

RESEARCH ARTICLE

Comparative phytochemical profiling of *Physalis longifolia* Nutt. leaves and fruits extracts: Insights from gas chromatography-mass spectrometry (GC-MS) analysis

Rohita Singla^{1*}, Pooja Bhatt², Vivek Sharma³ and Pamita Bhandari²

Received: 9 November 2024 | Revised: 10 August 2025 | Accepted: 14 August 2025

ABSTRACT

The aim of this study was to examine the bioactive compounds of *Physalis longifolia* Nutt. growing in northern part of India utilizing gas chromatography-mass spectrometry (GC-MS). The leaf extracts showed remarkable antioxidant activity in DPPH (2, 2-diphenyl-2-picrylhydrazyl hydrate) radical scavenging activity and FRAP (Ferric reducing anti-oxidant power assay) as compared to fruit extracts, evidenced by their lower IC₅₀ value. The leaves and fruit extracts of *P. longifolia* included 25 different compounds, including saturated fatty acid esters, organosilicons, mono- and poly-unsaturated omega-6 fatty acids. Methyl linoleate (11.89%), methyl palmitate (20.38%), methyl stearate (22.90%), cyclotetrasiloxane (octamethyl) (18.99%), and heptadecanoic acid methyl ester (0.18%) are among the most prominent bioactive substances. The phytochemical and GC-MS profiling of *P. longifolia* leaves and fruits revealed the presence of bioactive compounds with important medicinal properties.

Keywords: Antioxidant, Bioactive compounds, Gas chromatography-mass spectrometry, Medicinal properties, *Physalis longifolia*

INTRODUCTION

Physalis longifolia Nutt., wild tomatillo or long leaf groundcherry, is a perennial herb that occurs throughout the continental U.S. and into southern Canada and northern Mexico (Kindscher et al. 2012). Its habitat includes old fields, open woods, and prairies, but it thrives in disturbed sites, including roadsides. Plants form colonies through the spread of underground rhizomes and it is often considered to be a weed because the plant is so common. Originally brought to California, Physalis longifolia Nutt. is now regarded as invasive there because of its quicker seeds and rhizomes dissemination (USDA 2011, Kindscher et al. 2012).

Native to North America and South Asia, its adaptability allows it to grow in diverse conditions, including sandy soils during *Kharif* in India where it is often seen as a weed (Singh *et al.* 2019). Traditionally, native American tribes such as the Acoma, Hopi and Zuni consumed its berries fresh or

cooked. Historically, it has been used for various medicinal and culinary purposes, though species identification was often unclear, leading to confusion with related species Traditional uses of the herb *P. longifolia* are extensively reported as medicine, the Omaha and Ponca tribes generally using as local medicine to treat headache, stomach problems and to dress wounds (Kindscher *et al.* 2012). The fruits and flowers of the plant are also used in the stomach pain, constipation and herb paste is used in ear problems (Vipin and Ashok 2010).

Physalis longifolia is highlighted as a prominent weed due to its invasive nature and agricultural significance. This species spreads rapidly through both seeds and rhizomes, making it difficult to control in disturbed areas and cropping systems. Additionally, P. longifolia plays a key role in the transmission of zebra chip disease in potatoes. It serves as a preferred host for the potato psyllid (Bactericera cockerelli), which vectors the pathogen Candidatus Liberibacter solanacearum, posing a serious risk to solanaceous crops in the U.S. (Reyes Corral et al. 2021). Given its ecological impact and role in pest dynamics, P. longifolia is recognised as a prominent weed and hence effective management planning is necessary. The utilization of the weeds is considered to be one of the management approaches (Chandrasena and Rao 2018).

