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QUALITATIVE CHARACTERIZATION OF PHYTOCHEMICALS PRESENT IN THE BARK FROM SOME SELECTED TREE SPECIES.

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ABSTRACT

Plants have tremendous properties which are used in our day to day life. Majorly plant used as source of food, fabrics and medicine. Hence the identification of plants is very essential process. Generally, plants are identified by their primary structures of growth; such as leaves, flowers and fruits. In the absence of these organs bark is the only outermost region of the plants; through which identification of the particular plant can be identified. Bark is the secondary structure and which persist throughout the plant life. Bark is composed of varioustissues which are incorporated with many phyto-chemicals which are photosynthetic productsas well as bio products jointly known as photosynthates. Due to the presence of these photosynthates bark are used as medicine. Hence the preparation of data that includes information about phytochemical present in bark leads us to the new aspect towards bark foridentification as well as pharmacological purpose.

Keywords: *Pharmacognosy, Bark, Identification, Phyto-chemicals, Photosynthates.*

INTRODUCTION

Fig.5 ***Peltophorum pterocarpum***

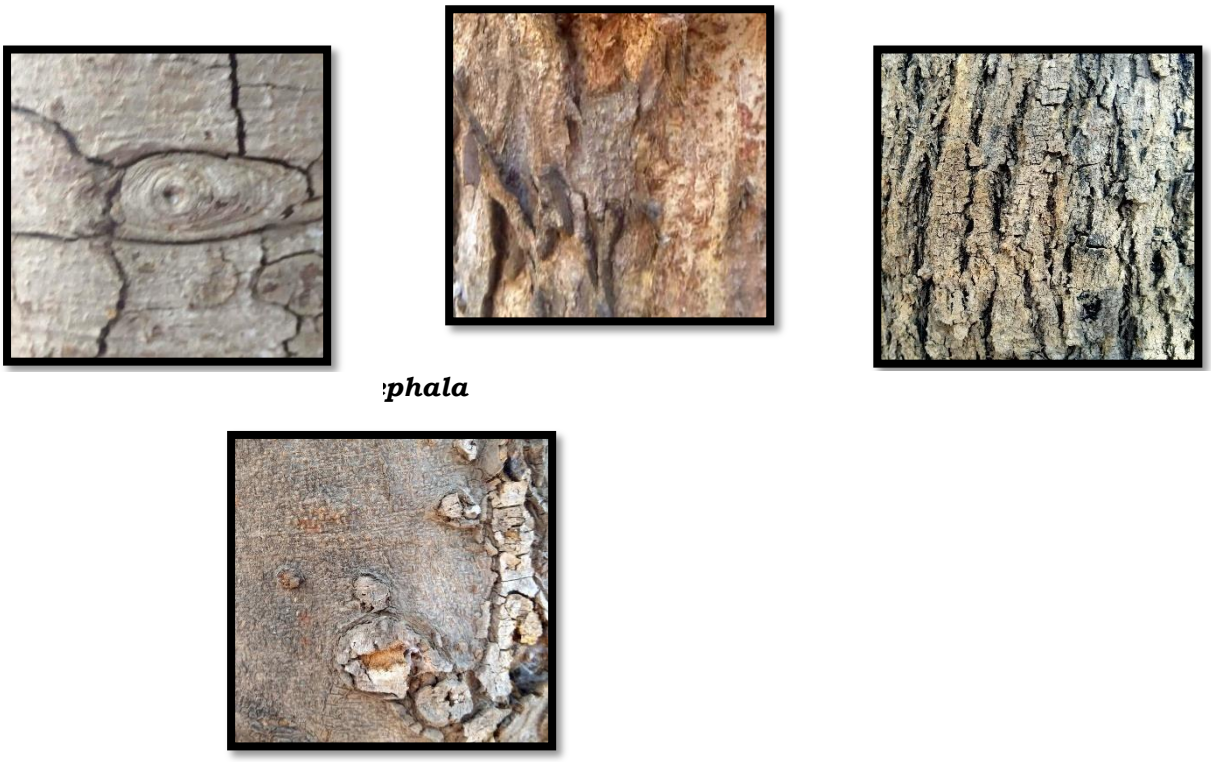
India is the country where Ayurveda play a vital role in health of society. From the ancient times to till the date of today, plants serve us in many manners especially in preparation of drug. Plants around us exhibit many roles in our day to day life. Every plant has its own properties and these properties are the result of their metabolic activity or their chemical composition. Trees around us have many known and unknown properties according to their various organs viz, leaf, stem, root, bark, flowers and fruits. Among all the parts Bark of tree have many medicinal uses and it is also showing variation in their morphology as well as chemical constituents. That's why plants which are commonly found in campus of M. K. Bhavnagar University were chosen for the qualitative characterization of bark. Trees which are selected are as follows: **(1) *Bauhinia purpurea* L., (2) *Calliandra haematocephala* Hassle, (3) *Dalbergia sissoo* Roxb., (4) *Derris indica* (Lam.)Bennet, (5) *Peltophrum pterocarpum* (DC) Backer**. These plants as well as their family membersare reported for medicinal uses. Like ***Bauhina pupurea*** and other ***Bauhinia*** species plant parts used in bone fracture, foot and mouth disease and also utilized as a tonic (Pullaiah, T. *et. al.,* 2016).On the other hand, ***Calliandra haematocephala*** used traditionally as antioxidant and blood purifier(Moharran, 2006). It also used traditionally as antibacterial (Nia *et. al.*, 1999) (Tiwari, 2016). Different plant parts of ***Dalbergia*** species ethno-medicinally used in jaundice, fever and leucorrhoea, skin eruptions. ***Derris indica*** is being used by ancient times in many diseases like body-ache, piles, itching, ringworm infection, snakebites, toothache, etc. (Pullaiah, T. *et. al, 2016*).Different parts of this tree ***Peltophorum pterocarpum*** are used to treat many diseases like stomatitis, insomnia, skin troubles, constipation, ringworm and its flower extract is known to be agood sleep inducer and used in insomnia treatment. Bark is used as medicine for dysentery, as eyelotion, embrocating for pains and sores. (Jash, *et. al.*

