularly notable, comprising up to 2.29% and 1.68%, while tetradecanoic acid accounted for up to 1.24%. Dodecanoic acid was also detected in significant amounts in *I. alberti* at 3.83%. The presence of polyunsaturated fatty acids and their esters was exceptionally high. 9,12,15-octadecatrienoic acid reached concentrations of 4.77% in the *I.sogdiana* and 1.88% in the *I.pallasii*, whereas 9,12-octadecadienoic acid represented 2.77% and 0.92%. Their ethyl esters were also abundant; for example, the ethyl ester of 9,12,15-octadecatrienoic acid accounted for 2.32% in *I.sogdiana* and 0.85% and 1.04% across the other species. Other significant sterols included stigmasterol, which comprised 3.84% and 6.49% in *I. sogdiana* and *I. pallasii*, and campesterol, which contributed 1.48%, 3.28%, and 26.51% in *I. alberti*.

The study also considered hydrocarbons and aldehydes, such as pentacosane, heptacosane, nonacosane, octacosane, and triacontanal, as well as esters such as phytyl decanoate. A total of sterols, triterpenoids, tocopherols, and isoprenoid-type compounds were identified across all three analysed samples (Tables 3 and 4). While the overall qualitative composition was broadly similar, the relative proportions of individual metabolites varied considerably. Sterols were the predominant class of compounds across all species. γ -Sitosterol and stigmasterol accounted for a substantial portion of the sterol profile, with the highest concentration in *I. alberti* (28.49% and 3.67%), nearly double that in *I. sogdiana* and *I. pallasii*. Campesterol increased from 1.48% in *I.sogdiana* to 3.28% in *I. pallasii*, peaking at 6.51% in *I.alberti*. Ergostan series sterols were observed in the studied species: γ -ergosterol (*I. sogdiana*, 0.63%), ergostanol (*I. pallasii*, 0.62%), and ergosta-4,6,22-trien-3 α -ol (*I. alberti*, 0.69%) (Supplementary Tables S1–S6).

Table 4. Comparative Analysis of Sterols, Terpenes, and Tocopherols in the CH₂Cl₂ extracts of leaf samples from the studied *Iris* species (RT, LRI and area, %).

Compound	RT	LRI	<i>I. sogdiana</i>	<i>I. pallasii</i>	<i>I. alberti</i>
Phytosterols and Derivatives					
Stigmasta-5,24(28)-dien-3-ol	33.51	3381	—	1.38	1.67
Stigmast-7-en-3-ol, (3 α ,5 α ,24S)-	34.09	3426	1.56	0.59	—
γ -Ergosterol	32.78	3325	0.63	—	—
Ergostanol	32.20	3277	—	0.62	—
Ergosta-4,6,22-trien-3 α -ol	34.31	3443	—	—	0.69
Triterpenes and Isoprenoids					
β -Amyrin	33.94	3414	0.5 (β -Amyrone)	0.66 (with 4-Campestene-3-one)	1.08
Lup-20(29)-en-3-one	34.27	3440	0.70	—	—
Lupeol	36.18	3576	0.21	—	—
Tocopherols					
α -Tocopherol	30.76	3154	5.75	3.58	4.27
γ -Tocopherol	29.92	3080	0.80	0.41	0.33

Tocopherol accounted for 3.58–5.75% of the total GC–MS profiles among the three *Iris* species. The highest tocopherol level was observed in *I. sogdiana* and was approximately 1.6 times greater than in *I. pallasii*, indicating moderate interspecific variation in tocopherol accumulation within the genus. Triterpenoids, including lupeol and lup-20(29)-en-3-one, have been identified as exclusive to *I. sogdiana*.

GC-MS Analysis of the TMS Derivatives of Nonpolar Leaf Extracts from Three Iris Species

The analysis showed that all three species possess complex but variable chemical profiles, comprising more than 70 identified compounds (Supplementary Tables S2, S4 and S6). These compounds predominantly belong to the fatty acids, sterols, long-chain alcohols, and hydrocarbons.

The studied species contain key phytosterols, including β -sitosterol, stigmasterol, and campesterol, as well as a wide range of common saturated fatty acids, such as palmitic, stearic, and myristic acids. Additionally, all samples contain long-chain alkanes (e.g., nonacosane and heptacosane) and alcohols such as 1-octacosanol. For example, the sterol composition differed significantly among the three species. *I. alberti* exhibited the highest overall sterol content, with β -sitosterol, which represents the main peak (16.85%) of the GC-MS profile. Furthermore, campesterol had the highest relative abundance (2.95%), and stigmasterol was present at 0.30%. In comparison, *I. pallasii* also contained substantial amounts of β -sitosterol (12.08%) and showed the highest relative concentration of octacosanol among the studied species (28.23%) (Figure 4). Meanwhile, *I. sogdiana* displayed the lowest relative abundance of β -sitosterol (3.39%). This species was characterised by the presence of stigmasterol (0.30%) and lup-20(29)-en-3-one (0.25%).

