

Phytochemical Profiling and Anti-Gastric Cancer Activity of *Akebia quinata* Root Extract

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ABSTRACT

Gastric cancer is a highly prevalent malignancy worldwide and remains associated with poor prognosis, highlighting the urgent need for effective and low-toxicity therapeutic strategies. *Akebia quinata*, a plant widely used in traditional medicine, contains diverse bioactive compounds, yet its anti-gastric cancer activity and molecular basis remain poorly understood. In this work, the inhibitory activity of ethanol extracts of *A. quinata* obtained from different plant organs (leaves, stems, and roots) and seasons (summer and autumn) was tested in the human gastric cancer cell line MKN74 using an MTT assay, revealing that the summer root extract exhibited the strongest anti-proliferative activity (76.46% inhibition). Comprehensive chemical profiling using GC-MS and UPLC-QTOF-MS identified multiple compounds with potential anti-cancer activity, including dihydrocucurbitacin F and anemoside A3. To further investigate the potential mechanisms, network pharmacology analysis was conducted to construct a component-target-disease network, identifying AKT1 and STAT3 as key putative targets; molecular docking also suggested stable compound-protein interactions with the corresponding targets, with predicted binding free energies below -5.0 kcal/mol. Overall, these findings indicate that *A. quinata* root extract exerts notable *in vitro* anti-gastric cancer activity and may act through multi-target modulation of cancer-related signaling pathways, providing a basis for further investigation of this plant as a potential source of anti-cancer compounds.

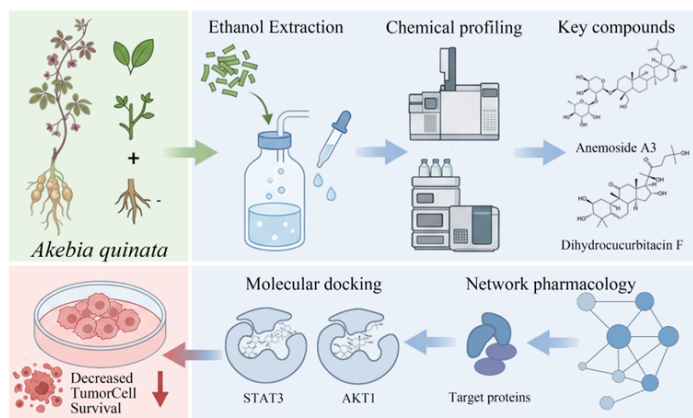
Keywords

Akebia quinata, Gastric cancer, Chemical profiling, Network pharmacology, AKT1 signaling, Molecular docking.

Research highlights

Summer root extract showed the strongest anti-proliferative activity against MKN74 cells.
Ethanol extracts varied markedly in anti-gastric cancer activity by organ and season.
GC-MS and UPLC-QTOF-MS identified multiple putative anti-cancer compounds.
Dihydrocucurbitacin F and anemoside A3 were highlighted as key active constituents.
AKT1 and STAT3 were identified as potential targets underlying the anti-cancer effect.

Graphical Abstract



Introduction

Cancer represents a substantial global health burden, with more than 18 million newly diagnosed cases and nearly 10 million cancer-related deaths reported each year [1]. Among all cancer types, gastric cancer is the fifth most frequently diagnosed malignancy and the fifth leading cause of cancer-related death worldwide [2]. Due to the absence of obvious symptoms during the early stages, gastric cancer is often identified only at an advanced stage, which limits therapeutic options and contributes to an unfavorable clinical outcome [3]. Current treatment strategies, including endoscopic resection, surgery, radiotherapy, chemotherapy, targeted therapy, and immunotherapy [4], have improved patient outcomes; however, their effectiveness remains limited in many cases. They remain associated with significant limitations, including therapeutic resistance, adverse effects, tumor recurrence, and metastasis [5]. These challenges underscore the urgent need to develop novel, effective, and low-toxicity therapeutic approaches.

Natural plants provide a valuable reservoir of bioactive molecules with diverse pharmacological functions. A growing body of evidence suggests that phytochemicals, including tannins, flavonoids, terpenoids, and alkaloids, exhibit significant anti-cancer activity [6]. Notably, a substantial proportion of currently used anti-cancer drugs are derived from or inspired by natural products, underscoring their significance in drug discovery [7]. In the context of gastric cancer, increasing evidence indicates that plant-derived extracts have been reported to inhibit tumor cell proliferation, induce cell cycle arrest, and modulate key molecular pathways. For example, tannin-rich extracts have shown strong inhibitory effects on AGS cells [8], while extracts from *Blumea lacera*, *Ardisia gigantifolia*, and *Allium* species have been reported to regulate cancer cell growth, cell cycle progression, and gene expression in gastric cancer models [9-11]. Collectively, these findings highlight the potential of plant-derived compounds as candidates for further investigation in gastric cancer therapy.

Akebia quinata, a plant used in traditional medicine, contains diverse bioactive constituents, including calceolarioside B, which has been reported to exhibit anti-bacterial, anti-inflammatory, antioxidant, and anti-cancer activities [12]. In traditional Chinese medicine, it has been used as a diuretic, anti-inflammatory, and analgesic, while modern studies have further highlighted its antioxidant and anti-aging potential, supporting its potential relevance for pharmaceutical and cosmetic applications. These reported bioactivities suggest that *A. quinata* may represent a potential source of bioactive compounds for further pharmacological investigation. Although its antioxidant activity has been preliminarily characterized [13], its anti-cancer potential, particularly against gastric cancer, remains insufficiently investigated.

To date, systematic investigations into the inhibitory effects of *A. quinata* extracts on gastric cancer cells, particularly in the MKN74 cell line, are limited, and the underlying mechanisms remain unclear. This lack of mechanistic understanding limits the rational evaluation and potential application of this plant in cancer therapy.

Therefore, this study aims to evaluate the inhibitory effects of *A. quinata* extracts on MKN74 gastric cancer cells and to explore their potential mechanisms through integrated chemical analysis and computational approaches. This work provides preliminary insights into the pharmacological potential of *A. quinata* and supports its further development as a plant-derived candidate for gastric cancer therapy.

Materials and Methods

Plant Material Collection and Extraction

Samples of different tissues of *A. quinata* (Thunb.) Decne. were collected from Madao Forest Farm, Zhumadian City, Henan Province, China, on two separate dates: summer samples on August 4, 2024, and autumn samples on November 1, 2024. Voucher specimens were assigned the following numbers: summer leaf (100X1), summer stem (100X2), summer root (100X3), autumn leaf (100Q1), autumn stem (100Q2), and autumn root (100Q3), and were stored in a refrigerator at 4 °C. Immediately after collection, the samples were subjected to pretreatment, including surface cleaning, air-drying, and sectioning. The dried materials were ground into a homogeneous powder under controlled low-temperature conditions using a high-speed grinder, and the powders were stored in the dark until use. For extraction, 10 g of each tissue powder was mixed with 300 mL of absolute ethanol (solid-to-solvent ratio 1:30, w/v) and subjected to reflux extraction using a constant-temperature water bath at 75 °C for 5 h. The extracts were then concentrated to approximately 20 mL using rotary evaporation, and the concentrated solutions were sealed and stored at 4 °C for subsequent analyses.

MTT Assay

To determine the concentration of extract residues, 1 mL of each sample extract was transferred into a pre-weighed vial, the extract aliquots were dried under vacuum at 50°C, frozen at -80°C for 1 h, and subsequently lyophilized. The vial was reweighed to calculate the mass of solid content per mL of extract. Based on this value, each extract was redissolved in DMSO to prepare a 100 mg/mL stock solution, which was then diluted into culture medium to achieve a final treatment concentration of 1 mg/mL.

Human gastric cancer MKN74 cells (CL-0730, Procell, Wuhan, China) were maintained in RPMI-1640 medium (Procell) supplemented with 10% fetal bovine serum (FBS, Sangon Biotech) without antibiotics. Cells were seeded into 96-well plates at a density of 5×10^4 cells/mL (5×10^3 cells/well, 100 μ L/well) and incubated for 24 h at 37 °C in a humidified atmosphere containing 5% CO₂. The medium was then replaced with fresh medium containing the plant extract at the indicated final concentration, while control wells received the same volume of culture medium containing an equivalent concentration of DMSO and 10% FBS. After a further 24 h of incubation, each treatment was performed with three technical replicates.

Subsequently, 50 μ L of MTT solution (1 mg/mL) was added to each well, and the cells were incubated for an additional 4 h. The supernatant was carefully removed, and 100 μ L of DMSO

was added to dissolve the formazan crystals. The plate was gently shaken until the formazan crystals dissolved completely, then incubated at 37 °C for 10–15 min under gentle shaking. Absorbance was measured at 570 nm using a microplate reader. The cell inhibition rate was calculated as Eq. (1):

$$\text{Cell inhibition (\%)} = (1 - A_t/A_c) \times 100 \quad (1)$$

where A_t and A_c represent the absorbance of the treated and control groups, respectively.