2014) as ethnomedicines. Qualitative tests forCarbohydrate, Proteins, Phenols, Alkaloids, Amino acids, Flavonoids are taking place from bark samples. These compounds are responsible for their medicinal value. Qualitative analysis is the first step towards the characterization of chemical constituents present in the bark and utilization of this information for the identification purpose.

MATERIALS AND METHOD

Sample Collection:

Bark from each species collected in the month of March, 2019. The bark peeled from the tree trunkof the size 6 x 6 inches (Fig. 1 to 5). These bark pieces were dried for twenty days and then finelygrinded. The powder collected and stored in air-tight containers at dry place (Fig. 6).



phala

Fig.4 ***Derris indica***

Extraction:

Extraction was taken place in distilled water according decoction method (Handa S. S. *et. al.,* 2008)30 gm of bark powder was dissolved in 5000ml distilled water and then volume is brought down to ¼ to its original volume by boiling during the extraction procedure. The concentrated extract isfiltered and used as a sample (Fig.7).

Fig.6 Drying of Collected Bark Fig.7 Extraction of Bark Sample bydecoction Method

Qualitative Phytochemical Analysis:

Various qualitative tests for different aspect take place viz. for, Carbohydrate, Amino acids, Alkaloids, Flavonoids, Tannin, Phenolic compound and Protein (Kokate *et. al.,* 2001; ThimmaiahS.K., 2016)
Carbohydrates:

The qualitative analysis of carbohydrates is carried out with standard biochemical tests described by Thimmaiah S.K. in 2016 which are Molisch Test, Fehling test, Benedict Test, Barfoed Test, Bial Test, Seliwanoff Test, Mucic acid test, Osazone formation and Iodine test.
Amino acids

Detection of various Amino acids is taken place by different tests like Ninhydrin test, Millons test,Xanthoproteic test, Hopkin - Cole test, Ehrlich test, Pauly test, Sakiguchi test as well as Nitroprusside test (Thimmaiah S.K., 2016).

