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Research Article

# Investigation of Antimicrobial and Antioxidant Potential of *Tinospora cordifolia* by In-vitro Methods

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## Abstract

Antimicrobial and Antioxidant Potential of *Tinospora Cordifolia* were performed by *In-vitro* Methods. For this study stem of plant *Tinospora cordifolia* where collected and dried under shade. Morphological characteristics and Physio-chemical parameters of *Tinospora cordifolia* stem were investigated. Plant materials were extracted by Soxhlet extraction method then Pharmacognostical evaluation of extract was subjected to various phytochemical tests for preliminary identification of various phytoconstituents. The free radicals scavenging potential of standard and extract tested by DPPH method are depicted in with a characteristic absorption at 517nm. Effects of methanolic extract of *Tinospora cordifolia* was evaluated against gram positive (*S. aureus)* and gram-negative (*E. coli)* bacteria and antifungal activity was obtained against *Candida Albicans* and *Aspergilus Niger*. Antimicrobial activities were estimated by disk diffusion method. Antimicrobial activities of methanolic extract of stem of tinospora were evaluated in the term of zone of inhibition. Methanolic steam extract of *Tinospora cordifolia* was significantly against both gram positive (*Staphylococcus aureus*) and gram-negative bacteria (*E. coli*). it was observed that the extract was not significantly effective against both gram positive and gram negative bacteria, while, Methanolic stem extract of *Tinospora cordifolia* was more effective against *Candida Albicans and A niger*.

**Keyword:** *Tinospora cordifolia,* antioxidant, Antimicrobial,

## INTRODUCTION

Herbal remedies have been employed in medical systems for the treatment and management of different diseases. The plant *tinospora cordifolia* has been used in different system of traditional medication for the treatment of diseases and ailments of human beings[1]. Antibiotics refer to the secondary metabolites produced by microorganisms or higher animals and plants in the course of life that have anti-pathogen or other activities and can interfere with the development of other living cells[2]. According to research findings, antibiotics can promote cancer apoptosis, inhibit cancer growth and prevent cancer metastasis[3]. For these reasons, antibiotics are increasingly being used to assist in the treatment of cancers[4]. There is a lot of need of development of new antibiotics[5]. Application of phytoconstituents is frequently considered nowadays because plant constituents are having high potential to be developing a drug[6]. The development of antibiotic agents, there preclinical evaluation on animal paradigm is very much essential[7].

*Tinospora cordifolia* possess different important pharmacological activity viz the notable medicinal properties are Anti-diabetic activity, Anticancer activity, Anti-spasmodic activity, Anti-malarial activity, Anti-inflammatory activity, Anti-arthritic activity, Anti-oxidant activity, Anti-allergic activity, Anti-stress activity, Anti-leprotic activity, Hepatoprotective activity, immunomodulatory & antineoplastic activity[8]. Recent studies stated that the *Tinospora cordifolia belong* to different classes such as alkaloids, glycosides, steroids, phenolics, aliphatic compounds, polysaccharides, leaves are rich in protein (11.2%), calcium and phosphorus, The stem contains clerodane furono diterpene glucoside. so that it may be used as anticancer and antioxidant medicines[9]. It is herbal that have less side effects, easily available and economic[10]. The aim and objective of this study highlights the *in-vitro* antioxidant and anticancer effects *of Tinospora cordifolia.*

## MATERIALS AND METHOD

**Plant Materials:** The stem,leaf and root of *Tinospora cordifolia* were collected Gwalior, Madhya Pradesh, India.

**Drying and size reduction of plant material:** Giloy (*Tinospora cordifolia*) were collected dried shade in college laboratory. They were pulverized to make coarse powder. The stem, leaf and root were harvested in the early morning hours in dry climate. The stems were kept in plastic bags and then carried to the experimental laboratory.

**Screening of powder (physiochemical Analysis):** Physiochemical screening of powdered steam was done by the Standard reported methods for determination of Loss on Drying, Total ash Value, Acid Insoluble Ashe value, Water Soluble Ash Value etc.

**Extraction of parts of *Tinospora cordifolia*:** Extraction of steam of *Tinospora cordifolia* was done by Soxhlet extraction method.

**Phytochemical screening of Giloy stem powder:** Phytochemical screening of powdered steam was done by the

Standard reported methods for determination of Alkaloids, Glycosides, Saponins, phytosterols, tannins and Resins.

