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Phytochemical Profiles, *In Vitro* Antioxidant, Anti Inflammatory and Antibacterial Activities of Aqueous Extract of *Terminalia catappa* L. leaves

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Abstract

The aim of the study was to analyze the antioxidant, antibacterial and anti-inflammatory potential of aqueous extract of *Terminalia catappa* leaves. Antioxidant capability was studied using DPPH, Nitric oxide, superoxide scavenging assays and *in vitro* anti-inflammatory activity was evaluated using Protein denaturation assay, protease inhibition assay and were subjected for gas chromatography-mass spectrometry (GC-MS) analysis to identify its bioactive components. Antibacterial potential was analysed using agar well diffusion method against Gram negative and Gram positive bacteria. Aqueous leaves extract of *T.catappa* was assessed for its antioxidant and anti-inflammatory activity by *in vitro* method. Preliminary phytochemical screening confirmed the presence of alkaloids, flavonoids, phenols, tannins, triterpinoids, Amino acids, Carbohydrate and absence of glycosides and sterols. Remarkable free radicals scavenging ability was observed in all the tested radicals namely, DPPH**.** (IC50 = 272.76±3.41µg/ml), nitric oxide (NO**.**) (IC50 = 126.96±0.35µg/ml) and Superoxide ($O_2^{\cdot-}$) (IC50 = 336.23±6.26µg/ml) and anti-inflammatory assay also observed Protein denaturation (IC50 = 384.02±4.81µg/ml), Protease inhibition (IC50 = 384.02±4.81µg/ml). In the antibacterial assay, the zone of inhibition was recorded as 9.5 ± 0.5, 12.0 ± 1.21, 6.5 ± 0.35, 5.5 ± 0.5 and 8.5 ± 1.0 for the strains of *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas* spp., *Staphylococcus aureus*, and *Bacillus* spp. respectively. The gas chromatography–mass spectrometry (GC–MS) profiling was exhibited the presence of 19 major components highly accountable for its pharmaceutical activities.

Keywords: *Terminalia catappa* L., Antioxidant, Antibacterial, Anti-inflammatory, DPPH.

1. INTRODUCTION

Oxidative stress is initiated due to disparity amid formation of free radicals and detoxification potential of a biological system [1,2]. Both enzymatic and non-enzymatic reactions in the cell can incessantly produce the free radicals instigating wide-ranging impairment to tissues and also in several medical ailments [3]. However, the cells are shielded from the injury produced by uninhibited radicals by antioxidants [4]. Phenolic acids, polyphenols and flavonoids have the potential antioxidants properties and aid in scavenging free radicals [5].

Inflammatory abnormalities underlie a vast variety of human diseases. The immune system is often involved in inflammatory disorders and is demonstrated in both allergic reactions and myopathies. Non-immune diseases with inflammation include atherosclerosis and ischemic conditions [6]. Anti-inflammatory drugs are divided into two classes: Non steroids anti-inflammatory drugs (NSAIDs) and Corticosteroids. NSAIDs are known analgesics, anti pyretics and anti-inflammatory drugs. Their analgesic and anti-inflammatory effects are mainly due to inhibition of prostaglandin synthesis in the inflamed tissues and therefore it is on a peripheral level. Aspirin is the oldest NSAID and is very effective. It is used at far lower doses to inhibit clotting of blood and prevent strokes and heart attacks. However, up to 50% of patients are unable to tolerate the adverse effects i.e. nausea, vomiting, epigastric pain and tinnitus that are caused by high doses of Aspirin that often administered to produce effective anti-inflammatory activity. Due to the serious side effects produced by the synthetic drugs a trend from the usage of synthetic drugs to herbal medicine is observed among mankind which can be called 'Return to Nature'. Antioxidant and anti-inflammatory principles present in the natural resources are providing enormous scope in herbal medicine. Therefore, in the recent years, research interest is centered on phytochemicals that are derived from herbal sources in view of their therapeutic benefits [7]. Phytochemicals are commonly available with less toxic effect and serving as medicinal components have been suggested to reduce threat of adverse effects of ROS [8].