FTIR Analysis

FTIR spectra were acquired on a Nicolet iS10 FTIR spectrometer (Thermo Fisher Scientific, USA) using the KBr pellet coating method. KBr powder (80 mg) was dried at 105 °C for 24 h, finely ground in an agate mortar, and pressed into a transparent pellet. Then, 10 μ L of undiluted *A. quinata* ethanol extract was applied to the pellet surface and dried under ambient conditions. Spectral acquisition was performed over 4000–400 cm^{-1} at a resolution of 4 cm^{-1} with 32 scans per sample. Before each measurement, the background spectrum of pure KBr was collected to correct for atmospheric H_2O and CO_2 . The spectra were processed in OMNIC 8.0 software using baseline correction and smoothing. Peak assignments were tentatively made by comparison with published literature.

GC-MS Analysis

Six ethanol extracts of *A. quinata* were analyzed by GC–MS using an Agilent 7890B gas chromatograph coupled with an Agilent 5977A mass spectrometer (Agilent Technologies, USA). Chromatographic separation was achieved on an HP-5MS fused-silica capillary column (60 m $\times$ 0.25 mm $\times$ 0.25 μ m). High-purity helium was used as the carrier gas at a constant flow rate of 1.5 mL/min. A 1 μ L aliquot of each sample was injected in splitless mode, and the inlet temperature was maintained at 250 °C. The oven temperature was programmed as follows: 50 °C initially, ramped to 250 °C at 8 °C/min and held for 2 min, then increased to 280 °C at 5 °C/min and held for 5 min. The mass spectrometer was operated in electron impact ionization mode at 70 eV. The ion source and quadrupole temperatures were set to 230 °C and 150 °C, respectively. Mass spectra were acquired across an m/z range of 30–600. Tentative compound identification was performed by comparing the acquired mass spectra with those in the NIST17 library. The relative abundance of each tentatively identified component was estimated using the peak-area normalization method.

UPLC–QTOF–MS Analysis of Non-Volatile Compounds

Non-volatile compounds in the ethanol extract of *A. quinata* summer roots were profiled by UPLC–QTOF–MS using a 1290 UPLC coupled with a 6550 QTOF mass spectrometer (Agilent Technologies, USA). Chromatographic separation was performed on an ACQUITY UPLC BEH C18 column (2.1 $\times$ 100 mm, 1.7 μ m) maintained at 40 °C. The injection volume was 5 μ L, and the flow rate was 0.30 mL/min. The mobile phase consisted of solvent A (0.1% formic acid in water) and solvent B (acetonitrile). The

gradient elution program was as follows: 0–2 min, 5% B; 2–20 min, 5–100% B; 20–25 min, 100% B; 25–26 min, 100–5% B; 26–27 min, 5% B, followed by re-equilibration.

Mass spectrometric analysis was performed using an electrospray ionization source in positive-ion mode. The drying gas flow rate and temperature were set at 15 L/min and 150 °C, respectively. The nebulizer pressure was set at 40 psi, and the sheath gas temperature and flow rate were 350 °C and 12 L/min, respectively. The capillary voltage was set to 4 kV. Data were acquired over a mass range of m/z 50–1200. Compounds were tentatively annotated by matching mass spectral data and retention information to entries in the Waters UNIFI Traditional Chinese Medicine database, and only putative annotations with a database-matching score > 90 were accepted. Relative abundances were estimated using peak-area normalization.

Network Pharmacology Analysis

Compounds detected in GC–MS and UPLC–QTOF–MS profiles and previously reported to possess anti-cancer activity were selected for network pharmacology analysis. The candidate compounds were screened based on their detection in the analytical profiles and reported bioactive relevance. The corresponding chemical structures were retrieved in SMILES format and submitted to the Swiss TargetPrediction database for prediction of potential protein targets, with *Homo sapiens* selected as the species. Duplicate targets were removed to obtain a non-redundant target list.

Gastric cancer-associated genes were retrieved from the Gene Cards and OMIM databases using the keyword “gastric cancer” as the search term, and *Homo sapiens* was selected as the species setting throughout the analysis. The overlapping targets between predicted compound targets and disease-associated genes were considered potential candidate targets for further analysis.

An interaction network linking active compounds, targets, and gastric cancer was built and visualized in Cytoscape. The overlapping targets were then imported into the STRING database to construct a protein–protein interaction (PPI) network, using a confidence score threshold of 0.4 and removing isolated nodes. Subsequently, key targets were identified *via* topological analysis in Cytoscape, and nodes with degree values greater than 70 were defined as hub nodes.

The overlapping targets were further analyzed for Gene Ontology (GO) functional enrichment and Kyoto Encyclopedia of Genes and Genomes (KEGG) pathway enrichment using the Metascape database. The enrichment analysis was performed with a significance threshold of $P < 0.01$, corrected for false discovery rate. Terms and pathways with adjusted P-values below 0.01 were considered significant, and enrichment findings were visualized to identify the major biological processes and signaling pathways involved.

Molecular Docking Analysis

Based on the network topology analysis, candidate target proteins

with the highest degree scores were selected for molecular docking. The corresponding protein crystal structures were obtained from the Protein Data Bank (PDB) based on their UniProt IDs. Only protein structures with high resolution and good structural completeness were selected for further analysis. The selected protein structures were retrieved in PDB format.

The chemical structures of the selected compounds were retrieved from the PubChem database in SDF format. Prior to docking, the ligands were subjected to energy minimization to optimize their geometries and were subsequently converted to PDB format.

For protein preparation, water molecules and heteroatoms were removed using PyMOL. The cleaned protein structures were then imported into AutoDockTools 1.5.7, where polar hydrogens were added, and Gasteiger charges were calculated. Ligand files were prepared using the same workflow.

Molecular docking was carried out with AutoDockTools 1.5.7. The docking search space was defined using a grid box covering the entire protein surface to enable broad sampling of potential ligand-binding regions. The grid box dimensions were set to cover the entire protein region, and docking calculations were carried out with 100 runs for each protein–ligand pair. Binding affinity values were interpreted according to previously reported literature thresholds. Multiple docking poses were generated and the pose with the lowest binding energy, along with other representative conformations, was selected for detailed interaction analysis. The docking results were visualized in PyMOL to illustrate ligand-binding modes and potential protein–ligand interactions.

Statistical Analysis

All data were first compiled in Excel 2016 and then analyzed in SPSS 27.0. For continuous variables, normally distributed data were reported as mean $\pm$ standard deviation (SD), whereas non-normally distributed data were summarized as median with interquartile range [M (P_{25} , P_{75})]. Categorical data were presented as counts and proportions [n (%)]. When comparing two independent groups, an independent-samples t-test was used for normally distributed data with equal variances; otherwise, the Mann–Whitney U test was used. For comparisons involving three or more independent groups, the Kruskal–Wallis H test was applied. Statistical effects were interpreted according to the corresponding P values. Statistical significance was defined as a two-tailed adjusted P value < 0.05 when multiple comparisons were performed. GraphPad Prism was used to visualize the inhibitory effects of different extracts on cell viability.

Results and Discussion

Cytotoxicity in MKN74 Cells and FTIR Analysis

The cytotoxicity of six *A. quinata* ethanol extracts on MKN74 human gastric cancer cells was assessed using the MTT assay, with blank controls and three technical triplicate. As shown in Table 1, all extracts exhibited measurable inhibition of MKN74 cell viability, with inhibition rates ranging from $62.68 \pm 2.56\%$ to $76.46 \pm 0.47\%$. The summer root extract showed the highest inhibitory

effect on MKN74 cell viability ($76.46 \pm 0.47\%$), followed by the autumn root extract ($74.14 \pm 3.96\%$) and the autumn stem extract ($71.13 \pm 1.07\%$), whereas the autumn leaf extract showed the lowest inhibitory effect on cell viability ($62.68 \pm 2.56\%$).

To assess the effects of seasonal variation and plant organ on the inhibitory effects on MKN74 cell viability, statistical analyses were performed. As summarized in Table 2, no significant difference was observed between summer and autumn extracts overall (summer: $69.51 \pm 6.18\%$ vs. autumn: $69.32 \pm 5.69\%$; $t = 0.067$, $P = 0.947$). In contrast, MTT-derived inhibition rates differed significantly among plant organs (Table 3; $H = 12.772$, $P = 0.002$). Root extracts showed the highest median inhibition rate of MKN74 cell viability [76.24 (73.84 , 77.04%)], followed by stem extracts [70.70 (66.89 , 71.73%)], whereas leaf extracts exhibited the lowest inhibition rate [63.46 (60.03 , 66.05%)]. These results suggest that the plant organ had a greater influence than season on the observed inhibition of MKN74 cell viability under the tested conditions.

