

# Phytochemical Analysis of *Gongronema latifolium* Benth Leaf Using Gas Chromatographic Flame Ionization Detector

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## Abstract

The aim of this study is to analyze the phytochemicals present in the leaf of *Gongronema latifolium* (Benth). The leaf of *Gongronema latifolium* (Benth) was harvested and sun-dried. The dried leaf was milled to fine powder. The milled leaf powder was weighed and used for the phytochemical analysis. Gas chromatographic flame ionization detector was used to quantify the phytochemicals analyzed. The results show the presence of different types of Alkaloids, Flavonoids, Total phenolic compound, Lignan, Terpenes, Sterol, Allicin, Hydroxycinnamic acids, Saponin and Carotenoid. Some of the phytochemicals detected in high quantities among the different groups of phytochemicals analyzed in 100g of *Gongronema latifolium* (Benth) leaf include Cinchonidine 52.47mg, Oxoasosanine 43.51mg, Lupanine 35.65mg and Buphanidrine 33.33mg (Alkaloids), Hyperoside 37.54mg, Quercetin 31.03mg and Kaemferol 24.80mg (Flavonoids), Tannic acid 116.60mg, Ferulic acid 82.26mg and Vanillic acid 64.17mg (Total phenolic compound), Retusin 4.40mg and Galgravin 4.33mg (Lignan), Nerol (geraniol) 33.05mg and Beta pinene 32.79mg (Terpenes), 5-avenasterol 9.42mg and Stigmasterol 4.89mg (Sterol), Chlorogenic acid 48.87mg and Caffeic acid 23.01mg (Hydroxycinnamic acids), Saponine 59.11mg and Sapogenin 50.79mg (Saponin) and Beta-crytoxanthin 433.14mg, Xanthophylls 311.36mg and Carotene 158.36mg (Carotenoid). The result shows that *Gongronema latifolium* (Benth) leaf possess an appreciable level of phytochemicals. It could be a good raw material for the production of some medicinal drugs and can be used in folk medicine for the treatment of some diseases.

## Keywords

Phytochemicals, Medicine, *Gongronema latifolium* Benth, Diseases

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## 1. Introduction

*Gongronema latifolium* (Benth) is a perennial edible plant with soft and pliable stem. It is widely used in the West African sub-region for a number of medicinal and nutritional purposes. It belongs to the family of asclepiadaceae. *Gongronema latifolium* (whose leaves are bitter) is commonly called “utazi” and “arokeke” in South Eastern and South Western parts of Nigeria respectively. It is a tropical rainforest plant primarily used as spice and vegetable in traditional folk medicine [1, 2, 3].

According to Nwanjo *et al.* [4], phytochemical studies of *Gongronema latifolium* leaves show the presence of Glycosides, Alkaloids, Saponin, Tannin and Flavonoids. Egbung *et al.* [5] reported the presence of phytochemicals (tannins, saponins, alkaloids, flavonoids and hydrocyanide), mineral elements (Cr, Cu, Se, Zn and Fe) and vitamins (A, C, riboflavin, niacin and thiamine) in the root bark and twig extracts of *Gongronema latifolium*. However, the concentration of these phytochemicals varies among these plant parts. Tiwari and Rao [6] reported that the different composition of the active components in plants give medicinal plants an edge as better therapeutic agents than

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chemotherapy in management of different ailments such as atherosclerosis, hypertension and diabetes.

A cold infusion of the pounded stem of *Gongronema latifolium* is used to manage colic and intestinal symptoms usually associated with worms. The leaves are used to prepare food for nursing mothers, where it is believed to stimulate appetite, reduce post-partum contraction and enhance the return of the menstrual cycle. *Gongronema latifolium* not only possesses hypoglycemic activity, but also hypotensive, hepatoprotective and hypolipidemic effects [7, 2, 8]. *Gongronema latifolium* leaf exhibits biochemical and histological changes in acetaminophen-induced hepatic toxicity in albino rats and can be used against some hepatic inflammations [9].

Phytochemicals are defined as the substances found in edible fruits and vegetables that exhibit a potential for modulating human metabolism in a manner beneficial for the prevention of chronic and degenerative diseases [10]. Phytochemicals, as plant components with discrete bio-activities towards animal biochemistry and metabolism are being widely examined for their ability to provide health benefits [11]. Phytochemicals could provide health benefits as: substrates for biochemical reactions; cofactors of enzymatic reactions; inhibitors of enzymatic reactions; absorbents/sequestrants that bind to and eliminate undesirable constituents in the intestine; ligands that agonize or antagonize cell surface or intracellular receptors; scavengers of reactive or toxic chemicals; compounds that enhance the absorption and or stability of essential nutrients; selective growth factors for beneficial gastrointestinal bacteria; fermentation substrates for beneficial oral, gastric or intestinal bacteria; and selective inhibitors of deleterious intestinal bacteria [11]. Research supporting beneficial roles for phytochemicals against cancers, coronary heart disease, diabetes, high blood pressure, inflammation, microbial, viral and parasitic infections, psychotic diseases, spasmodic conditions, ulcers, etc is based on chemical mechanisms using *in vitro* and cell culture systems, various disease states in animals and epidemiology of humans.