Department of Botany, Eternal University, Baru Sahib, Himachal Pradesh 173101, India

² CSIR-Institute of Himalayan Bioresource Technology, Palampur, Himachal Pradesh 176061, India

³ Department of Phytomedicine, Baba Farid University of Health Sciences, Punjab 151203, India

^{*} Corresponding author email: singlarohita7@gmail.com

Phytochemical studies have identified antioxidants in *Physalis* fruits, including anthocyanins in *P. ixocarpa* (Gonzalez-Mendoza *et al.* 2010), and carotenoids and withanolides in *P. peruviana* (Ramadan 2011). Similar research is recommended for related species like *P. longifolia*, which has shown anticancer, anti-proliferative, and anti-inflammatory effects due to its phytochemicals. It may also help manage chronic conditions like diabetes and neurological disorders (Huang *et al.* 2020).

Gas chromatography-mass spectrometry (GC-MS) effectively identifies and quantifies bioactive compounds in plant extracts by comparing mass spectra to reference databases, detecting volatile and semi-volatile compounds like alkaloids, flavonoids, terpenoids, and phenolics (Grover and Ptani 2013). It supports plant-based drug discovery in nutraceuticals and pharmaceuticals. *P. longifolia* remains underexplored, with no Indian GC-MS studies on its phytochemicals or bioactivity. Thus, this study was conducted with an objective to assess the phytochemical and antioxidant properties of its leaf and fruit extracts.

MATERIALS AND METHODS

Leaves and fruits of Physalis longifolia Nutt. were collected from Yamuna Ghats, Paonta Sahib (Himachal Pradesh, India) in September to October 2021. The species was authenticated at CSIR-IHBT, Palampur, and a voucher specimen (PLP#22087) was deposited. Samples were air-dried in shade, coarsely ground, 10 g of each was soaked for 24 hours at room temperature in 250 mL of methanol: distilled water (80:20). 2,2'-azino-bis-(3-ethylbenzothiazoline-6-sulphonic acid) diammonium salt (ABTS), hydrogen peroxide (H₂O₂), aluminium chloride (AlCl₃), potassium persulfate (K₂S₂O₈), 2,2-diphenyl-1-picrylhydrazyl (DPPH), free radical antioxidant powder (FRAP), 2,4,6-tripyridyl-s-triazin were used, with most chemicals sourced from Merck Limited (Mumbai, India) in analytical grade.

Antioxidant activity of *Physalis longifolia* extracts was evaluated through standard assays. Total phenolic content (TPC) and total flavonoid content (TFC) were measured using the Folin–Ciocalteu and aluminium chloride methods, respectively. Total antioxidant capacity (TAC) was assessed via the phospho-molybdenum assay. DPPH radical scavenging and FRAP assays were conducted to determine free radical inhibition and reducing power. Gallic acid, quercetin, ascorbic acid, and FeSO₄ were used as standards. Results were expressed in standard equivalents per gram of extract (Singla and Pradhan 2019, Banothu *et al.* 2017).

DPPH scavenging activity (%) = [(($A_{Control}$ -(A_{Sample} - A_{Sample} blank))/ $A_{Control}$] x 100

GC-MS analysis was performed using a Shimadzu GC 2010 with an AOC-5000 auto-injector and SH-Rxi-5Sil MS column (30/ m \times 0.25/ mm). The temperature was ramped from 40°C to 220°C, then held for 21 minutes. Samples in HPLC-grade dichloromethane were injected using helium (1.28/



Figure 2. Map displaying the precise location of *Physalis* longifolia plant sample collection

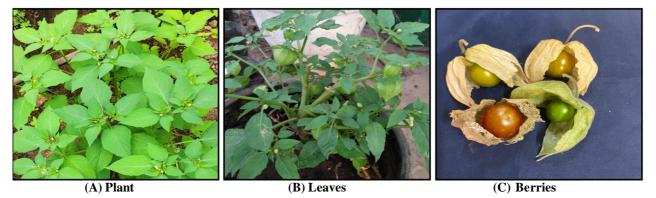


Figure 1. Physalis longifolia in its native environment

mL/min, split 1:10) as the carrier gas. Components were identified via mass spectra comparisons with NIST and Wiley libraries. Chromatograms were generated for hydroalcoholic leaf and fruit extracts. Data were analysed using GraphPad Prism v7.01.