**Anti-oxidant Activity by DPPH method**

**Preparation of extract from *Tinospora cordifolia***: The dried powdered stem of *Tinospora cordifolia* was allowed to pass through ss sieve (20 mesh). It was defatted by treating with petroleum ether (60-80oC) and then extracted to exhaustion (Soxhlet) with methanol. The solvent was removed under vacuum to get solid mass.

**DPPH Free radical scavenging activity:** Free radical scavenging activity of the extract and fractions of *T cordifolia* was evaluated using the stable free radical i.e., DPPH. 1.0 mL of standard/extract solution at different concentrations was added to 1.0 mL of 0.1 mM DPPH solution in methanol and the absorbance of mixture recorded at 517 nm after 20 min of incubation. Ascorbic acid was used as positive control.

**Antibacterial Activity by Disc Diffusion Method**

The antibacterial activity of the fractions eluted from *T. cordifolia* extract was tested in vitro using disc diffusion methods. A small aliquot (10 µl) of bacterial culture (*E. coli* and *S. aureus*) was transferred to the Mueller-Hinton agar plates aseptically. The required sterile discs were placed on the agar medium. The The maximum antibacterial activity of *T. cordifolia* was observed with a volume of 40 µl at 2% concentration with a zone of inhibition of 19 mm. A 30 µl volume of 0.2% chlorhexidine showed a zone of inhibition of 28 mm, and no zone of inhibition was observed with dimethylformamide prepared fraction was added to the discs in different concentrations and then incubated at 37°C for 24 h.

## RESULTS

**Table 1: Morphological characteristics of *Tinospora cordifolia***

| characters | Male plant stem | Female plant stem |
|---|---|---|
| General appearance | Succulent, ridged, studded with warty tubercles as a result of the development of vertical and longitudinal rows of lenticels, with characteristics three beaked nodal swelling | Similar characters as that of male one but more succulent and less flaky in nature |
| Shape | Slender, dextrorotatory twinned | Similar |
| Size (2years mature plant) | 1.38-2.06 cm | 1.78-2.32 cm |
| Surface | Jagged due to longitudinal fissures of cracks along the rows of lenticels | Similar |
| Mucilage content on cut surface | Less | More |
| Pith proportion of cut surface (pith indirectly proportion/age) | More than half in young plant | Comparatively large pith area |
| Touch | Rough | Similar |
| Color (of outer exfoliating bark) | Light- grey to creamish-white | Dark green to creamish-white |
| Odor | Not specific but characteristics bitter smell after removal of outer bark | Comparatively strong |

**Physio-chemical parameters of *Tinospora cordifolia***

**Table 2: Physio-chemical parameters of *Tinospora cordifolia* stem**

| Parameters | Results |
|---|---|
| Description brown powder | Light brown to dark |
| Loss on drying at 105$^{O}$C (%W/W) | 3.31 |
| Total ash content (%W/W) | 7.6 |
| Acid insoluble ash (%W/W) | 1.06 |
| Water soluble extractive values (%W/W) | 2.84 |
| Alcohol soluble extractive value | 6.03 |
| Extractive value | 16.4 |
| Forge in matter analysis | 0.12 |
| Ash values | 1.7 |

**Phytochemical screening of *Tinospora cordifolia***

**Table 3: Phytochemical screening of methanolic stem extracts of *Tinospora cordifolia***

| S. No | Phytochemical Test | Methanolic Extract of *Tinospora Cordifolia* |
|---|---|---|
| 1 | Carbohydrates | + |
| 2 | Proteins and Amino acids | - |
| 3 | Alkaloids | + |
| 4 | Flavonoids | + |
| 5 | Steroids | + |
| 6 | Triterpenoids | - |
| 7 | Glycosides | - |
| 8 | Tannins | + |
| 9 | Cholesterol | - |
| 10 | Phenols | + |
| 11 | Saponins | - |