The World Health Organization has also recognized the importance of traditional medicine and has created strategies, guidelines and standards for botanical medicines. Over the past decade, there has been a resurgence of interest in the investigation of natural materials as a source of potential drug substance [12]. In recent times, developed countries are turning to the use of traditional medicinal systems that involve the use of herbal drugs and remedies and according to the World Health Organization (WHO), almost 65% of the world's population has incorporated the value of plants as a methodology of medicinal agents into their primary modality of health care [12].

The objectives of this study were to evaluate the phytochemical composition of the leaf of *Gongronema latifolium* Benth and to create the public awareness of the possible uses or importance of *Gongronema latifolium* Benth leaf.

## 2. Material and Methods

### 2.1. Plant Material

The leaf of *Gongronema latifolium* (Benth) was harvested at Itaja-Amaegbu in Olokoro, Umuahia, Abia State, Nigeria. The plant was identified at the Department of Plant Science and Biotechnology, Abia State University, Uturu and voucher specimen deposited at the herbarium of the same department. The plant material was sun-dried and milled to fine powder. The milled leaf powder was weighed and used for the phytochemical analysis.

### 2.2. Determination of Phytochemicals

Phytochemical analysis of the leaf of *Gongronema latifolium* (Benth) was carried out with the use of gas chromatographic flame ionization detector. Identification was based on comparison of retention times.

### 2.3. Determination of Alkaloids

Alkaloids determination was carried out using the method of AOAC [13]. After the filtration and evaporation at reduced pressure, the resultant crude extract was treated with 5% aqueous HCl of about 7.5ml.

### 2.4. Determination of Flavonoids

The crude methanolic extract was obtained by pouring 100ml of the boiling methanol and (70:30) unto 10g of the plant material.

### 2.5. Determination of Phenolic Compounds

Phenolic compounds were extracted from pulverized sample according to the method described by AOAC [13].

### 2.6. Determination of Lignan

Extraction was carried out using the method of AOAC [13]. The lignin was removed by suction filtration and the filtrate shaking over night with hexane dichloromethane.

### 2.7. Determination of Terpenes

Extraction was carried out by method of AOAC [13]. The sample was pulverized and the terpenes constituents extracted with redistilled chloroform. The terpenes were removed with 10ml of the solvent for 15 minutes. The mixture was filtered and concentrated to 1ml in the vial for

gas chromatography analysis and 1ml was injected into the injection pot of GC.

## 2.8. Determination of Sterols

Sterols analysis was carried out following the modified method of AOAC [13]. The sample was saponified at 95°C for 30 minutes by using 3ml of 10% KOH in ethanol to which of benzene has been added to ensure miscibility. Deionized water (3ml) was added, 2mls of hexane was used in extracting non saponified particles (sterols).

## 2.9. Determination of Allicin

Allicin extraction was carried out using the modified method of Chehregani *et al.* [14].

## 2.10. Determination of Hydrocinnamic Acid

Extraction was carried out using the method of AOAC [13]. The sample was pulverized and the hydroxycinnamic acid constituents extracted with methanol. The hydroxycinnamic acids were removed with 10ml of the solvent for 15 minutes. The mixture was filtered and concentrated to 1ml in the vial for gas chromatographic analysis and 1ml was injected into the injection pot of GC.

## 2.11. Determination of Saponins

Extraction was carried out by method of AOAC [13]. The combined extract were concentrated to syrup under reduced pressure and then suspended in air. The suspension was extracted with petroleum ether, chloroform and 1-butanol saturated with water, successively to give the respective extract after removal of the solvent.

## 2.12. Determination of Carotenoids

The carotenoid extractions were carried out following the modified method of Takagi [15]. The extract were combined and evaporated under reduced pressure and the residue was re-extracted by a mixture of diethyl ether and petroleum ether in equal ration.

## 3. Results and Discussion

The results/findings of the phytochemical analysis are as presented in the tables below:

Alkaloids are naturally occurring chemical compounds that contain mostly basic nitrogen atoms. Alkaloids have a wide range of pharmacological activities including antimalarial (e.g. quinine), antiasthma (e.g. ephedrine), anticancer (e.g. homoharringtonine) [16], cholinomimetic (e.g. galantamine) [17], vasodilatory (e.g. vincamine), antiarrhythmic (e.g. quinidine), analgesic (e.g. morphine) [18], antibacterial (e.g. chelerythrine) [19], and antihyperglycemic activities (e.g.

piperine) [20]. Cinchonidine is believed to be used for treating malaria and reducing fever. Cinchonidine, Oxoasosanine, Lupanine and Buphanidrine among others were present in appreciable concentration in the leaf of *Gongronema latifolium* (Benth).