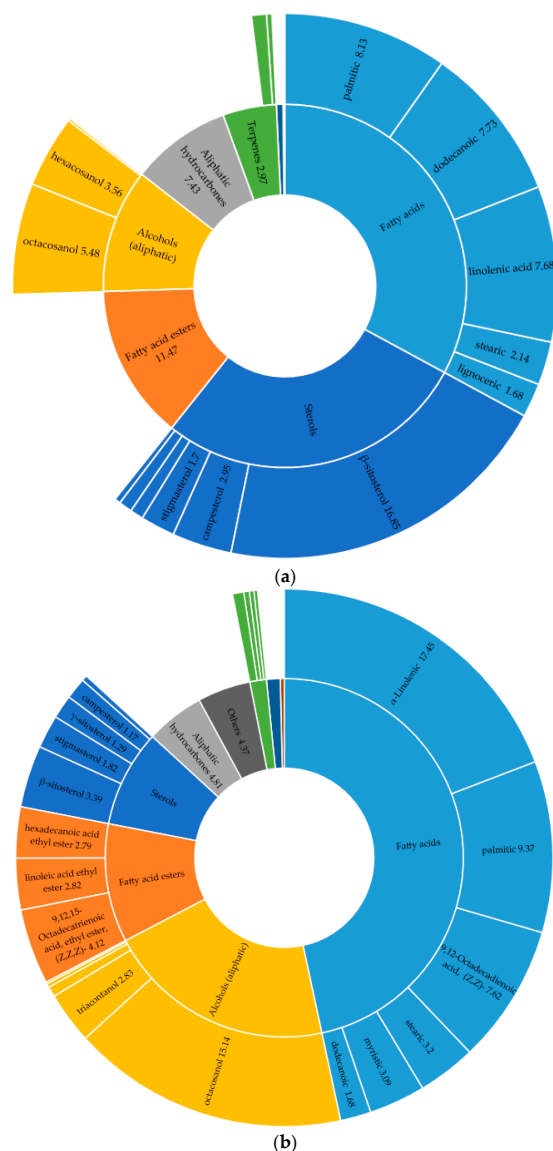


Figure 4. Cont.

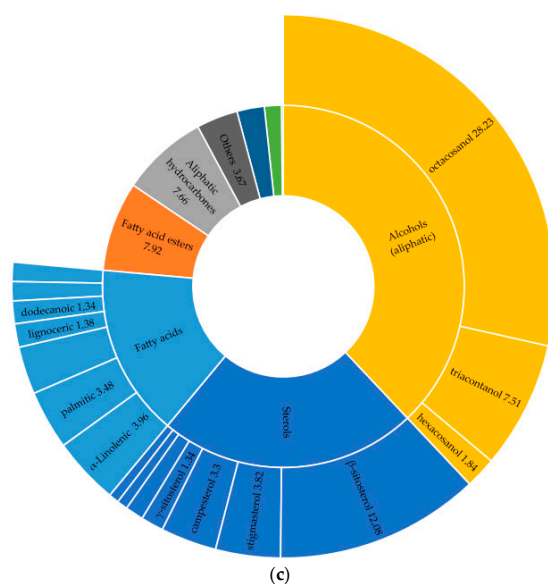


Figure 4. Percentage distribution of identified compounds by major chemical classes in three *Iris* species (TMS-derivatised of native CH_2Cl_2 extracts): (a)—*I. alberti*; (b)—*I. sogdiana*; (c)—*I. pallasii*.

A comparative analysis of fatty acids (FAs) and their derivatives revealed notable differences between species in both the amount and types of fatty acids present, although several key fatty acids were shared among all three species. *Iris sogdiana* showed the highest accumulation of unsaturated fatty acids, with α -linolenic acid as the main component of the fatty acid profile (17.45%), along with palmitic acid (9.37%) and a significant amount of fatty acid esters, suggesting active fatty acid metabolism. In contrast, *I. pallasii* had relatively low levels of α -linolenic acid (3.96%) and palmitic acid (3.48%), which aligns with the dominance of long-chain alcohols in this species. *Iris alberti* consistently contained dodecanoic acid (7.73%), and unique to this species were branched-chain (iso-) fatty acids, including isopentadecanoic and iso-palmitoleic acids, as well as oleic acid (Supplementary Table S2).