Pairwise comparisons shown in Figure 1A further confirmed these differences in MTT-derived inhibition rates. In summer, root extracts showed significantly higher MTT-derived inhibition rates than leaf extracts ($P = 0.0007$) and stem extracts ($P = 0.0157$). In autumn, root extracts also exhibited significantly higher MTT-derived inhibition rates than leaf extracts ($P = 0.0015$), and stem extracts showed significantly higher inhibition rates than leaf extracts ($P = 0.0137$). No significant difference in MTT-derived inhibition rates was observed between summer- and autumn-collected extracts within the same plant organ. Collectively, these findings demonstrate that root-derived extracts exert the strongest inhibitory effects on MKN74 cell viability.

FTIR spectroscopy was used to provide a preliminary characterization of the functional groups in the ethanol extracts of *A. quinata*. The FTIR spectra of the six extracts are presented in Figure 1B. The spectra exhibited several common absorption bands at approximately 3380, 2970, 1650, 1450, 1050, and 880 cm^{-1} . The broad band in the 3200–3600 cm^{-1} was attributed to O–H stretching vibrations, indicating the presence of hydroxyl-containing groups [14,15]. Peaks in the 2800–3000 cm^{-1} region were attributed to C–H stretching, suggesting the presence of aliphatic components [16,17]. The band near 1650 cm^{-1} was attributed to C=O stretching vibrations, suggesting the possible presence of carbonyl-containing functional groups [18,19]. The peaks around 1400–1450 cm^{-1} were attributed to C–H bending vibrations [20]. The absorption in the range of 1020–1100 cm^{-1} corresponded to C–O stretching vibrations, suggesting oxygenated functional groups [21]. Additionally, the bands observed between 800–900 cm^{-1} were associated with out-of-plane C–H bending vibrations, which may be associated with aromatic or substituted hydrocarbon structures [22,23].

Overall, the FTIR results suggest that the ethanol extracts of *A. quinata* contain diverse types of functional groups, but FTIR alone does not allow definitive identification of specific compound classes or confirmation of compound classes. Therefore, these tentative

assignments should be interpreted together with complementary analytical data, such as GC–MS and UPLC–QTOF–MS, to obtain more comprehensive information on the chemical composition of the extracts.

Table 1: Cytotoxic activity of ethanol extracts from different organs of *A. quinata* collected in summer and autumn against MKN74 cells.

Season	Organ	n	Cytotoxicity (%)
Summer	Leaf	3	63.87±4.47
Summer	Stem	3	68.19±3.10
Summer	Root	3	76.46±0.47
Autumn	Leaf	3	62.68±2.56
Autumn	Stem	3	71.13±1.07
Autumn	Root	3	74.14±3.96

Table 2: Comparison of cytotoxicity between summer and autumn extracts.

Season	n	Cytotoxicity (%)	t	P
Summer	9	69.51±6.18	0.067	0.947
Autumn	9	69.32±5.69		

Table 3: Comparison of cytotoxicity among ethanol extracts from different organs of *A. quinata* against MKN74 cells.

Organ	n	Cytotoxicity (%)	H	P
Leaf	6	63.46 (60.03,66.05)b	12.772	0.002
Stem	6	70.70 (66.89, 71.73) b		
Root	6	76.24 (73.84,77.04)a		

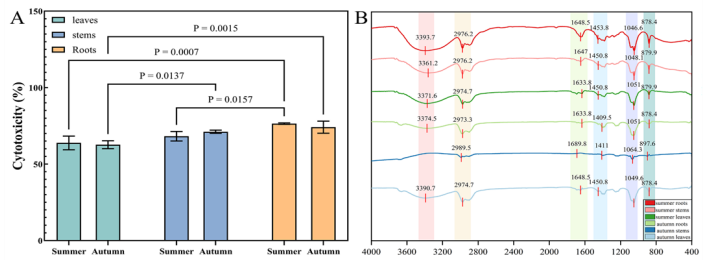


Figure 1: (A) Comparison of the inhibitory effects of six *A. quinata* extracts on MKN74 human gastric cancer cells. (B) FTIR spectra of ethanol extracts obtained from different organs of *A. quinata*.

Construction of an Active Compound Library GC–MS Chemical Profiling of Extracts

To investigate the chemical complexity of the ethanol extracts from *A. quinata*, GC–MS analysis was performed following preliminary FTIR characterization to investigate major constituents, compositional variation, and potential functional significance. GC–MS analysis was conducted on six ethanol extracts, and the total ion chromatograms are shown in Figure S1. A total of 71, 79, 67, 63, 55, and 46 peaks were detected in summer leaves, summer stems, summer roots, autumn leaves, autumn stems, and autumn roots, respectively, from which 69, 38, 65, 59, 28, and 20 chemical constituents were tentatively annotated. Compound annotation was performed by comparison with the NIST mass spectral library, and only matches with a similarity index >80% were accepted. The detected compounds were not confirmed with authentic standards. These results demonstrate substantial variation in both

chemical diversity and composition across different plant organs and seasons, highlighting the complexity of secondary metabolite distribution in *A. quinata*.

Analysis of compound categories in the six ethanol extracts (Figure 2B) further revealed distinct distribution patterns across plant organs and seasons. Compound classes were assigned according to the chemical functional groups and structural features inferred by the GC–MS library annotations. Ester compounds were widely present across all plant parts, although their relative contents varied significantly, with the highest levels observed in autumn leaves (44.62%) and relatively high levels also in summer leaves (34.00%), suggesting that leaves represent an important source of detected ester-type compounds. In stem tissues, ester content exhibited clear seasonal variation, with higher levels in summer (34.10%) than in autumn (17.30%), indicating compositional differences between growth periods. In contrast, alcohol and phenolic compounds showed their highest relative abundance in autumn roots (27.68%), exceeding those in summer roots (9.07%).

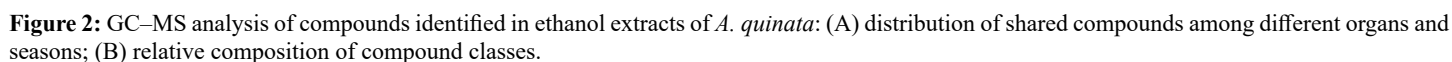
Hydrocarbon compounds showed pronounced organ specificity, being predominantly detected in leaf tissues—particularly summer leaves (24.16%)—while remaining minimal or absent in stems. Ether compounds were relatively evenly distributed but were more abundant in autumn stems (20.58%). Aldehyde and ketone compounds were generally present at low levels and mainly detected in leaves. Notably, the “other compounds” category was particularly abundant in stem tissues, especially in autumn stems (48.26%), suggesting the presence of structurally diverse or less-characterized metabolites that may warrant further investigation.

Further intersection analysis of specific compounds (Figure 2A) provided deeper insight into the chemical composition of different plant parts. Leaf tissues showed higher relative abundance of terpenoid-related compounds such as squalene, which has been reported in the literature to exhibit antioxidant activity and potential anti-cancer effects [24]. Stem tissues showed higher relative abundance of fatty acid-related compounds such as palmitic acid and linoleic acid, which are commonly associated with plant lipid structures [25]. Root tissues showed enrichment of sterol compounds such as β -sitosterol and stigmasterol, which have been reported to possess diverse biological activities [26,27]. In addition to organ-specific variation, seasonal effects were also evident, with summer samples generally exhibiting higher chemical diversity, particularly in terpenes and esters, whereas autumn samples contained several unique or enriched compounds, such as diisooctyl phthalate in root tissues. These observations collectively reflect the compositional plasticity of *A. quinata* and provide a more refined understanding of its chemical diversity without implying direct biological activity of the extracts.

Building on these compositional patterns, selected compounds tentatively annotated by GC–MS were further analyzed for their reported biological activities (Table S1). It should be emphasized that these activities are derived from previous literature. Compounds such as dibutyl phthalate and ethyl palmitate were commonly

Overall, the GC–MS analysis demonstrates that ethanol extracts of *A. quinata* contain a wide range of chemically diverse compounds,

Among the detected compounds, 22 components exhibited relative abundances exceeding 1%, including dimethylcamphorate (7.28%), phthalic anhydride (7.22%), (2,3)trans-N-(p-hydroxyphenethyl) ferulamide (6.34%), anemoside A3 (5.33%), tetrahydrobungeanol (4.53%), (24Z)-27-hydroxy-3-oxo-7,24-tirucalladien-21-al (4.46%), 10-O-acetylmonotropein (4.02%), 4-hydroxymethyl-2-furaldehyde (3.36%), dihydrocucurbitacin F (3.28%), 5-O-p-methoxy cinnamoyl scandoside methyl ester (2.91%), 2''-O-beta-D-xylopyranosylsaikosaponin b2 (2.45%), dibutyl phthalate (2.44%), desmodilactone (2.36%), 21-hydroxy-30-norhopan-22-one (2.04%), tribulusamide A (1.99%), neohancoside B (1.88%), 6xi-methoxypiperidin-2-one (1.54%), androst-4,6-diene-3,17-dione (1.43%), acetoin (1.31%), candidoside A (1.25%), wattoside



C (1.15%), and amarasterone B (1.01%).