**Pharmacological screening**

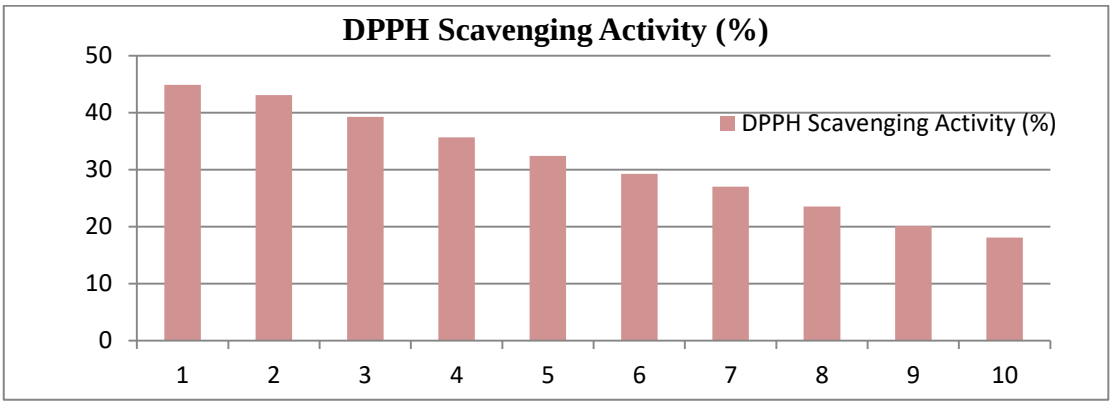


**Figure 1: Anti-oxidant activity of *Tinospora cordifolia* by DPPH assay**

**Table 4: Antibacterial activity of *Tinospora cordifolia***

| **Sample applied** | **Diameter of zone of inhibition (mm)** | |
|---|---|---|
| **Concentration of methanolic fraction** | ***E. coli*** | ***S. aureus*** |
| A –Tetracycline (30 mg) | 15± 0.21 | 19±1 |
| B – Control 5 µg/ml | - | - |
| C – 10 µg/ml | 2±0.02 | 1.5±0.2 |
| D – 15 µg/ml | 3 ±0.29 | 3±0.81 |
| E – 20 µg/ml | 4±0.58 | 4±0.92 |
| F – 25 µg/ml | 5.5±0.5 | 5.3±05.8 |
| G – 30 µg/ml | 6±0.91 | 6.3±0.29 |
| H- 50 µg/ml | 6.4 ±0.39 | 6.6 ± 0.19 |