**Table 1.** Alkaloids analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Choline	1.07	4.74
2	Theophylline	0.57	5.61
3	Dillapiole	0.32	6.01
4	Angustifoline	18.86	6.61
5	Sparteine	6.49	7.02
6	Ellipicine	4.99	7.87
7	Lupanine	35.65	8.74
8	13-Alpha-hydroxyhombifoline	5.18	9.04
9	9-Octadecenamine	3.54	10.51
10	Dihydro-oxo-demethoxyhaemanthamine	2.64	11.84
11	Augustamine	21.83	12.60
12	Oxoasosanine	43.51	12.72
13	Shogaol	0.70	12.92
14	Piperline	0.90	13.08
15	Gingerdione	0.10	13.50
16	Capsaicin	2.79	13.72
17	Cinchonidine	52.47	13.93
18	Cinchonine	0.72	14.05
19	Crinane-3alpha-01	4.91	14.14
20	Buphanidrine	33.33	14.35
21	Indicine-N-oxide	25.36	15.23
22	Powelline	9.53	16.27
23	Undulatine	14.25	16.52
24	Ambelline	8.70	17.37
25	6-Hydroxybuphanidrine	13.65	18.15
26	Acronycine	0.88	18.78
27	Monocrotalline	1.90	19.01
28	6-Hydroxypowelline	0.03	19.50
29	Nitidine	0.41	20.04
30	Crinamidine	16.90	21.65
31	Ibeta, 2-beta-Epoxyambelline	0.86	22.41
32	6-Hydroxyundulatine	0.60	22.47
33	Epoxy-3, 7-dimethoxycrinane-11-one	2.85	23.16
34	Akuamidine	12.30	24.52
35	Echitamidine	9.96	24.63
36	Voacangine	12.17	24.75
37	Mitraphylin	4.53	25.33
38	Camptothecin	2.48	25.87
39	Echitamine	1.18	26.32
40	Colchicine	0.84	26.87
41	Emetine	0.22	27.25
42	Tetrandrine	0.88	27.44
43	Paclitaxel	1.43	27.74

Total = 368.38 (mg/100g)

These phytochemicals may be responsible or contributing to the acclaimed analgesic, anti-pyretic, protective, antibacterial, antiulceration and ameliorating effects of the leaf of

*Gongronema latifolium* (Benth). Many of these alkaloids have found use in traditional or modern medicine, or as starting materials for drug discovery. Other alkaloids possess psychotropic (e.g. psilocin) and stimulant activities (e.g. cocaine, caffeine, nicotine), and have been used in entheogenic rituals or as recreational drugs. Alkaloids can be toxic too (e.g. atropine, tubocurarine) [21].

**Table 2.** Flavonoids analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Catechin	8.05	16.72
2	Resveratrol	3.82	18.01
3	Genistein	3.75	18.48
4	Daidzein	3.71	18.77
5	Apigein	3.17	19.65
6	Butein	8.96	19.79
7	Naringenin	2.95	20.07
8	Biochanin	10.83	20.75
9	Luteolin	6.10	21.03
10	Kaempferol	24.80	22.50
11	Epicatechin	3.60	23.45
12	Epigallocatechin	5.88	24.80
13	Quercetin	31.03	25.34
14	Gallocatechin	6.95	25.83
15	Epicatechin-3-gallate	2.97	26.21
16	Epigallocatechin-3-gallate	2.30	26.95
17	Isorhamnetin	3.51	27.22
18	Robinetin	6.34	27.59
19	Ellagic acid	2.55	27.77
20	Myricetin	5.12	28.46
21	Baicalin	5.32	28.67
22	Nobicalin	10.82	28.85
23	Kaempferol-3,7,4,-trimethyl ether	4.07	29.02
24	Quercetin-3,7,4,-trimethyl ether	2.48	29.28
25	Baicalin	8.50	29.02
26	Tangeretin	4.63	29.49
27	Quercetin-3,7,3',4'-tetramethyl ether	4.21	29.72
28	Artemetin	3.97	29.81
29	Hyperoside	37.54	29.93
30	Silymarin	5.65	30.04
31	Kaempferol-3-Arabinoside	4.96	30.27
32	Quercitrin	5.58	30.46
33	Naringin	3.14	30.62
34	Isoquercetin	3.16	30.79
35	Oriebtin	3.28	30.90
36	Rutin	19.81	31.08
37	Isoorientin	2.42	31.22

Total = 278.71 (mg/100g)

Flavonoids have been reported to exert wide range of biological activities. These include: anti-inflammatory, antibacterial, antiviral, antiallergic, cytotoxic antitumour, treatment of neurodegenerative diseases and vasodilatory action [22]. In this study, flavonoids were found in an appreciable level.