Proteins:

As per Thimmaiah (2016) test for protein detection performed viz. Biuret test, Folin- Ciocalteu reaction (FCR) or Lowry test.
Secondary metabolites:

Qualitative analysis of secondary metabolites like Alkaloids, Phenolic compounds, Flavonoids andtannin are done by using procedure described by Kokate in 1994. Detection of Alkaloid is carriedout with Mayer test, Wagner test, and Hager test. While detection of Phenolic compound and Flavonoids done by Lead acetate test. On the other hand, Ferric chloride test reveals the presenceor absence of Tannin (Kokate *et al.,* 2001; Rathinam *et. al,* 2012).

RESULT

Carbohydrates:

All five plants show positive results for Molisch test, Fehling test, Benedict test, Seliwanoff test and Phenyhydrazine test. On the other hand, Barfoed test is positive ***C. haematocephala*** and ***P. pterocrpum*** (Fig. 8) (Table No.1).
Proteins:

Both Biuret test and Folin- Lowery's test are positive for all he five plants:***B. purpurea***, ***C. haematocephala***, ***D. sissoo, D. indica***, ***P. pterocarpum*** (Fig. 9) (Table No.2).
Amino acids:

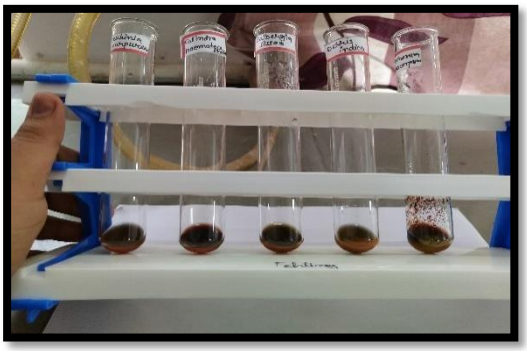
Included all five plants are show positive results for Ninhydrin test, Xanthoproteic test,

Hopkin- Cole test, Ehrlich test and Pauly's test. While ***C. haematocephala*** and ***P. pterocrpum*** express positive results for Millon's Test. (Fig. 10) (Table No. 3)

Secondary Metabolites:

B. purpurea, ***C. haematocephala***, ***D. sissoo***, and ***D. indica*** show positive results for all the tests for alkaloids Viz. Mayer's test, Wagner's test and Hagers test. All five plants showed positive results of Lead acetate test (Phenolic compounds and Flavonoids).
P. pterocrpum express positive result of Gelatin test for phenolic compounds. ***B. purpurea***,

C. haematocephala, ***D. sissoo,*** P. ***pterocarpum*** give positive result for presence of Tannin. (Fig. 11 to 13) (Table No. 4 to 7)



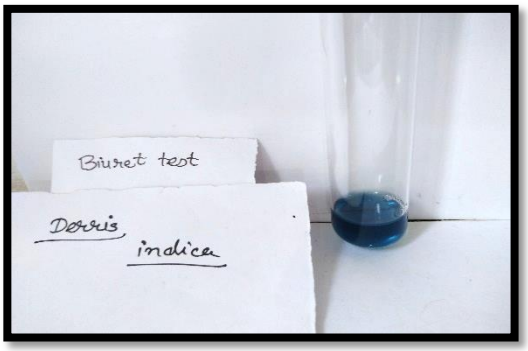


Fig. 8 Fehling's test for Carbohydrates

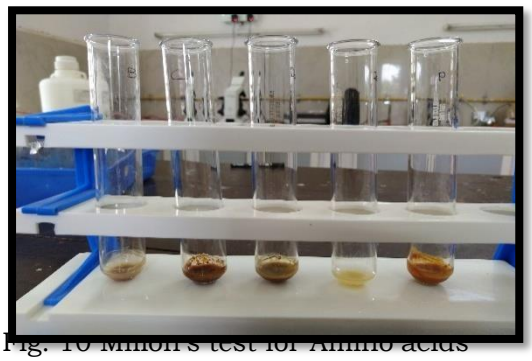


Fig. 10 Millon's test for Amino acids
Fig. 9 Biuret test for Proteins (***D. indica***)