**Figure 2: Antibacterial activity of *Tinospora cordifolia* against *E. coli***

**Figure 3: Antibacterial activity of *Tinospora cordifolia* against *S. Aureus***

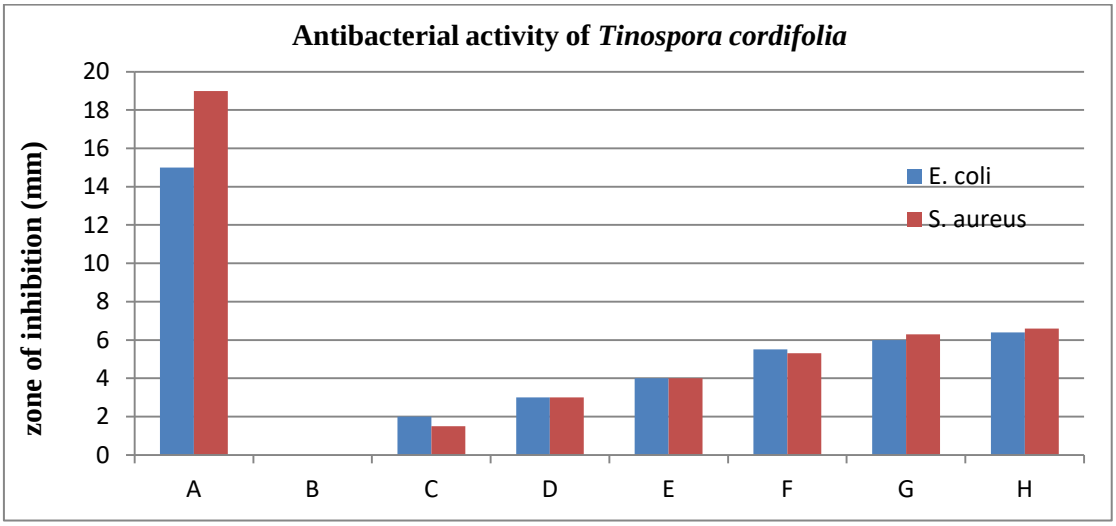


**Figure 4: Antimicrobial activity of *Tinospora cordifolia***

**Table 5: Antifungal activity of *Tinospora cordifolia***

| **Sample applied** | **Diameter of zone of inhibition (mm)** | |
|---|---|---|
| **Concentration of methanolic fraction** | ***C. Albicans*** | ***A. Niger*** |
| A – Chlotrimazole(30 mg) | 16 ± 0.11 | 18 ±1 |
| B – Control 5 µg/ml | - | - |
| C – 10 µg/ml | 6 ±0.01 | 5 ±0.2 |
| D – 15 µg/ml | 7 ±0.31 | 7 ±0.81 |
| E – 20 µg/ml | 9 ±0.1 | 8 ±0.92 |
| F – 25 µg/ml | 5 ±0.61 | 7.5 ±05.8 |
| G – 30 µg/ml | 6 ±0.72 | 6.2 ±0.29 |
| H- 50 µg/ml | 15 ±0.13 | 7.7 ± 0.22 |

