Evaluation of Antioxidant Potential of Stem and Leaf Extracts of Himalayan *Tinospora cordifolia* Hook.f. & Thomson

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### Abstract:

**Background:**
Medicinal plants are considered a rich source of ingredients, which can be used in drug development and synthesis. *Tinospora cordifolia* (Wild.) Hook.f. & Thomson, commonly known as guduchi, heart-leaved moonseed and giloya is a herbaceous vine of the family Menispermaceae, has several beneficial properties including antioxidant activity.

**Aim:**
The present study was carried out to analyze the antioxidant activity of leaf and stem extracts of *Tinospora cordifolia* by using DPPH (1,1-Diphenyl-2-picrylhydrazyl) and ABTS (2,2’-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) free radical scavenging assays.

**Materials and Methods:**
Dried and powdered leaves and stem of *T. cordifolia* were extracted with methanol. Ascorbic acid was taken as standard. Total phenolic content was estimated by using Folin-ciocalteu's reagent while total flavonoid content by aluminium chloride reagent to find the correlation of polyphenols with antioxidant activity. ABTS assay of methanolic leaf and stem extracts showed the highest scavenging activity as compared to the DPPH assay.

**Results:**
Methanolic stem extract showed higher phenolic and flavonoid content along with antioxidant activity as compared to the methanolic leaf extract.

**Conclusion:**
The stem extract exhibited more antioxidant activity than the leaf extract with regards to all parameters analyzed.

**Keywords:** *Tinospora cordifolia*, Antioxidant activity, Phenolic content, Flavonoid content, Leaf extract, Stem extract.

### Article History

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### 1. INTRODUCTION

Plants are one of the most important sources of medicines. They can synthesize different bioactive molecules such as phenols, flavonoids, vitamins, alkaloids, terpenoids, tannins, glycosides, quinones and many others. Most of the plants used for medicinal purposes have been identified and their uses are well documented and described by various authors [1 - 3]. Medicinal plants have been used as natural medicines and they came into existence since prehistoric times. There are different ways in which plants have been found useful in medicines. The crude extract of plants has been used directly because of the presence of natural chemical constituents such as berberine, morphine, psilocin and vincristine [4]. Medicinal plant research has increased all over the world and collected the immense potential of medicinal plants used in various traditional...
systems. The extraction and characterization of bioactive compounds from medicinal plants have resulted in the introduction of new drugs with high medicinal value [5]. The parts of medicinal plants that may be used are different types of seeds, root, leaf, fruit, flowers or even the whole plant [6].

*Tinospora cordifolia* (Wild.) Hook.f. & Thomson commonly known as guduchi, heart-leaved moonseed and giloy, indigenous to the tropical areas of India, Myanmar and Sri Lanka, is a herbaceous vine of the family Menispermaceae. Guduchi is widely used in veterinary folk medicine/ ayurvedic system of medicine for its genera tonic, antiperiodic, antispasmodic, anti-inflammatory, anti-arthritic, anti-allergic and anti-diabetic properties [7, 8]. *Tinospora cordifolia* is a rich source of alkaloids, furan diterpenoids, clerodane, norditerpenoids, sequiterpenoids, phenolics, lignans, sterols, aliphatic compounds, polysaccharides, essential oil and fatty acids. In its pharmacological actions, *Tinospora cordifolia* targets body organs, mainly kidney, liver, and spleen [9]. The genus *Tinospora cordifolia* has been widely investigated by a number of workers and reported to contain a number of phytochemicals with marked therapeutic activity [10-26]. Free radicals form in our body as a result of biological oxidation. Oxidation is a natural process in organisms for the production of energy to fuel biological cycles. Oxidation by-products of normal metabolism cause extensive damage to DNA, protein and lipids, constituting a major contribution to ageing and also to degenerative disease. Oxidative damage is associated with chronic degenerative diseases, including cancer, coronary artery disease, hypertension and diabetes [14]. An antioxidant is a chemical that prevents the oxidation of other chemicals. They protect the key cell components by neutralizing the damaging effects of free radicals, which are natural by-products of cell metabolism [27]. Antioxidants occur naturally in many fruits and are able to neutralize free radicals by donating an electron and convert them into harmless molecules [28].

