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# Antioxidant activity of different parts of *Lysimachia laxa* and *Gymnocladus assamicus*, a comparison using three different solvent extraction systems

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**ABSTRACT**

*Lysimachia laxa is a perennial herb belonging to the family Primulaceae and Gymnocladus assamicus is a critically endangered species of the family Caesalpinioideae contain biologically active substances. The present investigation deals with the evaluation of antioxidant activity of the different parts of these two plants and to determine the best solvent for extracting the antioxidant components. In our research three different solvents (acetone, methanol and aq. methanol ) extracts from different parts of these two plants were used to examine the effects of extraction solvent on total phenolics, flavonoid, flavonol content, reducing power and 1,1-Diphenyl-2-picrylhydrazyl (DPPH) radical scavanging activity. Results showed that extraction solvent had significant effects on antioxidant activity of these two plants. The highest content of total phenolics, flavonoids and DPPH scavenging activities were found in the aq. methanol extract. The methanol had the highest extraction capacity for the flavonol components from these two plants. The highest reducing power was found in the aq. methanol extract of L. laxa and the same was observed in the methanol extract of G. assamicus. Results also showed that the extraction yields of antioxidant components is depending on the polarity of solvent. With increased in solvent polarity from acetone to aq.methanol, amount of extractive materials increased in both the plants. The overall results showed that the aq. methanol extracts of whole parts of L. laxa and G. assamicus showed potent antioxidant activity.*

**Key words**: *Lysimachia laxa* and *Gymnocladus assamicus*; different solvent extraction; antioxidant activity.

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## INTRODUCTION

Antioxidants are vital compound that may function as free radical scavengers, complexing agents for pro-oxidant metals, reducing and quenchers of singlet oxygen formation [1-3]. Antioxidants are compounds that can delay or inhibit the oxidation of lipids or other molecules by inhibiting the initiation or propagation of oxidative chain reactions [4]. Plants are potential source of diverse bioactive component like phenolic, flavonoid, flavonol, saponin, etc. Such heterogeneous bioactive compounds having significant antioxidant potency protect from highly unstable free radical bombardment or metabolized in cellular microenvironment. Antioxidants prevent and cure several human diseases like aging, cancer, atherosclerosis, ischemic injury and neurodegenerative [5-7]. Antioxidant properties have been screened in several medicinal plants for the herbal drugs formulation and in the areas of nutrient, medicine and cosmetic formulations [8].

Plant materials are the rich source of active constituents of varied chemical characteristics and polarities and complete extraction of active components, responsible for antioxidant activities, are strongly dependant on the nature of solvents and plant parts used. The most current research on antioxidant action focuses on phenolic compounds such as flavonoids. Flavonoids, tannins and other phenolic constituents present in food of plant origin are also potential antioxidants [9]. During the extraction of plant material, the selection of solvents and plant parts is very much important to minimize interference from compounds that may co-extract with the chemicals and to avoid the contamination of the extract. Polar solvents are frequently employed for the recovery of polyphenols from a plant matrix. Solvents, such as methanol, ethanol, acetone, chloroform and ethyl acetate have been widely used for the extraction of antioxidant compounds from various plants and plant based foods and medicines. Results of previous studies showed that the extraction yield of phenolic and flavonoid content is greatly depending on the polarity of the solvent [10-11]. Bonoli et al., 2004 [12] reported that maximum phenolic compounds were obtained from barley flour with the mixture of ethanol and actone. The aq methanol was found to be more effective solvent to extract the phenolic compounds from rice bran and *Moringa oleifera* leaves [13-14]. The extraction of high content of antioxidant compounds with 80 % aq. methanol (methanol: water 80: 20) were found from various plant materials like rice bran, wheat bran, oat groats and hull, coffee beans, citrus peel and guava leaves as reported by Anwar et al., 2006 [15]. The highest extract yields were obtained from polar alcohol based solvents. Addition of water to ethanol improves the extraction rate but too high water content may leads to the extraction of other compounds.The highest level of phenolic compounds was found with 50% acetone from wheat, whereas ethanol is the least effective solvent to isolate phenolics from wheat bran [16]. It is still not clear which type of solvent is more effective for extracting the antioxidant components from plant.