2.4. HPLC Analysis of Extracts from Rhizomes

The phytochemical profiling of the summary 70% water–alcohol extracts from rhizomes belonging to the species of *I. alberti*, *I. pallasii*, and *I. sogdiana*, as well as ethyl acetate extracts from *I. pallasii* (EA-IP) and *I. sogdiana* (EA-IS), revealed significant qualitative and quantitative variations in their phenolic composition. Only 11 compounds were identified across the analysed samples (Table 5). While certain marker compounds were consistently present, the extracts exhibited distinct chemical signatures, especially in the distribution of simple phenolics, flavonoids, and phenolic esters. These compositional differences were also evident in the antioxidant activities assessed through DPPH and ABTS assays.

The HPLC-DAD analysis revealed that *I. alberti* exhibited a profile dominated by coumarin (35.92 mg/g) and trans-2-hydroxycinnamic acid (15.82 mg/g), which together represent the highest concentration of simple phenolics across all samples. Additionally, the presence of genistein (3.84 mg/g) introduces a flavonoid component to the overall profile, although at comparatively low levels.

In contrast, *I. pallasii* was characterised by flavan-3-ols, most notably catechin (4.72 mg/g) and epicatechin (1.98 mg/g), which were absent from other extracts. These compounds, commonly associated with potent radical scavenging activity, suggest a mechanistic basis for the observed antioxidant potency of this extract.

Table 5. Retention times, calibration parameters ($R^2 = 0.99$), linearity ranges, limits of detection (LOD), recoveries, and phenolic composition were determined for rhizome extracts from three Iris species using HPLC–DAD at 254 nm (mg/g extract). Analyses were conducted on 70% water–alcohol extracts of *I. alberti*, *I. pallasii*, and *I. sogdiana*.

Compound	RT	Calibration Equation	Linear Range	LOD	LOQ	Recovery (%)	RSD (n = 7)	<i>I. alberti</i>	<i>I. pallasii</i>	<i>I. sogdiana</i>	EA-IP	EA-IS
Protocatechuic acid	24.625	$y = 76,181x - 88,801$	3.13–100	3.42	10.35	102.35 ± 4.21	3.19	-	-	-	0.47 ± 0.01	-
Catechin	30.274	$y = 3865.1x + 32,660$	15.6–500	3.29	9.96	102.11 ± 4.08	3.78	-	4.72 ± 0.18	-	-	-
4-hydroxy benzaldehyde	33.367	$y = 34,376x + 4239.6$	1.25–50	1.33	4.44	99.01 ± 2.78	4.76	-	-	-	-	0.04 ± 0.002
Vanillic acid	34.758	$y = 74,653x - 9634.1$	1.56–100	1.56	4.68	103.58 ± 4.43	5.06	-	-	-	0.43 ± 0.02	0.92 ± 0.05
Epicatechin	35.278	$y = 2097.6x + 7998.2$	3.9–500	0.95	2.86	101.25 ± 1.65	4.00	-	1.98 ± 0.08	-	-	-
Caffeic acid	35.28	$y = 67,972x - 32,965$	3.00–30.0	4.54	13.75	102.67 ± 4.92	4.01	-	-	-	tr	0.64 ± 0.02
Ferulic acid	42.564	$y = 35,737x + 12,977$	2.34–300	3.96	11.99	100.99 ± 3.54	3.20	-	-	-	-	0.61 ± 0.02
Coumarin	45.178	$y = 36,021x - 23,215$	3.13–100	2.21	6.69	101.74 ± 4.83	3.59	35.92 ± 1.19	-	-	-	-
Propylgallate	46.984	$y = 29,731x - 12,781$	1.86–952.5	1.46	4.44	95.44 ± 3.81	3.83	-	-	250.83 ± 9.61	-	1.02 ± 0.04
trans-2-hydroxy-cinnamic acid	48.243	$y = 53,843x + 12,4308$	3.13–400	3.09	9.27	99.75 ± 3.75	2.85	15.82 ± 0.45	-	-	-	-
Genistein	57.739	$y = 69,160x + 5753.8$	4.74–615	2.20	10.14	99.00 ± 4.51	3.11	3.84 ± 0.11	-	-	0.58 ± 0.02	7.76 ± 0.24

Note: Linear range, LOD, and LOQ values are expressed in $\mu\text{g/mL}$.

Iris sogdiana showed an extraordinarily high propyl gallate content (250.83 mg/g), a well-known synthetic phenolic antioxidant often encountered as a natural artefact or an extraction-derived constituent. Its overwhelming abundance suggests either deliberate enrichment during sample processing or an intrinsic conversion of native gallic acid derivatives during extraction. The presence of trace amounts of genistein further supports the notion that the extract retains limited native phenolic diversity.