During the last few decades, there has been an increasing interest of scientists in the study of medicinal plants and surveys on their traditional use all over the world. Although some eminent work on *Tinospora cordifolia* has been reported from some areas of the world like Angat, Bulacan, Philippines [16] and some from different regions of India like Mysore [10, 11], Ranchi [13], Khandari Campus Agra [14, 15], Bengaluru [18], Gwalior (MP) [19], Udupi (Karnataka) [20], Chitrakoot region (MP) [22], West Singhbhum (Jharkhand) [23], Hazaribagh, and Bokaro (Jharkhand) [24]. However, comparative reports on Himalayan *Tinospora cordifolia* stem and leaf extracts so far have not been investigated. Therefore, keeping these points in mind, the present study was undertaken in order to identify the antioxidant potential and quantitative analysis of stem and leaf extracts of *Tinospora cordifolia* (Wild.) Hook.f. & Thomson, which were procured from the Haldwani regions of Kumaun Himalaya.

### 2. MATERIALS AND METHODS

#### 2.1. Collection and Processing of Plant Materials

The plant material (*Tinospora cordifolia* (Wild.) Hook.f. & Thomson) of the present study was collected from Haldwani, District Nainital, Uttarakhand, India (Latitude: 29.17558; Longitude: 79.51718; Altitude: 424m). The identity of the collected specimen was authenticated and submitted at the Taxonomy lab of D. S. B. Campus, Nainital. The plant material used for antioxidant activity was thoroughly washed off under running tap water to remove all the debris and soil and shade dried at room temperature for two to three weeks. The air-dried plant material was finely ground into powdered and packed in self-seal airtight polythene bags for further analysis [14].

#### 2.2. Extraction

Fivegram powder of each stem and leaf of *Tinospora cordifolia* (Wild.) Hook.f. & Thomson was soaked in 50 mL of methanol solvent in an orbital shaker at 37°C 120 rpm for 3 days. After that, the extracts were filtered with the help of Wallman No. 1 and solvent was evaporated in an incubator at 37°C to get the syrupy consistency, then 25% DMSO (Dimethyl sulphoxide) was added and after that the extracts were kept in a refrigerator at 0°C to determine the antioxidant activity [14].

#### 2.3. Antioxidant Activity Measurement

The scavenging activity of *Tinospora cordifolia* (Wild.) Hook.f. & Thomson stem and leaf extracts was determined using DPPH and ABTS assay.

##### 2.3.1. DPPH (1,1-Diphenyl -2-picrylhydrazyl) free radical scavenging assay

The method adopted for this study was developed by Blois (1958), which depends on the reduction of purple DPPH to yellow coloured diphenyl picrylhydrazine radical scavenger [29]. A solution of the radical was prepared by dissolving 1mg/mL DPPH in methanol. The determination of the disappearance of the free radical was done using a UV-visible spectrophotometer. Each plant extract sample stock solution (1mg/mL) was diluted in final concentration of 10, 20, 40, 80, 160, 320, 640µg/mL. Ascorbic acid was used as positive control and prepared in the same manner as an extract. As DPPH is sensitive to light, it is exposed to minimum sensitive light. These solutions were allowed to react at room temperature for 30 minutes in the dark [14].