Terminalia catappa L., a large spreading tree belongs to the family combretaceae, is distributed throughout the tropics in coastal environments. The dried leaves are used as an alternative to antibiotics to control fish pathogens [9]. The leaves are also reported to possess antioxidant and anti-clastogenic properties [10]. Various extracts of leaves and bark of *Terminalia catappa* have been reported to exhibit antibacterial [11], anti-inflammatory [12], antioxidant and anti-tumor [13], anti-HIV [14] and hepato-protective [15] and anti-diabetic properties [16] besides being aphrodisiac [17]. The moderate consumption of the seed kernel is useful in treating sexual dysfunction among men, primarily for premature ejaculation [17]. The ethanol extract of the leaves of *Terminalia catappa* inhibits osmotically-induced hemolysis of human erythrocytes in a dose-dependent manner [15]. Punicalagin and punicalin isolated from the leaves are used to treat dermatitis and hepatitis as both have strong anti-oxidative activity [12]. Hence, the present work is focused on evaluating the antioxidant and anti-inflammatory properties of phytochemical aqueous leaves extract obtained from Indian almond tree *Terminalia catappa* L.

2. MATERIALS AND METHODS

2.1 Plant Material Collection and Processing

The fresh leaves of *Terminalia catappa* were collected from Dr. N.G.P. Arts and Science College, Coimbatore and rinsed with tap water then the leaves were dried under shade at room temperature for 2 weeks. The dried leaves were

powdered using an electric grinder. 20 g of dry leaves powder with 1 L distilled water and heated at 80 °C for 20 min on a hot plate. This solution was filtered by filter paper (Whatman No 42, Maidstone, England) and stored at 4 °C for further experiments.

2.2 Phytochemical Analysis

Qualitative Phytochemical Analysis The phytochemical screening of *T. catappa* Leaves aqueous extract is assayed by standard methods (Harborne and Trease 1978) [18]. The screening was carried out to discover the significant bioactive components such as tannins, saponins, flavonoids, phenols, terpenoids, alkaloids, glycosides, cardiac glycosides, coumarins, and steroids.

2.2.1Quantitative Phytochemical Analysis

2.2.1.1. Determination of Total Phenol Content -The total phenolic content of *T. catappa* Leaves aqueous was determined according to the method described by Siddhuraju and Becker (2003) [19].

2.2.1.2 Determination of Total Tannin Content -Tannins were estimated after treatment with polyvinylpolypyrrolidone (PVPP) according to Siddhuraju and Manian (2007) [20].

Determination of Total Flavonoid Content The flavonoid contentwas determined by the use of a slightly modified colorimetry method described previously by Zhishen *et al*. (1999) [21].

2.2.2 In-Vitro Antioxidant Activities

Different concentrations (100–500 µg/ml) of *T. catappa* Leaves aqueous extract (GFPAE) were tested for various types of radicals scavenging potential. Ascorbic acid was used as standard reference compounds for all in vitro antioxidant assays.

2.2.2.1 Free Radical Scavenging Activity on DPPH.

The antioxidant activity of the sample was determined in terms of hydrogen donating or radical scavenging ability, using the stable radical DPPH, according to the method of Blois (1958) [22].

2.2.2.2 Superoxide Radical Scavenging Activity

Superoxide radicals were generated by a modified method of Beauchamp and Fridovich (1971) [23].

2.2.2.3 Nitric Oxide Radical Scavenging Activity

The nitric oxide scavenging activity of the sample was measured according to the method of Sreejayan and Rao (1997) [24].

2.2.3 In-Vitro Antiinflammatory Activity of T. catappa Leaves aqueous extract

2.2.3.1 Inhibition of Protein Denaturation

Protein denaturation assay was done according to the method described by Gambhire *et al*. (2018) [25].

2.2.3.2 Protease Inhibitory Activity

Proteinase inhibitory activity of the leaf extracts was performed according to the method of Sakat *et al*., [26].

