IJPSR (2013), Vol. 4, Issue 12 **(Research Article)**

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 14 April, 2013; received in revised form, 10 June, 2013; accepted, 14 August, 2013; published 01 December, 2013

EVALUATION OF *LEONOTIS NEPETAEFOLIA* FOR ITS PHYTOCHEMICAL AND HEAVY METAL ANALYSIS

M. Kamalam*, C. Saraswathi and U. Umadevi

PG and Research Department of Botany, PSGR Krishnammal College for women, Coimbatore, Tamil Nadu, India

Keywords:
Leonotis nepetaefolia, Phytochemical, HPTLC studies, Heavy metal analysis

Correspondence to Author:

Dr. M. Kamalam

Associate Professor, PG and Research Department of Botany, PSGR Krishnammal College for women, Coimbatore, Tamil Nadu, India
E-mail: kamaluma12@gmail.com

ABSTRACT: Phytochemical, HPTLC and Heavy metal analysis of Leaf of *Leonotis nepetaefolia* was done. Qualitative phytochemical screening of Leaf was done using different solvents which showed the presence of various vital secondary metabolites. Neutraceutical studies revealed the presence of carbohydrates, proteins and aminoacids. HPTLC studies confirmed the presence of flavonoids, diterpene, phenolic class of compounds. Heavy metal analysis showed the presence of Lead, Cadmium, Chromium and copper in all the samples analyzed.

INTRODUCTION: *Leonotis nepetaefolia* is a little known medicinal plant, belonging to the family Lamiaceae, consisting of 220 genera. It is found almost along the hotter parts of India. Traditionally, the leaf is used for rheumatic problems and also serves as a tonic. Flower heads are used against scalds, burns, ringworm and some skin diseases.

Crushed root is applied locally for facilitating breast milk and also used in the treatment of rheumatism, rickets, headache and wounds [1]. It has got anticancer, anti-inflammatory, anti-diarrheal, antioxidant, antibacterial properties [2]. In Ayurvedic system it is used to treat fever, bronchial asthma, malaria and influenza [3]. Since the plant is highly medicinal it has to be explored more scientifically

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.4(12).4591-96

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.4(12).4591-96

MATERIAL AND METHODS: The plant material was collected from wild condition, Salem District of Tamil Nadu. The leaves collected were shade dried, powdered mechanically and stored in an airtight container for further studies. The dried leaf powder was defatted with Petroleum ether and further extracted successively with benzene, chloroform, acetone, ethanol and methanol using Soxhlet apparatus.

Qualitative phytochemical analysis: Qualitative Phytochemical analysis was done by following the standard procedures of Harborne[4]. Presence of alkaloids, flavonoids, glycosides, Diterpenes, Phenolic and tannins, carbohydrates, proteins and amino acids were qualitatively analyzed.

HPTLC analysis: Secondary metabolites like alkaloids, flavonoids, diterpenes and phenolic compounds and their derivatives were analyzed by using HPTLC techniques. 2µl of ethanolic extract was loaded as 8mm band length in the 5×10 silica gel 60 F_{254} TLC plate using Hamilton Syringe and CAMAG LINOMAT 5 instrument.

The sample loaded plate was kept in TLC twin trough developing chamber with respective mobile phases for alkaloids, flavonoids, Phenolic compounds and Diterpene and the plates were developed up to 90mm. The developed plate was dried by Hot air to evaporate solvents from the plate and kept in Photo-documentation chamber and sprayed with respective spray reagent for each compound and dried at 110^{o}C in hot air oven. The plate was photo documented in white light and UV 366 nm using photo-documentation chamber.

Heavy Metal Analysis: The plants grown along the road side were collected with soil samples. Then the materials were shade dried powdered and about 1gm of dried Plant material and 1 gm of soil samples were used for heavy metal analysis. The samples were digested in microwave digestion system (Milestone model 1200) with an exhaust model (EM -45/A) using 10ml of HNO_3 (69) %, 1ml of $HClO_4$ (70 %) for 5 min and 5ml of H_2O_2 (30%) for 10 min at 250 WV power setting. The digested samples were analyzed for various metals like lead, cadmium, chromium and copper in double beam Atomic absorption Spectrophotometer.

