# THERAPEUTIC POTENTIALS OF Garcinia cambogia (L.) Roxb.: PHYTO-CHEMICAL CHARACTERIZATION AND BIOLOGICAL ACTIVITIES

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### ABSTRACT

Garcinia cambogia is widely used in traditional medicine, yet scientific validation of its pharmacological properties and bioactive compounds remains not fully explored. The present study aimed to bridge this gap by characterizing the phytochemical composition of G. cambogia hydro-alcoholic fruit extract and evaluating its antioxidant, anti-inflammatory, and antidiabetic properties. In fruit extract GC-MS and LC-MS analyses identified 19 and 18 bioactive compounds, respectively. The extract exhibited high total phenolic (6.58 mg g<sup>-1</sup> gallic acid equivalent) and flavonoid content (5.82 mg g<sup>-1</sup> quercetin equivalent). The antioxidant assay showed significant activity with IC50 value of 36.22 µg mL-1, in DPPH assay, outperforming ascorbic acid (40.62 µg mL<sup>-1</sup>). Enzyme inhibition assays demonstrated  $\alpha$ -amylase (IC<sub>50</sub> = 82.87 µg mL<sup>-1</sup>) and  $\alpha$ -glucosidase (IC<sub>50</sub> = 67.73 µg mL<sup>-1</sup>) inhibitory activities, indicating its antidiabetic potential. The anti-inflammatory efficacy was evaluated using a Carrageenan-induced paw edema model in rats, with the extract at 300 mg kg<sup>-1</sup> reducing inflammation by 76.5% at 3 h, comparable to diclofenac (78.2%). The study findings support the therapeutic potential of G. cambogia and its application in oxidative stress management, inflammation, and diabetes.

Keywords: Bioactive compounds, *Garcinia cambogia*, hydro-alcoholic extract, phytochemical profiling, therapeutic potential

#### INTRODUCTION

Medicinal plants play crucial role in traditional healthcare systems such as Ayurveda, Unani, and Siddha (Perveen *et al.*, 2024). They continue to be relevant in modern medicines, with approximately 125 prescription drugs derived from over 100 plant species (Agri *et al.*, 2024). Their therapeutic potential lies in bioactive phytochemicals such as flavonoids, alkaloids, and phenols, which exhibit antioxidant, anti-inflammatory, and antimicrobial properties (Kruttika *et al.*, 2023; Abdelkhalek *et al.*, 2024). Advanced techniques like GC-MS and LC-MS have significantly improved phytochemical identification, aiding drug discovery (Waris *et al.*, 2022; Vij and Pathania, 2023).

*Garcinia cambogia* (L.) Roxb. (syn. *Garcinia gummi-gutta*), a tropical fruit tree, has traditionally been used to treat gastrointestinal issues, inflammation, and rheumatism (Thandayamparambil *et al.*, 2023). Its therapeutic properties are largely attributed to hydroxycitric acid (HCA), garcinol, and xanthochymol, with studies highlighting its anti-obesity, hepatoprotective, and antimicrobial effects (Noreen *et al.*, 2023). However, research has primarily focused on HCA, leaving the pharmacological potential of other bioactive compounds largely unexplored. Limited studies have quantitatively

assessed its antioxidant, anti-inflammatory, and antidiabetic properties using modern analytical methods. This study was aimed to characterize the phytochemical composition of *G. cambogia* using GC-MS and LC-MS, evaluate its antioxidant activity through DPPH and hydrogen peroxide scavenging assays, assess its enzyme inhibition potential against  $\alpha$ -amylase and  $\alpha$ -glucosidase, and determine its anti-inflammatory efficacy using a carrageenan-induced paw edema model in rats.

### **MATERIALS AND METHODS**

#### Collection and extraction of plant material

The fruits of *G. cambogia* were collected from Sorakoppa village, Bagalkot, Karnataka (India) and its identity got authenticated by the Department of Botany, Basaveshwar Science College, Bagalkot under voucher specimen code HSKCOP-01/04-22 in the College Herbarium. The fruits were washed with water and potassium permanganate solution, air-dried, and ground into a coarsely powder. The powder was then maceration in 50:50 hydro-alcoholic solution (water and ethanol), followed by solvent separation through distillation and evaporation at room temperature. The concentrated extract was lyophilized using a Mini Lyotrap (LTE Scientific Ltd, Great Britain), yielding 56.58 g dark green solid mass, which was stored under refrigeration for further anti-diabetic activity evaluation.

### Phytochemical analysis

The hydro-alcoholic extract of *G. cambogia* was subject to the qualitative phytochemical screening to identify bioactive compounds by using established protocols (Harborne, 1973; Trease and Evans, 1987). Alkaloids were detected using iodine test, Wagner's test, and Dragendorff's tests, while flavonoids were confirmed by Shinoda and NaOH tests. Glycosides were identified using Keller-Killani, Liebermann's, and Molisch tests and phenols and lignins were confirmed via their respective tests. Saponins were detected by Foam and Hemolysis tests, and sterols by Liebermann-Burchard's test, and reducing sugars using the Reducing sugar test. The total phenolic content was quantified using Folin–Ciocalteu colorimetric method (Singleton *et al.*, 1999), with slight modifications like reducing the incubation time from 120 to 60 min and adjusting the reagent-to-sample ratio for improved sensitivity. The total flavonoid content was measured by using the aluminum chloride method (Zhishen *et al.*, 1999).

### Analytical analysis of compounds

**Gas chromatography-mass spectrometry (GC-MS):** The hydro-alcoholic extract of *G. cambogia* was analysed using GC-MS to identify its constituent compounds. A 1  $\mu$ L aliquot of sample was injected into Agilent 7890B GC system coupled with Agilent 5977A MSD using 30 m HP-5MS capillary-column. The separation was performed with helium as the carrier gas at 1.0 mL min<sup>-1</sup>, with injector temperature set at 250°C. The oven temperature program started at 60°C for 2 min, followed by ramping at 10°C min<sup>-1</sup> to 300°C and holding for 10 min. The mass spectrometer operated in an electron impact mode (70 eV), scanning a mass range of 50–550 m/z. Compounds were identified by comparing their mass spectra with those in NIST Mass Spectral Library (NIST 17) using Agilent Mass Hunter Workstation software.