Among the ethanol extracts, samples EA-IP and EA-IS showed broader but lower-intensity phenolic profiles. EA-IP contained protocatechuic and vanillic acids, as well as caffeic acid in trace amounts, while both incorporated genistein in concentrations of 0.58 mg/g and 7.76 mg/g, respectively. Sample EA-IS additionally contained low levels of vanillic acid, caffeic acid, and ferulic acid, indicating partial hydrolysis of phenolic glycosides or liberation of bound phenolics during ethanol extraction.

Overall, the phenolic landscape clearly demonstrates that the samples differ not only in specific phenols present but also in the structural classes they represent—simple phenolic acids in the extracts of *I. alberti* and EA-IP, flavan-3-ols in sample 2, phenolic esters in *I. sogdiana*, and mixed phenolics with flavonoids in EA-IS.

Correlation Between Phenolic Composition and Antioxidant Activity

The antioxidant assays revealed significant differences in activity across samples, with IC₅₀ values closely correlating with the presence of specific phenolic groups (Table 5). In the DPPH assay, *Iris alberti* exhibited the lowest IC₅₀ value among the native extracts (16.90 ± 6.04 µg/mL), whereas *I. pallasii* showed a slightly higher IC₅₀ (28.77 ± 2.93 µg/mL), and *I. sogdiana* demonstrated negligible activity (IC₅₀ > 400 µg/mL). In the ABTS assay, *I. pallasii* displayed the lowest IC₅₀ (10.93 ± 7.29 µg/mL), *I. sogdiana* showed moderate activity (114.42 ± 3 µg/mL), and *I. alberti* was inactive under the tested conditions (NA).

Ethyl acetate (EA) extracts generally exhibited lower radical scavenging potential than the native extracts. EA-*I. pallasii* had IC₅₀ values of 64.38 µg/mL (DPPH) and 150.21 µg/mL (ABTS), while EA-*I. sogdiana* ranged from 120 to >300 µg/mL in both assays. These results are consistent with the lower total phenolic content in the EA fractions.

Across all samples, IC₅₀ values corresponded with the relative abundance of phenolic compounds. Extracts enriched in flavan-3-ols, coumarins, and cinnamic acid derivatives generally had lower IC₅₀ values, reflecting higher radical scavenging potential, whereas extracts with lower phenolic content or dominated by other compounds showed higher IC₅₀ values and reduced activity. Differences between DPPH and ABTS assays indicate that both hydrogen-atom transfer and electron-transfer mechanisms contribute to the observed antioxidant activity, depending on each extract's phenolic composition.

Taken together, these results demonstrate that antioxidant profiles are species-dependent and influenced by both the type and abundance of phenolic constituents. Native extracts consistently exhibited higher radical scavenging potential than the EA fractions, highlighting the importance of the complete phenolic matrix. All IC₅₀ values were higher than those of the reference antioxidant BHA (5.09 ± 0.22 µg/mL for DPPH, 2.21 ± 0.09 µg/mL for ABTS), indicating lower activity than the standard.

3. Discussion

The complex taxonomy of *Iris* Tourn. ex L. remains a critical consideration when interpreting phytochemical data [28,34]. Notably, *Iris sogdiana* Bunge is frequently treated as a synonym of *I. halophila* var. *sogdiana* or *I. spuria* L. [25,35], while *I. pallasii* Fisch. ex Trevir. is often regarded as a synonym of *I. lactea* Pall. [25,28]. Despite these taxonomic ambiguities, and while retaining the species names as recognised in the flora of Kazakhstan [2–4,12,15],

these species exhibit pronounced adaptations to local eco-geographical conditions, as evidenced by their successful acclimatisation at the MBG and the Ili Botanical Garden (IBG) [19,36].

A herbarium review of *Iris* collections from southeastern Kazakhstan at the Institute of Botany and Phytointroduction (herbarium AA) confirmed at least 65 species across five floristic regions: Balkhash-Alakol, Dzungarian Alatau, Zailiysky and Kungey Alatau, Ketmen and Terskey Alatau, and Chu-Ili mountains. *I. sogdiana* was found in eight floristic regions. *I. pallasii* was recorded in Dzungar Alatau, Zailiysky Alatau, Toraygyr mountains, and Balkhash-Alakol. *I. alberti* was identified in Zailiysky Alatau, the Kazakh part of the Western Tien-Shan, and other Tien-Shan regions in Kyrgyzstan (Appendix B, Table A2). These studies independently verify species identity and geographic distribution, supporting the relevance of the phytochemical and antioxidant analyses presented here.

This study is the first report on the quantitative (flavonoids, tannins, and alkaloids) and phytochemical profiles of CH₂Cl₂ extracts and their TMS derivatives from leaves, along with HPLC-DAD analysis of phenolic compounds and the antioxidant potential of summary extracts from rhizomes for the Kazakhstani three *Iris* species.