The major chemical constituents of the summer root extract were categorized into glycosides (23.60%), organic acid esters and lactones (22.07%), terpenoids and steroids (20.12%), and alkaloids (15.37%). Together, these four classes accounted for more than 80% of the total relative abundance, indicating that they form the dominant chemical framework of the extract (Figure 3B). This distribution reflects a structurally diverse metabolite profile in which multiple secondary metabolite classes coexist, potentially influencing both physicochemical properties and biological activities. Notably, while glycosides are the most abundant fraction, terpenoids, steroids, and alkaloids are widely reported to exhibit various bioactivities. However, relative abundance does not necessarily correlate with biological effect, and further investigation is needed to determine the contributions of individual compound classes to the observed cytotoxic activity.

Glycosides are the dominant tentatively assigned class of constituents in the extract and have been shown to exert a range of biological activities, including anti-bacterial and anti-inflammatory effects. For example, Anemoside A3 has been reported to possess potential anti-breast cancer activity through dual regulation of tumor-associated macrophage polarization. Specifically, it promotes M1 polarization via activation of the TLR4/NF- κ B pathway and inhibits M2 polarization by suppressing STAT3 phosphorylation, which may contribute to remodeling of the tumor immune microenvironment and limitation of tumor growth, angiogenesis, and metastatic progression [38,39]. In addition, although present at lower abundance, Cimicide E has been reported to induce concentration-dependent cell cycle arrest and apoptosis in human gastric cancer AGS cells, with an IC_{50} of 14.58 μ M at 24 h [40].

The relatively high proportion of organic acids and esters may influence the physicochemical properties of the extract and potentially contribute to characteristics such as stability and bioavailability. For instance, dibutyl phthalate, isolated from marine sources, has demonstrated anti-bacterial activity against *Bacillus subtilis*, *Bacillus cereus*, and *Staphylococcus aureus*, as well as antifungal activity against *Candida albicans* [41]. Furthermore, the presence of terpenoids, steroids, and alkaloids suggests potential pharmacological relevance. For example, dihydrocucurbitacin F has been found to inhibit the growth of prostate cancer cell lines (DU145, PC3, and LNCaP) with IC_{50} values of 15, 7, and 5 μ M for these cell lines, respectively. Its mechanism involves cofilin dephosphorylation, disruption of the actin cytoskeleton, G₂/M phase arrest, and induction of apoptosis [42].

In contrast, flavonoids and polyphenols (2.42%), alcohols (5.98%), and aldehydes (3.79%) were present at relatively lower levels. However, these compounds may still contribute to antioxidant properties or aroma characteristics. Quinones were detected at very low levels (0.37%) and are therefore unlikely to represent major active constituents. For example, 6-Shogaol is a ginger-

derived bioactive molecule with anti-inflammatory, antioxidant, neuroprotective, and anti-cancer activities. Studies have shown that 6-shogaol, particularly in combination with hyperthermia, enhances ROS generation and modulates heat shock proteins, thereby inducing apoptosis and cell-cycle arrest in cancer cells with minimal impact on normal cells [43,44].

Overall, the UPLC–QTOF–MS results present a broad profile of the chemical composition of the summer root extract of *A. quinata*, which is dominated by glycosides, organic acids, terpenoids/steroids, and alkaloids. These compositional features provide important insights into the chemical composition of the extract and may help guide future investigations into its potential pharmaceutical or functional applications. However, it should be emphasized that compound identification is tentative and that additional experimental validation is needed to verify the biological roles of these constituents. In particular, bioactivity-guided fractionation and targeted validation will be required to pinpoint the major compounds contributing to the observed inhibitory effects on cell viability. Additionally, integrating chemical profiling with mechanistic assays will be essential to define the relationship between chemical composition and biological function more clearly.

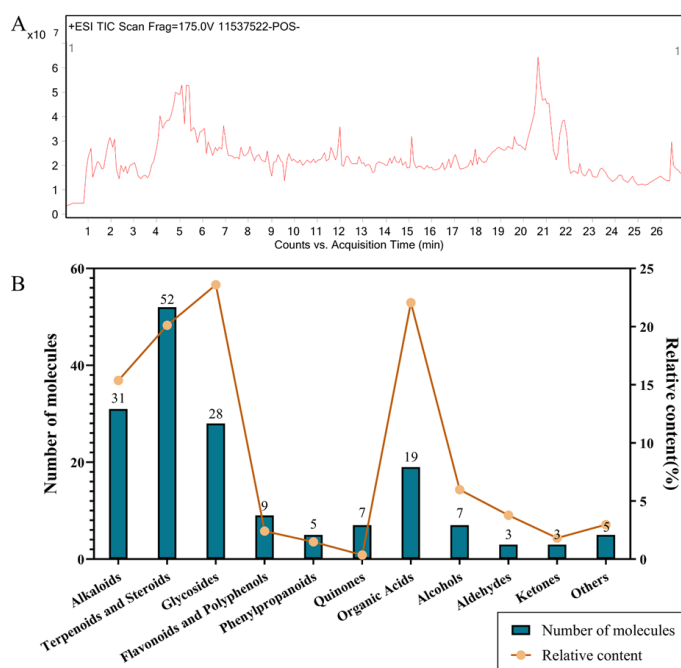


Figure 3: UPLC–QTOF–MS total ion chromatogram of the ethanol extract from *A. quinata* roots collected in summer.

Study on the Anti-Cancer Mechanism

Core Target Identification and Enrichment Analysis

Through screening of compounds with reported anti-cancer activity in the summer root ethanol extract, components with relative abundances exceeding 1% and reported anti-cancer relevance were selected, including propanoic acid, anhydride; 13-docosenamide (Z)-; (R)-(-)-14-methyl-8-hexadecyn-1-ol; hexadecanoic acid,

ethyl ester; dihydrocucurbitacin F; and anemoside A3. A total of 272 potential targets were predicted, among which 206 overlapped with gastric cancer-associated genes.

Figure 4A illustrates the component–target–disease interaction network, in which green squares represent targets shared with gastric cancer and yellow circles represent non-overlapping targets. The selected compounds shared 12, 8, 66, 37, 79, and 57 targets with gastric cancer, respectively. Figure 4B displays the PPI network of common targets, containing 206 protein nodes and 1,750 interaction edges, with an average node degree of 17 and a clustering coefficient of 0.525. Figure 4C presents the PPI network for MKN74-related targets, comprising 14 protein nodes linked by 73 interaction edges, with an average node degree of 10.4 and a clustering coefficient of 0.876. Based on topological analysis of the PPI networks, six key targets with the highest degree values were identified: AKT1, STAT3, CASP3, BCL2, PTGS2, and MTOR. These targets have been reported to be involved in apoptosis regulation, cell survival, and stress-response signaling pathways.

In the GO enrichment analysis of overlapping targets, 1,715 biological process (BP), 215 molecular function (MF), and 113 cellular component (CC) terms were significantly enriched ($P < 0.01$). The top 10 terms for each category are presented in Figures 5A–C. The BP analysis indicated enrichment in cellular stress and adaptive response processes, including responses to nitrogen compounds (GO:1901699), alcohol (GO:0097305), lipids (GO:0071396), hormonal stimuli (GO:0032870), and xenobiotic stimuli (GO:0009410). In addition, inflammatory response (GO:0006954) was prominently enriched, suggesting potential involvement of inflammation-associated biological processes in the predicted target network rather than confirming a direct anti-inflammatory effect.

The CC analysis showed that enriched terms were associated with neuron-related subcellular components, including postsynapse, dendrite, dendritic tree, and glutamatergic synapse, which are key sites of signal transmission. The MF analysis revealed enrichment in functions related to signal recognition and transduction. These included G protein-coupled amine receptor activity (GO:0008227), steroid binding (GO:0005496), hormone binding (GO:0042562), nuclear receptor activity (GO:0004879), and kinase-related activities such as protein kinase activity (GO:0004672) and serine/threonine kinase activity (GO:0004674). These findings suggest that the active compounds may interact with receptor systems and kinase-mediated signaling pathways.