**Liquid chromatography-mass spectrometry (LC-MS)**: LC-MS analysis of *G. cambogia* extract was performed by using a Waters Acquity UPLC with a Xevo G2-XS QT of mass spectrometer. Separation occurred on an Accucore C18 column with a gradient mobile phase of 0.1% formic acid in water and acetonitrile. The gradient shifted from 95% A to 50% A over 6 min, then reversed and was held for 5 min. System parameters included a 3.0 kV capillary voltage, 20-90 V collision energy, and source and desolvation temperatures of 150 and 450°C, respectively. Gas flows were set to 50 L h<sup>-1</sup> (cone) and 800 L h<sup>-1</sup> (desolvation).

## Anti-oxidation analysis

**DPPH** (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity: The antioxidant activity of hydroalcoholic extract of *G. cambogia* was assessed by DPPH assay. A 0.1 mM DPPH solution in methanol was prepared, and 2 mL DPPH solution mixed with 1 mL plant extract at varying concentrations (10-100  $\mu$ g mL<sup>-1</sup>). The mixtures were vortexed and incubated in dark at room temperature for 30 min. Absorbance was measured at 517 nm using a UV-visible spectrophotometer (Labman). The percentage scavenging activity was calculated as:

% Scavenging activity = 
$$\frac{Ao - A1}{Ao} x 100$$

where  $A_0$  is the absorbance of control and  $A_1$  is the absorbance of sample.

The IC<sub>50</sub> value, representing the concentration required to scavenge 50% of DPPH radicals, was determined from a linear regression plot of scavenging activity against concentration. Ascorbic acid served as positive control.

**Hydrogen peroxide-scavenging activity** ( $H_2O_2$ ): The  $H_2O_2$  scavenging activity of hydro-alcoholic extract of *G. cambogia* was assessed using the method of Ruch *et al.* (1989). The plant extract was dissolved in 0.1 M phosphate buffer (pH 7.4) and mixed with 43 mM  $H_2O_2$  solution. The reaction mixture was incubated and its absorbance measured at 230 nm using a UV-visible spectrophotometer (Labman) at 10 min intervals for 40 min. Blank samples for each concentration were tested to correct the background interference. The decrease in absorbance indicated the neutralization of  $H_2O_2$ , with low levels of residual  $H_2O_2$  signifying higher antioxidant activity. This method allowed for quantification of the scavenging ability of the plant extract against  $H_2O_2$ .

**<u>Reducing power assay</u>:** The reducing power of hydro-alcoholic extract of *G. cambogia* was assessed by mixing varying concentrations of extract with 2.5 mL phosphate buffer (200 mM, pH 6.6) and 2.5 mL 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min, followed by the addition of 2.5 mL trichloroacetic acid (10%). After centrifugation at  $650 \times g$  for 10 min, 5 mL supernatant was mixed with 5 mL distilled water and 1 mL 0.1% ferric chloride. The absorbance of resulting solution was measured at 700 nm using a UV-visible spectrophotometer (Labman), with phosphate buffer used as the blank and butylated hydroxytoluene (BHT) as standard.

## Enzyme inhibition studies

<u>a-amylase inhibitory assay</u>: The  $\alpha$ -amylase inhibitory activity of *G. cambogia* hydro-alcoholic fruit extract was assessed by mixing 1 mL phosphate-buffer saline (PBS, pH 7.4) with 0.5 mL extract at varying concentrations (50–250 µg mL<sup>-1</sup>), and 200 µL of 0.5 mg mL<sup>-1</sup>  $\alpha$ -amylase solution in an Eppendorf tube. Acarbose was used as standard inhibitor. After adding 200 µL of 5 mg mL<sup>-1</sup> starch solution, the mixture was incubated at room temperature for 10 min. The reaction was stopped by adding 400 µL dinitrosalicylic acid (DNS) solution, followed by heating the mixture in a boiling water bath for 5 min. The absorbance was measured at 540 nm using a Labman UV-visible spectrophotometer. Controls with and without  $\alpha$ -amylase were included for accuracy. The experiment was conducted in triplicate to ensure data reliability and minimize random errors.

The percentage inhibition of α-amylase activity was calculated as:

% inhibition in  $\alpha$ -amylase activity =  $\frac{AC - AS}{AC} \times 100$ 

Where AC = Absorbance of control, AS = Absorbance of sample

<u>a-Glucosidase inhibition assay</u>: The  $\alpha$ -glucosidase inhibitory activity of *G. cambogia* hydro-alcoholic fruit extract was assessed using  $\rho$ -nitrophenyl- $\rho$ -D-glucopyranoside (PNPG) as substrate. The assay was conducted in 50 mM phosphate buffer (pH 6.5), with extract concentrations (50, 100, 150, 200, and 250 µg mL<sup>-1</sup>) prepared in 5% dimethyl sulfoxide (DMSO). For each reaction, 30 µL extract was mixed with phosphate buffer (pH 6.5) and 30 µL of 2 mM PNPG solution, followed by incubation at 37°C for 5 min. Subsequently, 30 µL  $\alpha$ -glucosidase enzyme (0.15 U mL<sup>-1</sup>) was added, and the mixture

incubated for an additional 15 min. The reaction was terminated by adding 50  $\mu$ L 1 M sodium carbonate (Na<sub>2</sub>CO<sub>3</sub>), and the absorbance was measured at 405 nm by using a UV-visible spectrophotometer (Shimadzu UV-1900). Acarbose was used as a positive control, and the percentage inhibition of  $\alpha$ -glucosidase activity was calculated by using the formula:

% inhibition in  $\alpha$ -glucosidase activity =  $\frac{OD \text{ of blank} - OD \text{ of sample}}{OD \text{ of Blank}} x 100$ 

The IC<sub>50</sub> value, depicting the concentration inhibiting the 50% of  $\alpha$ -glucosidase activity, was determined from a concentration-inhibition curve. The experiment was performed in triplicate across three independent trials.