RESULTS:

Habit and inflorescence of *Leonotis nepetaefolia*:

Qualitative Phytochemical Analysis: The dried leaf powder of *Leonotis nepetaefolia* was extracted with different solvents namely petroleum ether, benzene, chloroform**,** acetone, ethanol and methanol and subjected to qualitative phytochemical analysis. The extracts of acetone ethanol and methanol showed the presence of flavonoids, glycosides, phenols, and tannins. Diterpenes were reported in petroleum ether, benzene and ethanol extract. Alkaloids were absent in all the solvent extracts used. The neutraceutical studies revealed the presence of Carbohydrates, Proteins and amino acids in alcoholic extract (**Table 1**) and it was absent in petroleum ether and benzene extracts.

TABLE 1: QUALITATIVE PHYTOCHEMICAL ANALYSIS OF LEAF POWDER OF *L. NEPETAEFOLIA*

S. No.	Tests	Petroleum ether extract	Benzene extract	Chloroform extract	Acetone extract	Ethanol extract	Methanol extract
1.	Alkaloids	-	-	-	-	-	-
2.	Flavonoids	-	-	-	+	+	+
3.	Glycosides	-	-	+	+	+	+
4.	Diterpenoids	+	+	-	-	+	-
5.	Phenols & Tannins	-	-	-	+	+	+
6.	Carbohydrate	-	-	+	+	+	+
7.	Proteins	-	-	-	-	+	+
8.	Amino acids	-	-	-	-	+	+

HPTLC studies of leaf powder of *L. nepetaefolia* showed the absence of alkaloids (**Plate 1**) and presence of four types of flavonoids, two types of Phenolics and one type of diterpene class of compounds (**Plate 2- 4**).