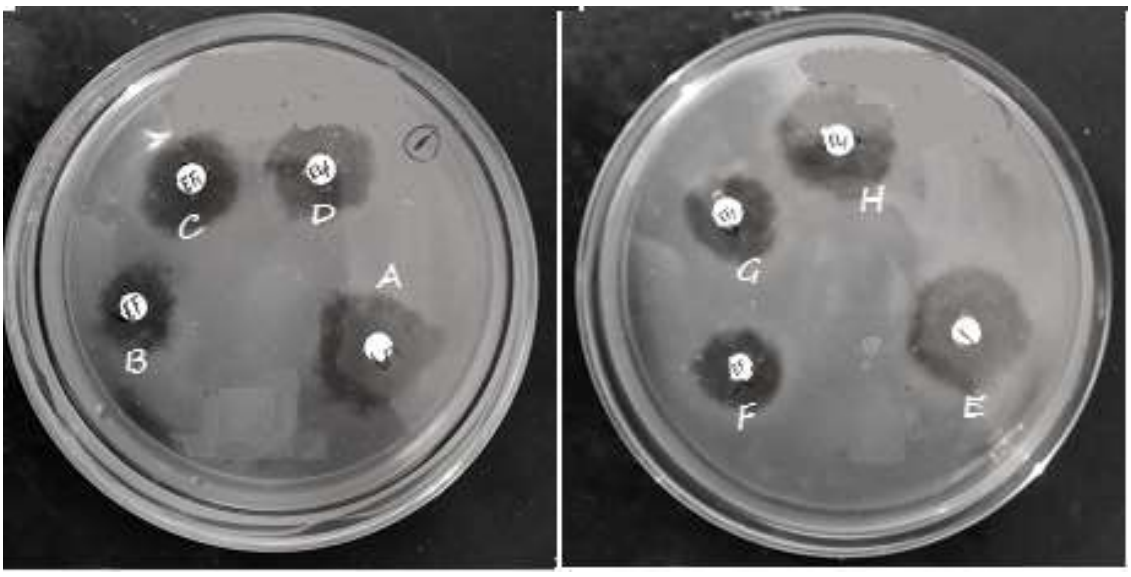


**Figure 5: activity of *Tinospora cordifolia* against *C. Albicans***

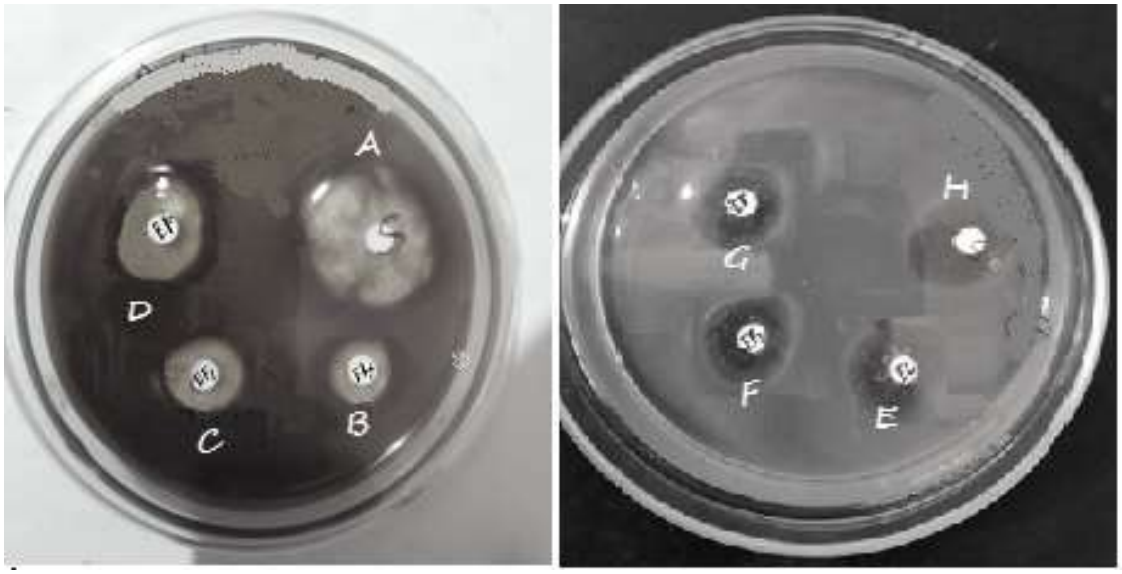


**Figure 6: activity of *Tinospora cordifolia* against *A. Niger***

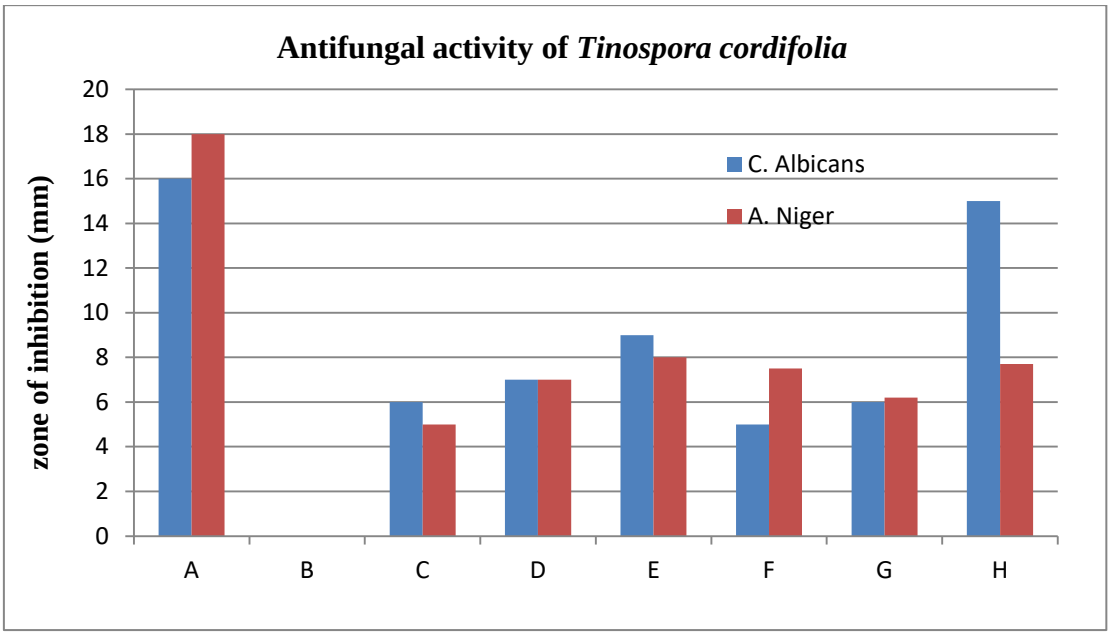


**Figure 7: Antifungal activity of *Tinospora cordifolia***

## DISCUSSION

Stem of plant *Tinospora cordifolia* where collected and dried under shade. Dry plant part pulverized and screened to get uniform sized. Morphological characteristics of *Tinospora cordifolia* revealed as Male plant stem was Succulent, ridged, studded with warty tubercles as a result of the development of vertical and longitudinal rows of lenticels, with characteristics three beaked nodal swelling while Female plant stemmore succulent and less flaky in nature, Slender, dextrorotatory twinned shaped, 1.38-2.06 cm in size, Jagged due to longitudinal fissures of cracks along the rows of lenticels, contains mucilage on surface cut, Light- grey to creamish-white in color and Not specific but characteristics bitter smell after removal of exploiting outer bark in odor. Physio-chemical parameters of *Tinospora cordifolia* stem found as Description, brown powder, Loss on drying (3.31), Total ash content (7.6), Acid insoluble ash (1.06), Water soluble extractive values (2.84), Alcohol soluble extractive value (6.03), Extractive value (16.4), Forge in matter analysis (0.12) and Ash values (1.7).