KEGG analysis showed 192 significantly enriched pathways ($P < 0.01$), with the top 20 displayed in Figure 5D. Among them, the ‘Pathways in cancer’ pathway showed prominent enrichment, which may reflect the multi-target profile and pathway diversity of the predicted target set. Pathways related to neuroactive ligand–receptor signaling and serotonergic synaptic transmission were also enriched, consistent with the GO enrichment results, and may represent shared ligand–receptor signaling modules relevant to cancer-related regulatory processes. In addition, enrichment of the

“EGFR tyrosine kinase inhibitor resistance” pathway suggests that the predicted targets may be associated with signaling networks involved in therapeutic resistance. Notably, the PI3K–Akt pathway was also enriched and is directly linked to the core target AKT1 [45], whereas STAT3 is functionally related to cytokine-mediated signaling, inflammatory regulation, and growth factor-associated pathways [46]. Overall, these enrichment results suggest that the overlapping targets are associated with multiple signaling pathways related to stress response, inflammation, metabolism, therapeutic resistance, and cell death. These findings provide a basis for further mechanistic investigation and require experimental validation.

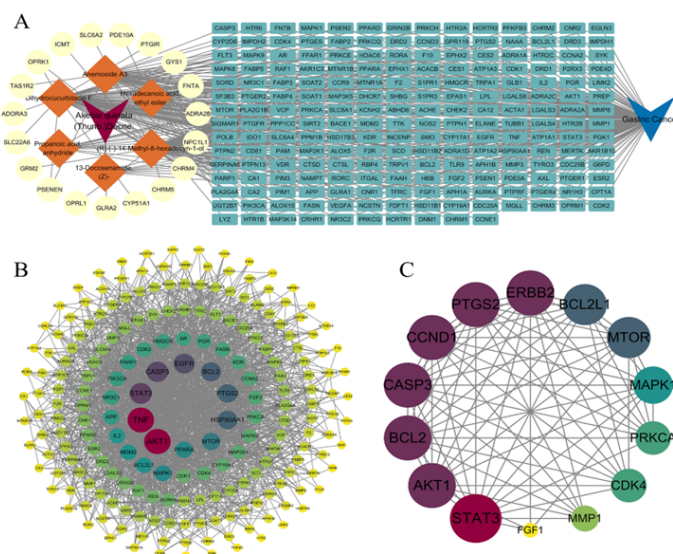


Figure 4: (A) Component–target–disease interaction network of the summer root ethanol extract. (B) PPI network of predicted targets associated with gastric cancer. (C) PPI network of predicted targets associated with the MKN74 cell line. Node size is proportional to degree value.

Molecular Docking Analysis of Candidate Components with Core Targets

Molecular docking with AutoDock was carried out to examine the interactions between the two candidate hub proteins, AKT1 and STAT3, which exhibited the highest degree values in the PPI network, and the selected active compounds, dihydrocucurbitacin F and anemoside A3. Figure 6 presents the docking results between the candidate compounds and the target proteins. Figures 6A–B illustrate the binding mode of dihydrocucurbitacin F with AKT1. The docking results indicate that dihydrocucurbitacin F forms three hydrogen bonds with the residues TYR-350, ALA-409, and ILE-411, and the binding free energy was calculated to be -5.06 kcal/mol. Figures 6C–D show the interaction between Anemoside A3 and STAT3, with the calculated binding free energy being -5.44 kcal/mol. In this complex, anemoside A3 forms hydrogen bonds with HIS-281, LYS-84, and GLN-100.

Predicted binding free energy is commonly used as an indicator of ligand–protein binding affinity, and values below -5.0 kcal/mol are generally interpreted as indicating favorable binding between

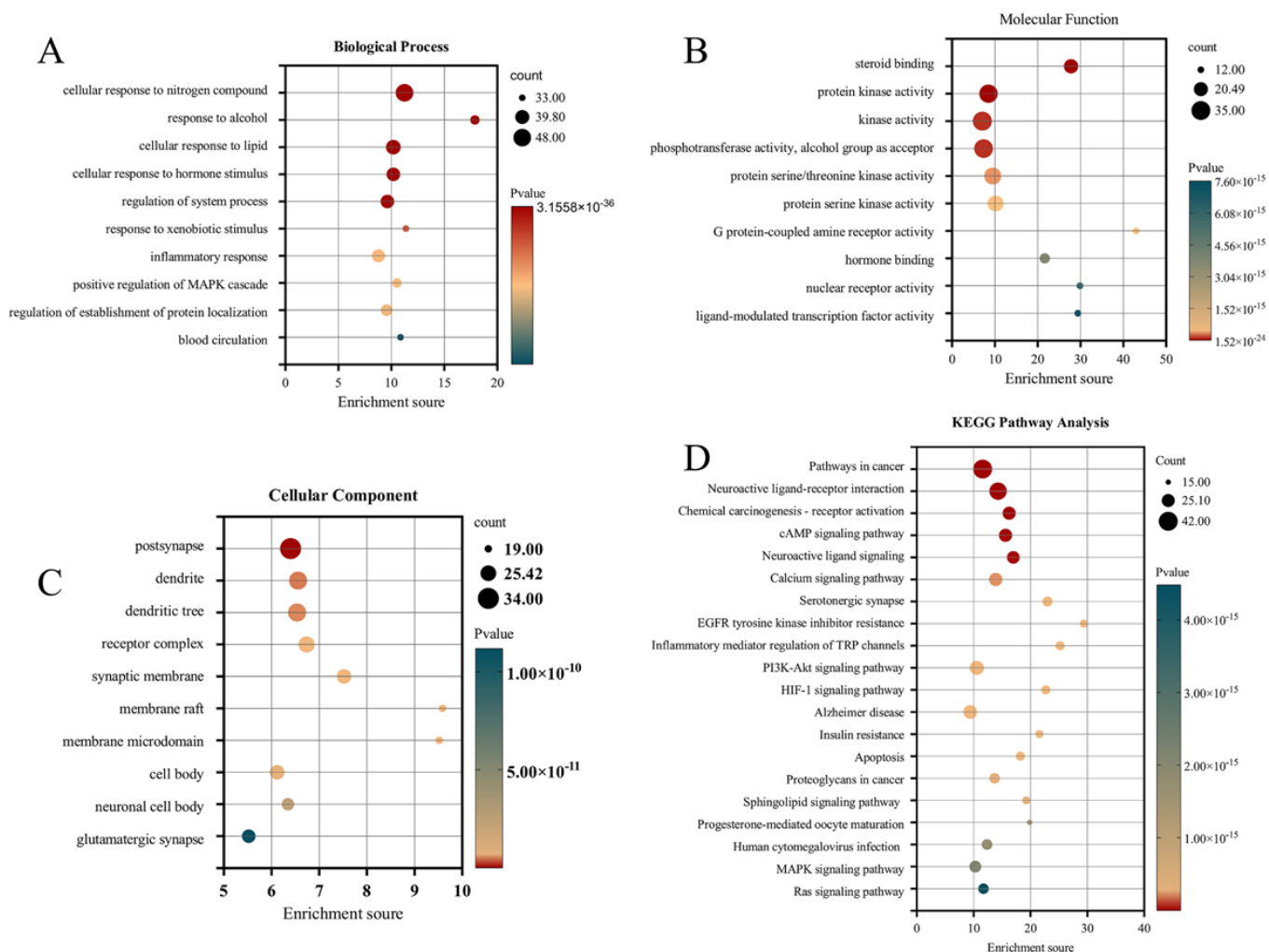


Figure 5: Bubble plots of GO enrichment and KEGG pathway analyses of predicted targets: (A) BP; (B) CC; (C) MF; (D) KEGG pathway analysis.

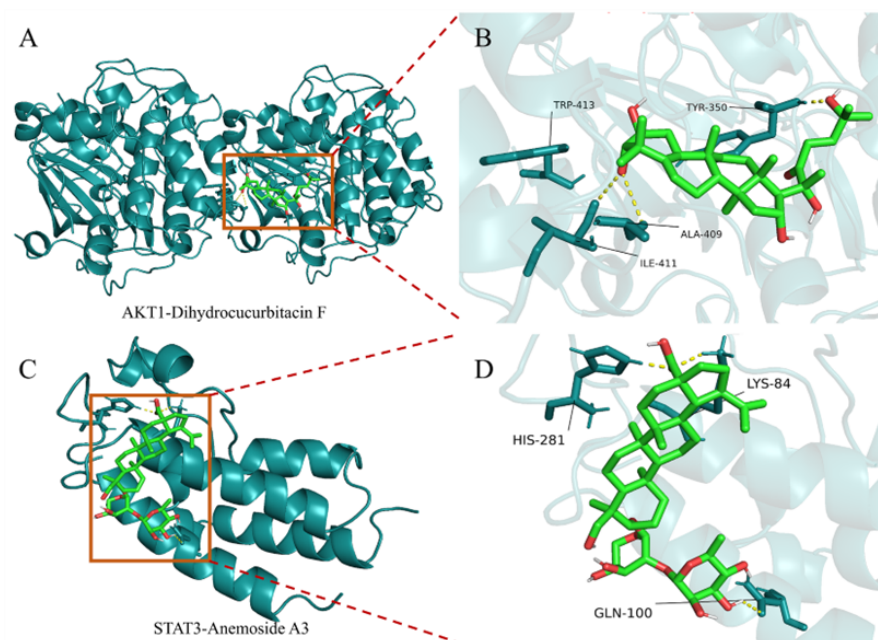


Figure 6: Molecular docking interactions between selected active compounds and key target proteins.

ligands and proteins [47]. The docking results, therefore, suggest that both compounds may interact with the selected candidate targets, with anemoside A3 showing a slightly stronger predicted binding affinity toward STAT3 than dihydrocucurbitacin F toward AKT1. It should be noted that molecular docking provides a theoretical prediction of ligand–protein interactions based on static structures and simplified scoring functions. Therefore, while the observed interactions, including hydrogen bonding and possible hydrophobic contacts, indicate potential binding modes, they do not confirm biological activity or inhibitory effects. Further experimental validation, such as enzyme inhibition assays or cellular pathway analysis, is required to verify these interactions.