Flavonoids are known to inhibit lipid-peroxidation, platelet aggregation, capillary permeability and fragility, cyclooxygenase and lipoxygenase enzyme activities. They exert these effects as antioxidants, free radical scavengers and chelators of divalent cation [23]. They are also reported to inhibit variety of enzymes like hydrolases, hyaluronidase, alkaline phosphatase, arylsulphatase, cAMP phosphodiesterase, lipase,  $\alpha$ -glucosidase, kinase [24]. Apigenin, luteolin, quercetin are known to possess anti-inflammatory activity. These phytochemicals are present in *Gongronema latifolium* (Benth) leaf, and therefore suggest the anti-inflammatory activity of the leaf of the plant. Kaempferol, quercetin, myricetin, fisetin are reported to possess COX and LOX inhibitory activities [25]. Kaempferol and myricetin are also found in the leaf of *Gongronema latifolium* (Benth). This suggests that the *Gongronema latifolium* leaf possesses COX and LOX inhibitory activities. In this study, hyperoside was found to be the highest (flavonoid) in concentration. Hyperoside has been reported to contribute to the antibacterial and antioxidant [26] properties of plant.

**Table 3.** Total phenolic compound analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	4-hydroxy benzaldehyde	5.81	10.65
2	4-hydroxybenzoic acid	16.26	13.87
3	4-hydroxybenzoic acid methyl ester	12.89	15.36
4	Vanillic acid	64.17	17.00
5	Gallic acid	10.38	18.08
6	Ferulic acid	82.26	19.89
7	Capsaicin	5.69	21.51
8	Rosmarinic acid	24.29	22.44
9	Tannic acid	116.60	24.44

Total= 338.36 (mg/100g)

Plant phenolic compounds are classified as simple phenols or polyphenols based on the number of phenol units in the molecule. Thus, plant phenolics comprise simple phenols, coumarins, lignins, lignans, condensed and hydrolyzable tannins, phenolic acids and flavonoids. The antioxidant properties of phenolic compounds are important in determining their role as protective agents against free radical-mediated disease processes [27]. Tannic acid and ferulic acid levels were high in the leaf analyzed. Tannins have been reported to exert many physiological effects, such as acceleration of blood clotting, reduction of blood pressure, decreasing the serum lipid level and modulating immunoresponses [28]. Ferulic acid, like many natural phenols, is an antioxidant *in vitro* in the sense that it is reactive towards free radicals such as reactive oxygen species (ROS). ROS and free radicals are implicated in DNA damage, cancer, and accelerated cell aging. Animal studies and *in vitro*

studies suggest that ferulic acid may have direct antitumor activity against breast cancer [29] and liver cancer [30]. Ferulic acid may have pro-apoptotic effects in cancer cells, thereby leading to their destruction [30]. Ferulic acid may be effective at preventing cancer induced by exposure to the carcinogenic compounds benzopyrene [31] and 4-nitroquinoline 1-oxide [32]. These suggest the beneficial biochemical effects of the leaf of *Gongronema latifolium* (Benth).

**Table 4.** Lignan analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	2-allyl-5ethoxy-4-methoxyphenol	1.91	9.47
2	(9E, 12E, 15E) -9, 12, 15-Octadecatrien-1-01	1.91	11.8
3	Apigenin-4, 7-methyl ether	2.34	13.82
4	Dehydroabietic acid	1.93	15.39
5	Retusin	4.40	16.66
6	Galgravin	4.33	18.21
7	Epieudesmin	2.04	20.11
8	Sakuranin	2.00	21.51

Total = 20.85 (mg/100g)

**Table 5.** Terpenes analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Alpha pinene	10.15	7.28
2	Beta pinene	32.79	8.63
3	Cis ocimene	2.67	9.04
4	Myrcene	1.65	9.85
5	Allo ocimene	2.38	10.09
6	Limonene	1.69	10.49
7	Camphene	1.71	10.95
8	Sabinene	1.42	11.33
9	Alpha thujene	1.50	11.85
10	Camphor	1.42	12.48
11	Neral	1.42	13.02
12	1, 8-cineole	10.71	13.23
13	Borneol	1.41	13.69
14	Linalool	15.96	13.79
15	Nerol (geraniol)	33.05	14.14
16	Alpha terpineol	1.42	14.34
17	Terpinen-4-ol	1.42	14.62
18	Citronellol	1.43	15.64
19	Ethyl cinnamate	2.01	15.35
20	Borneol acetate	1.71	15.72
21	Neryl acetate	1.41	16.39
22	Geranyl acetate	1.41	16.49
23	Taraxeron	1.41	17.35
24	Alpha amyirin	1.41	17.94
25	Beta amyirin	1.41	18.44
26	Lupeol	1.41	18.81
27	Alpha bergamotene	1.67	19.46

Total=138.08 (mg/100g)

Plant lignans are polyphenolic substances derived from phenylalanine via dimerization of substituted cinnamic alcohols, known as monolignols, to a dibenzylbutane skeleton. This reaction is catalysed by oxidative enzymes and

is often controlled by dirigent proteins. Lignans serve an antioxidant role in the plant's defense against biotic and abiotic factors, and have shown anti-inflammatory and antioxidant activity in basic research models of human diseases [33]. Lignans may also have anticarcinogenic activities. Some epidemiological studies have shown that lignan exposure associates with lower risk of breast cancer [34]. Experimental studies have confirmed that enterolactone, the intestinal metabolite of many dietary plant lignans, has anti-carcinogenic activity in experimental models of breast cancer [35, 36]. Retusin and Galgravin were high in concentration among the lignans present in the leaf. These and other lignan support the antioxidant activities of *Gongronema latifolium* (Benth) leaf.