The absorbance values were measured at 517nm and converted into percentage antioxidant activity using the following equation:

\[
\text{Abs (Control)-Abs (Sample)} \times 100
\]

\[
\text{Abs (Control)}
\]

Free radical scavenging activity (%) =
2.3.2. ABTS (2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) Free Radical Scavenging Assay

This method depends on the reduction of sea green coloured ABTS to transparent ABTS [30]. ABTS + cation radical was produced by a reaction between 7mM ABTS and 2.45mM potassium persulfate stored in the dark at room temperature for overnight before use. The solution was diluted with 50% methanol. Each plant extract sample stock solution (1mg/mL) was diluted in a final concentration of 10, 20, 40, 80, 160, 320, 640µg/mL. Ascorbic acid was used as positive control and prepared in the same manner as above. Then 2mM ABTS added to different concentrations of extract and incubate in the dark for 30 min at room temperature. As ABTS is sensitive to light, it is exposed to minimum sensitive light at room temperature. The determination of the disappearance of the free radical was done using a UV-visible spectrophotometer. All the measurements were carried out at least three times [20]. Percent inhibition of absorbance at 745nm was calculated using the formula:

\[
\text{Free radical scavenging activity (\%)} = \frac{\text{Abs (Control)} - \text{Abs (Sample)}}{\text{Abs (Control)}} \times 100
\]

(where Abs = Absorbance)

All the determinations were performed in triplicate.

The concentration of any antioxidant, which shows 50% inhibition, is called as IC₅₀ value. In the experiment, IC₅₀ values for extracts and standards were calculated from the regression plot between serial dilutions and the % inhibition.

2.4. Quantitative Analysis

2.4.1. Total Phenolic Content

Total phenolics were estimated by following Folin-Ciocalteu’s phenol reagent (FCR) method of Upadhyay et al. with little modification [14]. Different concentrations (0.5, 1, 2, 4, 8mg/mL) of standard and test samples were prepared in methanol from the stock solution. 0.5mL of each concentration was mixed with 0.2 mL of FCR and 0.5 mL Na₂CO₃ (20%). After 30min of incubation at room temperature, absorbance was measured at 750nm against blank (methanol). The concentration of total phenolic content was calculated as mg gallic acid equivalent/g extract from the calibration curve of the standard solution of gallic acid.

2.4.2. Total Flavonoid Content

Total flavonoid content was estimated by aluminium chloride (AlCl₃) method. Different concentrations (0.5, 1, 2, 4, 8mg/mL) of standard and test samples were prepared in methanol from the stock solution. 0.5mL of each concentration was mixed with 0.2 mL of AlCl₃, 0.5mL NaNO₂ (20%). The concentration of total flavonoid content was calculated as mg quercetin acid equivalent/g extract, from the calibration curve of the standard solution of quercetin acid. Then allow the solution at room temperature for 30 min. The absorbance was measured at 506nm by spectrophotometer [22].

3. RESULTS AND DISCUSSION

3.1. Antioxidant Activity

3.1.1. DPPH (1,1-Diphenyl-2-picrylhydrazyl) Free Radical Scavenging Activity of Stem and Leaf Extracts

Both the stem and leaf extracts of Tinospora cordifolia (Wild.) Hook.f. & Thomson showed effective scavenging activity against the free radicals. The DPPH standard (ascorbic acid) had a maximum (96.58%) free radical scavenging activity at 640µg/mL and with the lowest IC₅₀ value of 22.38 µg/mL (Tables 1-3). The methanolic stem extract showed the highest scavenging activity (55.94%) at 640µg/mL and lowest (25.32%) at 10µg/mL (Table 1). It was found that when the concentration of the extract increased, the absorbance value gets decreased regularly. The methanolic leaf extract of T. cordifolia showed comparatively lesser activity with the highest scavenging activity (48.03%) at 640 µg/mL and the lowest (25.01%) at 10µg/mL (Table 1) as compared to the methanolic stem extract. Similar results were also observed by Upadhyay et al. [14] in the methanolic stem extract of T. cordifolia and found that at a concentration of 10 mg/mL, DPPH free radical scavenging activity was low (44%). In contrast, Praveen et al. [31] reported that methanolic leaf extract showed the highest scavenging activity. Scavenging effect on DPPH radical was observed to be higher in methanolic leaf extract as compared to the methanolic extract of stem of T. cordifolia procured from the local market of Mysore [11]. The leaf and stem extracts of the methanolic solvent of T. cordifolia showed greater percentage inhibition with IC₅₀ values of 0.54 mg/mL and IC₅₀ 0.74 mg/mL, respectively, which was more than the present study [11]. In a previous study, the methanolic stem extract of T. cordifolia from the Philippines exhibited good free radical scavenging activity potential with an IC₅₀ value of 0.861 µg/µL [16].
3.1.2. **ABTS (2,2′-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)) Free Radical Scavenging Activity of Stem and Leaf Extracts**