Quantitative analysis of rhizome extracts revealed relatively low total flavonoid content (0.29–0.33%) and tannin content (1.06–1.88%) across all three species, whereas their qualitative profiles exhibited clear species-specific differences.

This research provides the first detailed phytochemical profile of *I. alberti* rhizomes and leaves. HPLC-DAD analysis revealed that simple phenols, particularly coumarin (35.92 mg/g) and trans-2-hydroxycinnamic acid (15.82 mg/g), are the predominant compounds. According to [27,29], the leaves of *I. alberti* contain xanthenes (mangiferin and isomangiferin) and phenolic acids (sinapic and ferulic), along with elevated tannin [29]. While GC-MS analysis showed that the leaves contain saturated fatty acids (dodecanoic acid, *n*-hexadecanoic acid) and the unsaturated fatty acid 9,12,15-octadecatrienoic acid, a profile characteristic of the lipophilic fractions of numerous *Iris* species, such as *I. lactea* and *I. sibirica* [28,37]. Squalene, terpenes and phytosterols, including campesterol (6.51%) and γ -sitosterol (28.49%), were also detected in *I. alberti*. Analysis of sterols in other species, including *I. lactea*, *I. germanica*, and *I. pallida*, revealed the presence of stigmasterol-3-O- β -D-glucopyranoside and β -sitosterol [30]. The occurrence of β -amyrin in *I. alberti* aligns with previous reports of this pentacyclic triterpenoid in the rhizomes of *I. germanica* [38]. Additionally, α -iron (0.11%) was detected in the lipophilic fraction of *I. alberti*; irons, including α -, β -, and γ -isomers, are characteristic of irises in the *Iris* section (bearded irises such as *I. germanica* and *I. florentina*) and contribute to their distinctive violet aroma [27,39].

While the literature on *Iris spuria* (a syn. *I. sogdiana*) has focused on isoflavonoids (tectorigenin, irisolone) and rotenoids (12 α -hydroxirotenoid) [27,34], our analysis revealed an exceptionally high content of propyl gallate (250.83 mg/g) in the summary extract of *I. sogdiana*. Additionally, in the EA extract, six phenolic compounds were identified and quantified: vanillic acid (0.92 mg/g), caffeic acid (0.64 mg/g), ferulic acid (0.61 mg/g), and genistein (7.76 mg/g). Furthermore, *I. sogdiana* exhibited the highest potassium content (538.36 mg/100 g) among the studied species. GC-MS analysis of the non-polar extracts confirmed that sterols and triterpenoids are the predominant classes of compounds across all three species, with γ -sitosterol (53.24%) and stigmasterol (16.77%).

In the literature, *Iris lactea* s.l. is the most frequently described species, and, if considered a synonym of *I. pallasii*, it is among the most extensively studied taxa within the genus (Appendix A). Nevertheless, the studied phenolic profile reveals the presence of catechin (4.72 mg/g) and epicatechin (1.98 mg/g). The EA extracts contained protocatechuic, vanillic, and caffeic acids, which have also been identified in other species of the genus [17,28]. Compared to the sterol and terpenes profiles of the rhizome, which con-

tains only stigmasterol derivatives according to [28], we semi-quantified the presence of campesterol (1.17–3.30%), γ -, β -sitosterol (1.29 and 16.85%), and fucosterol (0.66%).

This study presents the first comprehensive phytochemical and quantitative analysis of Kazakhstani Iris species, examining flavonoids, tannins, alkaloids, phenolic compounds, and antioxidant activity in rhizomes and leaves. Comparative analysis of *I. albertii*, *I. pallasii*, and *I. sogdiana* identifies both shared structural compounds characteristic of the genus and unique metabolites reflecting species-specific ecological adaptations. Sterols, triterpenoids, fatty acids, and phenolic compounds emerged as the dominant constituents across all studied species.

4. Materials and Methods

Quantitative analyses were performed in accordance with the protocols described in the State Pharmacopoeia of the Republic of Kazakhstan [40] and procedures described in [41].

4.1. Sample Preparation

The dried, ground leaves of the studied samples (0.2 g) were extracted with 2 mL of dichloromethane. Extraction was carried out for 2 h at room temperature, consisting of two ultrasonic treatments (5 min each) at 25 °C. The extract was decanted and evaporated to dryness without heating; the dry residue was redissolved in dichloromethane and analysed by GC–MS. The extractive yield was 10.33%.

4.2. Summary Extract Preparation

The summary extracts were prepared using 50% water–ethanol, and each sample underwent three extractions to ensure maximal recovery of phenolic constituents. The combined extracts were concentrated to a minimal volume and then sequentially partitioned with chloroform and ethyl acetate to obtain the corresponding solvent fractions.