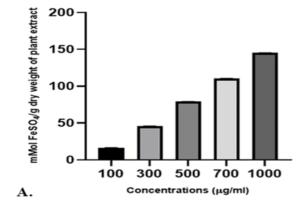
RESULTS AND DISCUSSION

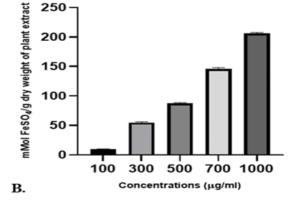
Total phenolic (TPC) and flavonoid content (TFC) in P. longifolia were expressed as gallic acid and quercetin equivalents (mg/g DW). Leaf extracts showed higher TPC (27.5 mg GAE/g) and TFC (136.7 mg QE/g) than fruit extracts (TPC: 13.1 mg GAE/g; TFC: 80.48 mg QE/g). These compounds contribute to antioxidant activity by neutralizing free radicals via hydroxyl and methyl groups. Elevated phenolic levels in leaves are linked to environmental and stress-related factors. Flavonoids, the second most abundant phenolics in leaves, are concentrated in outer tissues and provide UV protection. Their accumulation, particularly of quercetin and kaempferol, increases with UV-B exposure, while fruit shading reduces flavonoid synthesis and gene expression, especially in skins. These trends align with prior studies on related species (Treutter 2005, Pillai *et al.* 2022, Nathiya and Dorcus 2012).

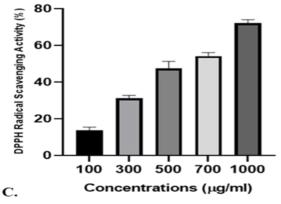
Leaf extracts exhibited higher total antioxidant capacity (29.4 ± 0.174 mg AAE/g DW) than fruit extracts (26.3 ± 0.105 mg AAE/g DW) (**Table 1**). DPPH assay results (**Figure 3**) demonstrated dose-dependent radical scavenging, with leaf extracts showing significantly greater inhibition ($72.1 \pm 1.14\%$) than fruits ($45 \pm 0.0964\%$) at $1000 \mu g/mL$, along with lower IC₅₀ values, indicating superior antioxidant potential. FRAP assay results also revealed concentration-dependent activity. While fruits showed slightly higher activity than leaves at $100 \mu g/mL$, leaf extracts displayed lower IC₅₀ values overall. At $1000 \mu g/mL$, fruits reached $206 \pm 1.00\%$, and leaves $145 \pm 0.196\%$. These findings highlight

Table 1. Total phenolic, flavonoid, and antioxidant content of different plant parts of *Physalis longifolia*

	TPC	TFC	TAC
Plant parts	(mg GAE/g	(mg QE/g	(mg AA/g
	DW)	DW)	DW)
Leaves	27.5 ± 0.605	136.7 ± 2.034	29.4 ± 0.174
Fruits	13.1 ± 0.782	80.48 ± 0.238	26.3 ± 0.105







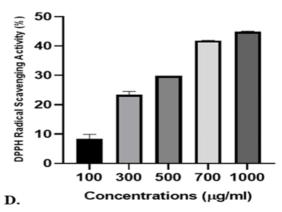


Figure 3. Antioxidant activities of hydroalcoholic extracts of *Physalis longifolia* leaves and fruits, *in vitro*. Data represent mean ± standard error (SE). (A) FRAP assay for leaf extract; (B) FRAP assay for fruit extract; (C) DPPH radical-scavenging leaf extract; and (D) DPPH radical-scavenging fruit extract