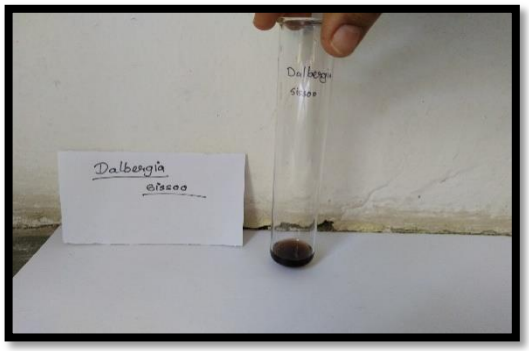


Fig. 11 Mayer's test for alkaloids (***D. sissoo***)

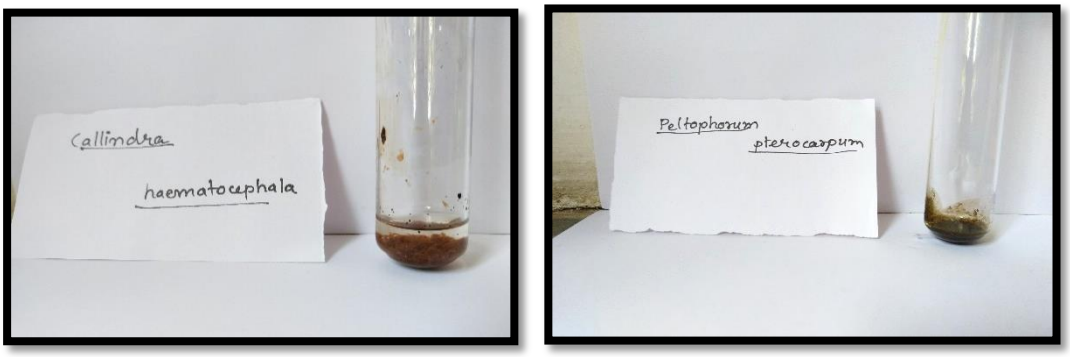


Fig. 12 Lead acetate test for Phenolic and Flavonoids(***C. haemtocephala***)
Fig. 13 Ferric Chloride test forTannins (***P. pterocarpum***)

Table No.1 Carbohydrates test:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	Molisch	+	+	+	+	+
2.	Fehling	+	+	+	+	+
3.	Benedict	+	+	+	+	+
4.	Barfoed	-	+	-	-	+
5.	Bial	-	-	-	-	-
6.	Seliwanoff	+	+	+	+	+
7.	Mucic acid	-	-	-	-	-
8.	Phenylhydrazine	+	+	+	+	+
9.	Iodine	-	-	+	-	-

Table No.2 Proteins test:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	Biuret	+	+	+	+	+
2.	Folin - Lowery	+	+	+	+	+

Table No.3 Amino acids test:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	Ninhydrin	+	+	+	+	+
2.	Millon	-	+	-	-	+
3.	Xanthoproteic	+	+	+	+	+
4.	Hopkin- Cole	+	+	+	+	+
5.	Ehrlich	+	+	+	+	+
6.	Pauly	+	+	+	+	+
7.	Sakaguchi	-	-	-	-	-
8.	Nitroprusside	-	-	-	-	-

Table No.4 Alkaloids test:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	Mayer	+	+	+	+	-
2.	Wagner	+	+	+	+	-
3.	Hager	+	+	+	+	-