Powdered plant materials were subjected for extraction by Soxhlet extraction method, after the extraction Pharmacognostical evaluation of extract was subjected to various phytochemical tests for preliminary identification of various phytoconstituents. The stems extract contains, Carbohydrates, Alkaloids, Flavonoids, Steroids, Tanninsand Phenols. The free radical scavenging potential of standard and extract tested by DPPH method are depicted in with a characteristic absorption at 517nm. Extract showed gradual increasing percentage inhibition with increasing concentration at 517 nm in spectrophotometer as antioxidant by DPPH assay.

In this study, effects of methanolic extract of *Tinospora cordifolia* was evaluated against gram positive (*S. aureus)* and gram-negative (*E. coli)* bacteria and antifungal activity was obtained against *Candida Albicans* and *Aspergilus Niger*. Antimicrobial activities were estimated by disk diffusion method. Antimicrobial activities of methanolic extract of stem of tinospora were evaluated in the term of zone of inhibition as described in tables. Methanolic steam extract of *Tinospora cordifolia* was significantly against both gram positive(*Staphylococcus aureus*)and gram-negative bacteria (*E. coli*). It was observed that the extract was not significantly effective against both gram positive and gram negative

bacteria, while, Methanolic stem extract of *Tinospora cordifolia* was more effective against *Candida Albicans and A niger*.

## CONCLUSION

In conclusion, the present study antioxidant activity and antimicrobial activity. The methanolic stem fraction of *T. cordifolia* possesses therapeutic activity against the UTI pathogens such as *E. coli and S. aureus* and fungus. A further exploration in the isolation and characterization such as plant-derived phytoconstituents would open up new ventures in the field of antibacterial drug discovery. Antioxidant activity and antimicrobial activity property of the extracts attributed to the phytoconstituents they contain, which may be either due to their individual or additives effect that Fastens the process activities. At this stage, it is difficult to say which component(s) of the extract are responsible for the above activities. However, further phytochemical studies are needed to isolate the active compound(s) responsible for these pharmacological activities. Further investigations also needed for evolution these actions.

## CONFLICTS OF INTERESTS

There are no conflicts of interests.

## REFERENCES

1. Koch AL. Bacterial wall as target for attack: past, present, and future research. Clinical Microbiology Reviews. 2003:16 (4):673-87. https://doi.org/10.1128/CMR.16.4.673-687.2003

2. Chancey ST, Zähner D, Stephens DS. Acquired inducible antimicrobial resistance in Gram-positive bacteria. Future Microbiol. 2012; 7:959-978. https://doi.org/10.2217/fmb.12.63

3. Blair JM, Richmond GE, Piddock LJ. Multidrug efflux pumps in Gram-negative bacteria and their role in antibiotic resistance. Future Microbiol. 2014; 9:1165-1177. https://doi.org/10.2217/fmb.14.66

4. Blair JM, Webber MA, Baylay AJ, et al. Molecular mechanisms of antibiotic resistance. Nat Rev Microbiol. 2015; 13:42-51. https://doi.org/10.1038/nrmicro3380

5. Ekor M. The glowing use of herbal medicines: issues relating to adverse reactions and challenges in monitoring safety. Front pharmacol. 2014; 4:177. https://doi.org/10.3389/fphar.2013.00177

6. George, J.& Ling, Mark & Drewes, S E. Phytochemical research in South Africa. South African journal of science.2001; 97:93-105.

7. Dwivedi Sumeet, Dwivedi Abhishek & Dwivedi S.N. Folklore uses of some plants by the tribales of Madhya Pradesh with special reference to their conservation. Ethnobotanical leaflets. 2008; 12:742-743.

8. Sharma U, Bala M, Kumar N, Singh B, Munshi RK, Bhalerao S. Immunomodulatory active compounds from Tinospora cordifolia. J Ethnopharmacol. 2012; 14; 141(3):918-26. https://doi.org/10.1016/j.jep.2012.03.027

9. Patil S. Potential Application of an Aqueous Extract of Tinospora Cordifolia (Thunb.) Miers (Giloy) in Oral Submucous Fibrosis-An In Vitro Study. Materials (Basel). 2021; 14(12):3374. https://doi.org/10.3390/ma14123374

10. Ali H, Ali D, Almutairi BO, Kumar G, Karga GA, Masi C, Sundramurthy VP. Synergistic Effect of Conventional Medicinal Herbs against Different Pharmacological Activity. Biomed Res Int. 2022; 29:7337261. https://doi.org/10.1155/2022/7337261