All the concentration of the test solution more or less scavenged the free radicals. Both the stem and leaf extracts of *Tinospora cordifolia* showed effective scavenging activity against ABTS free radical. The ABTS standard (Ascorbic acid) had maximum free radical scavenging activity (91.55%) at 80µg/mL concentration with the least IC₅₀ value of 9.36 µg/mL as represented in Table 2. The methanolic stem extract showed the highest scavenging activity (74.21%) at 640µg/mL concentration and the lowest (4.51%) at 20µg/mL (Table 2). The methanolic leaf extract of *T. cordifolia* showed comparatively lesser activity with the highest scavenging activity (73.86%) at 640 µg/mL concentration and the lowest (6.33%) at 10µg/mL concentration as compared to the methanolic stem extract (Table 2). The stem extract of *T. cordifolia* showed better IC₅₀ value in ABTS (126.93 µg/mL) and DPPH (500.40 µg/mL) assays as compared to the leaf extract of the plant with IC₅₀ values if 379.31 µg/mL and 677.78µg/mL in ABTS and DPPH assays respectively (Table 3). With all the extracts (leaf and stem), dose-dependent radical scavenging was observed, i.e., when the concentration of the extract was increased, the free radical scavenging activity also increased. These results are in agreement with reports of Upadhyay et al. [14]. ABTS scavenging assay of leaf and stem extracts of the methanolic solvent of *T. cordifolia* from Mysore showed greater inhibition as compared to the present study with the least IC₅₀ values of 95µg/mL and 107 µg/mL [11]. The ABTS scavenging assay of both leaf and stem extracts was the most potent method as compared to the DPPH scavenging assay with the highest scavenging activity and least IC₅₀ values.

### 3.2. Quantitative Analysis

#### 3.2.1. Total Phenolic Content (TPC)

TPC was measured by folin-ciocalteu's phenol reagent method and gallic acid was taken as standard in different concentrations (mg/mL). TPC of methanol extract of *Tinospora cordifolia* leaves and stem was calculated as gallic acid equivalent (GAE) (mg GAE/g), using the calibration curve, which was based on the following equation: Y= 1.5118x−0.1533 R² =0.9664 shown in Fig. (1). It was clear that the phenolic content in methanolic leaf extract (8.51 mg GAE /g) was lower as compared to the methanolic stem extract (17.44 mg GAE /g) as shown in Table 4. The assay supported that there was a positive relationship between total phenolic content and antioxidant activity. The TPC of methanolic stem extract was less than the TPC in the

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**Table 1. DPPH radical scavenging activity of methanolic stem and leaf extracts of *Tinospora cordifolia*.**

| Concentration (µg/mL) | Ascorbic Acid (% Scavenging Activity) | Stem Extract (% Scavenging Activity) | Leaf Extract (% Scavenging Activity) |
|-----------------------|---------------------------------------|---------------------------------------|--------------------------------------|
| 10                    | 21.42                                 | 25.32                                 | 25.01                                |
| 20                    | 53.57                                 | 25.54                                 | 26.64                                |
| 40                    | 80.43                                 | 26.69                                 | 27.92                                |
| 80                    | 84.00                                 | 27.48                                 | 28.94                                |
| 160                   | 89.05                                 | 33.76                                 | 30.66                                |
| 320                   | 93.78                                 | 44.10                                 | 39.28                                |
| 640                   | 96.38                                 | 55.94                                 | 48.03                                |