A range of terpenes have been identified as high-value chemicals in food, cosmetic, pharmaceutical and biotechnology industries [37]. They are often strong-smelling. They may protect the plants that produce them by deterring herbivores and by attracting predators and parasites of herbivores [38]. Many terpenes are aromatic hydrocarbons and thus may have had a protective function [39]. Many terpenes were found in *Gongronema latifolium* (Benth) leaf. Some are alpha-pinene, beta pinene, nerol and linalool. Terpenes are believed to exhibit anti-inflammatory activity and antibiotic effect. It has been reported that, at low exposure levels, alpha-pinene is a bronchodilator in humans, and is highly bioavailable with 60% human pulmonary uptake with rapid metabolism or redistribution [40].

**Table 6.** Sterol analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Cholesterol	3.51	8.96
2	Cholestanol	1.87	17.05
3	Ergosterol	1.97	18.16
4	Campesterol	3.46	18.94
5	Stig-masterol	4.89	19.43
6	5-avenasterol	9.42	20.63
7	Sitosterol	3.26	21.56

Total= 28.38 (mg/100g)

Phytosterols are compounds that may be found in a great variety of different food products. Many studies have demonstrated their ability to reduce blood cholesterol levels in hyper- and normocholesterolemic subjects. Investigators report that phytosterol intakes of 2 to 3 g/ reduce low-density lipoprotein (LDL) cholesterol levels by about 10% in human subjects [41]. Other beneficial effects from phytosterols include anti-inflammatory and antipyretics. The most frequently encountered phytosterols belong to the group of 4-desmethyl sterols:  $\beta$ -sitosterol, campesterol and stigmasterol. These phytochemicals are present in *Gongronema latifolium* (Benth) leaf. This confirm the plant as a hypocholesterolemic and anti-inflammatory agent.

**Table 7.** Allicin analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Diallylthiosulphate	0.89	10.47
2	Methylallylthiosulphate	0.89	11.22
3	Allylmethylthiosulphate	0.89	12.82

Total = 2.67 (mg/100g)

Allicin is an oily, slightly yellow liquid that gives garlic its unique odor. It is a thioester of sulfenic acid and is also known as allyl thiosulfinate [42]. Its biological activity can be attributed to both its antioxidant activity and its reaction with thiol containing proteins [43]. Several animal studies indicate that allicin may reduce atherosclerosis and fat deposition [44], normalize the lipoprotein balance, decrease blood pressure [45], have anti-thrombotic and anti-inflammatory activities, and function as an antioxidant to some extent [46]. The three allicins present in *Gongronema latifolium* (Benth) leaf may suggest its antioxidant activity and lipoprotein balance, though they were of low concentrations in the leaf.

**Table 8.** Hydroxycinnamic acids analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	p-coumarin	1.29	5.72
2	p-coumaric acid	7.73	8.66
3	Caffeic acid	23.01	11.94
4	Scopoletin	2.28	13.41
5	Chlorogenic acid	48.87	15.82
6	Chicoric acid	4.49	17.21

Total = 87.67 (mg/100g)

Hydroxycinnamic acids (such as ferulic, caffeic, sinapic, and p-coumaric acids) is a group of compounds highly abundant in food that may account for about one-third of the phenolic compounds in our diet [47]. Hydroxycinnamic acids have gained an increasing interest in health because they are known to be potent antioxidants. Hydroxycinnamic acids, such as p-coumaric, caffeic, ferulic, and sinapic acids, are known to play an important role in nature. Research data have revealed that Hydroxycinnamic acids can be used for preventive and/or therapeutic purposes in several diseases related to oxidative stress (e.g., atherosclerosis, inflammatory injury, cancer, and cardiovascular diseases) [47]. Caffeic acid, Chlorogenic acid, p-coumaric acid and chicoric acid were all present in the leaf of *Gongronema latifolium* (Benth). Chlorogenic acid, caffeic acid and p-coumaric acid are known as antioxidants and exhibit immunomodulatory and anti-inflammatory activities. Onakpoya *et al.* [48] report modest, but significant, blood pressure lowering effects from chlorogenic acid administration.

**Table 9.** Saponin analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Hispogenin	7.11	20.48
2	Solagenin	9.91	21.05
3	Diosgenin	4.19	22.22
4	Tigogenin	6.90	22.91
5	Neochlorogenin	15.11	23.59
6	Hecogenin	8.54	24.59
7	Sapogenin	50.79	25.72
8	Tribuloin	4.89	26.35
9	Yanogenin	7.89	27.09
10	Conyzorgin	6.77	27.91
11	Saponine	59.11	29.41

Total = 181.21 (mg/100g)