Conclusions

This study integrated cell-based assays, chemical profiling, and computational analysis to evaluate the inhibitory effects of *A. quinata* extracts on gastric cancer MKN74 cells and to explore their potential mechanisms of action. The MTT assay showed that ethanol extracts from different plant parts exhibited inhibitory effects on MKN74 cell viability, with the summer root extract demonstrating the strongest inhibitory effect (76.46% inhibition rate). The results revealed clear differences among plant organs (roots > stems ≈ leaves).

Comprehensive chemical characterization using GC–MS and UPLC–QTOF–MS detected a wide variety of compounds in the summer root extract, including several compounds with previously reported anti-cancer activities, such as dihydrocucurbitacin F and anemoside A3. Network pharmacology analysis suggested that multiple targets and pathways could contribute to the effects observed, with AKT1 and STAT3 identified as major nodes in the predicted interaction network. KEGG enrichment analysis further suggested the involvement of cancer-related and signal transduction pathways.

Molecular docking results suggested that the selected compounds may exhibit favorable predicted interactions with AKT1 and STAT3 with favorable predicted binding energies (−5.06 and −5.44 kcal/mol, respectively), supporting their potential association with these targets. However, these interactions are based on computational predictions and do not confirm direct biological activity or mechanism. Overall, these results suggest that the summer root extract shows *in vitro* anti-proliferative activity against gastric cancer cells and contains multiple bioactive compounds that may contribute to its effects through multi-target interactions. Nevertheless, further experimental validation, including bioactivity-guided isolation, animal experiments, and mechanistic investigations, is necessary to clarify the roles of specific compounds and pathways.

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Appendix

Table S1: Major chemical constituents identified in ethanol extracts of *A. quinata* by GC–MS analysis.

Active component	Relative content (%)					
	Summer leaves	Summer Stems	Summer Roots	Autumn leaves	Autumn Stems	Autumn Roots
Phytol	5.47%	-	-	7.06%	-	-
2-Amino-1,3-propanediol	-	9.35%	-	1.01%	-	-
Neophytadiene	15.21%		0.74%	6.25%	-	-
Squalene	4.90%	-	-	7.87%	-	-
Propanoic acid, anhydride	0.49%	-	1.99%	-	-	-
2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-, (R)-	0.87%	-	-	-	-	-
Carvone	0.90%	-	-	-	-	-
Oleic Acid	-	0.75%				
2-Imidazolidinethione	-	-	2.49%	-	-	-
3,4-Dimethoxycinnamic acid	-	-	0.16%	-	-	-
Dibutyl phthalate	2.77%	5.83%	3.54%	4.28%	-	-
Hexa-1,3,5-triyne	-	-	3.87%	0.91%	1.62%	2.04%
β-D-Glucopyranose, 1-thio-, 1-[N-hydroxy-5-(methylthio)pentanimidate	-	1.97%	-	-	1.76%	0.77%
2,2-Dimethyl-6-methylene-1-[3,5-dihydroxy-1-pentenyl]cyclohexan-1-perhydrol	-	2.26%	-	-	6.89%	2.93%
2,4,5-Trihydroxypyrimidine	-	-	1.68%	-	-	1.56%
Melezitose	-	2.80%	-	-	1.19%	-
4-Methoxy-6-methyl-6,7-dihydro-4H-furo[3,2-c]pyran	0.20%	-	-	-	-	0.35%
9,12-Octadecadienoyl chloride, (Z,Z)-	-	-	-	-	1.34%	2.73%
Benzenemethanol, 4-hydroxy-.alpha.-[1-(methylamino)ethyl]-, (R*,S*)-	-	-	-	-	0.97%	0.56%
13-Docosenamide, (Z)-	-	-	11.48%	-	-	-
(R)-(-)-14-Methyl-8-hexadecyn-1-ol	-	-	2.97%	-	-	-
Hexadecanoic acid, ethyl ester	2.89%	2.73%	3.18%	4.67%	-	-

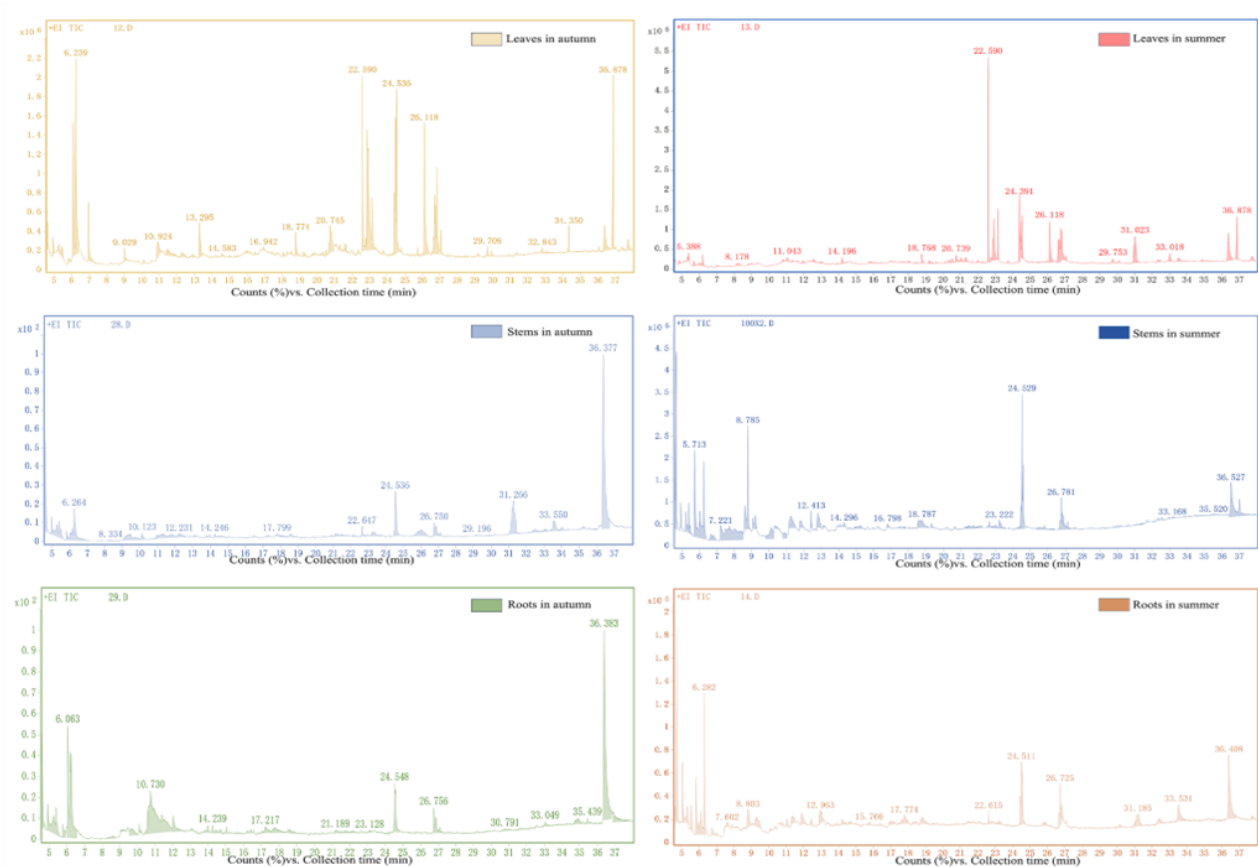


Figure S1: GC–MS total ion chromatograms of ethanol extracts derived from different organs of *A. quinata* collected in summer and autumn

Food Sci Nutr Res, 2026 Volume 9 | Issue 3 | 13 of 17

Table S2: Compounds tentatively identified in the summer root ethanol extract of *A. quinata* by UPLC–QTOF–MS analysis.