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**Table 2. ABTS scavenging activity of methanolic stem and leaf extracts of *Tinospora cordifolia***

| Concentration (µg/mL) | Ascorbic Acid (% Scavenging Activity) | Stem Extract (% Scavenging Activity) | Leaf Extract (% Scavenging Activity) |
|-----------------------|---------------------------------------|---------------------------------------|--------------------------------------|
| 5                     | 30.46                                 | -                                     | -                                    |
| 10                    | 58.49                                 | -                                     | 6.33                                 |
| 20                    | 85.41                                 | 4.51                                 | 12.79                                |
| 40                    | 90.55                                 | 11.89                                 | 18.81                                |
| 80                    | 91.55                                 | 26.89                                 | 30.05                                |
| 160                   | -                                     | 56.50                                 | 49.77                                |
| 320                   | -                                     | 73.43                                 | 73.43                                |
| 640                   | -                                     | 74.21                                 | 73.86                                |

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**Table 3. IC₅₀ values of leaf and stem extracts against DPPH and ABTS activities in, µg/mL.**

| Antioxidant Activity | Ascorbic Acid (Standard) | Stem Extract | Leaf Extract |
|----------------------|--------------------------|--------------|--------------|
| DPPH                 | 22.38                    | 500.40       | 677.78       |
| ABTS                 | 9.36                     | 126.93       | 379.31       |
methanolic stem extract (51.86 mg GAE /g), as reported by Upadhyay et al. [14]. Similar results were reported by Hiiyaraja and Khanum [11] with total phenolic content of 33.93 mg GAE /g and 52.17 mg GAE /g in the methanolic stem and methanolic leaf extracts of T. cordifolia, respectively. The total phenolic content of methanolic leaf extract of T. cordifolia was reported to be 2.00 mg GAE /g by Shwetha et al. [18], which was less than the present findings.

3.2.2. Total Flavonoid Content (TFC)

The TFC was measured by aluminium chloride reagent method and quercetin was taken as standard. TFC of methanol extract of Tinospora cordifolia leaves and stem extracts was calculated as Quercetin Equivalent (QE) (mg/g), using a calibration curve, which was based on the following equation: 

\[ Y = 0.2216x + 0.1955 \]

\[ R^2 = 0.9808 \]

as shown in Fig. (2). The flavonoid content of the methanolic leaf extract (20.94 mg QE/g) was lower as compared to the methanolic stem extract.

Table 4. Quantitative analysis of stem and leaf extracts of Tinospora cordifolia.

| Samples               | TPC (mg GAE /g of extract) | TFC (mg QE /g of extract) |
|-----------------------|-----------------------------|---------------------------|
| Methanolic stem extract | 17.48±0.01                  | 23.83±0.00               |
| Methanolic leaf extract | 8.51±0.012                  | 20.94±0.02               |

Fig. (1). Standard curve of gallic acid for total phenolic content.

Fig. (2). Standard curve of quercetin for total flavonoid content.
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(23.82 mg QE/g) of T. cordifolia as shown in Table 4. The high flavonoid content was found to be positively correlated with the antioxidant activity. The TFC in the methanolic leaf extract of T. cordifolia was 0.7 mg QE/g reported by Shwetha et al. [18], which was less than the present findings. The methanolic stem extract showed higher antioxidant activity and higher phenol and flavonoid content as compared to the Methanolic leaf extract. The similar results were observed by Garg and Garg [22] showing lesser flavonoid content in methanolic stem extract (2.14 mg QE/g) as compared to the methanolic leaf extract (4.53 mg QE/g).

CONCLUSION

The plant kingdom is a treasure of potential drugs and the drugs from the plants are easily available, less expensive, safe, efficient and rarely have side effects. In the present study, Tinospora cordifolia (Wild.) Hook.f. & Thomson, which was collected from Haldwani (Nainital), was assessed for its antioxidant activity and quantitative analysis. The stem extract exhibited more antioxidant activity than the leaf extract with regards to the all parameters analyzed.