2.2.4 In-Vitro Antibacterial Activity of T. catappa Leaves aqueous extract

The antibacterial efficacy of the *T. catappa* Leaves aqueous extract 30 µl was tested against the organism like *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumonia*, *Pseudomonas* spp. And *Bacillus* spp. by disc diffusion according to Rodriguez- Carpena *et al*. (2011) [27] with minor changes. A 30 µl of sterile distilled water and standard antibiotic (Tetracyclin-20 µg/ml) were loaded in separate disc denoting the negative and positive control respectively.

2.3 Gas Chromatography–Mass Spectrometry (GC–MS) Analysis

The main phytocomponents of *T. catappa* Leaves aqueous extract (GFPAE) were identified by using GC–MS detection system. The samples were suspended with ethanol and subjected to GC–MS analysis. Elucidation of phytocomponents was assayed by comparison of their retention times and mass with their regular authentic standard spectra using computer searches in NIST0. L and Wiley7n.l libraries [28].

3. Results And Discussion

The genus Terminalia harbours a number of species with phyto pharmaceutical significance such as *T. chebula*, *T. arjuna* and *T. bellerica*, *T. chebula* and *T. bellerica* along with *Emblica officinal* is constitute triphala - an Ayurvedic medicine with much therapeutic and rejuvenating potential. All the parts such as leaves, fruits, seeds, wood and bark of the selected species *Terminalia catappa* are found to be useful in treating various human ailments. But still, intensive research needs to be carried out to provide scientific evidence. Little or no evidences are available validating the pharmacological activities of aqueous leaves extracts. Hence in the present work, attempts were taken to evaluate the antioxidant, antibacterial and anti-inflammatory potentials of aqueous leaves extracts *T. catappa*.

3.1 Phytochemical Screening

Phytochemical screening of *T. catappa* Leaves aqueous extract (TCLAE) confirmed the presence of alkaloids, flavonoids, phenols, tannins, glycosides, Amino acids and Carbohydrate in different qualitative ranges. The negative sign depicted the absence of Triterpenoids and sterols (Table 1). Among the nine phytoconstituents, phenol, flavonoid, and tannin showed a strong presence depicting the pharmaceutical property of the *T. catappa* Leaves aqueous extract. Hence all these secondary metabolites in the extracts were quantified further to assay the best extraction possessing the significant levels of bioactive components (Table 2).

Table .1 Qualitative phytochemical screening of *T. catappa* Leaves aqueous extract

S.NO	Test	Inference [presence (+); absence (−)]
1	Alkaloids	+
2	Flavoniods	+
3	Tannins	+
4	Phenols	+
5	Aminoacids	+
6	Carbohydrates	+
7	Proteins	+
8	Phlobatannins	+
9	Saponins	+
10	Glycosides	-
11	Triterpenoids	+
12	Steroids	-

Table.2 Total phenol, tannin and flavonoid content of *T. catappa* Leaves aqueous extract

Phytochemicals	Content
Total phenols	77.78±8.39 (mg/g Gallic acid equivalent)
Tannins	13.23±0.27 (mg/g Gallic acid equivalent)
Total flavonoids	1.39±0.01 (mg/g Quercetin equivalent)

Values are expressed as mean ± SD of triplicates

Total phenolics content in the *T. catappa* Leaves aqueous extract was 77.78±8.39 mg of Gallic Acid equivalents (GAE)/g. Whereas, total tannin content in the TCLAE was 13.23±0.27 mg of Tannic acid equivalents (TAE)/g. Total flavonoid content in the TCLAE was 1.39±0.01 mg of Quercetin equivalent (RE)/g (Table 2).

Plant materials rich in phenolics are increasingly being used in the food industry because they retard oxidative degradation of lipids and improve the quality and nutritional value of food [29]. Phenolic compounds are considered secondary metabolites and these phytochemical compounds derived from phenylalanine and tyrosine occur ubiquitously in plants and are diversified [30]. Phenolic compounds of plants are also very important because their hydroxyl groups confer scavenging ability. Phenolic compounds of plants fall into several categories; chief among these are the flavonoids which have potent antioxidant activities [31].