PLATE 1 - CHROMATOGRAM OF ALKALOID USING HPTLC

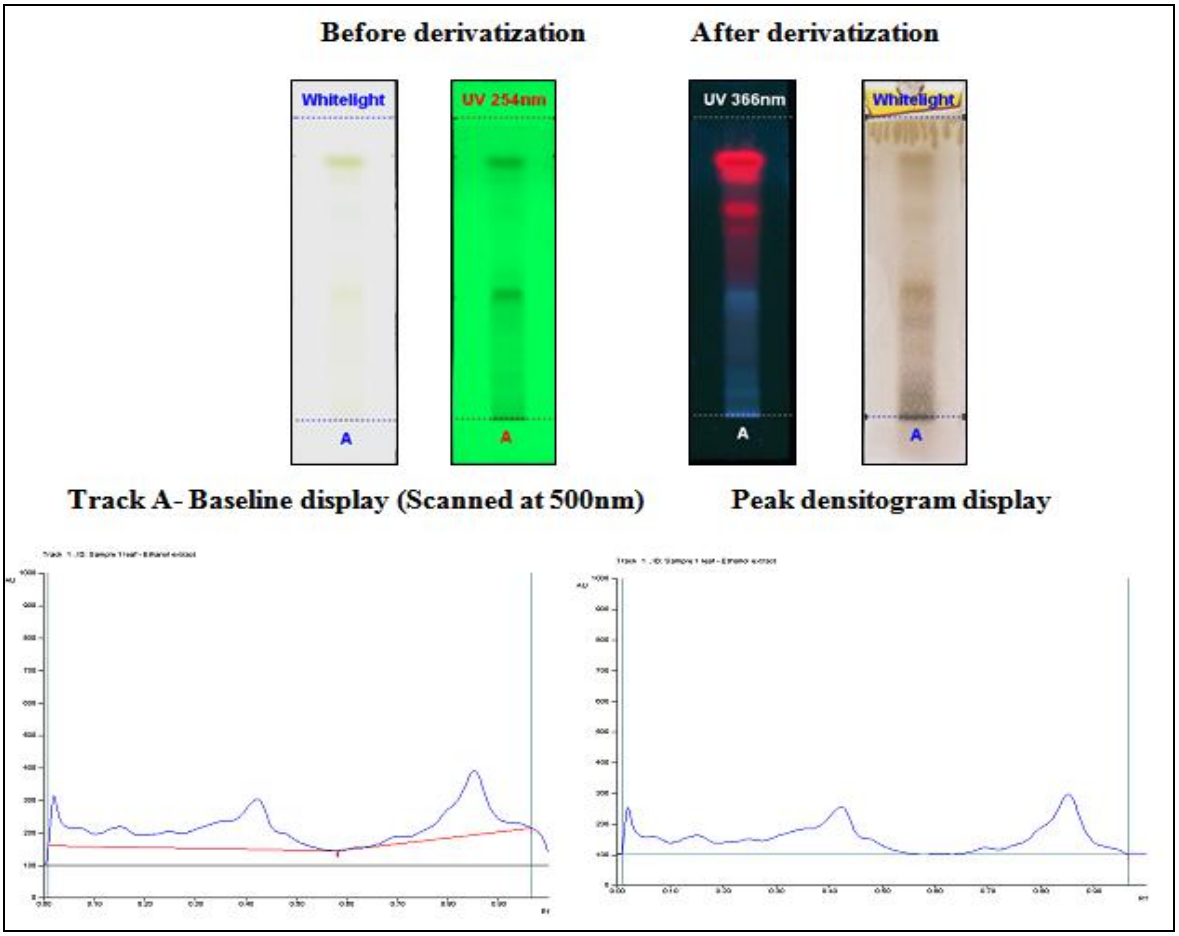


PLATE 2-CHROMATOGRAM OF FLAVONOID USING HPTLC

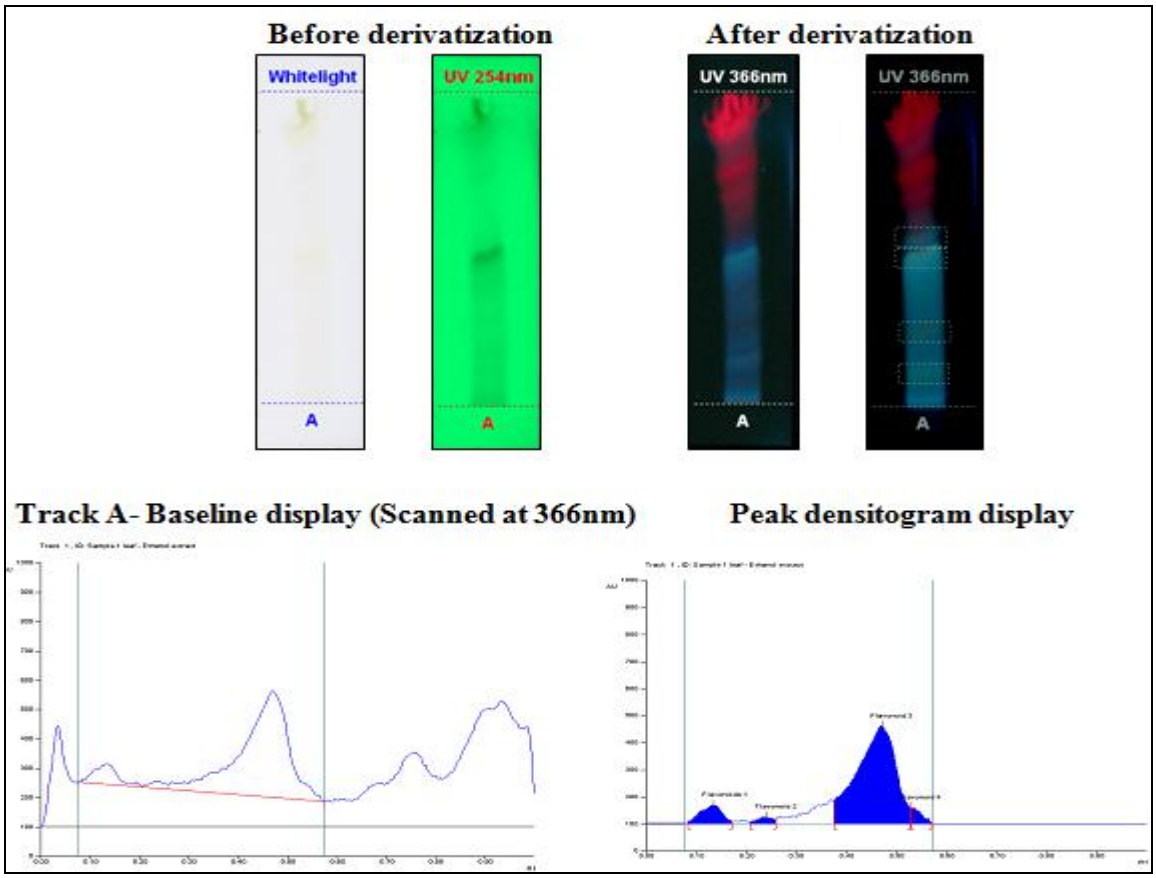


