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# Antimicrobial activity of *Tectona grandis* against MDR enteric pathogens

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**Abstract**
India is a rich source of medicinal plants and plant kingdom is valuable source of many new medicines which act as a natural remedies for certain diseases. In this study, an attempt had made to find out the antimicrobial activity of *Tectona grandis* leaves (pure and hybrid variety) against enteric bacterial pathogens. Plant extract was prepared using Chloroform and suspended in DMSO solution. Comparative study of leaf extract (Pure and hybrid) were evaluated by disc diffusion method against different pathogenic bacteria viz. *Staphylococcus aureus*, *Bacillus cereus*, *Salmonella typhi* and *Escherichia coli*. The result obtained revealed that plant showed maximum inhibition against gram positive bacteria rather than gram negative bacteria. Comparatively it was also observed that hybrid variety of *Tectona grandis* showed more antimicrobial activity than the pure variety. This study concludes that the phytochemicals obtained from *Tectona grandis* can be used in several gastrointestinal ailment and can be commercialised.

**Keywords:** Antimicrobial activity, solvent extraction, *Tectona grandis*, gastrointestinal pathogens

## Introduction
*Tectona grandis* Linn., commonly known as teak tree or sagwan (Hindi) is known in the world for its stability, extreme durability and hardness in timber production. It belongs to the family Verbenaceae. The other two species, *Tectona hamiltoniana* and *Tectona philippinensis*, are found in particular areas with small native distribution in Myanmar and the Philippines respectively. *Tectona grandis* is an excellent timber therefore it is used for boat ship building, bridge building and many other construction purpose, decoration, furniture, cabinets, musical instruments, handicrafts. *Tectona grandis* wood is found useful in the treatment of many disease like constipation, biliousness, hyperacidity, leprosy, headache, burning sensation, worms, cough, piles, microbial, fungal, dysentery infection, liver related problems and leukoderma. *Tectona grandis* leaf extract are used for the treatment of various kind of wounds, burning sensation, and pain. The oil obtained from the flower and nuts promotes hair growth and useful in the treatment of scabies while roots are used in the treatment of urinary related problems (Aboaba *et al*. 2013) [1]. *Tectona grandis* is a tropical hardwood trees and placed under the flowering plant. It is a large deciduous tree that occurs in mixed hardwood forest. Teak is one of the most valuable tree of the tropics and is extensively used for the various purposes, owing to its timber qualities it is so valuable. It is resistance towards termites, resistance towards fungi, resistance towards weather and having non-corrosive properties. (Kaosa-ard, 1986) [8]. *Tectona grandis* is large tree reaching up to 30m height under favourable condition. Having crown which open with many small branches up to 15m long. Stem is cylindrical and strength at the base. Bark is light brown, fibrous shallow or longitudinal crevice. The root system is superficial, having depth not more than 50cm, but may extend up to 15m laterally from the shoot. The leaves are shiny from the above and hairy from the lower, showing clearly vein network. The leaves are opposite, elliptic or ovate, entire, acute. Young leaves are up to 1m long. Flowers are small, white and arranged in large, flowering heads, found on the topmost branches in the unshaded part of the crown. Fruits is drupe having 4 chambers which are round, hard and woody, pale green when young and become brown at maturity. (Nidavani and Mahalakshmi 2014) [9]. Traditionally, *Tectona grandis* used globally for the treatment of various disease but Bitchoga *et al*. (2015) studied the use of *Tectona grandis* in the Northern region of Cameroon, the leaves obtained from teak are used for their laxative properties while in the Western region, leaves are used for the treatment of skin diseases. Phytochemical investigation of the teak led to the isolation of the triterpenoids, flavonoids (Ragasa *et al*. 2007) [10], chromomoric acid derivatives, anthroquinones-naphthoquinones (Aguinalda *et al*. 1997), apocarotenoids and lignans (Lacret *et al*. 2012) [12].