Silylation: To the dry extract obtained using the previously described method, add 50 μ L of N, O-bis(trimethylsilyl)acetamide (BSA). Incubate the mixture at 50 °C with constant stirring using a magnetic stirrer until the residue has fully dissolved and the silylation reaction is complete. Once the reaction is finished, dilute the resulting solution of silylated derivatives with dichloromethane and analyse it using GC–MS.

4.3. GC-MS Analysis

GC–MS analysis was performed using a Maestro gas chromatograph (“Interlab”, Moscow, Russia) coupled with a quadrupole mass spectrometer detector (Agilent 5975) and a capillary quartz column (HP-25, 30 m, “Interlab”, Moscow, Russia). The carrier gas was helium (1 mL/min), and the oven temperature was programmed from 50 °C (3 min) to 280 °C over 10 min, followed by 20 min isothermal at 280 °C (total run time 45 min). The injector temperature was 270 °C, injection volume 1 μ L, with electron-impact ionisation at 70 eV. Components were identified using the NIST-2011 and Wiley-2008 libraries (match factor $\geq$ 85%) and confirmed by calculating linear retention indices (LRIs) from a homologous series of n-alkanes (C8–C37) under the same chromatographic conditions. The LRI for each compound was determined according to the Van den Dool and Kratz equation:

$$\text{LRI} = 100 \times n + 100 \times \frac{t_x - t_n}{t_{n+1} - t_n}$$

Here, t_x is the retention time of the compound, t_n and $t_{(n+1)}$ are the retention times of the n-alkanes eluting immediately before and after the compound, and n is the carbon number of the preceding n-alkane. LRIs, together with mass spectral data, were used to confirm compound identities by comparing them with literature values and reference databases. Relative abundances were expressed as the percentage of peak area in the total

ion chromatogram (TIC), providing a semi-quantitative comparison of constituents within and between samples.

4.4. HPLC–DAD Analysis

Phenolic compounds in the extract were analysed together with 43 standard phenolics [42] using a Shimadzu HPLC–DAD system (Shimadzu, Kyoto Japan) consisting of an LC-20AT pump and an SPD-M20A diode array detector, controlled by LC-Solution software 5.117 software (CBM-20A). Separation was achieved on an Inertsil ODS-3 column (GL Sciences Inc., Shinjuku-ku, Tokyo, 163-1130 Japan) (4 µm, 4.0 × 150 mm) with a guard column at 35 °C. The mobile phase comprised 0.1% acetic acid in water (A) and 0.1% acetic acid in methanol (B) and was applied in gradient mode at a flow rate of 1.0 mL/min. Samples and standards (8 mg/mL) were filtered through a 0.45 µm MN filter, and 20 µL was injected. Detection was performed at 254 nm. Phenolic compounds were identified by comparing retention times, and results were expressed as mg/g dry extract.

4.5. Antioxidant Activities

The antioxidant activity of the extracts was evaluated spectrophotometrically using DPPH free radical scavenging and ABTS cation radical scavenging assays with a 96-well microplate reader (SpectraMax 340PC384, Molecular Devices, Silicon Valley, CA, USA). SoftMax Pro v5.2 software (Molecular Devices, Silicon Valley, CA, USA) was used for data analysis and bioactivity calculations. Stock solutions of the samples were prepared at a concentration of 4000 µg/mL. Radical scavenging activity was calculated using the following formula:

$$\text{Scavenging activity (\%)} = [(\text{Abs control} - \text{Abs sample}) / \text{Abs control}] \times 100,$$

where Abs control represents the initial absorbance of DPPH or ABTS, and Abs sample represents the absorbance of the remaining radicals in the presence of the extract or positive control. Antioxidant activity was expressed as IC₅₀ values (µg/mL), defined as the concentration of the extract required to inhibit 50% of DPPH or ABTS radicals. BHA was used as a positive control.

4.6. DPPH Radical Scavenging Assay

The free radical scavenging activity of the extracts was evaluated using the DPPH assay [43]. A 0.1 mM DPPH solution was prepared by dissolving 39.4 mg of DPPH in methanol. Aliquots of 40 µL of the extracts at various concentrations were mixed with 160 µL of DPPH solution. After incubation in the dark at room temperature for one hour, absorbance was measured at 517 nm. Butylated hydroxyanisole (BHA) was used as the reference antioxidant. All measurements were performed in triplicate.