PLATE 3-CHROMATOGRAM OF PHENOLIC COMPOUND USING HPTLC

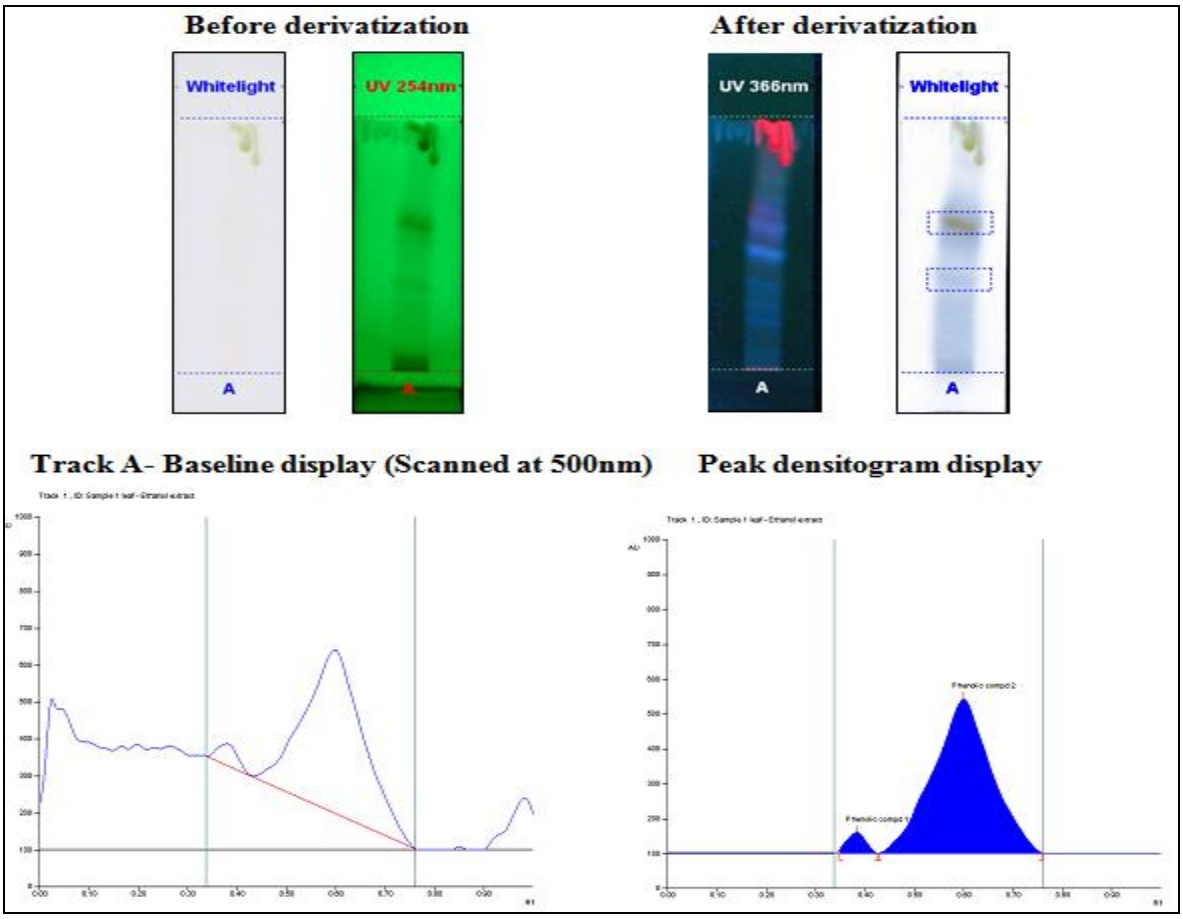
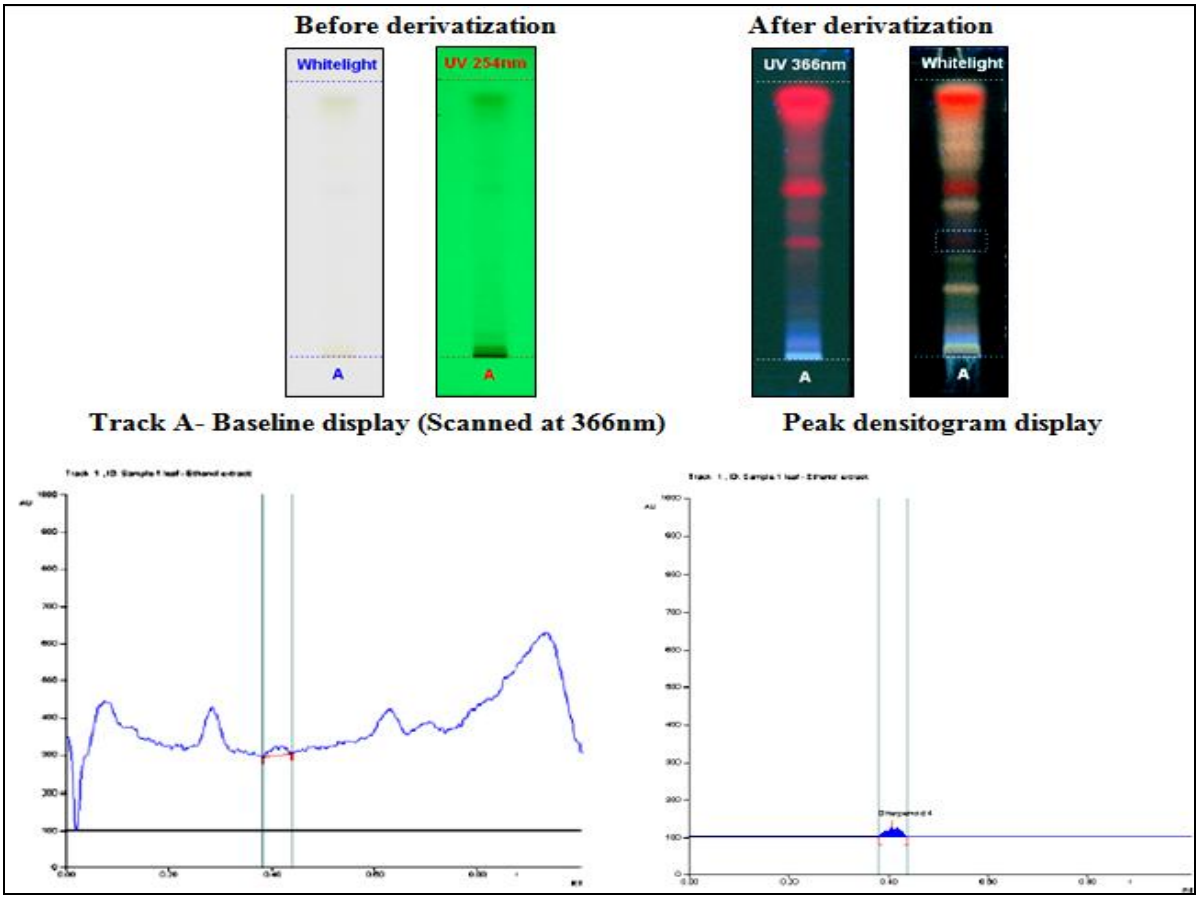