Saponins are low molecular weight secondary plant constituents containing either a tetracyclic steroidal or a pentacyclic triterpenoid aglycone with one or more sugar chains [49]. Saponins have been variously attributed with a diverse range of properties, some of which include both beneficial and detrimental effects on human health, pesticidal, insecticidal and molluscicidal activity, allelopathic action, antinutritional effects, sweetness and bitterness, and as phyto-protectants that defend plants against attack by microbes and herbivores [50]. The saponins are also responsible for lowering cancer risks by lowering the blood cholesterol levels. A high saponin diet can be used in the inhibition of dental caries and platelet aggregation, in the treatment of hypercalciuria in humans, and as an antidote against acute lead poisoning. In epidemiological studies, saponins have been shown to have an inverse relationship with the incidence of renal stones. They are also responsible for many other important activities- Molluscicidal, Anthelmintic, Antiulcerogenic, Anticancer, Antioxidant, Immunomodulatory, Anti-malarial, Anti-bacterial, Analgesic, Anti-nociceptive, hepatoprotective [51]. It is possible that the saponins present in *Gongronema latifolium* (Benth) contribute to these activities. Some of the saponins present include saponine, sapogenin, neochlorogenin and solagenin.

The carotenoid have been extensively studied as antioxidants [52]. These compounds are highly pigmented, being yellow, orange and red, and present in fruits and vegetables. Carotenoids comprise two types of molecules, carotenes and xanthophylls. As reviewed by Bendich [53], the carotenes, including  $\gamma$ -carotene, lycopene and lutein, protect against uterine, prostate, breast, colorectal and lung cancers. Carotene, beta-cryptoxanthin, xanthophylls, lutein and lycopene were all present in the leaf of *Gongronema latifolium* (Benth). Therefore, they may be responsible for these activities. They may also protect against risk of digestive tract cancer [54]. To be effective, lycopene must be absorbed and distributed to the tissues. The xanthophyll type



of carotenoids offer protection to other antioxidants, and they may exhibit tissue specific protection [55]. Zeaxanthin, cryptoxanthin and astaxanthin are members of the xanthophyll group.

**Table 10.** Carotenoid analysis of *Gongronema latifolium* (Benth) leaf.

S/NO	NAME (PHYTOCHEMICAL)	AMOUNT (mg/100g)	RETENTION TIME (min)
1	Malvidin	2.84	16.10
2	Carotene	158.36	17.11
3	Lycopene	2.94	18.01
4	Beta-crytoxanthin	433.14	18.94
5	Lutein	31.45	19.18
6	Anther-xanthin	2.86	21.46
7	Asta-xanthin	1.56	22.19
8	Viola-xanthin	26.69	22.93
9	Neo-xanthin	43.86	23.59
10	Xanthophylls	311.36	23.61

Total= 1,015.06 (mg/100g)

## 4. Conclusion

The leaf of *Gongronema latifolium* (Benth) examined show the presence of different types of Alkaloids, Flavonoids, Total phenolic compound, Lignan, Terpenes, Sterol, Allicin, Hydroxycinnamic acids, Saponin and Carotenoid; with some in an appreciable concentrations. These phytochemicals contribute to the acclaimed medicinal properties of *Gongronema latifolium* leaf. The result shows that *Gongronema latifolium* (Benth) leaf is a good raw material for the production of some medicinal drugs and can be used in folk medicine for the treatment and prevention of some diseases.