4.7. ABTS Cation Radical Scavenging Assay

ABTS cation radical scavenging activity was determined according to [44] with slight modifications [45]. The ABTS cation radical was generated by reacting 7.0 mM ABTS with 2.45 mM potassium persulfate and incubating the mixture in the dark at room temperature for 12 h. Prior to analysis, the solution was diluted with distilled water to an absorbance of 0.70 ± 0.01 at 734 nm. In each well, 160 µL of ABTS solution was mixed with 40 µL of the extract at different concentrations. Solvent for the extract served as the negative control, and BHA as the standard antioxidant. Absorbance was recorded at 734 nm after 10 min of incubation at room temperature. All analyses were conducted in triplicate.

4.8. Statistical Analyses

HPLC and activity assay data represent the mean of three independent replicates and are expressed as mean $\pm$ SEM. Statistical analysis was performed using ANOVA, and significant differences between means were determined by Student's *t*-test ($p < 0.05$).

5. Conclusions

The taxonomy of *Iris* species remains complex, with *I. sogdiana* and *I. pallasii* often considered synonyms of other taxa. Nevertheless, all studied species demonstrate clear local adaptations to Kazakhstani conditions. This study provides the first comprehensive phytochemical and quantitative analysis of these species, encompassing flavonoids, tannins, alkaloids, phenolic compounds, and antioxidant activity in rhizomes and leaves.

The results reveal species-specific phytochemical profiles: *I. alberti* is rich in xanthenes, phenolic acids, fatty acids, squalene, terpenes, and phytosterols, including β -amyrin and α -iron, contributing to its aroma; *I. sogdiana* exhibits exceptionally high propyl gallate, multiple phenolic acids, and elevated potassium, with sterols and triterpenoids predominant in non-polar extracts; *I. pallasii* contains diverse phenolics and rhizome sterols, including campesterol, γ - and β -sitosterol, and fucosterol.

Overall, sterols, triterpenoids, fatty acids, and phenolic compounds are the dominant constituents across all Kazakhstani *Iris* species studied, reflecting both common genus-level features and species-specific adaptations to their ecological environments.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/molecules31040643/s1>, Table S1: GC–MS peak assignments of native leaves extract of *Iris alberti* (RT, compound name, peak area %); Table S2: GC–MS peak assignments of TMS-derivatized extract of *Iris alberti* (RT, compound name, peak area %); Table S3: GC–MS peak assignments of native leaves extract of *Iris pallasii* (RT, compound name, peak area %); Table S4: GC–MS peak assignments of TMS-derivatized extract of *Iris pallasii* (RT, compound name, peak area %); Table S5: GC–MS peak assignments of native leaves extract of *Iris sogdiana* (RT, compound name, peak area %); Table S6: GC–MS peak assignments of TMS-derivatized extract of *Iris sogdiana* (RT, compound name, peak area %).

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Appendix A

Table A1. Component composition and biological activity of the species *Iris* L.

Species	Species (syn.)	Plant Part	Compounds	Ref.
<i>Iris alberti</i> Regel	<i>I. alberti</i>	Leaves	Phenolic acids: sinapic and ferulic; vitamin C; tannins (2.99 and 3.52%)	[29,30]
		Rhizome	Xanthones: mangiferin, and isomangiferin	
	<i>I. halophila</i> Pall.	Leaves	Ascorbic acid 250–950.6 mg%.	[30,31,34]
		Seeds	Stilbenes: halophilol A, halophilol B, resveratrol, ϵ -viniferin, γ -2-viniferin. Iridals: Oxo-23-hydroxyiridal-3- $[\beta$ -D-Glcp-(1 $\rightarrow$ 6)- β -D-Glcp]-16- β -D-Glcp, Oxo-23-hydroxyiridal-3,16-di- β -D-Glcp, Oxo-23-isoiridal-3,16,23-tri- β -D-Glcp, Oxo-23-hydroxy-isoiridal-3- $[\beta$ -D-Glcp-(1 $\rightarrow$ 6)- β -D-Glcp]-16- β -D-Glcp, Oxo-23-hydroxy-isoiridal-3,16-di- β -D-Glcp, Dihydroxy-iridal-3,16-di- β -D-Glcp, 22,23-Dihydroxy-isoiridal-3,16-di- β -D-Glcp;	
<i>Iris sogdiana</i> Bunge	<i>Iris spuria</i>		isoflavonoes: 7-O-,4'-O—D-glucopyranosylisoflavones; 7-O- β -D-Glcp(tectoridin), isoflavone-4'-O- β -D-Glcp; isoflavone disaccharides: 5,4'-dihydroxy-6,7-dimethoxyisoflavone(7-methyltectorigenin); IF-7-O- β -Glcp-(1 $\rightarrow$ 6)- β -Glcp; 7-O- β -D-Glcp-4'-O- β -D-Glcp; Rotenoid, 1,11-dihydroxy-9,10-methylenedioxy-12a – dehydrorotenoid; tectorigenin-7-O- β -glucosyl-4'-O- β -glucoside	[35–37]
		Rhizome Roots	Isoflavones: irilin A; iristectorigenin B; iristectorigenin B 4'-O- β -D-glucopyranoside; iristectorigenin B 4'-[O- β -D-glucopyranosyl-(1''' $\rightarrow$ 6'')— β -D-glucopyranoside; flavonoids alpinone; sterol: β -daucosterol	
	<i>Iris halophila</i> var. <i>sogdiana</i>		Flavonoids and isoflavanoid: 5,7,3'-Trihydroxy-6,4'-dimethoxyflavanone; 5,8,2'-Trihydroxy-7-methoxyflavanone; Tectorigenin; 7-O-glucoside; Iristectorigenin A.	[36]
			Xanthones: mangiferin and isomangiferin; Flavonoids (including C-glycosylflavones): apigenin- and luteolin-type compounds (orientin, isoorientin/homoorientin, embinin and acetylated derivatives, swertiajaponin); Phenolic acids: hydroxybenzoic and hydroxycinnamic acids (gallic, protocatechuic, vanillic, syringic, caffeic, chlorogenic, ferulic, p-coumaric, trans-cinnamic). (C12–C18).	
<i>Iris pallasii</i> Fisch. ex Trevir.	<i>I. lactea</i> Pall.	Rhizomes Roots	Xanthones: mangiferin and isomangiferin; Flavonoids (including C-glycosylflavones): apigenin- and luteolin-type compounds (orientin, isoorientin/homoorientin, embinin and acetylated derivatives, swertiajaponin); Phenolic acids: hydroxybenzoic and hydroxycinnamic acids (gallic, protocatechuic, vanillic, syringic, caffeic, chlorogenic, ferulic, p-coumaric, trans-cinnamic).	[30,32,33]
		Aerial parts		