PLATE 4- CHROMATOGRAM OF DITERPENOID USING HPTLC



Heavy Metal Analysis: The result of heavy metal analysis showed that lead and chromium was found at a higher concentration in soil followed by leaf, root and stem. Cadmium is at higher level in root and stem and equal amount of cadmium being accumulated in soil and leaf. In case of copper soil accumulates higher level and followed by root, leaf and stem (**Table 2**).

TABLE 2: METAL CONTAMINATION IN SOIL AND PLANT PARTS OF *L. NEPETAEFOLIA*

Sample	Pb (ppm)	Cr (ppm)	Cd (ppm)	Cu (ppm)
Soil	14.975	18.175	0.4	19.1
Root	8.725	4.125	0.525	16.025
Stem	5.75	2.675	0.5	4.3
Leaf	14.05	4.35	0.4	11.1

DISCUSSION: In the present study, qualitative phytochemical analysis of alcoholic extract of leaf materials showed the presence of many secondary metabolites like flavonoids, glycosides, phenols, tannins and diterpenes. Similar result was reported in the same plant which conforms the presence all the above mentioned phytochemicals except flavonoids [5].

In another report, presence of flavonoids is conformed in this plant [6]. The medicinal property of this plant is enhanced mainly due to the presence of the secondary metabolites and the neutraceutical components [7].

Hence, more phytochemical analysis is needed to identify and quantify the bioactive compounds of the plant.