The metabolites like quinones shows antimycobacterial, antifungal, antibacterial and allelopathic. Since many times are the source of medicine and it is safe and effective drugs prepared from the extracts which obtained from the different parts of the plants. Plants are the rich source of phytochemicals and it is used for making different drugs having medicinal property. 25% of the medicinal drugs prepared in the developed countries are based on the plants and their derivatives. *Tectona grandis* is one of the most famous timber plants and major exotic species found in the tropical regions and it has been known from decades. It is found mainly in the South East Asian countries and in India. The quality of timber due to mainly quinones and quinones derivatives and therefore it is used in making different traditional medicines. The extract obtained from the plant used for analgesic, diuretic activity, gastroprotective activity, hair growth activity, Antioxidant activity and also used for treating swelling. (Suseela Lanka, 2017) [13]. India has a rich heritage of traditional plants and this have been using as medicine from a long time. The earliest use of medicinal plants is found in the *Rigveda* which is one of the oldest repositories of human knowledge known. (Chopra, 1958) [14]. The local people or folklore use frontal leaves of *Tectona grandis* for the treatment of various kinds of disease or wounds, especially used for burn treatment. It is also used in the treatment of various kind of skin disease, anti-inflammatory, leprosy, ulcer, haemorrhages and haemoptysis reported by (Purushotham and Shankar, 2013) [15]. It is studied that accumulation of certain compounds like Quinones and Quinones derivatives in *Tectona grandis* are mainly responsible for its wood quality. (Kokutse *et al*. 2006) [16]. Lapachal, a napthoquinone which is isolated from the Tectona grandis is reported to have anti-ulcer (Ferreira *et al*. 2003) [17] and nitric oxide scavenging activity (Jagetia and Baliga, 2004) [18]. Therefore keeping in view the significance of *Tectona grandis* the present work was conducted to study the antimicrobial activity of tectona plant against selected bacterial pathogens.

## Materials and Methods

**Place of work:** The present study entitled "Antimicrobial activity of *Tectona grandis* against enteric pathogens", was carried out in the Centre for Microbiology, Department of Botany, Ewing Christian College (ECC), Prayagraj, Uttar Pradesh, India.

**Study sample:** The study was conducted on the leaves of *Tectona grandis* of both pure and hybrid variety by disc diffusion method using Chloroform and Dimethyl Sulfoxide (DMSO) to prepare the leaf extract.

**Collection of plant material:** Fresh leaves of both pure and hybrid variety were collected from Botanical Garden of ECC, Prayagraj, Uttar Pradesh, India.

**Preparation of plant extract:** Fresh plant material was washed under tap water followed by drying it into the hot air oven at the temperature of 72 degree Celsius for 3 days and then dried leaves homogenised into the fine powder with the help of mortar and pestle and stored it in the air tight container and preserve in the Microbiology Lab of ECC. The leaf extract were prepared by using chloroform as a solvent. For this 20gms of dried powder of plant material (pure and hybrid) was taken in the different test tubes and was soaked in 40ml of chloroform in each test tubes, covered it and left it for 72 hours at room temperature. Now this chloroform soaked dried powder is used for further extraction. Now 2gms of chloroform soaked dried powder (pure and hybrid) taken in different test tubes and to this 5ml of DMSO was added in each test tubes and then this solutions were filtered using Whatman's no.1 filter paper. The filtrate is collected in different test tubes for pure and hybrid varieties. Disc was prepared using same filter paper and added to the filtrate (Purushottam and Sankar, 2013) [15].

**Microorganism used:** Four microorganism were used in the practical and they were two gram negative bacteria i.e., *Escherichia coli* and *Salmonella typhi* and two gram positive bacteria i.e., *Staphylococcus aureus* and *Bacillus cereus*. All bacterial strain used in the practical was maintained on nutrient agar medium in Microbiology lab of ECC.

**Preparation of bacterial suspension:** Colonies of different strains of bacteria were aseptically transferred to the individual flasks containing fresh nutrient broth and further incubated at 37 ˚C for 24 hours. This prepared inoculum was used to spread onto Muller Hinton Agar (MHA) using sterile cotton swab to make a lawn of bacteria on MHA culture media.

**Antimicrobial assay using disc diffusion method:** The antibacterial activity of the test extracts was determined by the disc diffusion method. To perform antibacterial assay, initially the stock culture of different bacteria were revived by inoculating the broth media and incubated at 37˚C for 24 hours. The agar plates of MHA media was prepared. Each plate was swabbed with bacterial suspension using sterile cotton swab which was spread evenly on the plate under aseptic condition. Suspension in each plate was allowed to dry for 20-25 minutes. Then disc was prepared from Whatman no.1 filter paper using paper puncher having diameter of 5mm. Now the disc were dipped in the plant extract (pure and hybrid) and placed in the plates which has been prepared by swabbing the bacteria. To this same plates control disc of chloroform and DMSO were placed. All the plates were incubated at 37 ˚C for 24 hours. The antibacterial activity of each extract was assessed by measuring the diameter of the zone of inhibition (in mm) around each disc. (Purushotham and Sankar 2013) [15].