*Lysimachia laxa* (Syn.: *Lysimachia ramosa* Wall ex Duby) (Primulaceae) is a perennial herb. About 180 species are reported in temperate and subtropical parts of northern hemisphere, but with a few species in Africa, Australia and South America [17], whereas a few species of genus *Lysimachia* reported in a few localities up to 1000-2000 meter altitude in Arunachal Pradesh, Nagaland and Meghalaya in India. The fresh leaf is being widely used to cure gastrointestinal worm infection by tribal people of Meghalaya [18-19]. The plant extract have lethal anthelmintic efficacy to distortion of the tegumental surface of helminthes parasite [18]. The other species of genus *Lysimachia* has reported pharmacologically bioactive compounds such as triterpenoid, saponins, organic acid, flavones and flavanoids [20-21]. Therefore presence of these bioactive principles lead to the presumptions that like any other medicinal plant, *Lysimachia laxa* may also possess the antioxidant activity.

The genus *Gymnocladus* is a primitive genus of the family Caesalpinioideae. In India, the species *G. assamicus* is reportedly a critically endangered species in Northeast India [22]. Fleshy pods rich in saponin are used as a substitute for soap, detergent or antidandruff for washing hair by the tribal people of Monpa and Khasis. Monpa tribe community consumes roasted seeds as substitute for groundnut and coffee for its similar aroma like coffee, while excessive consumption of roasted seeds cause dizziness, nausea and vomiting. Peoples also make use of the pods for removing/expelling leeches of their domestic animals [23].

However, the objective of present study was to investigate the effect of different extracting solvents with different polarity on the antioxidant activities of the different parts of two high altitude plant species from North-East India viz *Lysimachia laxa* and *Gymnocladus assamicus* which has not been studied till date. Hence present research would be enabling to develop a commercial product with antioxidant for external herbals saop or detergent and shampoo as well as further platform for chemical profiling of two species.

## EXPERIMENTAL SECTION

### *Plant materials*

*Lysimachia laxa* : The plant material of *L. laxa* was collected in 2008 from the Talle valley, Lower Subansiri district, Arunachal Pradesh in India at an altitude of 1800-2000 m. The specimen (field number Mao & Gupta-19205), was identified and deposited in the 'ARUN' Harberium of Botanical Survey of India, Arunachal Pradesh Regional Center, Itanagar, Arunachal Pradesh, India. The collected plant material was also dried at room temperature and made into powder and stored for antioxidant potential and biochemical studies.

*Gymnocladus assamicus* : The mature seed pods of *Gymnocladus assamicus* were collected in 2008 from the Dirang Valley, west Kameng district in Arunachal Predesh, India at an altitude of 1700-2000 m. The specimen (field number- 111310), was identified and deposited in the Kanjilal Harberium 'ASSAM' of BSI, Shillong Regional

Center, Shillong, India. The plant materials and seed was carefully detached, dried in shade and stored for antioxidant potential and biochemical studies.

***Chemicals***

1,1-Diphenyl-2-picrylhydrazyl (DPPH), butylated hydroxytoluene (BHT), ascorbic acid, rutin, quercetin were purchased from Sigma Chemical Co. (St. Louis, MO, USA)., Folin-Ciocalteus's phenol reagent, gallic acid, potassium ferricyanide, Aluminium chloride, $FeCl_3$ sodium acetate and sodium carbonate were from Merck Chemical Supplies (Damstadt, Germany). All the chemicals used including the solvents, were of analytical grade.

***Extraction of plant material ( Methanol, Aqueous methanol (1:1) and Acetone)***

One gram of each plant material were extracted with 20 ml each of methanol, aqueous methanol (50%, v/v) and acetone, with agitation for 18 -24 h at ambient temperature. The extracts were filtered and diluted to 50 ml and aliquot were analyzed for their total phenolic, flavonoid and flavonol content, reducing power and their free radical scavenging activity.

***Estimation of total phenolic content***

The amount of total phenolic content of crude extracts was determined according to Folin-Ciocalteu procedure [24]. 20 - 100 µl of the tested samples were introduced into test tubes; 1.0 ml of Folin-Ciocalteu reagent and 0.8 ml of sodium carbonate (7.5%) were added. The tubes were mixed and allowed to stand for 30 min. Absorption at 765 nm was measured (UV-visible spectrophotometer Hitachi U 2000 Japan). The total phenolic content was expressed as gallic acid equivalents (GAE) in miligram per gram (mg/g) of extract using the following equation based on the calibration curve $y = 0.0013x + 0.0498$, $R^2 = 0.999$ where y was the absorbance and x was the Gallic acid equivalent (mg/g).