Table No.5 Tests for phenolic compounds:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	**Lead acetate**	**+**	**+**	**+**	**+**	**+**
2.	**Gelatine test**	**-**	**-**	**-**	**-**	**+**

Table No.6 Tests for Flavonoids:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	**Lead acetate**	**+**	**+**	**+**	**+**	**+**

Table No.7 Tests for Tannins:

Sr. no.	Name of test	*Bauhinia purpurea*	*Callindra haematocephala*	*Dalbergia sissoo*	*Derris indica*	*Peltophorum pterocarpum*
1.	**Ferric chloride**	**+**	**+**	**+**	**-**	**+**

DISCUSSION

Sample of all the five tree barks show positive result for benedict test which indicate the presenceof reducing sugars. ***C. haemetocephala*** and ***P. peltophorum*** show positive result for barfoed test which indicate presence of monosaccharides while other three ***B. purpurea***, ***D. sissoo***, ***D. indica*** show negative result which show presence of disaccharides. Positive results of all sample for Seliwanoff's test indicate presence of the fructose in the bark. While negative results for bial's testshows presence of aldohexoses. Presence of starch indicated by Iodine test for ***D. sissoo*** (Fig 8) (Table No.1).

All plants show positive results folin- lowery and biuret's test that indicate presence of proteins (Fig. 9) (Table No.2). In all plant, ninhydrin test is positive which indicate presence of all amino acids except proline and hydroxyproline. Positive results for xanthoproteic test and Pauly's test indicate the possibilities for presence of phenylalanine, tyrosine, tryptophan and histidine. While positive results of Millon's test (Fig. 10) and Hopkin-cole's test and Ehrlich's test confirm the presence of tyrosine and tryptophan respectively. Amino acids like tryptophan is used to treat patient with insomnia, anxiety, grinding teeth during sleep and improve sleep/wake cycle in adults.(Bravo R, *et. al.* 2013). While tyrosine has antioxidant properties and also found to be useful duringconditions of stress, cold, fatigue (in mice), (Hao S, *et. al.* 2001) prolonged work and sleep deprivation, with reductions in stress hormone levels also improved metal health, alertness and memory. (Neri D. F., *et. al.* 1995) (Magill R. A., *et. al.* 2003). The negative result of Sakaguchi and Nitroprusside test indicates the absence of Arginine, cysteine and methionine (Table No. 3).

All plants except ***P. pterocarpum*** show positive result for all the tests for alkaloids (Fig. 11) (TableNo.4). As per Perviz, S. e*t. al.* (2016) alkaloids are the secondary metabolites which are effective as antidepressant. Positive results of all five plants for lead acetate test indicate the

presence of Flavonoids and phenolic compound (Fig.12) (Table No.5,6), presence of flavonoids and phenolic compound make these barks useful as antioxidant, antibacterial and anti-inflammatory compoundin medicines. They are shows anticancer activity, cardioprotective effects and Anti glycemic activity etc. (Cheplick *et. al.* 2010) (Tungmunnithum D, *et. al.* 2018) Among all the plants except *P. peltophorum* shows negative result for gelatin test which indicate presence of phenolic compound (Table No.5). All the plant except *D. indica* indicate shows the positive result for ferric chloride test which indicate the presence of tannin in bark (Fig. 13) (TableNo. 7) which is match with the sentence quoted by Tibiri, *et. al.* (2007) that the bark exhibited thehighest Total Phenol Content (TPC) which is understandably due to the high content of tannins normally found in barks.
Presence of chemicals like phenols, alkaloids, tannins, proteins, amino acids indicate these plant bark can used as a medicine.

CONCLUSION

The tree bark is the result of secondary growth in plants and also plays an important role in the identification of tree species. Although it is tough to classify plants with bark morphology, the chemical composition of bark lead to the preparation of identification key for tree with medicinal values. As noted in results that the chemicals present in bark have different medicinal value and itcan be characterized by qualitative as well as quantitative analysis.