## Results and Discussions

Antimicrobial activity of *Tectona grandis* of both pure and hybridi variety were evaluated by using disc diffusion method against different pathogenic bacteria. The observation was taken after 24 hours and following results were obtained. The results of pure and hybrid variety taken separately. In pure variety of *Tectona grandis* the *Staphylococcus aureus* showed the maximum antimicrobial activity ZOI=4mm while, *Salmonella typhi* showed ZOI=3mm and minimum antimicrobial activity was shown by *Bacillus cereus* ZOI=2mm while *Escherichia coli* showed no antimicrobial activity. In comparison with the work conducted by Purushotham and Sankar (2013) [15] it was observed that *Tectona grandis* leaves extract prepared using chloroform showed antimicrobial activities against *Staphylococcus aureus*, *Escherichia coli* and *Salmonella typhi*. The efficiency of *Salmonella typhi* was very less. Purushotham *et al*. (2010) tested four plant extract and only *Tectona grandis* leaves showed inhibition, leaves were extracted using methanol which showed high antibacterial activity against

*Mycobacterium tuberculosis* even at low concentration. The result suggested that presence of active compounds at high concentration like tectoleafquinone, tectoquinone, juglone, naphthaquinone, beulinac acid could be the reason. In other study Krishna and Nair (2010) reported that plant extract obtained using chloroform shows activity against *Staphylococcus aureus*. They further reported that only leaves extract prepared using chloroform showed antibacterial activity at low concentration. Since leaves of the plant are the site for the synthesis of bioactive compounds and therefore they have high concentration of active compounds in leaves and hence they showed antibacterial activity in the leaves. Nayeen and Karvekar (2011) suggested that some phenolic compounds like gallic acid, ellagic acid, rutin, sitosterol, quercetin etc from the leave extract of *Tectona grandis* are responsible for its anti – allergic, anti – microbial, anti – oxidant, cardioprotective and vasodilator effect. In hybrid variety of *Tectona grandis* the *Staphylococcus aureus* showed the maximum antimicrobial activity ZOI=9mm while, *Salmonella typhi* showed ZOI=5mm and minimum antimicrobial activity was shown by *Bacillus cereus* ZOI=3mm while *Escherichia coli* showed no antimicrobial activity. It can be assumed that hybrid plant have some genetic modification that lead to the more antimicrobial activity in hybrid plant as compared to pure variety of *Tectona grandis.* In comparative way of both pure and hybrid variety of *Tectona grandis*, it concluded that hybrid variety of plant showed more antiit might be microbial activity as compared to pure variety of plant. Like *Staphylococcus aureus* in hybrid plant extract showed ZOI=9mm whereas in pure variety it showed ZOI=4mm. Likewise *Salmonella typhi* in hybrid plant extract showed ZOI=5mm whereas in pure variety it showed ZOI=3mm. Same *Bacillus cereus* in hybrid plant extract showed ZOI=3mm while in pure variety it showed ZOI=2mm. The common between the two is that in both plant extract (pure and hybrid) antimicrobial activity was absent in *Escherichia coli* (gram negative bacteria). It might be due the complex cell wall of *E.coli* that makes this bacteria resistant against antimicrobial compounds. It can be said that highest ZOI was shown *by Staphylococcus aureus* followed by *Salmonella typhi* and *Bacillus cereus* respectively in both hybrid and pure varieties of *Tectona grandis.* And no antimicrobial activity in *Escherichia coli.*

**Table 1:** Antimicrobial Activity of *Tectona grandis* (pure and hybrid) against different pathogens in (mm)

| Sample | *S. aureus* | *B. cereus* | *S. typhi* | *E. coli* |
|---|---|---|---|---|
| Plant extract (pure) | 4mm | 2mm | 3mm | _ |
| Plant extract (hybrid) | 9mm | 3mm | 5mm | _ |
| Control | _ | _ | _ | _ |

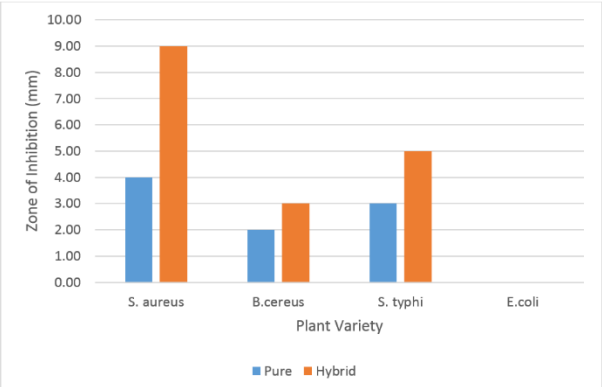


**Fig 1:** Antimicrobial Activity of *Tectona grandis* (pure and Hybrid) against different pathogens in (mm)

## Conclusion

In view of the increasing trend of multidrug resistance among pathogenic bacteria it is pertinent to evaluate and reassess the antibacterial compounds of traditional plants that have been used as herbal medicines. The potential activity of *Tectona grandis* was demonstrated in this study. Further evaluation of minimum inhibitory values and screening of antibacterial phytochemical compounds may pave the path for development of inhibitory agents in the race against drug resistant pathogens.

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