## References

- [1] Ugochukwu NH, Babady NE. Antioxidant effects of *Gongronema latifolium* in hepatocytes of rat models of non-insulin dependent diabetes mellitus. *Fitoterapia*, 2002;73:612-618.
- [2] Ugochukwu NH, Babady NE, Cobourne M, Gasset SR. The effect of *Gongronema latifolium* extracts on serum lipid profile and oxidative stress in hepatocytes of diabetic rats. *J. Biosci.*, 2003;20(1):1-5.
- [3] Chinedu I, Uhegbu FO, Imo CK, Ifeanchi NG. Ameliorating effect and haematological activities of methanolic leaf extract of *Gongronema latifolium* in acetaminophen-induced hepatic toxicity in wistar albino rats. *International Journal of Biosciences*, 2013;3(11):183-188.
- [4] Nwanjo HU, Okafor MC, Oze GO. Anti-lipid peroxidative activity of *Gongronema latifolium* in streptozotocin induced diabetes rats. *Niger. J. Physiol. Sci.* 2006;221(2):61-65.
- [5] Egbung GE, Atangwho IJ, Iwara IA, Eyong UE. Micronutrient and phyto-chemical composition of root bark and twig extracts of *Gongronema latifolium*. *Journal of Medicine and Medical Sciences*, 2011;2(11):1185-1188.
- [6] Tiwari AK, Roa M. Diabetes mellitus and multiple therapeutic approaches of phytochemical. Present status and future prospects. *Current Science*, 2002;83:30-38.
- [7] Ugochukwu NH, Babady NE. Antihyperglycaemic of effect aqueous and ethanolic extracts of *Gongronema latifolium* leaves on glucose and glycogen metabolism in livers of normal and streptozotocin induced diabetic rats. *Life Sci.*, 2003;73(150):1925-1938.
- [8] Nwanjo HU, Alumanah EO. Effect of aqueous extract of *Gongronema latifolium* leaf on some indices of liver function in rats. *Global J. Med. Sci.*, 2005;4(1):29-32.
- [9] Imo C, Friday OU, Ifeanchi NG, Egbeigwe O, Ezekwe AS. Biochemical and histological changes associated with methanolic leaf extract of *Gongronema latifolium* in acetaminophen-induced hepatic toxicity in wistar albino rats. *International Journal of Biomolecules and Biomedicine*, 2014;4(2):1-7.
- [10] Tripoli E, Guardia ML, Giammanco S, Majo DD, Giammanco M. Citrus flavonoids: Molecular structure, biological activity and nutritional properties: A review. *Food Chemistry*, 2007;104:466-479.
- [11] Cora JD, Bruce GJ. Review Phytochemicals: nutraceuticals and human health. *Journal of the Science of Food and Agriculture*, 2000;80: 1744-1756.
- [12] Manas KM, Pratyusha B, Debjani N. Phytochemicals – biomolecules for prevention and treatment of human diseases-a review. *International Journal of Scientific and Engineering Research*, 2012;3(7):1-32.
- [13] Association of official Analytical Chemist. Official method of analysis. Washington DC, USA, 2009.
- [14] Chehregani AH, Sabounchi SJ, Jodian V, Pak J. Antibacterial effect of N-NaphtylenDiamine Platinum (II) chloride as novel compound. *Journal of Biological Sciences*, 2007; 10(4): 641-644.
- [15] Takagi S. Determination of green leaf carotenoids by HPLC. *Agric Bio Chem.*, 1985; 49: 1211-1213.
- [16] Kittakoop P, Mahidol C, Ruchirawat S. Alkaloids as important scaffolds in therapeutic drugs for the treatments of cancer, tuberculosis, and smoking cessation. *Curr Top Med Chem.*, 2014;14 (2): 239–252.
- [17] Russo P, Frustaci A, Del Bufalo A, Fini M, Cesario A. Multitarget drugs of plants origin acting on Alzheimer's disease. *Curr Med Chem.*, 2013;20(13): 1686–1693.
- [18] Raymond SS, Jonathan SJ, Michael WJ. The Essence of Analgesia and Analgesics. Cambridge University Press. 2010;pp.82–90.
- [19] Cushnie T, Cushnie B, Lamb A. Alkaloids: An overview of their antibacterial, antibiotic-enhancing and antivirulence activities. *Int J Antimicrob Agents*, 2014;44(5):377–386.
- [20] Qiu S, Sun H, Zhang A, Xu H, Yan G, Han Y. Natural alkaloids: basic aspects, biological roles, and future perspectives. *Chin J Nat Med.*, 2014;12(6):401–406.