Peak No.	Retention time (min)	Chemical compound	Formula
1	0.437	1-Ethyl-beta-D-galactoside	C ₈ H ₁₆ O ₆
2	0.913	Macranthoin F	C ₂₆ H ₂₆ O ₁₂
3	1.009	Trigonelline	C ₇ H ₇ NO ₂
4	1.104	Pyroglutamic acid	C ₅ H ₇ NO ₃
5	1.295	Gentianol	C ₁₀ H ₁₁ NO ₃
6	1.295	6-Shogaol	C ₁₇ H ₂₄ O ₃
7	1.39	Nicotinamide	C ₆ H ₆ N ₂ O
8	1.39	5alpha-O-(3'-Amino-3'-phenylpropionyl)nicotaxine	C ₃₉ H ₄₆ N ₂ O ₁₀
9	1.771	Neohancoside B	C ₂₁ H ₃₆ O ₁₁
10	1.771	(+)-gamma-Hydroxy-L-homoarginine	C ₇ H ₁₆ N ₄ O ₃
11	1.962	Phlorin	C ₁₂ H ₁₆ O ₈
12	2.153	Dimethyl camphorate	C ₁₂ H ₂₀ O ₄
13	2.153	Arecaidine	C ₇ H ₁₁ NO ₂
14	2.344	Histamine	C ₅ H ₉ N ₃
15	2.534	Sesamoside	C ₁₇ H ₂₄ O ₁₂
16	2.534	Peristrophamide	C ₉ H ₉ NO ₄
17	2.534	Levulinic acid	C ₅ H ₈ O ₃
18	2.725	6xi-Methoxypiperidin-2-one	C ₆ H ₁₁ NO ₂
19	2.82	Gentianadine	C ₈ H ₇ NO ₂
20	3.011	gamma-Guanidinobutyric acid	C ₅ H ₁₁ N ₃ O ₂
21	3.106	Coronarian	C ₁₂ H ₁₈ N ₂ O ₁₂
22	3.106	2,3,4-Trihydroxy-benzenepropanoic acid	C ₉ H ₁₀ O ₅
23	3.106	Chelerythrine methanolate	C ₂₂ H ₂₁ NO ₅
24	3.202	Genipinic acid	C ₁₁ H ₁₄ O ₆
25	3.297	Betaine	C ₅ H ₁₁ NO ₂
26	3.583	Daturametelin E	C ₂₉ H ₄₂ O ₈ S
27	3.678	6-Methoxygeniposidic acid	C ₁₇ H ₂₄ O ₁₁
28	4.346	Acteoside	C ₂₉ H ₃₆ O ₁₅
29	4.536	(-)-Istanbulin A	C ₁₅ H ₂₀ O ₄
30	4.822	4-Hydroxymethyl-2-furaldehyde	C ₆ H ₆ O ₃
31	4.918	Aucubigenin	C ₉ H ₁₂ O ₄
32	5.013	Retusine	C ₁₆ H ₂₅ NO ₅
33	5.108	10-O-Acetylmonotropein	C ₁₈ H ₂₄ O ₁₂
34	5.204	Desmodilactone	C ₈ H ₁₃ NO ₃
35	5.204	5-Hydroxy-2-pyridinemethanol	C ₆ H ₇ NO ₂
36	5.204	3,4-Dioxymethylene-5-methoxy-1-(1-oxopropyl)benzene	C ₁₁ H ₁₂ O ₄
37	5.299	D-8Acetoxycarvotanacetone	C ₁₂ H ₁₈ O ₃
38	5.68	Dehydromorroniaglycone	C ₁₁ H ₁₄ O ₅
39	5.776	2-Methoxymollugin	C ₁₈ H ₂₀ O ₅
40	5.776	Indole-3-acetonitrile-6-O-beta-D-glucopyranoside	C ₁₆ H ₁₈ N ₂ O ₆
41	6.062	Galocatechin-(4alpha-->8)epicatechin	C ₃₀ H ₂₆ O ₃
42	6.157	Artemisinin I	C ₁₃ H ₁₈ O ₂
43	6.252	Adouetine Y	C ₃₄ H ₄₀ N ₄ O ₄
44	6.443	Artemisin	C ₁₅ H ₁₈ O ₄
45	6.538	Ecgonine	C ₉ H ₁₅ NO ₃
46	6.538	trans-3-Ethylidene-2-pyrrolidone	C ₆ H ₉ NO
47	6.538	Nicotinic acid	C ₆ H ₅ NO ₂
48	6.634	Cascaroside A	C ₂₇ H ₃₂ O ₁₄

49	6.729	alpha,alpha-Dimethylbenzene methanol	C ₉ H ₁₂ O
50	6.92	1-Phenyl-1,3-butanedion	C ₁₀ H ₁₀ O ₂
51	6.92	Isosakuranetin-7-rutinoside	C ₂₈ H ₃₄ O ₁₄
52	6.92	2-Methyl-5-acetonyl-7-hydroxychromone	C ₁₃ H ₁₂ O ₄
53	7.015	5-O-p-Methoxy cinnamoyl scandoside methyl ester	C ₂₇ H ₃₂ O ₁₃
54	7.301	Plaunol E	C ₂₂ H ₂₄ O ₈
55	7.682	Abrusin-2"-o-apioside	C ₂₈ H ₃₄ O ₁₅
56	7.682	8-O-Angeloyl-9-O-acetylanthemolide B	C ₂₂ H ₂₈ O ₉
57	7.778	2,4-Dimethoxybenzaldehyde	C ₉ H ₁₀ O ₃
58	8.064	(2,3)trans-N-(p-Hydroxyphenethyl)ferulamide	C ₁₈ H ₁₉ NO ₄
59	8.064	3,3'-Diacetyl-4,4'-dimethoxy-2,2',6,6'-tetrahydroxy diphenyl methane	C ₁₉ H ₂₀ O ₈
60	8.159	Terrestriamide	C ₁₈ H ₁₇ NO ₅
61	8.159	11-O-Acetyl haemanthamine	C ₁₉ H ₂₁ NO ₅
62	8.159	Bujeine	C ₂₀ H ₂₃ NO ₆
63	8.159	(Z)-4,5-Dihydro-6,7-cis-dihydroxy-3-butyldiene phthalide	C ₁₂ H ₁₆ O ₄
64	8.35	Aschantin	C ₂₂ H ₂₄ O ₇
65	8.445	3,24-Dioxo-fridelan-29-oic acid	C ₃₀ H ₄₆ O ₄
66	8.445	12-Tetradecanoylphorbol 13-acetate	C ₃₆ H ₅₆ O ₈
67	8.54	(-)-5'-Methoxyisolariciresinol-2a-O-beta-D-xylopyranoside	C ₂₆ H ₃₄ O ₁₁
68	8.636	Dianoside G	C ₄₈ H ₇₆ O ₂₀
69	8.636	3-O-[beta-D-Glucopyranosyl-(1-->3)-beta-D-glucopyranosyl] phytolaccagenic acid 28-O-beta-D-glucopyra	C ₄₉ H ₇₈ O ₂₁
70	8.731	Spinoside A	C ₃₉ H ₅₆ O ₁₂
71	8.826	Candidoside A	C ₄₁ H ₆₆ O ₁₃
72	8.826	7-(5',6'-Dihydroxy-3',7'-dimethylocta-2',7'-dienyloxy)-coumarin	C ₁₉ H ₂₂ O ₅
73	8.826	Lactucasterol	C ₂₉ H ₄₆ O ₂
74	8.826	Lanyuamide I	C ₁₈ H ₃₁ NO ₂
75	8.826	16beta-Hydroxy-18beta-H-oleanolic acid-28-O-beta-D-glucopyranoside	C ₃₆ H ₅₈ O ₉
76	8.922	Dianoside A	C ₄₂ H ₆₆ O ₁₅
77	9.017	(-)-Isolariciresinol-2a-O-beta-D-xylopyranoside	C ₂₅ H ₃₂ O ₁₀
78	9.017	Doederleinic acid	C ₇ H ₈ O ₄
79	9.208	1'-Acetoxyeugenol acetate	C ₁₄ H ₁₆ O ₅
80	9.303	10-Methoxyheptadeca-1-ene-4,6-diyne-3,9-diol	C ₁₈ H ₂₈ O ₃
81	9.303	Aphylline	C ₁₅ H ₂₄ N ₂ O
82	9.303	6-Methylgingediol	C ₁₈ H ₃₀ O ₄
83	9.303	Cucurbitic acid	C ₁₂ H ₂₀ O ₃
84	9.398	7,9-Deacetylbaccatin IV	C ₂₈ H ₄₀ O ₁₂
85	9.589	Picrasinoside A	C ₂₇ H ₃₈ O ₁₁
86	9.78	Matatabiether	C ₁₁ H ₁₈ O
87	9.875	1-Carboethoxy-beta-carboline	C ₁₄ H ₁₂ N ₂ O ₂
88	9.875	(+)-Medioresinol	C ₂₁ H ₂₄ O ₇
89	9.875	(+)-Epipinoresinol	C ₂₀ H ₂₂ O ₆
90	9.97	Tribulusamide A	C ₃₆ H ₃₆ N ₂ O ₈
91	9.97	Baohuoside III	C ₃₂ H ₃₈ O ₁₄
92	10.066	3-Hydroxy-25-norfriedel-3,1(10)-dien-2-one-30-oic acid	C ₂₉ H ₄₂ O ₄
93	10.256	12-Keto-porrigenin	C ₂₈ H ₄₄ O ₅
94	10.256	10beta-Hydroxy-6beta-isobutyryl furanoeremophilane	C ₁₉ H ₂₈ O ₄
95	10.352	1-Ethyl-4,8-dimethoxy-beta-carboline	C ₁₅ H ₁₆ N ₂ O ₂
96	10.447	10-Hydroxyacetylbaccatin VI	C ₃₇ H ₄₆ O ₁₅
97	11.114	Cimisine E	C ₃₅ H ₅₄ O ₈