***Estimation of total flavonoids***

Total flavonoids were estimated using the method of Ordonez et al., 2006 [25]. To 0.5 ml of sample, 0.5 ml of 2% $AlCl_3$ ethanol solution was added. After one hour, at room temperature, the absorbance was measured at 420 nm (UV-visible spectrophotometer Hitachi U 2000 Japan). A yellow color indicated the presence of flavonoids. Total flavonoid contents were calculated as rutin (mg/g) using the following equation based on the calibration curve : $y = 0.0182x - 0.0222$, $R^2 = 0.9962$, where y was the absorbance and x was the Rutin equivalent (mg/g).

***Estimation of total flavonols***

Total flavonols in the plant extracts were estimated using the method of Kumaran and Karunakaran, 2006 [26]. To 2.0 ml of sample (standard), 2.0 ml of 2% $AlCl_3$ ethanol and 3.0 ml (50 g/L) sodium acetate solutions were added. The absorption at 440 nm (UV-visible spectrophotometer Hitachi U 2000 Japan) was read after 2.5 h at 20°C. Total flavonol content was calculated as quercetin (mg/g) using the following equation based on the calibration curve: $y = 0.0049x + 0.0047$, $R^2 = 0.9935$, where y was the absorbance and x was the quercetin equivalent (mg/g).

***Measurement of reducing power***

The reducing power of the extracts was determined according to the method of Oyaizu, 1986 [27]. Extracts (100 µl) of plant extracts were mixed with phosphate buffer (2.5 ml, 0.2 M, *p*H 6.6) and 1% potassium ferricyanide (2.5 ml). The mixture was incubated at 50°C for 20 min. Aliquots of 10% trichloroacetic acid (2.5 ml) were added to the mixture, which was then centrifuged at 3000 rpm for 10 min. The upper layer of the solution (2.5 ml) was mixed with distilled water (2.5 ml) and a freshly prepared ferric chloride solution (0.5 ml, 0.1%). The absorbance was measured at 700 nm (UV-visible spectrophotometer Hitachi U 2000 Japan). Reducing power is given in ascorbic acid equivalent (AAE) in milligram per gram (mg/g) of dry material using the following equation based on the calibration curve : $y = 0.0023x - 0.0063$, $R^2 = 0.9955$ where y was the absorbance and x was the ascorbic acid equivalent (mg/g).

***Determination of free radical scavenging activity***

The free radical scavenging activity of the plant samples and butylated hydroxyl toluene (BHT) as positive control was determined using the stable radical DPPH (1,1-diphenyl-2-picrylhydrazyl) [28] . Aliquots (20 - 100 µl) of the tested sample were placed in test tubes and 3.9 ml of freshly prepared DPPH solution (25 mg $L^{-1}$) in methanol was added in each test tube and mixed. 30 min later, the absorbance was measured at 517 nm (UV-visible spectrophotometer Hitachi U 2000 Japan). The capability to scavenge the DPPH radical was calculated, using the following equation:

DPPH scavenged (%) = {(Ac – At)/Ac} x 100

Where Ac is the absorbance of the control reaction and At is the absorbance in presence of the sample of the extracts. The antioxidant activity of the extract was expressed as $IC_{50}$. The $IC_{50}$ value was defined as the concentration in mg of dry material per ml (mg / ml) that inhibits the formation of DPPH radicals by 50%. Each value was determined from regression equation.

Values are presented as mean ± standard error mean of three replicates. The total phenolic content, flavonoid content, flavonol content, reducing power and $IC_{50}$ value of each plant material was calculated by using Linear Regression analysis.

## RESULTS AND DISCUSSION

***Extractive value***

The extractive value of the different parts of the tested plants with three different solvents are depicted in table 1. The amount of extracts in the different parts of *L. laxa* varied from 1.93±0.02 to 40.50±0.29 g/100g dry material and in the different parts of *G. assamicus* ranged from 3.17±0.02 to 72.83±0.17 g/100g dry material, using three solvents *viz*. methanol, Aq. methanol (1:1) and acetone. Aq. methanol extract from both the plants exhibited highest extract yield (leaf of *L. laxa*, 40.50±0.29 g /100g & seed pod of *G. assamicus* 72.83±0.17 g/100g dry material). The differences in the extractive value of the plants may be due to the varying nature of the components present and the polarities of the solvent used for extraction [29] .