### Anti-inflammatory effect

The anti-inflammatory potential of hydro-alcoholic extract of *G. cambogia* was evaluated by using the Carrageenan-induced paw edema model in rats. Male Wistar rats (age: 8-10 weeks, weight: 180-220 g) were divided into five groups (n = 6 per group). Group I served as vehicle control and received saline orally. Group II was treated with diclofenac @ 10 mg kg<sup>-1</sup> body weight (positive control), and groups III, IV, and V were orally administered *G. cambogia* fruit extract @ 100, 200, and 300 mg kg<sup>-1</sup> body weight, respectively. Carrageenan (0.1 mL 1% solution in saline) was injected into the plantar region of left hind paw of each rat to induce inflammation, and paw volumes were measured at predetermined intervals (0, 1, 3, 6, 12, and 24 h) by using a plethysmometer. Paw edema was quantified as difference in volume between injected (left) and non-injected (right) paws. The anti-inflammatory effect of extract was assessed by calculating percent inhibition of edema, using formula:

% inhibition of edema = 
$$\frac{\text{Change in control group} - \text{Change in treatment group}}{\text{Change in control group}} x 100$$

The change in paw thickness values was assessed by comparing the difference between the volumes of left and right paws.

#### Statistical analysis

The experiments were conducted in a completely randomized design and the data presented as mean  $\pm$  SEM. Data was statistically analysed using one-way analysis of variance (ANOVA) to assess differences between the groups. Post-hoc analysis was performed using Tukey's multiple comparison test to identify specific group differences. Statistical analysis was conducted using appropriate software (e.g., SPSS or GraphPad Prism), and significance set at p < 0.05 (Motulsky, 2018).

## **RESULTS AND DISCUSSION**

#### Phytochemical analysis

The phytochemical analysis of hydro-alcoholic extract of *G. cambogia* fruits revealed the presence of diverse bioactive compounds (Table 1). The alkaloids were detected through iodine and Wagner's tests, aligning with findings of Nchiozem-Ngnitedem (2023) and Thandayamparambil *et al.* (2023). Flavonoids, detected by Shinoda test and NaOH test were also consistent with earlier studies highlighting their significant role in plant's medicinal properties. Glycosides were confirmed by Keller-Killani, Liebermann's, and Molisch tests, further supporting previous reports by Priyadharisini (2019) that emphasized the importance of glycosides in *G. cambogia*. Interestingly, lignins and anthraquinones were absent in the extract while reducing sugars and glycosides were present in the extract contrasting with Priyadharisini (2019) who used chloroform-based extract. This highlights the significance of solvent selection on the extraction of bioactive compounds. The presence of phenols, saponins, sterols, and tannins in the hydro-alcoholic extract further supports its established medicinal uses, such as antioxidant, anti-inflammatory, and antimicrobial activities.

	yuro-alconolic extract of	l O. cambogia					
Phytochem	Hydro-alcoholic						
-	extract						
Alkaloid:	Iodine test	+					
	Wagnore's test	+					
	Dragendroff's test	-					
Flavonoids	+						
	NaOH test	++					
Glycosides	: Keller- Killani test	++					
	Liebermann's test	++					
	Molisch test	++					
Phenols:	Phenol test	++					
Lignins:	Lignin test						
Saponins:	Foam Test	+					
	Heamolysis Test	+					
Sterols: Lil	bermann-Burchard's test	+					
Tannins:	Lead acetate test	++					
	Gelatin test	++					
Anthraquinone: Bomtrager's test							
Reducing s	+						
+: Present; ++: High;: Absent							
1							
	y = 0.8272x + 0.021						
0.8 -	$R^2 = 0.9901$						
8							
- 9.0 Absorbance at 750 mm	×	r					
eat	. /						
0160 0.4							
losq							
< 0.2							
0.2							
0 0 0	.2 0.4 0.6 0	.8 1 1.2					
Concentration (mg/ml)							

 
 Table 1: Qualitative analysis of phytochemicals in hydro-alcoholic extract of G. cambogia

Fig. 1: Standard curve of gallic acid for estimating the total phenol content

### Total phenol content

The total phenolic content of G. cambogia hydro-alcoholic fruit extract was found to be 6.58 mg g<sup>-1</sup> gallic acid equivalent, revealing strong antioxidant potential (Fig. 1). This value is notably higher than 0.32 mg g<sup>-1</sup> reported by Joseph and Tangavel (2023), highlighting the impact of extraction methods on phenolic yield. Phenolic compounds like quercetin, rutin, catechin, and gallic acid, known for their antioxidant properties, have previously been identified in G. cambogia (Jamal et al., 2017). The high linearity of calibration curve  $(r^2 =$ reliable 0.9901) ensures quantification. Previous studies have shown that methanol is highly effective in extracting the phenolics from Garcinia species (Aati et al., 2022), though the hydro-alcoholic extract also yielded significant phenolic content. The antioxidant activity of G. cambogia, confirmed through DPPH and hydrogen peroxide scavenging assays, reveals the role of phenolics in reducing the oxidative stress (Jantan et al., 2011), thus highlights the therapeutic potential of G. cambogia.

#### Total flavonoid content

The total flavonoid content in hydro-alcoholic fruit extract of *G. cambogia* was 5.82 mg g<sup>-1</sup> quercetin equivalent, which was higher than  $0.35 \pm 0.016$  mg g<sup>-1</sup> reported for its ethanolic extract by El-Mesallamy *et al.* (2024). This variation highlights the role of hydro-alcoholic solvent in enhancing flavonoid extraction due to their balanced polarity. Comparisons with other species, like *G. indica* (137.27 µg g<sup>-1</sup> in methanolic fruit extract (Singh *et al.*, 2011) and

*G. schomburgkiana* (214.00  $\pm$  7.32 mg rutin equivalent mg<sup>-1</sup> in leaf ethanol extract (Thummajitsakul and Silprasit, 2022) revealed the species, plant part, and solvent-specific differences. The findings revealed the importance of optimized extraction methods as well as highlight the potential of *G. cambogia* as a source of bioactive flavonoids for therapeutic applications.