98	11.114	2-Acetyl-3-(p-coumaroyl)-meso-tartaric acid	C ₁₅ H ₁₄ O ₉
99	11.305	2''-O-beta-D-Xylopyranosylsaikosaponin b2	C ₄₇ H ₇₆ O ₁₇
100	11.305	1-Methyl-acetylshikonin	C ₁₉ H ₂₀ O ₆
101	11.4	Gitoxin	C ₄₁ H ₆₄ O ₁₄
102	11.4	(12R)-12-Hydroxy cascarill one	C ₂₀ H ₃₀ O ₃
103	11.496	Anemoside A3	C ₄₁ H ₆₆ O ₁₂
104	11.686	alpha-Solamarine	C ₄₅ H ₇₃ NO ₁₆
105	11.782	2-(1-Ethoxy-2-hydroxy)propyl-4-methoxyphenol-2-methyl-butyrate	C ₁₇ H ₂₆ O ₅
106	11.877	Wattoside C	C ₄₆ H ₇₆ O ₂₀
107	11.972	Oleanolic acid-28-O-beta-D-glucopyranoside	C ₃₆ H ₅₈ O ₈
108	11.972	25D-Spirosta-3,5-diene	C ₂₇ H ₄₀ O ₂
109	11.972	Ilexolide A	C ₃₅ H ₅₄ O ₇
110	11.972	cis-alpha-Irone	C ₁₄ H ₂₂ O
111	12.163	(-)-2beta-Hydroperoxykolavelool	C ₂₀ H ₃₄ O ₃
112	12.163	beta-Solamarine	C ₄₅ H ₇₃ NO ₁₅
113	12.258	3-O-alpha-L-Glucopyranosyl-(1-->3)-alpha-L-rhamno-pyranosyl-(1-->2)-alpha-L-arabinopyranosyloleancli	C ₄₇ H ₇₆ O ₁₆
114	12.258	Tuliposide A	C ₁₁ H ₁₈ O ₈
115	12.258	3'-O-beta-glucopyranosyl plumbagic acid	C ₁₇ H ₂₂ O ₁₀
116	12.354	Elenolide	C ₁₁ H ₁₂ O ₅
117	12.449	Armillaripin	C ₂₄ H ₃₀ O ₆
118	12.64	1-Undecylenyl-3,4-methylenedioxybenzene	C ₁₈ H ₂₈ O ₂
119	12.64	Triptofordin A	C ₃₁ H ₃₆ O ₆
120	13.021	(24S)-Ethylcholesta-5,22,25-trans-3beta-ol	C ₂₉ H ₄₆ O
121	13.307	Androst-4,6-diene-3,17-dione	C ₁₉ H ₂₄ O ₂
122	13.402	(-)-3,4-Di-epi-3,7-trifara-9,14-diene	C ₁₅ H ₂₄
123	13.498	4-(1,5-Dimethyl-1,4-hexadienyl)-1-methyl-cyclohexene	C ₁₄ H ₂₂
124	13.498	Aspidoside A	C ₃₃ H ₅₂ O ₁₂
125	13.784	Annuionone E	C ₁₃ H ₂₂ O ₃
126	14.07	Acetoin	C ₄ H ₈ O ₂
127	14.26	(Z,Z,Z)-9,12,15-Octadecatrienoic acid	C ₁₈ H ₃₀ O ₂
128	14.356	Tetrahydrobungeanol	C ₁₈ H ₃₃ NO ₂
129	14.546	Vindoline	C ₂₅ H ₃₂ N ₂ O ₆
130	14.642	Dihydrocucurbitacin F	C ₃₀ H ₄₈ O ₇
131	14.642	3-Phenylundecane	C ₁₇ H ₂₈
132	15.118	Phthalic anhydride	C ₈ H ₄ O ₃
133	15.118	Dibutyl phthalate	C ₁₆ H ₂₂ O ₄
134	15.118	(Z)-5-Hydroxy-3-butyldiene-phthalide	C ₁₂ H ₁₂ O ₃
135	15.118	alpha-Dichroine	C ₁₆ H ₁₉ N ₃ O ₃
136	15.404	DELTA6-Dehydroferruginol	C ₂₀ H ₂₈ O
137	15.881	Amarasterone B	C ₃₀ H ₅₀ O ₇
138	16.072	12-Methoxy-6,8,11,13-abietatraen-11-ol	C ₂₁ H ₃₀ O ₂
139	16.262	13-Epi-2-oxo-kolavelool	C ₂₀ H ₃₂ O ₂
140	16.453	(24Z)-27-Hydroxy-3-oxo-7,24-tirucalladien-21-al	C ₃₀ H ₄₆ O ₃
141	16.548	8-Gingerol	C ₁₉ H ₃₀ O ₄
142	16.93	(-)-2beta-Hydroxykolavelool	C ₂₀ H ₃₄ O ₂
143	17.311	Tamarixinol	C ₃₀ H ₅₄ O ₂
144	17.406	26,27-Dihydroxy-lanosta-7,9(11),24-trien-3,16-dione	C ₃₁ H ₄₆ O ₄
145	17.502	Deoxymorellin	C ₃₃ H ₃₈ O ₆
146	17.978	alpha-Camphorene	C ₂₀ H ₃₂

147	17.978	Triptofordin D2	C ₃₆ H ₄₂ O ₁₁
148	18.264	4-Epiisocembrol	C ₂₀ H ₃₄ O
149	18.741	Jurubine	C ₃₃ H ₅₇ NO ₈
150	18.836	Ormosanine	C ₂₀ H ₃₅ N ₃
151	18.932	12-O-Cinnamoyl-20-O-tigloyl sarcostin	C ₃₅ H ₄₆ O ₈
152	19.122	Irisoquin F	C ₂₉ H ₅₀ O ₄
153	19.122	3,4',5-Trihydroxy-7-methoxy-8-isopentenylflavone	C ₂₁ H ₂₀ O ₆
154	19.885	3alpha-Hydroxy-6-oxo-5alpha-cholanic acid	C ₂₄ H ₃₈ O ₄
155	20.934	Regelin C	C ₃₃ H ₅₀ O ₆
156	21.315	21-Hydroxy-30-norhopan-22-one	C ₂₉ H ₄₈ O ₂
157	21.315	3beta-Methoxy-9beta,19-cyclolanost-23(E)-en-25,26-diol	C ₃₁ H ₅₂ O ₃
158	21.41	Myxoxanthophyll	C ₄₁ H ₆₀ O ₇
159	21.41	Agastachin	C ₄₇ H ₄₄ O ₂₂
160	21.601	2'-Hydroxycinnamaldehyde	C ₉ H ₈ O ₂
161	21.601	(22Z,24S)-Cerevisterol	C ₂₈ H ₄₆ O ₃
162	22.078	beta-Sitosterol-3-O-beta-D-xylopyranoside	C ₃₄ H ₅₈ O ₅
163	22.745	(+)-Gynunone	C ₁₃ H ₁₄ O ₄
164	23.698	Embelin	C ₁₇ H ₂₆ O ₄
165	24.175	14alpha-Methyl-5alpha-ergosta-9(11),24(28)-dien-3beta-ol	C ₂₉ H ₄₈ O
166	24.747	1-Friedelen-3-one	C ₃₀ H ₄₈ O
167	26.272	4-Methyl-7-ergosta-8,24(28)-diene	C ₂₉ H ₄₈
168	26.463	6-Hydroxy-7-methylesculetin	C ₁₀ H ₈ O ₃
169	26.558	Coniine	C ₈ H ₁₇ N