- [21] Robbers JE, Speedie MK, Tyler VE. Alkaloids. Pharmacognosy and Pharmacobiotechnology. Philadelphia: Lippincott, Williams & Wilkins. 1996;pp.143-185
- [22] Tsuchiya H. Structure-dependent membrane interaction of flavonoids associated with their bioactivity. Food Chemistry, 2010;120:1089-1096.
- [23] Cook NC, Samman S. (1996). Flavonoids: Chemistry, metabolism, cardioprotective effects and dietary sources. Nutritional Biochemistry, 1996;7:66-76.
- [24] Narayana KR, Reddy SR, Chaluvadi MR, Krishna DR. Bioflavonoids classification, pharmacological, biochemical effects and therapeutic potential. Indian Journal of Pharmacology, 2001;33:2-16.
- [25] Tapas AR, Sakarkar D, Kakde RB. Flavonoids as nutraceuticals: A Review. Tropical Journal of Pharmaceutical Research, 2008;7:1089-1099.
- [26] Ibtissem B. Antioxidant and antibacterial properties of *Mesembryanthemum crystallinum* and *Carpobrotus edulis* extracts. Advances in Chemical Engineering and Science, 2012;2(3):359.
- [27] de Beer D, Joubert E, Gelderblom WCA, Manley M. Phenolic Compounds: A Review of Their Possible Role as *In Vivo* Antioxidants. South African Journal for Enology and Veticulture, 2002;23:48-61.
- [28] Chung K, Wong TY, Wei C, Huang Y, Lin, Y. Tannins and Human Health: A Review. Critical Reviews in Food Science and Nutrition, 1998;38 (6): 421-464.
- [29] Bouftira I, Chedly A, Souad S. Antioxidant and Antibacterial Properties of *Mesembryanthemum crystallinum* and *Carpobrotus edulis* Extracts. Advances in Chemical Engineering and Science, 2012;2(3):359-365.
- [30] Gelinas P, McKinnon CM. Effect of wheat variety, farming site, and bread-baking on total phenolics. International Journal of Food Science and Technology, 2006;41(3):329.
- [31] Beejmohun V, Fliniaux O. Microwave-assisted extraction of the main phenolic compounds in flaxseed. Phytochemical Analysis, 2007;18(4):275-285.
- [32] Zory Q, Byung-Kee B. Phenolic Compounds of Barley Grain and Their Implication in Food Product Discoloration. J. Agric. Food Chem., 2006;54(26):9978-9984.
- [33] Korkina L, Kostyuk V, De Luca C, Pastore S. Plant phenylpropanoids as emerging anti-inflammatory agents. Mini reviews in medicinal chemistry, 2011;11(10):823-835.
- [34] Adlercreutz H. Lignans and human health. Critical reviews in clinical laboratory sciences, 2007;44 (5-6):483-525.
- [35] Bergman JM, Thompson LU, Dabrosin C. Flaxseed and its lignans inhibit estradiol-induced growth, angiogenesis, and secretion of vascular endothelial growth factor in human breast cancer xenografts in vivo. Clinical cancer research: an official journal of the American Association for Cancer Research, 2007;13 (3):1061-1067.
- [36] Lindahl G, Saarinen N, Abrahamsson A, Dabrosin C. Tamoxifen, flaxseed, and the lignan enterolactone increase stroma- and cancer cell-derived IL-1Ra and decrease tumor angiogenesis in estrogen-dependent breast cancer. Cancer research, 2011;71(1):51-60.
- [37] Thimmappa R, Geisler K, Louveau T, O'Maille P, Osbourn A. Triterpene biosynthesis in plants. Annu Rev Plant Biol., 2014;65:225-257.
- [38] Martin DM, Gershenzon J, Bohlmann J. Induction of Volatile Terpene Biosynthesis and Diurnal Emission by Methyl Jasmonate in Foliage of Norway Spruce. Plant Physiology, 2003;132(3):1586-1599.
- [39] Pichersky E. Biosynthesis of Plant Volatiles: Nature's Diversity and Ingenuity. Science, 2006;311(5762):808-811.
- [40] Russo EB. Taming THC: potential cannabis synergy and phytocannabinoid-terpenoid entourage effects. British Journal of Pharmacology, 2011;163(7):1344-1364.
- [41] Katan MB, Grundy SM, Jones P, Law M, Miettinen TA, Paoletti R. Efficacy and safety of plant stanols and sterols in the management of blood cholesterol levels. Mayo Clin Proc, 2003;78:965-978.
- [42] Nikolic V, Stankovic M, Nikolic LJ, Cvetkovic D. Mechanism and kinetics of synthesis of allicin. Pharmazie, 2004;59(1):10-14.
- [43] Rabinkov A, Miron T, Konstantinovski L, Wilchek M, Mirelman D, Weiner L. The mode of action of allicin: trapping of radicals and interaction with thiol containing proteins. Biochim Biophys Acta, 1998;1379 (2):233-244.
- [44] Abramovitz G, Gavri S, Harats D, Levkovitz H, Mirelman D, Miron T. et al. Allicin-induced decrease in formation of fatty streaks (atherosclerosis) in mice fed a cholesterol-rich diet. Coron. Artery Dis., 1999;10 (7):515-519.
- [45] Elkayam A, Mirelman D, Peleg E, Wilchek M, Miron T, Rabinkov A. et al. The effects of allicin on weight in fructose-induced hyperinsulinemic, hyperlipidemic, hypertensive rats. Am. J. Hypertens, 2003;16(12):1053-1056.
- [46] Lindsey JM, Bernhard HG, Veena V, Michael B, Samer RE, SunWook H. et al. The pungency of garlic: Activation of TRPA1 and TRPV1 in response to allicin. Current Biology, 2005;15(10):929-934.
- [47] José T, Alexandra G, Manuela GE, Jorge G, Fernanda B. Hydroxycinnamic Acid Antioxidants: An Electrochemical Overview. BioMed Research International, 2013;2013:1-11.
- [48] Onakpoya IJ, Spencer EA, Thompson MJ, Heneghan CJ. The effect of chlorogenic acid on blood pressure: a systematic review and meta-analysis of randomized clinical trials. Journal of Human Hypertension, 2015;29(2):77-81.
- [49] Vincken JP, Heng L, Groot AD, Gruppen H. Saponins, classification and occurrence in the plant kingdom. Phytochem., 2007;68:275-297.
- [50] Haralampidis, K., Trojanowska, M. and Osbourn, A. Biosynthesis of triterpenoid saponins in plants. Adv. Biochem. Eng. Biotechnol., 2002;75:31-49.
- [51] Tadros MM, Ghaly NS, Moharib MN. Molluscicidal and schistosomicidal activities of a steroidal saponin containing fraction from *Dracaena fragrans* (L.). J Egypt Soc Parasitol., 2008;38:585-598.
- [52] Liebler DC. Antioxidant reactions of carotenoids. Ann NY Acad Sci., 1996;691:20-31.
- [53] Bendich AJ. Carotenoids and the immune response. J Nutr., 1989;119:112-115.



- [54] Francheschi S, Bidoli E, La Vecchia C, Talamini R, D'Avanzo B, Negri E. Tomatoes and risk of digestive-tract cancer. *Int J Cancer*, 1994;50:181-184.
- [55] Parker RS. Carotenoids in human blood and tissues. *J Nutr.*, 1989;119:101-104.