Tannin levels were predominantly found high *T. catappa* Leaves aqueous extract showing the better extraction properties of water as a solvent (Table 2). Prasad *et al*., 2008 [32] have reported that tannins possess significant antimicrobial agents and contains water-soluble polyphenols and exist as precipitated proteins in many plant foods. It is also found to prevent the growth of microorganisms by precipitating the microbial proteins and also inhibits the growth of many fungi, yeasts, bacteria, and viruses. Gulcin *et al*., 2010 [33] have reported that high levels of tannins are responsible for free radical scavenging and antioxidant efficacy.

In the present analysis, the levels of flavonoids were considerably high in *T. catappa* Leaves aqueous extract elucidating the importance of the extract as a potent medicinal agent. Flavonoids are known to be synthesized by plants as a reflection to microbial infection. Hence it should not be surprising that they act as profound antibacterial substances against a wide range of infectious agents [34].Flavonoids are naturally occurring in plants and are thought to have positive effects on human health. Studies on flavonoidic derivatives have shown a wide range of antibacterial, antiviral, anti-inflammatory, anticancer, and anti-allergic activities [35,36]. Flavonoids have been shown to be highly effective scavengers of most oxidizing molecules, including singlet oxygen, and various free radicals [37] implicated in several diseases. So comparable with the findings in the literature for other extracts of plant products [38] our results suggested that phenolic acids, tannins and flavonoids may be the major contributors for the antioxidant activity.

3.2 *In-vitro* Antioxidant Activity

In the present study, *T. catappa* Leaves aqueous extract (TCLAE) was analyzed for an antioxidant activity via antioxidant assays (Table 3). The TCLAE, when subjected to DPPH assay, showed maximum free radical scavenging activity of 38.15±0.88% at a concentration of 500 µg when compared with an IC 50 value of 272.76±3.41 (µg/ml), thus elucidating the importance of the TCLAE as a potent anti-oxidant agent (Table 3).

Superoxide anion ($O_2^{\bullet-}$) is an extremely reactive compound synthesized when oxygen is reduced by a single electron and may be produced during the regular catalytic role of various enzymes. Studies report that superoxide anion is highly harmful to cellular components. Robak and Glyglewski, 1988 [35] studied that flavonoids are the most effective antioxidants because they scavenge a large range of superoxide anions. In the present study, the superoxide radical scavenging activities of the plant extract had markedly increased with concentrations. The results indicate that the radical scavenging ability of *T. catappa* Leaves aqueous extract was found to be 23.12±0.24% at a concentration of 500 µg with an IC 50 value of 336.23±6.26 (µg/ml) (Table 3).

Nitric oxide is a free radical component generated by endothelial cells, macrophages, and neuron, etc., and involved in the modulation and regulation of various physiological processes [36]. Excess concentration is connected with the onset of several diseases. It reacts with oxygen to produce its stable products of nitrate and nitrite through intermediates NO_2, N_2O_4, and N_3O_3. In the present analysis, nitric oxide radical scavenging ability of *T. catappa* Leaves aqueous extract was found to be 49.92±0.48% at a concentration of 500 µg with an IC 50 value of 126.96±0.35 (µg/ml) (Table 3).

Table.3 *In-vitro* antioxidant activity of *T. catappa* Leaves aqueous extract

Anti-oxidant assay	Sample concentration (µg/reaction volume)	Percentage inhibition %	IC50 (µg/ml)
DPPH radical scavenging activity	100	5.66±0.70	272.76±3.41
	200	10.97±0.28	
	300	22.75±1.45	
	400	31.09±1.04	
	500	38.15±0.88	
Super oxide radical scavenging activity	100	6.32±0.47	336.23±6.26
	200	8.78±0.74	
	300	13.74±0.12	
	400	17.76±1.06	
	500	23.12±0.24	
Nitric oxide radical scavenging activity	100	15.78±1.45	126.96±0.35
	200	21.94±1.78	
	300	31.99±1.56	
	400	42.86±1.77	
	500	49.92±0.48	