## DPPH assay

The DPPH assay demonstrated a dose-dependent increase in radical scavenging activity, with an IC<sub>50</sub> value of 36.22  $\mu$ g mL<sup>-1</sup>, outperforming the ascorbic acid (40.62  $\mu$ g mL<sup>-1</sup>) and indicating the potent antioxidant effects. This enhanced activity, attributed to flavonoids, phenols, and tannins, exceeds the IC<sub>50</sub> value of 39.45  $\mu$ g mL<sup>-1</sup> reported by Aati *et al.* (2022), thus suggesting synergistic effects of bioactive compounds. The concentration-dependent increase in inhibition (Fig. 2) showed that hydro-alcoholic extract consistently outperformed ascorbic acid, achieving over 90% inhibition at 250  $\mu$ g mL<sup>-1</sup>. The lower IC<sub>50</sub> (36.22  $\mu$ g mL<sup>-1</sup>) supports its stronger free radical scavenging potential. This superior antioxidant activity is linked to bioactive compounds that neutralize free radicals and chelate metals (Thummajitsakul *et al.*, 2016). Similar studies on *Garcinia* species (Rizikiyan *et al.*, 2022; Shetty

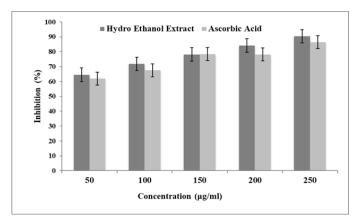
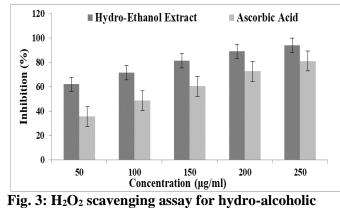


Fig. 2: DPPH scavenging assay for hydro-alcohol extract of *G. cambogia* 



fruit extract of G. cambogia

*et al.*, 2022) highlight the genus's antioxidant potential, emphasizing the importance of optimized extraction methods. *G. cambogia's* ability to neutralize free radicals suggests potential protective roles against oxidative stress-related diseases like neurodegeneration and cardiovascular disorders.

### $H_2O_2$ scavenging activity

The H<sub>2</sub>O<sub>2</sub> scavenging assay revealed a concentration-dependent increase in the scavenging activity of both hydroalcoholic extract of G. cambogia and standard ascorbic acid (Fig. 3). At 250  $\mu$ g mL<sup>-1</sup>, the extract achieved nearly 90% H<sub>2</sub>O<sub>2</sub> neutralization, indicating potent antioxidant effects. This activity is attributed to bioactive compounds like flavonoids, phenols, and tannins, which neutralize reactive oxygen species (ROS) and reduce oxidative stress. The hydroxyl (-OH) groups likely aids in electron donation to break down peroxides. These results are in agreement with previous studies showing xanthones of G. mangostana offering cytoprotection (Karim et al.,

2018), *G. cambogia*'s role in reducing ROS and lipid accumulation in liver cells (Prashanth *et al.*, 2011). Additionally, *G. cambogia* enhances antioxidant gene expression via NRF2 pathway (Sharma *et al.*, 2019) and activates SIRT3, promoting adipocyte browning (Han *et al.*, 2021).

### **Reducing power activity**

The reducing power activity assay demonstrated a concentration-dependent increase in both hydroalcoholic extract of *G. cambogia* and ascorbic acid (Fig. 4), with extract consistently showing superior activity across the concentrations tested. At 250  $\mu$ g mL<sup>-1</sup>, the extract exhibited significantly higher

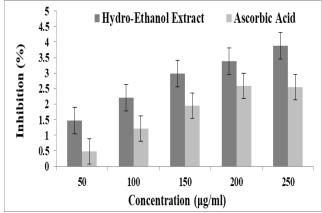


Fig. 4: Reducing power assay for hydro-alcoholic fruit extract of *G. cambogia* 

reducing power. This enhanced activity is attributed to its phytochemical composition including, terpenoids, tannins, saponins, flavonoids, phenols and steroids, which neutralize free radicals and reduce oxidative stress (Sharma et al., 2019). These results are in line with Thummajitsakul et al. (2016) and Bonesso dos Reis et al. (2009) who reported strong antioxidant activity in G. cambogia extracts and its ability to reduce oxidative stress rats. The extract surpassed BHT at higher concentrations, suggesting its role in preventing lipid peroxidation and maintaining redox balance.

## $\alpha$ -Amylase activity

The  $\alpha$ -amylase assay revealed a concentration-dependent increase in % inhibition for both the hydroalcoholic extract of *G. cambogia* and acarbose, with the extract consistently showing higher inhibitory

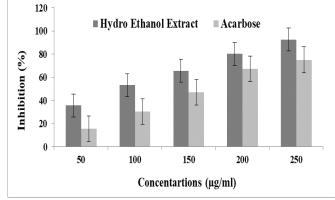


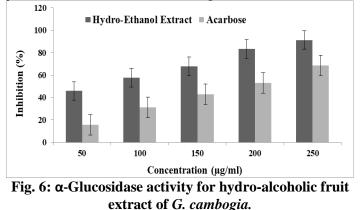
Fig. 5: α-Amylase activity for hydro-alcoholic fruit extract of *G. cambogia* 

activity (Fig. 5). At 250 µg mL<sup>-1</sup>, the extract exhibited lower IC<sub>50</sub> value (82.87 µg mL<sup>-1</sup>) as compared to acarbose (146.99 µg mL<sup>-1</sup>) indicating stronger inhibition. This enhanced activity may be attributed to bioactive compounds such as flavonoids, phenols, and tannins, which inhibit carbohydrate-hydrolysing enzymes. Similar findings in other Garcinia species, including G. mangostana, revealed strong alpha-amylase inhibition xanthones by and benzophenones with low IC<sub>50</sub> values

(Alhakamy *et al.*, 2022; Wairata *et al.*, 2023). The results suggest the therapeutic potential of *G. cambogia* extract as a natural alternative for managing diabetics and obesity by regulating carbohydrate metabolism and postprandial glucose levels.