**Table 1. Extractive value of the different plant extracts of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | Extractive value (g / 100g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 31.00±0.29 | 40.50±0.29 | 6.33±0.17 |
| 2 | *Lysimachia laxa* | Stem | 20.17±0.17 | 17.67±0.17 | 1.93±0.02 |
| 3 | *Lysimachia laxa* | Whole plant | 19.00±0.29 | 30.50±0.29 | 5.40±0.03 |
| 4 | *Gymnocladus assamicus* | Whole plant | 31.83±0.17 | 38.83±0.17 | 5.82±0.02 |
| 5 | *Gymnocladus assamicus* | Seed | 28.00±0.29 | 39.17±0.17 | 10.83±0.02 |
| 6 | *Gymnocladus assamicus* | Seed pod | 63.33±0.44 | 72.83±0.17 | 3.17±0.02 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ± SEM*

***Total phenol, flavonoid and flavonol content of the extracts***

Phenolic components are very important plant constituents with scavenging ability because of its hydroxyl group. Phenolic compounds are a class of antioxidant agents which can adsorb and neutralize the free radicals. It has been established that phenolic compounds are the major plant compounds with antioxidant activity and this activity is due to their redox properties [30] . Flavonoids and flavonols are regarded as one of the most widespread groups of natural constituents found in the plants. It has been recognized that both flavonoids and flavonols show antioxidant activity through scavenging or chelating process [31] .

The level of phenolic compounds in different solvent extracts (methanol, aq.methanol and acetone) of the different parts of *L. laxa* and *G. assamicus* are shown in table 2. The screening of the methanol, aq methanol and acetone extracts of different parts of *L.laxa* and *G. assamicus* revealed that there was a wide variation in the amount of total phenolics ranging from 0.75±0.09 to 15.11±0.15 mg GAE/g dry material and 0.874±0.07 to 10.71±0.11 mg GAE/g dry material respectively. The highest amount of phenolic content was found in the Aq. methanol extract of the whole plant *L. laxa* (15.11±0.15 mg GAE/g dry material), while least amount was observed in the acetone extract of this plant (0.75±0.09 GAE). The aq. methanol extract of the leaf of *G. assamicus* contain highest amount of phenolic compound (10.71±0.11 GAE) whereas least amount of phenolic was found in the acetone extract of the seed pod of this plant (0.874±0.07 GAE). In this study the content of phenolic components extracted by aq methanol was much higher than that extracted by methanol and acetone. This may be due to the fact that phenolics are often extracted in higher amounts in more polar solvents such as aqueous methanol/ethanol as compared with absolute methanol/ethanol or acetone [32-33].

**Table 2. Total phenolic content in the different extract of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | Total phenolic content (GAE equivalent mg/g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 9.99±0.32 | 7.61±0.10 | 1.83±0.10 |
| 2 | *Lysimachia laxa* | Stem | 10.21±0.27 | 7.73±0.07 | 3.62±0.18 |
| 3 | *Lysimachia laxa* | Whole plant | 13.47±0.24 | 15.11±0.15 | 0.75±0.09 |
| 4 | *Gymnocladus assamicus* | Whole plant | 8.36±0.45 | 10.71±0.11 | 4.39±0.22 |
| 5 | *Gymnocladus assamicus* | Seed | 5.71±0.33 | 1.24±0.03 | 1.72±0.31 |
| 6 | *Gymnocladus assamicus* | Seed pod | 2.66±0.07 | 2.35±0.11 | 0.874±0.07 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ±SEM*

Total flavonoids content of different plant materials, using three different solvent systems are presented in Table 3. The flavonoid content of the different extracts of *L. laxa* and *G. assamicus* in terms of Rutin equivalent were between 2.55± 0.03 to 7.29± 0.01 and 0.69±0.019 to 5.05±0.01 mg/g dry material respectively. Highest amount of flavonoid content was observed in the aq. methanol extract of the whole parts of both the plant under investigation. Results of the present study showed that the aq. methanolic extracts were better for flavonoid extraction.