Values are expressed as mean ± SD of triplicates

3.3 *In-vitro* Anti-inflammatory Activity

Most of the investigators have reported that denaturation of the protein is one of the cause of rheumatoid arthritis [39]. Production of auto-antigens in certain rheumatic diseases may be due to in vivo denaturation of proteins [40]. The mechanism of denaturation probably involves alteration in electrostatic, hydrogen, hydrophobic and disulphide

bonding. From the results of present study it can be stated that *T. catappa* Leaves aqueous extract is capable of controlling the production of auto-antigens due to in vivo denaturation of proteins in rheumatic diseases. Hence, our finding justifies the usefulness of *T. catappa* Leaves for the management and treatment of inflammation associated diseases like arthritis. Based on our results, obtained in the present studies, it can be concluded that compound form *T. catappa* Leaves aqueous extract possess significant in- vitro anti-arthritic activity which is comparable to synthetic anti-inflammatory agents. Protein denaturation inhibitory assay showed that the IC50 value of *T. catappa* Leaves aqueous extract 384.02±4.81 (Table 4).

Table.4 *In-vitro* anti-inflammatory activity of *T. catappa* Leaves aqueous extract

Anti-oxidant assay	Sample concentration (µg/reaction volume)	Percentage inhibition %	IC50 (µg/ml)
Inhibition of Protein Denaturation	100	5.05±0.12	384.02±4.81
	200	8.62±0.25	
	300	12.81±0.35	
	400	15.65±0.23	
	500	19.38±0.28	
Protease Inhibitory Activity	100	5.05±0.12	384.02±4.81
	200	8.62±0.25	
	300	12.81±0.35	
	400	15.65±0.23	
	500	19.38±0.28	

Values are expressed as mean ± SD of triplicates

Proteases are also secreted from synovial fibroblasts as the pannus invades contiguous bone and cartilage. The proteases act enzymatically to degrade the collagen and proteoglycan matrix of bone and cartilage. This destructive effect is further compounded by IL1 (and TNF) which suppresses synthesis of these matrix molecules. Thus, IL1 provides a "double insult" to connective tissue by both promoting its degradation by inducing synthesis of proteases and preventing its repair by suppressing synthesis of collagen and proteoglycans [41]. Proteinases have been implicated in arthritic reactions. Neutrophils are known to be a rich source of proteinases which carry in their lysosomal granules many neutral serine proteinases. It was previously reported that leucocyte proteinases play an important role in the development of tissue damage during inflammatory reactions and significant level of protection was provided by proteinase inhibitors. *T. catappa* Leaves aqueous extract exhibited significant anti-proteinase activity.

Our results showed that *T. catappa* Leaves aqueous extract inhibited the activity of trypsin with IC50 values 384.02±4.81µg/ml (Table 4). This was suggested that these TCLAE may be specific inhibitors of proteases. As per the earlier report showed that human trypsin is activated in certain forms of rheumatoid arthritis [42]. Therefore, the trypsin inhibitory activity of the isolated compound may contribute to its chondroprotective activity. This point is reinforced by reports stating that all four classes of proteases [43], namely, the zinc MMPs, serine proteases, cysteine proteases and the disintegrin containing metalloproteinases with thrombospondin motifs (ADAMTS) proteases [44] contribute to the degradation of cartilage matrix, bone resorption and inflammation in chronic arthritis and rheumatism.

3.4 *In-Vitro* Antibacterial Activity of *T. catappa* Leaves aqueous extract

Inhibition zone diameters exerted in disc diffusion method was recorded as evidence for the antimicrobial nature of *T. catappa* leaves aqueous extract against screened strains. In general, a diverse range of antimicrobial activity of TCLAE was observed against all the bacteria tested in this study and significant variance were recorded among Gram-positive bacteria (*Staphylococcus* spp. and *Bacillus* spp.) were generally known to be more sensitive than Gram-negative bacteria (*E. coli*, *Klebsiella* spp. and *Pseudomonas* spp.) (Table 5). The highest inhibitory effect was observed as 12.0 ± 1.21 mm for *Staphylococcus* spp. in Gram-positive group and 9.5 ± 0.5 for *E. coli* in the Gram-negative group tested. *Pseudomonas* spp. was identified as the most resistant bacteria (5.5 ± 0.5 mm) from the group of bacteria screened in this report (Table 5). A previous study by Sumitra Chanda *et al* 2013 [45], evaluated that 63% of Gram negative and 70% of the total Gram positive bacteria studied were inhibited by the methanol, acetone and N, N-dimethylformamide extracts of *T. catappa.* Our results have shown the best antibacterial effect of the extracts obtained from the aqueous extract leaves of *Terminalia catappa.*