ETHICAL STATEMENT

The plant material (Tinospora cordifolia (Wild.) Hook.f. & Thomson) of the present study was collected from Haldwani, District Nainital, Uttarakhand, India (Latitude: 29.17558; Longitude: 79.51718; Altitude: 424m). The identity of the collected specimen was authenticated and submitted at the Taxonomy lab of D. S. B. Campus, Nainital.

CONSENT FOR PUBLICATION

Not applicable.

AVAILABILITY OF DATA AND MATERIALS

Not applicable.

FUNDING

None.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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REFERENCES

[1] Nadkarni KM. India material medica. India: Bombay popular Prakashan 1976.
[2] Dastur JF. Medicinal plants of India and Pakistan. Bombay: D.B. Taraporewala Sons & Co Private Ltd 1985.
[3] Saradamma I. All India co-ordinated research project on ethnoanobiology Final technical report RRI Drug Research, CCRAS, Government of India, Pojupara. Trivandum: Kerela, India 1990.
[4] Mittal J, Sharma MM, Batra A. Tinospora cordifolia a multipurpose medicinal plant- A review. J Med Plants Stud 2014; 2: 32-47.
[5] Gangola S, Khati P, Parul, Bhatt P, Sharma A. India as the heritage of medicinal plant and their use. Curr Trends Biomedical Eng & Biosci 2017; 4(4): 50-1.
[6] Jumaldi KF, Lorigoinzi Z, Amini KH. Medicinal plant: Past history and future perspective. J Herbed Pharmacol 2018; 7(1): 1-7. [http://dx.doi.org/10.15171/jhp.2018.01]
[7] Spandan Hu, Shailk LA, Nirmala T, Santhi M, SD Sipai B. A review on Tinospora cordifolia. Int J Cur Pharma Res Rev 2013; 4(2): 61-8.
[8] Zhao TF, Wang X, Rimando AM. Che. Folkloric medicinal plants: Tinospora sagittatavar. cravaniana and Mahonia baeule. Planta Med 1991; 57: 507. [http://dx.doi.org/10.1055/-s-2006-960188]
[9] Singh D, Chaudhuri PK. Chemistry and pharmacology of Tinospora cordifolia. Nat Prod Commun 2017; 12(2): 299-308. [http://dx.doi.org/10.1177/1934578X1701200240] [PMID: 30428235]
[10] Premananth R, Lakshmi D. Studies on antioxidant activity of Tinospora cordifolia leaves using in vitro models. J Am Sci 2010; 6(10): 736-43.
[11] Ilayaraja N, Kanaum F. Antioxidant potential of Tinospora cordifolia extracts and their protective effects on oxidation of biomolecules. Pharmcogn J 2011; 3(20): 56-61. [http://dx.doi.org/10.22270/pj.2011.20.11]
[12] Bhalerao BM, Kasote DM, Nagarkar BF, et al. Comparative analysis of radical scavenging and immunomodulatory activities of Tinospora cordifolia growing with different supporting trees. Acta Biol Szeged 2012; 56(1): 65-71.
[13] Kumar A, Kumar M, Dandapat S, Sinha MP. Antioxidant activity and pharmacological screening of Tinospora cordifolia (Thunb.). Biocean 2013; 8(2): 689-93.
[14] Upadhyay N, Ahmad G, Agnihoti K, Sharma R. Studies on antioxidant activity and total phenolic content of Tinospora cordifolia stem using in vitro models. Am J Pythom Clinical Therapeutics 2013; 8: 617-27.
[15] Upadhyay N, Showkat A, Ahmad G, Agnihoti RK, Sharma R. Free radical scavenging activity of Tinospora cordifolia (Wild) Miers. J Pharmacogn Phytochem 2014; 3(2): 63-9.
[16] Castillo AL, Yuen MC, Cira JCD, et al. Free radical scavenging activity of Tinospora cordifolia stem methanolic fraction. Int J Biol Pharm Allied Sci 2015; 4(10): 6241-54.