## a-Glucosidase assay

The  $\alpha$ -glucosidase assay revealed a concentration-dependent increase in % inhibition for both the hydroalcoholic extract of *G. cambogia* and acarbose, with extract consistently showing significantly



inhibition at all concentrations (Fig. 6). At 250  $\mu$ g mL<sup>-1</sup>, the extract showed highest inhibition, with a lower IC<sub>50</sub> value (67.73  $\mu$ g mL<sup>-1</sup>) as compared to acarbose (180.22 µg mL<sup>-1</sup>). indicating its superior efficacy. This potent inhibitory attributed activity is to phytochemicals like flavonoids. tannins, and phenolic acids, which inhibit carbohydrate-digesting enzymes and regulate postprandial glucose levels. These results align

with previous studies on *Garcinia* species, such as *G. mangostana*, *G. mckeaniana*, and *G. fusca*, which also exhibited strong  $\alpha$ -glucosidase inhibition (Xu *et al.*, 2022; Artanti *et al.*, 2023). The results revealed the *G. cambogia's* therapeutic potential as a natural antidiabetic agent, capable of delaying carbohydrate digestion and reducing hyperglycemia.

## Anti-inflammatory activity

The anti-inflammatory activity of *G. cambogia* extract, assessed by using the Carrageenan-induced paw edema model in rats, was significantly reduced in dose-dependent manner in paw volume across all time points. At 300 mg kg<sup>-1</sup>, the extract exhibited highest activity, with paw volume reduced to  $0.3021 \pm 0.016$  mL at 3 h and  $0.284 \pm 0.014$  mL at 24 h (Table 2). These results are in line with Kumar *et al.* (2023) who reported anti-inflammatory effects of hydroxycitric acid, a major component of *G. cambogia*; while Chantree *et al.* (2023) identified polyisoprenylated benzophenones inhibiting nitric oxide (NO) production as well as NF- $\kappa$ B pathway. Similar effects have been observed in other *Garcinia* species, such as *G. dulcis*, *G. humilis*, and *G. indica*, known for modulating proinflammatory

Treatment	Paw volume (% of oedema inhibition)					
groups	0 h	1 h	3 h	6 h	12 h	24 h
Control (saline)	$1.210\pm0.112$	$0.989 \pm 0.125$	$0.897 \pm 0.062$	$1.176\pm0.049$	$1.18\pm0.05$	$1.185 \pm 0.015$
Diclofenac	$0.852\pm0.030$	$0.735\pm0.023$	$0.568 \pm 0.037$	$0.335\pm0.028$	$0.323 \pm 0.012^{***}$	$0.311 \pm 0.013^{***}$
(10 mg kg <sup>-1</sup> )						
GC (100 mg kg <sup>-1</sup> )	$0.963 \pm 0.102$	$0.732 \pm 0.078$	$0.622\pm0.042$	$0.652{\pm}0.038$	$0.600 \pm 0.031 **$	$0.590 \pm 0.025^{**}$
GC (200 mg kg <sup>-1</sup> )	$0.961\pm0.083$	$0.751 \pm 0.096$	$0.505\pm0.125$	$0.431 \pm 0.126$	$0.391 \pm 0.23^{***}$	$0.312 \pm 0.190^{***}$
GC (300 mg kg <sup>-1</sup> )	$0.912\pm0.091$	$0.612 \pm 0.095$	$0.302\pm0.016$	$0.302\pm0.047$	$0.302 \pm 0.021^{***}$	$0.284 \pm 0.014^{***}$

Table 2: Effect of G. cambogia (GC) extract on Carrageenan-induced paw oedema volume in rats

All values are expressed as mean  $\pm$  SEM, n = 6, ANOVA followed by multiple comparison Tukey's test; \*p<0.05, \*\*p<0.01, \*\*\*p<0.001 as compared to the control group.

cytokines and antioxidant activity (Happi *et al.*, 2022). These findings demonstrate the antiinflammatory potential of *G. cambogia* as an effective natural agent particularly at higher doses.

#### GC MS analysis

The GC-MS analysis of hydro-alcoholic extract of *G. cambogia* fruits revealed 19 bioactive compounds (Table 3) with distinct pharmacological properties. The key compounds included (3,3'-Bi-1H-1,2,4-triazole)-5,5'-diamine, known for its antibacterial, antifungal, anticancer, and anti-inflammatory effects (Ajmal *et al.*, 2024), and benzene derivatives like 1-ethoxy-2-methoxy-4-methylbenzene, showing immunosuppressive applications in managing autoimmune diseases (Machinaga *et al.*, 2012). The other compounds like 2-amino-5H-pyrrolo(3,4-d) pyrimidine-4,7(3H,7H)-dione have anti-neurodegenerative and antitumor properties (Liang *et al.*, 2023), while phosphoric acid dimethyl 1-propenyl ester is valued in cosmetics for its low toxicity. Additionally, alloaromadendrene oxide-(1) and tricycle (6.3.3.0) tetradec-4-ene demonstrated antimicrobial, anticancer, and neuro-protective properties. Santalol and humulenol-II offered cardiovascular and anti-inflammatory benefits (Sahoo *et al.*, 2022), while cis-Z- $\alpha$ -bisabolene epoxide exhibited potent antioxidant activity (Khojali and Mohammed, 2023). Arachidonoyl amides displayed anti-inflamatory and antimicrobial effects; while caryophyllene oxide contributed to the extract's anticancer potential. These findings reveal the *G. cambogia*'s therapeutics potential.