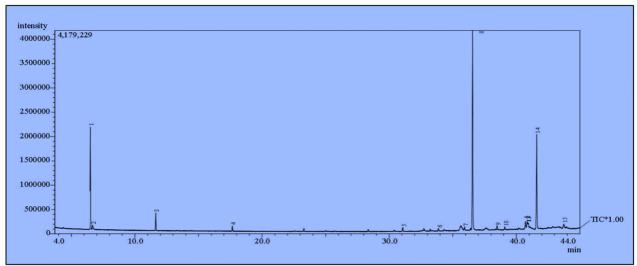


Figure 4. GC-MS Chromatogram of Physalis longifolia leaf extract

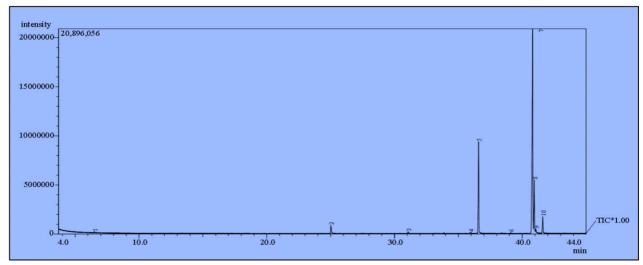


Figure 5. GC-MS Chromatogram of Physalis longifolia fruit extract

the strong antioxidant capacity of *P. longifolia* leaves and support further investigation into their bioactive constituents.

Previous research by Parikh et al. (2018) and Saeed et al. (2012a) demonstrated the use of total phenolic content (TPC), total flavonoid content (TFC), and total antioxidant capacity (TAC) as effective indicators of antioxidant potential in plants, underscoring their significance as natural sources of antioxidants. In this study, the antioxidant activity of two traditionally utilized parts of Physalis longifolia was assessed using FRAP and DPPH assays, which operate through distinct mechanisms-electron transfer in FRAP and both electron and hydrogen atom transfer in DPPH (Kýrca and Arslan 2008). The study revealed that leaf extracts contained higher TPC and TFC levels compared to fruits, which likely contributes to their greater antioxidant activity (Sroka and Cisowski 2003).

GC-MS analysis of *P. longifolia* leaf extract identified 15 bioactive compounds, notably Hexadecenoic acid methyl ester (43.52%), Methyl stearate (22.90%), and Cyclotetrasiloxane, octamethyl- (18.99%) (**Table 2**). The fruit extract revealed major constituents such as 9,12-Octadecadienoic acid methyl ester (59.89%) and Methyl palmitate (20.38%) (**Figure 5**). Both extracts exhibit diverse phytochemicals, including terpenes, fatty acids, and organosilicons, with potential medical and industrial applications.

Bioactive compounds like isoprenoids are recognized for their therapeutic benefits, including disease risk reduction. Terpenes, present in both plant parts, have uses in food, cosmetics, pharmaceuticals, and biofuels (Ponder and Hallmann 2019; Thimmappa *et al.* 2014). GC-MS analysis of *P. longifolia* leaves and fruits identified key bioactive compounds with therapeutic potential, including 9,12-octadecadienoic

Table 2. Chemical composition of methanolic-water fractions from Physalis longifolia