[17] Dwivedi SK. Enesspa. Tinospora cordifolia with reference to Biological and Microbial Properties. Int J Curr Microbiol Appl Sci 2016; 5(6): 446-65. [http://dx.doi.org/10.20546/ijcmmas.2016.506.052]
[18] Shwetha RJ, Tahareen, Myrene RD. Antioxidant and anti-inflammatory activity of Tinospora cordifolia using in vitro models. J Chem Biol Phys Sci 2016; 6(2): 497-512.
[19] Kumar V, Singh S, Singh A, Kumar DA, Srivastava, Sidhu G, Singh R, Meena, Singh R, Sushobh Varanasi, Prakash O. Phytochemical, antioxidant, antimicrobial and protein binding qualities of hydro-ethanolic extract of Tinospora cordifolia. J Bio Active Products from Nature 2018; 8(3): 192-200. [http://dx.doi.org/10.1080/22311866.2018.1485513]
[20] Polu RP, Nayanbhairama U, Khan S, Rajlexmi M. Assessment of free radical Scavenging and anti-proliferative activities of Tinospora cordifolia. BMC Complement Altern Med 2017; 17: 2-12. [http://dx.doi.org/10.1186/s12906-017-1953-3]
[21] Bharathi C, Harinatha R, Nageswar G. A review of medicinal properties of Tinospora cordifolia. Int J Sci Res Rev 2018; 7: 585-98.
[22] Garg P, Grag R. Qualitative and quantitative of leaves and stem of Tinospora cordifolia in different solvent extract. J Drug Deliv Ther 2018; 8(5): 259-64. [http://dx.doi.org/10.20227/ijdjt.v8i5.s.1967]
[23] Sinka R, Sinha MR. Preliminary phytochemical screening and phytochemical analysis of Tinospora cordifolia. Miers J Med Plant Sna 2018; 6(1): 177-80.
[24] Prasad B, Chauhan A. Antioxidant and antimicrobial studies of Tinospora cordifolia stem and roots under in vitro conditions. Int J Adv Micro Health Res 2019; 3(1): 1-10.
[25] Sharma P, Dwivedee BP, Bishi D, Dash AK, Kumar D. The chemical constituents and diverse pharmacological importance of Tinospora cordifolia. J Heliyon 2019; 1-8. [http://dx.doi.org/10.1016/j.heliyon.2019.e02437] [PMID: 31701036]
[26] Zeba S, Mishra BK. Phytochemical study of Tinospora cordifolia grown on three different soil conditions. Res J Life Sci, Bioinf Pharma Chem Sci 2019; 5(2): 810-5. [http://dx.doi.org/10.1055/s-0039-1688169]
[27] Miller HE, Rigelfo F, Marquet L, Prakash A, Kantar M. Antioxidant content of whole grain breakfast cereals and fruits and vegetables. J Am. Coll Nutr 2000; 19(3)(Suppl.): 312S-9S. [http://dx.doi.org/10.1080/07315724.2000.1071896] [PMID: 10875603]
[28] Markis DP, Boskou G, Andrikopoulos NK. Polyphenolic content and \textit{in vitro} antioxidant characteristics of wine industry and other agri-food solid waste extracts. J Food Compos Anal 2007; 20(2): 125-32. [http://dx.doi.org/10.1016/j.jfca.2006.04.010]

[29] Blois MS. Antioxidant determinations by the use of a stable free radical. Nature 1958; 181: 1199-200. [http://dx.doi.org/10.1038/1811199a0]

[30] Saeed N, Khan MR, Shabbir M. Antioxidant activity, total phenolic and total flavonoid contents of whole plant extracts \textit{Torilis leptophylla} L. BMC Complement Altern Med 2012; 12(1): 221. [http://dx.doi.org/10.1186/1472-6882-12-221] [PMID: 23153304]

[31] Praveen N, Thiruvengadam M, Kim HJ, Kumar JKP, Chung IM. Antioxidant activity of \textit{Tinospora cordifolia} leaf extract through non-enzymatic method. J Med Plants Res 2012; 6(3): 4790-5.

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