S.	Bioactive compounds identified	Peak	RT	Molecular
No.			(min)	formula
1.	[3,3'-Bi-1H-1,2,4-triazole]-5,5'-diamine	110	995	$C_4H_6N_8$
2.	Carbamic acid, (α-methylbenzyl)-, 1-ethyl-1-methylpentyl ester	105	994	$C_{17}H_27NO_2$
3.	Alanine, N-methyl-N-(2-chloroethoxy carbonyl)-, hexyl ester	164	992	$C_{13}H_{24}CINO_4$
4.	Benzene, 1-ethoxy-2-methoxy-4-methyl-	138	992	C10H14O2
5.	Alanine, N-methyl-N-(2-chloroethoxycarbonyl)-, isohexyl ester	164	989	$C_{13}H_{24}CINO_4$
6.	Alanine, N-methyl-N-(2-chloroethoxycarbonyl)-, nonyl ester	164	986	$C_{16}H_{30}CINO_4$
7.	2-Amino-5H-pyrrolo[3,4-d]pyrimidine-4,7(3H,7H)-dione	166	985	$C_6H_6N_4O_2$
8.	Phosphoric acid, dimethyl 1-propenyl ester	110	985	$C_5H_{11}O_4P$
9.	Alanine, N-methyl-N-(2-chloroethoxycarbonyl)-, heptyl ester	164	980	$C_{14}H_{26}CINO_4$
10.	Alanine, N-methyl-N-(2-chloroethoxycarbonyl)-, dodecyl ester	164	976	$C_{19}H_{36}CINO_4$
11.	Caryophyllene oxide	164	972	$C_{15}H_{24}O$
12.	Alloaromadendrene oxide-(1)	164	970	$C_{15}H_{24}O$
13.	Tricyclo[6.3.3.0]tetradec-4-ene,10,13-dioxo-	164	960	$C_{13}H_{16}O_2$
14.	Santalol, cis, a-	164	955	$C_{15}H_{24}O$
15.	Humulenol-II	164	950	$C_{15}H_{26}O$
16.	cis-Z-α-Bisabolene epoxide	164	945	$C_{15}H_{24}O$
17.	Arachidonoyl amide, N-(trifluoroacetyl)-	164	940	$C_{23}H_{37}NO_2$
18.	(1R,7S, E)-7-Isopropyl-4,10-dimethylenecyclodec-5-enol	164	935	$C_{15}H_{26}O$
19.	Lanceol, cis	164	930	$C_{15}H_{24}O$

Table 3: Bioactive compounds identified in the hydro-alcoholic extract of *G. cambogia* fruits

## LC-MS analysis

The LC-MS analysis of hydro-alcoholic extract of G. cambogia fruits showed 18 bioactive compounds having diverse pharmacological properties. These were hypotaurine, D-(-)-salicin, allicin, isoferulic acid, 2,6-dihydroxyanthracene-9,10-dione, alizarin, tryptophan, diallyl sulphide, (3R)-8-hydroxy-3-(4-methoxyphenyl)-3,4-dihydroisochromen-1-one (phyllodulcin), vasicinone, kawain, (4R)-4-((3R,5S,9S,10S,13R,14S,17R)-3-hydroxy-10,13-dimethylhexadecahydro-1H-cyclo penta[a] phenanthren-17-yl) pentanoic acid, quercetin-3-glucuronide, myricetin, stigmasterol, cyanidin-3-glucoside chloride, allantoin and fumarprotocetraric acid. Hypotaurine exhibits antioxidant, anti-aging, and analgesic effects; D-(-)-salicin has anti-inflammatory and antitumor properties; while allicin has anticancer, anti-inflammatory, and antimicrobial activity, including efficacy against MRSA (Talib et al., 2024). Kawain, a kavalactone, exhibits anxiolytic and neuroprotective effects, supporting its traditional use for anxiety relief. Other compounds, such as anthraquinones, tryptophan, and diallyl sulfide (DAS) reportedly provide additional anticancer, antimicrobial, and chemopreventive effects. Quercetin-3-glucuronide (Q3G) and myricetin exhibit strong antioxidant and anticancer activities (Carrillo-Martinez et al., 2024). Stigmasterol, cyanidin-3glucoside chloride (C3G), allantoin, and fumarprotocetraric acid contribute to the extract's neuroprotective, regenerative, and immunomodulatory properties, highlighting G. cambogia's broad therapeutic potential.

**Conclusion:** This study comprehensively analysed the hydro-alcoholic fruit extract of *G. cambogia*, highlighting its rich phytochemical profile and significant bioactive properties. GC-MS and LC-MS analyses identified numerous compounds, including (3,3'-Bi-1H-1,2,4-triazole)-5,5'-diamine and carbamic acid, with notable antimicrobial and anti-inflammatory activities. The extract demonstrated potent antioxidant capabilities, exceeding those of ascorbic acid, and showed strong  $\alpha$ -amylase and  $\alpha$ -glucosidase inhibitory activities, indicating potential antidiabetic benefits. Additionally, the anti-inflammatory effects were comparable to diclofenac, validating *G. cambogia* as a promising candidate for therapeutic applications in modern medicine. These findings support the traditional uses of *G. cambogia* and suggest its potential in developing novel treatments for oxidative stress, inflammation, and diabetes. Future research should focus on *in vivo* studies and clinical trials to further validate these findings and explore the mechanisms underlying these bioactivities.

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3-031-21973-3 12-2].