Table A1. Cont.

Species	Species (syn.)	Plant Part	Compounds	Ref.
<i>Iris pallasii</i> Fisch. ex Trevir.	<i>I. lactea</i> Pall.	Leaves	Xanthones (iriflophenone and mangiferin derivatives), quinones (irisquinone and irisoquin types), flavonoids and isoflavonoids (luteolin, apigenin, kaempferol, tectorigenin and their C- and O-glycosides), phenolic acids (hydroxybenzoic and hydroxycinnamic acids); terpenoids (iridal-type triterpenoids and sesquiterpenes), steroids (stigmasterol derivatives), and fatty acids (C12–C18).	[30,32,33]
	<i>I. oxypetala</i> Bunge (= <i>I. lactea</i> var. <i>chinensis</i> (Fisch.) Koidz., <i>I. lactea pallasii</i> Fischer var. <i>chinensis</i> , <i>I. pallasii</i> Fisch. var. <i>chinensis</i> Fisch.)	Seeds	Alkylphenols and alkylbenzenes (irisphenol and belamcandaphenol derivatives), benzofuran derivatives (belamcandone type), quinones (irisquinone and pallasone derivatives), flavan-3-ols and proanthocyanidins (catechin/epicatechin oligomers), oligostilbenes (viniferin-, vitisin-, and hopeaphenol-type compounds), and fatty acids, dominated by linoleic and oleic acids with minor saturated and long-chain components.	

Appendix B

Most herbarium specimens of the studied species were collected in the Zailiysky and Dzungarian Alatau regions, indicating that the greatest species diversity of irises in southeastern Kazakhstan is concentrated in the mountainous areas (Zailiysky, Kungey, and Dzungarian Alatau).

Table A2. The collection of iris species examined in this study, gathered from southeastern Kazakhstan and stored in the institute’s herbarium AA.

Species	Floristic Region, Gorges	Number of Specimens	Year of Collection	Herbarium Specimen
<i>Iris alberti</i> Regel	25. Zailiysky Alatau	5	1888, 1930, 1933, 1950, 2018	
<i>Iris pallasii</i> Fisch. ex Trevir.	18. Balkhash—Alakol	4	1939, 1966, 2013, 2014	
	24. Dzungarian Alatau	1	1964	
	25. Zailiysky Kungey Alatau	3	1951	
	25a. Ketmen, Terskey Alatau	1	2013	
<i>Iris sogdiana</i> Bunge	18. Balkhash -Alakol	3	1930, 1949, 1956	
	24. Dzungarian Alatau (6)	6	1927, 1950, 1956, 1959	
	25. Zailiysky Kungey Alatau	23	1937–1975, 1891, 1920	
	25a. Ketmen, Terskey Alatau	7	1961, 1962, 1972, 2013	
	26. Chu-Ili mountains	1	1940	

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