REFERENCES

1) Barbosa, A. & Silva, B. & Parente, J. P. (2012). Evaluation of the gastroprotective activity of Calliandra haematocephala extracts. Planta Medica. 78. 10.1055/s-0032-1321135.
2) Brahmachari A. K., Nath S., Brahmachari G., Batabyal S., Mandal T. K, Bandopadhyay S. K, Sinha B. P, Pandey A. K. (2016) Modulatory effect of semi purified fractions of *Bauhinia purpurea* L. bark extract on oxidative stress in STZ-induced diabetic rats. ExploreAnim Med Res 6(1): 26-36.
3) Bravo, R., Matito, S., Cubero, J., Paredes, S. D., Franco, L., Rivero, M., Rodríguez, A. B., & Barriga, C. (2013). Tryptophan-enriched cereal intake improves nocturnal sleep, melatonin, serotonin, and total antioxidant capacity levels and mood in elderly humans. Age (Dordrecht, Netherlands), 35(4), 1277–1285. https://doi.org/10.1007/s11357-012-9419-5.
4) Burkill H. M. The useful plants of West Tropical Africa. Royal Botanic Gardens, Kew, Second Edition, Vol. III, p. 100–143, 1995.
5) Chaturvedi P., Pipedi-Tshekiso M., Moseki B., Kwape T. (2010). Hepatoprotective potentials of water extract of Bauhinia purpurea bark against alcohol induced toxicity. SciRes Essays. 6. 10.5897/SRE10.825.
6) Cheplick, S., Kwon, Y. I., Bhowmik, P., & Shetty, K. (2010). Phenolic linked variation in strawberry cultivars for potential dietary management of hyperglycemia and related complications of hypertension. Bioresource Technology, 101, 404–413.
7) Handa, S.S., Khanuja, S., Longo, G., Rakesh, D.D. (2008). Extraction technologies for medicinal and aromatic plants. International centre for science and high technology. 21-25.
8) Hao S, Avraham Y, Bonne O, Berry EM (2001). "Separation-induced body weight loss, impairment in alternation behavior, and autonomic tone: Effects of tyrosine". *Pharmacol. Biochem. Behav.* **68** (2): 273–81. doi:10.1016/S0091-3057(00)00448-2
9) Hari, Balaji & Padmaja, T.K. & Parim, Brahmanaidu & Naik, Roopa & Meriga, Balaji. (2012). Anti-inflammatory and antioxidant activity of ethanolic extract of Bauhinia purpurea bark. International Journal of Drug Delivery. 4. 507-514.
10) Jash, S., Singh, R., Majhi, S., Sarkar, A. & Gorai, D. (2014). Peltophorum pterocarpum: Chemical and Pharmacological Aspects. International Journal of Pharmaceutical Sciences and Research. 5. 26-36. 10.13040/IJPSR.0975-8232.5(1).26-36.
11) Josephine I. G., Punnagai K. and Muthiah N.S. (2017) In vitro Antibacterial Activity Of Ethanolic Extract of 'Calliandra Haematocephala' against Selected Bacterial Strains. Biomedical & Pharmacology Journal Vol. 10(3), 1279-1284
12) Joshi, N., Bhatt, S., Dhyani, Dr, Nain, J. (2013). Phytochemical screening of secondary metabolites of Argemone mexicana Linn. Flowers. International Journal of Current Pharmaceutical Research. 5. 144-147.
13) Kokate C.K., (1994) Practical Pharmacognosy, 4th ed., Vallabh Prakasan, Delhi, 107-111.