**Table 3. Total flavonoid content in the different extract of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | Total flavonoid content (Rutin equivalent mg/g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 4.45±0.01 | 4.81±0.006 | 3.39±0.02 |
| 2 | *Lysimachia laxa* | Stem | 2.55±0.03 | 3.60±0.005 | 5.75±0.04 |
| 3 | *Lysimachia laxa* | Whole plant | 6.75±0.03 | 7.29±0.01 | 3.56±0.04 |
| 4 | *Gymnocladus assamicus* | Whole plant | 3.75±0.01 | 5.05±0.01 | 3.76±03 |
| 5 | *Gymnocladus assamicus* | Seed | 1.27±0.008 | 0.83±0.002 | 4.71±0.07 |
| 6 | *Gymnocladus assamicus* | Seed pod | 0.64±0.004 | 0.69±0.019 | 0.920±0.012 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ±SEM*

In case of flavonol, the highest amount was observed in the methanol extract of the whole plant of *L. laxa* (10.40±0.078 mg quercetin equivalent/g dry material and *G. assamicus* (8.43±0.011 mg quercetin equivalent/g dry material) (Table 4).

**Table 4. Total flavonol content in the different extract of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | Total flavonol content (Quercetin equivalent mg/g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 5.11±0.038 | 3.89±0.015 | 6.79±0.09 |
| 2 | *Lysimachia laxa* | Stem | 5.09±0.045 | 7.93±0.318 | 7.65±0.04 |
| 3 | *Lysimachia laxa* | Whole plant | 10.40±0.078 | 5.41±0.157 | 7.80±.25 |
| 4 | *Gymnocladus assamicus* | Whole plant | 8.43±0.011 | 7.66±0.061 | 7.89±0.12 |
| 5 | *Gymnocladus assamicus* | Seed | 7.19±0.056 | 4.04±0.140 | 5.18±0.031 |
| 6 | *Gymnocladus assamicus* | Seed pod | 3.43±0.023 | 2.42±0.178 | 1.38±0.04 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ±SEM*

The results strongly suggest that phenolics are important components of these plants. The other phenolic compounds such as flavonoids, flavonols, which contain hydroxyls are responsible for the radical scavenging effect in the plants. According to our study, aq. methanol is the best solvent for the extraction of phenolic components and flavonoids from the plant whereas very good amount of flavonol can be obtained using methanol as extracting solvent.

***Reducing power of the extracts***

The reducing capacity of a compound may serve as a significant indicator of its potential antioxidant activity. The reducing ability is generally associated with the presence of reductones which breaks the free radical chain by donating a hydrogen atom [34]. The reducing powers of these two plants were evaluated as mg AAE/g dry material as shown in Table 5. The highest reducing power was exhibited by the aq methanol extract of the whole plant of *L. laxa* (42.60±0.16mg/g AAE) which is also high in phenolic content (15.11±0.15 mg GAE/g dry material) and acetone extract of the leaf showed lowest activity (13.14±0.12 mg/g AAE) in terms of ascorbic acid equivalent. The methanol extract of the whole plant of *G. assamicus* which contain an appreciable amount of flavonol, exhibited highest reducing power (32.53±0.19 mg/g AAE) whereas minimum reducing power was observed in the acetone extract of the seed pod of this plant. In general, the aqueous methanol and methanol extracts of the tested plant materials, exhibiting greater phenol, flavonoids and flavonol content, also depicted good reducing power in the

present analysis. In this assay, the presence of antioxidants in the extracts reduced $Fe^{+3}$/ferricyanide complex to the ferrous form. This reducing capacity of the extracts may serve as an indicator of potential antioxidant activities through the action of breaking the free radical chain by donating hydrogen atom [35] .

**Table 5. Reducing power of the different extract of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | Reducing power (Ascorbic acid equivalent mg/g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 26.08±0.08 | 19.13±0.18 | 13.14±0.12 |
| 2 | *Lysimachia laxa* | Stem | 41.75±0.22 | 41.01± 0.26 | 22.04±0.18 |
| 3 | *Lysimachia laxa* | Whole plant | 41.11±0.06 | 42.60±0.16 | 21.17±0.39 |
| 4 | *Gymnocladus assamicus* | Whole plant | 32.53±0.19 | 21.40±0.08 | 15.94±.12 |
| 5 | *Gymnocladus assamicus* | Seed | 31.80±0.11 | 16.98±0.02 | 13.77±0.29 |
| 6 | *Gymnocladus assamicus* | Seed pod | 14.37±0.05 | 10.16±0.24 | 5.63±0.08 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ±SEM*

***DPPH radical scavenging activity of the extracts***

DPPH stable free radical method is an easy, rapid and sensitive way to survey the antioxidant activity of a specific compound or plant extracts [36] . The antioxidant effect is proportional to the disappearance of the purple colour of DPPH in test samples. Thus antioxidant molecules can quench DPPH free radicals by providing hydrogen atom or by electron donation and a colorless stable molecule 2,2- diphenyl-1-hydrazine is formed and as a result of which the absorbance ( at 517 nm) of the solution is decreased. Hence the more potent antioxidant, more decrease in absorbance is seen and consequently the $IC_{50}$ value will be minimum.