HPTLC studies confirmed the presence of flavonoids, diterpenoids and phenolic compounds which forms an identification and estimation of chemical marker in plant extract which can also be used for standardization of herbal formulation [8]. Results obtained in this study, may serve as an identification tool for the fingerprinting of any sample claimed as *L. nepetaefolia* in future.

The toxicity status of the plant is assessed by testing the concentration of heavy metals using AAS, revealed the presence of Pb, Cd, Cr and Cu in all the samples analyzed. The concentration of lead in leaves was higher than the permissible level of 10ppm. Accumulation of cadmium in root is higher than the permissible level of 0.3ppm set by WHO [9].

Higher amount of heavy metal accumulation in leaf may be due to external contamination from air and dust particles adhering to it. Usually plants grown in clean regions show lesser concentration of heavy metals than the plants collected from polluted area [10]. Plants present along road side are prone to contamination from vehicle exhaust [11]. Hence, the unsafe level of Pb and Cd is due to environmental pollution in addition to their presence in soil. It is also a fact cadmium concentration increases quickly in plants grown in polluted areas [12], which is also a reason for higher amount of Cadmium accumulation in *L nepetaefolia.* Cr and Cu are under permissible level set by various statutory agencies.

REFERENCE:

1. Ayanwuyi, L.O., Yaro, A.H., and Adamu, H.Y.S: Studies on anticonvulsant activity of methanol capitulum extract of *Leonotis nepetifolia* Linn. Nigerian Journal of Pharmaceutical sciences 2009; 8(1):74-78.
2. Syed Imran, S.S. Suradkar and Koche, K: Phytochemical analysis of *Leonotis nepetifolia* (L) R. BR. A wild medicinal plant of Lamiaceae. Bioscience Discovery 2012; 3(2): 196-197.
3. Gnaneswari, K and Venkatraju, R.R: Preliminary phytochemical screening and Antibacterial evaluation of *Leonotis nepetifolia* (L) R. Brazilian Journal of Natural Products, Plant Resources 2012; 62(6): 89-692.
4. Harborne, J.B: Methods of extraction and isolation. In: Phytochemical methods, (Chapman and Hall, London) 1998: 60.
5. Sinha sankar Narayan: Antibacterial potential of crude methanolic extract of *Leonotis nepetifolia* (L) R.BR. International research journal of Pharmacy 2012; 3(2): 277-278.
6. Nithya, V., Brinda, P and Anand, K.V: Wound healing activity of *Leonotis nepetifolia* (L) R.BR, in wistar albino rats. Asian Journal of Pharmaceutical and clinical research 2011; 4(2): 23-26.
7. Janat Akhanovna Mamyrbekova- Bekro., Kofi marcel Konan, Yves-Alain Bekro., Mare Gabin Djie bi. Tra Jerome Zomi bi., Veronique Mambo and Benson Boua, European Journal of Scientific research. 2008; 24(2): 219-228.
8. Trivedi Ashish, Sethiya Neeraj, K., and Misha, S.H: Preliminay pharmacognostic and phytochemcial analysis of "*Granthika*" (*Leonotis nepetifolia*): An Ayurvedic herb. Indian Journal of Traditional knowledge 2011; 10 (4): 682-688.
9. WHO: "*Quality control methods for medicinal plant materials*". WHO Geneva, Switzerland, 1998.

10. Unni, B.G., Archana Borah, Wann, S.B., Singh, H.R., Basabrani: Phytochemical and antimicrobial study of traditional medicinal plants of North east India on *E.coli*. Asian Journal of Exp.Sci. 2009; 23 (1): 103-108.
11. Koc, H., and Sari, H: Trace metal contents of some medicinal, aromatic plants and soil samples in the Mediterranean region Turkey. Journal of Applied Chemical research 2009; 8: 52-57.
12. Iqbal Hussain and Laber Khan: Comparitive study on heavy metal contents in *Taraxacum officinale*. International Journal of Pharmacognosy and Phytochemical research 2010; 2(1): 15-18.

How to cite this article:
Kamalam M, Saraswathi C and Umadevi U: Evaluation of *Leonotis nepetaefolia* for its phytochemical and heavy metal analysis. *Int J Pharm Sci Res* 2013; 4(12): 4591-96. doi: 10.13040/IJPSR. 0975-8232.4(12).4591-96