13.
14) Kokate C. K., Purohit A. P. and Gokhale S. B. (2001) Carbohydrate and derived Products, drugs containing glycosides, drugs containing tannins, lipids and protein alkaloids. Text book of Pharmacognosy, 7, edition: 133 -166, 167- 254, 255-2 69, 272- 310, 428-52 3. 14.
15) Magill R. A., Waters W. F., Bray G. A., Volaufova J., Smith S. R., Lieberman H. R., McNevin N., Ryan D. H. (2003). "Effects of tyrosine, phentermine, caffeine D- amphetamine, and placebo on cognitive and motor performance deficits during sleep deprivation". *Nutritional Neuroscience.* 6(4): 237-46 Doi:10.1080/1028415031000120552
16) Manaharan, T., Teng, L., Appleton, D. & Cheng, M., Masilamani, T.& Palanisamy, U. (2011). Antioxidant and antiglycemic potential of Peltophorum pterocarpum plant parts. Food Chemistry. 129. 1355-1361. 10.1016/j.foodchem.2011.05.041.
17) Moharram F. A., Marzouk M. S., Ibrahim M. T., Mabry T. J., (2006). Antioxidant Galloylated Flavonol Glycosides from Calliandra haematocephala; Natural Product Research, 20(10): 927 – 934.
18) Mopuri, R., Attitalla, I., Avinash, P., Meriga, B. (2010). Evaluation of Antilipidemic and Anti-Obesity Efficacy of Bauhinia purpurea Bark Extract on Rats Fed with High Fat Diet. Academic Journal of Plant Sciences.
19) Neri D. F., Wiegmann D., Stanny R. R., Shappell S. A., McCardie A., McKay D. L. (1995). "The effects of tyrosine on cognitive performance during extended wakefulness". *Aviation, Space, and Environmental Medicine*. **66** (4): 313–9.
20) Nia R, Adesanya S. A., Okeke I. N., Illoh H. C., Adesina S. J., (1999). Antibacterial Constituents of Calliandra haematocephala. Nigerian Journal of Natural Products and Medicine, 3:58-60.
21) Perviz S., Khan H., Pervaiz A. (2016) Plant Alkaloids as an Emerging Therapeutic Alternative for the Treatment of Depression. *Front Pharmacol.* 2016; 7:28. Published 2016 Feb 15. doi:10.3389/fphar.2016.00028
22) Pullaiah, T., Krishnamurthy, K., Bahadur, B. (2016). Ethnobotany of India, Volume 2. New York: Apple Academic Press, https://doi.org/10.1201/9781315366142
23) Rathinam, P., Kothapalli B., Chandrasekhar, Sekhar, D. (2012). Antimicrobial activity and phytochemical constituents of combined extracts of Cissus quadrangularis and Aegle marmelos. International Journal of Research in Pharmaceutical Sciences. 3.
24) Satish S., Mohana D. C., Ranhavendra M. P. and Raveesha K. A.(2007). Antifungal activity of some plant extracts against important seed borne pathogens of Aspergillus sp. Journal of Agriculture and Technology 2007; 3(1): 109–119.
25) Tiwari J, Shukla A. (2016). Investigations on Calliandra haematocephala flowers extract for in-vitro anthelmintic activity. Advance Pharmaceutical Journal, 1(1):17-20.
26) Siri S., Wadbua P., Wongphathanakul W., Kitancharoen N. and Chantaranothai P., (2008) Antibacterial and phytochemical studies of 20 Thai medicinal plants against catfish infectious bacteria, Aeromonas caviae. Khon Kaen University Science Journal 2008; 36(S.1): 1–10.
27) Thimmaiah, S. K. 1999. Standard Methods of Biochemical Analysis. Second Edition, Kalyani Publication, New Delhi, India.
28) Tibiri, A., Rakotonandrasana, O., Nacoulma, G. O., & Banzouzi, J. T. (2007). Radical scavenging activity, phenolic content and cytotoxicity of bark and leaves extracts of Entada Africana Guill. And Perr. (Mimosaceae). Journal of Biological Sciences, 7(6), 959–963.
29) Tungmunnithum D, Thongboonyou A, Pholboon A, Yangsabai A. Flavonoids and Other Phenolic Compounds from Medicinal Plants for Pharmaceutical and Medical Aspects: An Overview. *Medicines (Basel).* 2018;5(3):93. Published 2018 Aug 25. doi:10.3390/medicines5030093