The evaluation of anti-radical properties of the three different solvent extracts of different parts of two plants was performed by DPPH radical scavenging assay. The effect of solvent used for the extraction of antioxidant components on DPPH scavenging capacity by the different plant materials was shown in Table 6. A lower $IC_{50}$ value would reflect greater

**Table 6 . DPPH radical scavenging activity of the Methanol extract of *Lysimachia laxa* and *Gymnocladus assamicus***

| Sl No | Name of the plant | Parts used | DPPH radical scavenging activity ($IC_{50}$ mg/g dry material) | | |
|---|---|---|---|---|---|
| | | | Methanol | Aq. methanol | Acetone |
| 1 | *Lysimachia laxa* | Leaf | 6.58±0.40 | 3.37±0.06 | 7.80±1.37 |
| 2 | *Lysimachia laxa* | Stem | 5.83±0.26 | 2.62±0.06 | 3.35±0.14 |
| 3 | *Lysimachia laxa* | Whole plant | 2.35±0.06 | 2.05±0.003 | 3.80±0.51 |
| 4 | *Gymnocladus assamicus* | Whole plant | 1.30±0.002 | 1.26±0.01 | 2.34±0.09 |
| 5 | *Gymnocladus assamicus* | Seed | 11.47±2.23 | 8.48± 0.80 | 5.71±0.47 |
| 6 | *Gymnocladus assamicus* | Seed pod | 9.44±0.40 | 7.25±0.10 | 15.33±0.64 |

*Each value in the table was obtained by calculating the average of three experiments and data are presented as Mean ±SEM*

antioxidant activity of the sample. In the present study the highest radical scavenging activity was shown by the aq. methanol extract of the whole parts of both the plant [*L.laxa* ($IC_{50}$ = 2.05±0.003 mg dry material), *G. assamicus* ($IC_{50}$ = 1.26±0.01 mg dry material)]. The high radical scavenging property of the aq. methanol extract of *L. laxa* and *G. assamicus* may be due to the presence of highest amount of phenolic component and flavonoid content that can provide the necessary component as a radical scavenger. The free radical scavenging activity of antioxidant components is very much associated with their phenolic and flavonoid component [37]. The methanolic, aq. methanolic and acetone extracts of these two plants under investigation exhibited different extent of antioxidant activity. It can be concluded that the extracts obtained using high polarity solvents (aq. methanol) were considerably more effective radical scavengers than those using less polarity solvents (methanol, acetone), indicating that antioxidant or active compounds of different polarity could be present in the different parts of the plant under investigation. and thus provide a valuable source of nutraceutical supplements. Depending on the values, some plants are more important than some others.

## CONCLUSION

The result of present study showed that the aq. methanol extract of the whole parts of the plant *L. laxa* and *G. assamicus* which contain highest amount of phenolic compounds and flavonoids appreciable amount of and flavonols exhibited the greatest reducing power and also showed strong radical scavenging activity. The highest radical scavenging activity and very strong reducing power of the aq. methanol extract of these two plants may be

due to the presence of a very good amount of total phenolics, flavonoids content in these plant. The results showed that the yield and the antioxidant activity of these two plants depends on the type of solvent used for extraction of phenolics, flavonoids and flavonols as well as the part of the plant material used in the present investigation. For total phenolics, flavonoids and DPPH radical scavenging activity, the aq. methanol was more efficient compare to methanol and acetone extract of the whole part of these two plants. The radical scavenging activities of the selected plant extracts are still less effective than the commercial available synthetic like BHT. As the plant extracts are quite safe and the use of synthetic antioxidant has been limited because of their toxicity, therefore, these plants could be exploited as antioxidant additives or as nutritional supplements. However, further investigation is required to isolate and characterize the individual components from these plants which are actually responsible for their antioxidant activities and develop their applications for food and pharmaceutical industries.

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