Compound		RT (min)		% Peak Area		Molecular	
		PLF	PLL	PLF	(g/mol)	Formula	Nature of the compound
Cyclotetrasiloxane, octamethyl-	6.49	-	18.99	-	296.61	C8H24O4Si4	Organo-silicon
3-Oxo-Butyric Acid 2-Chloro-Ethyl Ester		-	1.00	-	86.17	C_6H_{14}	Long chain alkane
Cyclopentasiloxane, decamethyl-		-	3.22	-	370.77	$C_{10}H_{30}O_5Si_5$	Organo-silicon
Methane, dichloro- (CAS) Dichloromethane		6.51	-	0.19	84.93	CH_2Cl_2	Volatile organic compound
Dodecanoic acid, methyl ester (CAS) Methyl laurate	-	25.02	-	2.13	214.34	$C_{13}H_{26}O_2$	Saturated Fatty acid methyl ester
Cyclohexasiloxane, dodecamethyl	17.66	-	1.00	-	444.92	$C_{12}H_{36}O_6Si_6$	Organo-silicon
Heptadecanoic acid, methyl ester	31.05	39.10	0.89	0.18	284.48	$C_{18}H_{36}O_{2}$	Saturated Fatty acid ester
Tetra decanoic acid, methyl ester (CAS) Methyl myristate	-	31.07	-	0.39	242.40	$C_{15}H_{30}O_2$	Saturated Fatty acid ester
9-Hexadecenoic acid, methyl ester	-	35.96	-	0.22	270.45	$C_{17}H_{34}O_{2}$	Monounsaturated Fatty acid ester
Arachidic acid methyl ester (CAS) Eicosanoic acid	33.86	-	0.59	-	326.56	$C_{21}H_{42}O_2$	Monounsaturated Omega-9 Fatty acid ester
Pentacosane	35.91	-	0.71	-	352.68	$C_{25}H_{52}$	Aliphatic hydro-carbon
Hexadecanoic acid, methyl ester (CAS) Methyl palmitate	36.56	36.59	43.52	20.38	270.45	$C_{17}H_{34}O_2$	Monounsaturated Fatty acid ester
Triacontane	38.48	-	0.86	-	422.81	$C_{30}H_{62}$	Long chain alkane
Heptacosanoic acid, methyl ester	39.09	-	0.60	-	424.74	$C_{28}H_{56}O_{2}$	Saturated Fatty acid ester
9,12-Octadecadienoic acid (Z, Z)-, methyl ester	-	40.82	-	59.89	280.44	$C_{18}H_{32}O_2$	Polyunsaturated Omega-6 Fatty acid
9-Octadecanoic acid (Z)-, methyl ester	-	40.94	-	11.89	296.49	$C_{19}H_{36}O_2$	Monounsaturated Omega-9 Fatty acid ester
Hexadecadienoic acid, methyl ester	40.71	-	1.68	-	322.52	$C_{21}H_{38}O_2$	Saturated Fatty acid ester
8,11,14-Eicosatrienoic acid, methyl ester	40.86	-	1.95	-	320.51	$C_{21}H_{36}O_2$	Polyunsaturated Omega-6 Fatty acid ester
2-methyl tetracosane	43.73	-	1.14	-	352.68	$C_{25}H_{52}$	Tri- terpenoid
Methyl stearate	41.60	41.61	22.90	3.76	298.50	$C_{19}H_{38}O_2$	Saturated Fatty acid ester
Methyl oleate	-	41.06	-	0.96	296.49	$C_{19}H_{36}O_2$	Polyunsaturated Fatty acid

RIa = calculated retention indices; RIb = retention indices from literature; -: absent; % = relative percentages calculated from GC-FID

acid (Z, Z)-, hexadecanoic acid methyl ester, methyl stearate, and cyclotetrasiloxane octamethyl, also reported in traditional remedies (Ukwubile et al. 2019; Tulandi et al. 2021). Hexadecanoic acid methyl ester, found in both parts, exhibits antioxidant, antiinflammatory, antimicrobial, and anticancer activities. 9,12-Octadecadienoic acid methyl ester demonstrates antioxidant, neuroprotective, and anti-COVID effects (Zayed et al. 2019). Methyl stearate and cyclotetrasiloxane octamethyl possess antibacterial properties and applications in food and cosmetics (Keskin et al. 2012). Minor constituents such as dodecanoic and Eicosatrienoic acid methyl esters contribute antioxidant, antifungal, and anti-allergic effects (Table 2). The dominant fruit compound, 9,12-octadecadienoic acid methyl ester, along with others, also exhibits hypocholesterolmic activity.

Conclusions

This study is the first, to our knowledge, in India to use GC-MS to profile metabolites in *Physalis longifolia*, and it identified 15 phytochemicals in leaves and 10 in fruits from methanolic-water extracts. It also links these compounds to antioxidant activity, highlighting their therapeutic potential. The findings support the *Physalis longifolia*'s traditional medicinal use and suggest that it may serve as a potential source for drug/medicines development or health supplements. However, limited research exists on its biological functions, underscoring the need for

further studies to isolate compounds from various plant parts and assess their anticancer and other therapeutic properties.

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