### REFERENCES

Aati, H.Y., Ismail, A., Rateb, M.E., AboulMagd, A.M., Hassan, H.M. and Hetta, M.H. 2022. Garcinia cambogia phenolics as potent anti-Covid-19 agents: Phytochemical profiling, biological activities and molecular docking. Plants, 11(19): 2521 [https://doi.org/10.3390/plants11192521].
Abdelkhalek, S.T., Jin, S., Jin, M., Abdelgayed, S.S. and Wang, M. 2024. Classification of phytochemicals in plants with herbal value. Springer Nature, 1-39. [https://doi.org/10.1007/978-

- Agri, B.K., Verma, P.P.S., Tiwari, A.K., Singh, S., Kumar, D. and Padalia, R.C. 2024. Utility and conservation of medicinal and aromatic plants in India. *Futuristic Trends in Agriculture Engineering and Food Sciences*, 3(16): 39-53.
- Alhakamy, N.A., Mohamed, G.A., Fahmy, U.A., Eid, B.G., Ahmed, O.A.A., Al-Rabia, M.W., et al., 2022. New alpha-amylase inhibitory metabolites from pericarps of *Garcinia mangostana*. *Reproductive and Developmental Biology*, **12**(3): 384 [https://doi.org/10.3390/life12030384].
- Artanti, N., Dewijanti, I.D., Muzdalifah, D., Windarsih, A., Suratno, S. and Handayani, S. 2023. Alpha-glucosidase inhibitory activity of the combination of *Caesalpinia sappan* L. and *Garcinia mangostana* extract. *Journal of Applied Pharmaceutical Science*, **13**(5): 189-198.
- Bonesso dos Reis, S., de Oliveira, C.C., Acedo, S.C., da Conceição Miranda, D.D., Ribeiro, M.L., Pedrazzoli, J. and Gambero, A. 2009. Attenuation of colitis injury in rats using *Garcinia cambogia* extract. *Phytotherapy Research*, 23(3): 324-329.
- Carrillo-Martinez, J.E., Flores-Hernández, F.Y., Salazar-Montes, A.M., Nario-Chaidez, H.F. and Hernández-Ortega, L.D. 2024. Quercetin, a flavonoid with great pharmacological capacity. *Molecules*, 29(5): 1000. [https://doi.org/10.3390/molecules29051000].
- Chantree, P., Martviset, P., Thongsepee, N., Sangpairoj, K. and Sornchuer, P. 2023. Antiinflammatory effect of garcinol extracted from *Garcinia dulcis* via modulating NF-κB signaling pathway. *Nutrients*, **15**(3): 575. [*https://doi.org/10.3390/nu15030575*].
- El-Mesallamy, A., Alahwany, A., El-zaidy, M. and Hussein, S. 2024. Demonstration of phenolics content from *Garcinia cambogia* extract and evaluation of its cytotoxic activity. *Bulletin of Faculty of Science, Zagazig University*, **2024**(3): 156-162.
- Han, J.H., Park, M.H. and Myung, C.S. 2021. *Garcinia cambogia* ameliorates non-alcoholic fatty liver disease by inhibiting oxidative stress-mediated steatosis and apoptosis through NRF2-ARE activation. *Antioxidants*, **10**(8): 1226. [https://doi.org/10.3390/antiox10081226].
- Han, N., Park, H.J., Ko, S.G. and Moon, P.D. 2024. Stigmasterol exerts an anti-melanoma property through down-regulation of reactive oxygen species and programmed cell death ligand 1 in melanoma cells. *Antioxidants*, 13(3): 380. [https://doi.org/10.3390/antiox13030380].
- Happi, M.A.N., Djoko, E. and Ngandeu, S.M. 2022. Evaluation of the anti-inflammatory activity of Garcinia kola capsules on gonarthrosis and low back pain. International Journal of Biological and Pharmaceutical Sciences, 3(1): 25. [https://doi.org/10.53771/ijbpsa.2022.3.1.0025].
- Harborne, J.B. 1973. Phytochemical Methods. Chapman and Hall Ltd., London, UK.
- Jamal, P., Akbar, I., Jaswir, I., Zuhanis, Y. and Hashim, H.Y. 2017. Quantification of total phenolic compounds in papaya fruit peel. *Pertanika Journal of Tropical Agricultural Science*, **40**: 87-98.
- Jantan, I., Jumuddin, F.A., Saputri, C.S. and Rahman, K. 2011. Inhibitory effects of the extracts of *Garcinia* species on human low-density lipoprotein peroxidation and platelet aggregation in relation to their total phenolic contents. *Journal of Medicinal Plants Research*, **5**(13): 2699-2709.
- Joseph, M.K. and Thangavel, I. 2023. GC-MS/HPTLC analysis, antioxidant, anti-inflammatory, and antimicrobial efficacy of *Garcinia cambogia*. *Free Radicals and Antioxidants*, **13**(1): 10-20.
- Karim, N., Chanudom, L. and Tangpong, J. 2018. Cytoprotective and anti-genotoxic effects of xanthone derivatives from *Garcinia mangostana* against H<sub>2</sub>O<sub>2</sub> induced PBMC cell and blood leukocytes damage of normal and type 2 diabetes volunteers. *Walailak Journal of Science and Technology*, 16(3): 143-153.
- Khojali, I.O. and Mohammed, M.Y. 2023. Bisabolene compound extracted from *Cassia fistula* and docked as antioxidant and vitamin E alternative predicted drug design. *GSC Advanced Research and Reviews*, **15**(1): 69-75.
- Kruttika, J., Deshpande, K.M., Rebello, S.J., Manikantan, P., Chaudhary, A., Alagesan, K.P., *et al.*, 2023. Phytochemicals as weapons against drug resistance. *Plant Science Today*, **10**(sp2): 212-219.
- Kumar, S., Srivastava, P., Mujwar, S. and Gupta, S. 2023. The anti-inflammatory and anti-arthriti activity of *Garcinia travancorica* in a rat model: A proof of concept with computational studies.

*Combinatorial Chemistry & High Throughput Screening,* **26***e*120623217888. [*https://doi.org/10.2174/1386207326666230612121712*].

Machinaga, N., Chiba, J., Hashimoto, R., Otoyo, M. and Inoue, R. 2012. Phosphoric acid ester derivative. US patent no. JPS62103097

Motulsky, H. 2018. GraphPad Prism User Guide. GraphPad Software Inc., San Diego, CA, USA.