Table.5 *In vitro* antibacterial activity of *T. catappa* Leaves aqueous extract

Microbial culture	Zone of inhibition (mm)		
	TCLAE	Tetracyclin	Sterile distilled water
Escherichia coli	9.5 ± 0.5	20 ± 1.5	
Staphylococcus aureus	12.0 ± 1.21	23 ± 1.0	
Klebsiella pneumonia	6.5 ± 0.35	11 ± 1.0	No zone of inhibition
Pseudomonas spp	5.5 ± 0.5	17 ± 1.0	
Bacillus spp.	8.5 ± 1.0	20 ± 1.2	

Values are expressed as mean ± SD of triplicates

3.5 GC–MS Analysis

GC–MS results of aqueous extract leaves of *Terminalia catappa.* confirmed the presence of eighteen major components (Fig.1) highly responsible for its antioxidant, antibacterial and antioxidant properties. The active biomolecules and their retention time (RT), peak area, molecular formula, molecular weight (MW) and structures obtained from PubChem sources are presented in Table 6. The first compound identified with less RT (7.39min) was assigned as 3-methyl-3-butene-1-thiol and the compound which took long RT (23.31min) was identified as 5 methy l2 phenylindolizine. Specifically, Vitamins, Sterols and its derivative compounds possess a high percentage of peak area among the entire phytocompounds present in TCLAE (Fig.1, Table 6). The compounds elucidated in GC–MS spectra are profoundly known for their therapeutically properties.

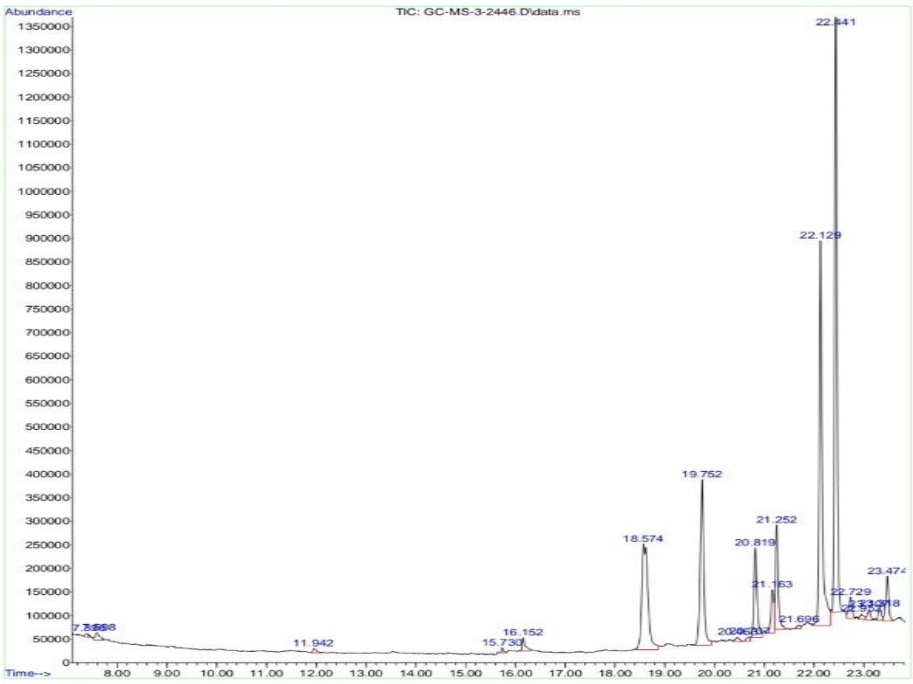
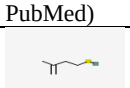
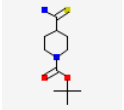
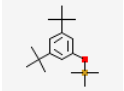
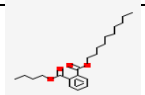
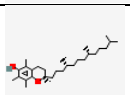
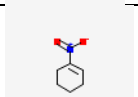
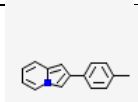
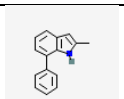
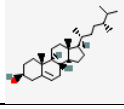
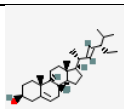
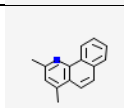
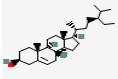