- Nchiozem-Ngnitedem, V.A., Mukavi, J., Omosa, L.K. and Kuete, V. 2023. Phytochemistry and antibacterial potential of the genus *Garcinia*. *Advances in Botanical Research*, **107**: 105-175.
- Noreen, S., Khan, M.N., Tufail, T., Hassan, F. and Awuchi, C.G. 2023. Nutraceutical, functional, and therapeutic properties of *Garcinia cambogia*: A review. *International Journal of Food Properties*, **26**(1): 729-738.
- Perveen, S., Kanwal, S., Ali, S., Alqahtani, F., Rao, F., Asghar, A., et al., 2024. Role of phytochemicals in management of schizophrenia. p. 101. In: New Approaches to the Management and Diagnosis of Schizophrenia. Intechopen, London, UK. [https://doi.org/10.5772/intechopen.1004847].
- Prashanth, B.G. and Kachole, M.S. 2011. Effect of exogenous hydrogen peroxide on peroxidase and polyphenol oxidase activities in *Cajanus cajan* (L.) Millsp. detached leaves. *International Journal of Current Research*, **3**(10): 61-65.
- Priyadharisini, J. 2019. Preliminary phytochemical screening and gas chromatography and mass spectrometry analysis of chloroform extract of *Garcinia cambogia* fruits. *Asian Journal of Pharmaceutical and Clinical Research*, **12**(7): 176-178.
- Rizikiyan, Y., Sulastri, L., Indriaty, S., Khofifah, Y. and Falya, Y. 2022. Formulasi dan uji aktivitas antioksidan lotion ekstrak etanol kulit buah manggis (*Garcinia mangostana* L.) dengan metode DPPH (2,2-difenyl-1-pikrylhidrazyl). *Medical Science: Scientific Journal of Pharmacy*, 7(2): 147-160.
- Ruch, R.J., Cheng, S.J. and Klaunig, J.E. 1989. Prevention of cytotoxicity and inhibition of intracellular communication by antioxidant catechins isolated from Chinese green tea. *Carcinogenesis*, **10**: 1003-1008.
- Sahoo, S., Sinha, S. and Singh, A. 2022. Molecular docking study reveals α-santalol from *Santalum album* Linn. as a promising inhibitor of CD36 for atherosclerosis. *Cardiometry*, **24**: 353-359.
- Sharma, K., Adhikari, D., Kim, H.J., Oh, S.H., Oak, M.H. and Yi, E. 2019. *Citrus junos* fruit extract facilitates anti-adipogenic activity of *Garcinia cambogia* extract in 3T3-L1 adipocytes by reducing oxidative stress. *Journal of Nanoscience and Nanotechnology*, **19**(2): 915-921.
- Shetty, P., Rai, M., Ravindran, A., Gopalakrishna, H.N., Pai, V.R. and Kalal, B.S. 2022. Hypoglycemic and hypolipidemic effects of *Garcinia cambogia* extracts in streptozotocinnicotinamide induced diabetic rat model. *International Journal of Clinical and Experimental Pathology*, **15**(9): 380-387.
- Silprasit, K. 2022. Analysis of FTIR spectra, flavonoid content, and anti-tyrosinase activity of extracts and lotion from *Garcinia schomburgkiana* by multivariate method. *Trends in Sciences*, **19**(18): 5780. [*https://doi.org/10.48048/tis.2022.5780*].
- Singh, T., Kasture, S.B., Jaliwala, Y. and Singh, M. 2011. *In vitro* antioxidative activity of phenolic and flavonoid compounds extracted from of *Garcinia indica*. *International Journal of Pharmacy* & *Life Sciences*, **2**(3): 613-616.
- Singleton, V.L., Orthofer, R. and Lamuela-Raventós, R.M. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Oxidants and Antioxidants Part A*, **299**: 152-178.
- Talib, W.H., Baban, M.M., Azzam, A.O., Issa, J.J., Ali, A.Y., AlSuwais, A.K. et al., 2024. Allicin and cancer hallmarks. *Molecules*, **29**(6): 1320. [https://doi.org/10.3390/molecules29061320].
- Thandayamparambil, A.A., Manoharan, S., Balasubramanian, S. and Perumal, E. 2023. *Garcinia gummi-gutta*: Phytochemicals and pharmacological applications. *Biofactors*, **49**(3): 584-599.
- Thummajitsakul, S. and Irulandi, K., Geetha, S., Mehalingam, P. and Nirmalkumar, N. 2016. Evaluation of antioxidant properties and antimicrobial activity of *Garcinia cambogia* Hort. Ex

Boerl (Clusiaceae) fruit extracts. *Journal of Advanced Applied Scientific Research*, **1**(3): [*https://doi.org/10.46947/joaasr13201621*].

- Trease, G.E. and Evans, W.C. 1987. *Pharmacognosy* (13<sup>th</sup> edn.). Brailliar Tiridel, Macmillan Publishers, UK.
- Vij, I. and Pathania, A. 2023. An overview: Advances in chromatographic techniques in phytochemistry. E3S Web of Conferences, 391: 101038. [https://doi.org/10.1051/e3sconf/202339101038].
- Wairata, J., Fadlan, A., Purnomo, A.S. and Ersam, T. 2023. α-amylase activities of fraction in dichloromethane extract from the stem bark of *Garcinia forbesii*. Nucleation and Atmospheric Aerosols, 2642: 070004. [https://doi.org/10.1063/5.0110781].
- Waris, M., Koçak, E., Gonulalan, E.M., Demirezer, L.Ö., Kır, S. and Nemutlu, E. 2022. Metabolomics analysis insight into medicinal plant science. *TrAC Trends in Analytical Chemistry*, **157**: 116795. [116795.https://doi.org/10.1016/j.trac.2022.116795].
- Xu, L., Zhang, L.Z., Li, Y.H., Li, L.Y., Xie, Z.H. and Tu, Z.C. 2022. Inhibitory activity and mechanism of guavinoside B from guava fruits against α-glucosidase: Insights by spectroscopy and molecular docking analyses. *Journal of Food Biochemistry*, **46**(7): e14101. [https://doi.org/10.1111/jfbc.14101].
- Zhishen, J., Mengcheng, T. and Jianming, W. 1999. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food Chemistry*, **64**(4): 555-559.