Fig.1 GC–MS chromatogram of *T. catappa* Leaves aqueous extract

Table.6 Phytocomponents of *T. catappa* Leaves aqueous extract detected by GC–MS

Peak no	Retention Time	Area %	Name of the compound	Molecular formula	2D structure (Source: PubMed)	Molecular weight	PubChem CID
1	7.39	0.23	3-methyl-3-butene-1-thiol	$C_5H_{10}S$		102.2	534532
2	7.59	0.70	1-piperidine thiocarboxamide	C6H12N2S.		144.2	135555447
3	11.94	0.34	phenol 2 4-bis(1 1-dimethylethyl)	$C_{17}H_{30}OSi$		278.5	528937
4	15.73	0.23	5 hexadecyne-	$C_{16}H_{30}$		222.41	557015
5	16.15	0.85	1,2-benzenedicarboxylic acid, butyl decyl ester	$C_{22}H_{34}O_4$		362.5	6963
6	18.57	13.33	Vitamin E	$C_{29}H_{50}O_2$		430.7	14985
7	19.75	11.76	Cyclohexene,1-nitro	$C_6H_9NO_2$		127.14	75713
8	20.46	0.35	Indolizine,2-(4-methylphenyl)-	$C_{15}H_{13}N$		207.27	346948

9	20.70	0.31	2methyl-7 phenylindole	C15H13N		207.27	610181
10	20.81	5.78	Campesterol	$C_{28}H_{48}O5$		400.68	173183
11	21.16	2.81	dl 4ethyl 5 methyl-3(-1-carboxyl)-delta(4)-thiazoline 2 thione	C6H9NS	Not Found	127.21	Not Found
12	21.25	5.48	Stigmasterol	$C_{29}H_{48}O$		412.702	5280794
13	21.69	0.29	benze[h]quinoline,2,4-dimethyl-	$C_{15}H_{13}N$		207.27	610182
14	22.12	22.50	Gamma sitosterol	$C_{29}H_{50}O$		414.7	457801
15	22.44	29.18	2(1H)-pyridinone	C_5H_7NO		97.12	8871
16	22.72	1.43	1,1,1,3,5,5,5-heptamethyltrisiloxane	$C_7H_{21}O_2Si_3$		221.5	6327366
17	22.95	0.39	2-ethylacridine	$C_{15}H_{13}N$		207.27	610161
18	23.10	0.67	cyclotrisiloxane hexamethyl-	$C_6H_{18}O_3Si_3$		222.46	10914
19	23.31	0.75	5methyl2 phenylindolizine	$C_{15}H_{13}N$		207.27	610180

4. Conclusion

The Present study highlights that the aqueous extracts of leaf from *T.catappa* serves as a potential antibacterial agent against Gram positive and Gram negative under *in-vitro* condition. Thereby the study concluded that antibacterial activity of aqueous extracts of leaf from *T.catappa* and its active constituents may be helpful in interacting with various kinds of plant disease and human allergies. *T.catappa* having better inhibitory effect on trypsin and good inhibitory effect on protein denaturation. Wherein, GC–MS analysis revealed the presence of 19 major components in *T.catappa* leaves aqueous extract. Based on that mechanism these compounds may be active in arthritic condition. So, these findings may helpful to find new lead molecule as antiarthritic drug.

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