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Comparative Antibacterial potential of Methanolic extract of the leaves of wild and cultivated *Ficus carica* L.

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Abstract
The Flora of Pakistan having a huge diversity in medicinal plants due to climatic, topographic, physiographic, altitudinal and edaphic conditions. The present study focus upon the *Ficus carica* to analyze its quantitative, qualitative, biological, pharmacological, phytochemical analysis and antioxidant properties of methanolic extract from leaves. The present exhaustive work revealed that *Ficus carica* has potential source of therapeutic agents in domestic health care practices and a significant source for development. The leaves of both wild and cultivated showed antibacterial activities and clear zone of inhibition in the methanol extracts. The comparative analysis showed that leaves of wild plant exhibits greater antibacterial activity against *Bacillus sabtilis* followed by *Kleisbella pnumoniae*.

Keywords: *Ficus carica*, antibacterial, methanolic, medicinal, zone of inhibition

Introduction
The flora of Pakistan due to its diverse climatic and soil conditions and many ecological regions is very rich in medicinal plants. According to a general survey of Pakistan about 6000 species of flowering plants have been exist, out of 6000 about 400-600 are medicinally important species. The history of plants to be utilized as medicines is thousands of years old (Irfan *et al.*, 2017). These plant materials initially took the form of crude drugs such as poultices, teas, powders tinctures, and many other herbal formulations. From near past it has been discovered that properties of medicinal plants are due to its active chemical compounds and therefore the isolation of active compounds and in the early 19th century morphine has been isolated from opium (Irfan *et al.*, 2018). The presence of phenols is considered to be containing substances that can be used for the potentially toxic to the growth and development of therapeutic purpose, are called medicinal plants. knowledge of the chemical constituents of plants is desirable, not only for the discovery of therapeutic agents, but also because such information may be of value in disclosing new sources of such economic materials as tannins, oils, gums, precursors for the synthesis of complex chemical substances, etc. In addition, the knowledge of the chemical constituents of plants would further be valuable in discovering the actual value of folkloric remedies (Irfan *et al.*, 2018) [1]. The history of medicine and surgery dates back perhaps to the origin of the human race. Use of plants as a source of medicine has been inherited and is an important component of the health care system in different countries of the world (Khan *et al.*, 2012) [11]. Medicinal plants are of great importance to the health of individuals and communities in general. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. Many of the indigenous medicinal plants are used as spices and food plants. They also sometimes added to foods meant for pregnant women and nursing mothers for medicinal purposes (Amin *et al*., 2013). In order to safe from much harmful infection, medicinal plants consumption increases. It is evident from the view of large population of developing countries become diverted to words the use of medicinal plants and traditional practitioners as to compete their urgent demand for primary health care needs. Phytomedicines have becoming popular as these have strong historical and cultural uses. Alterative and complementary therapies get attention of many peoples in developing countries in recent years, it also includes medicinal herbs (Devi *et al*., 2009) [4]. During the past decade, traditional systems of medicine have become increasingly important in view of their safety. New plants are adding to the flora of Pakistan which having great medicinal importance (Ali *et al.*, 2017) [1]. Current estimates suggest that in many developing countries, a large proportion of the population relies heavily on traditional practitioners and medicinal plants to meet primary health care needs. Although modern medicine may be available in these countries, herbal medicines have often maintained popularity for historical and cultural reasons. Concurrently, many people in developed countries have begun to turn to alternative or complementary therapies, including medicinal herbs. About 85000 valuable medicinal plant species worldwide are reported (Liu *et al*., 2007) [12]. Historically the development of novel drugs was primarily through the extraction of biologically active compounds from plants which were screening programs. (Hunter, 2001) [7] have examined long-

established uses of medicinal plants, but only a few studies have leads to these ethnobotanical findings with laboratory job to confirm the real antimicrobial property of these plants (Bhattarai *et al*., 2008) [3]. For many years, plants have been used as therapeutic resources either as herbal teas or other homemade remedies, as crude extracts, or as standard enriched fractions in pharmaceutical preparations such as tinctures, fluid extracts, powders, pills, and capsules (Dzhambazov *et al.,* 2002) [5]. Moraceae is often called the mulberry family or fig family is a family of flowering plants comprising about 40 genera and over 1000 species. Most are extensive in tropical and subtropical regions, less so in moderate climates. The only synapomorphy within Moraceae is presence of latecomers and milky sap in all parenchymatous tissues, but generally useful field characters include two carpels sometimes with one reduced, compound inconspicuous flowers and compound fruits. Included are well-known plants such as the fig, banyan, breadfruit, mulberry (Luna, 1984) [13]. The flower of Moraceae is often pseudanthia. In English "giving a fig" means to care about something. The word ficolin which appears similar to *Ficus* and refers to a lectin like compound combining the first parts of the words for fibrinogen and collagen. *Ficus carica* latex has been used as a shamanic inebriant by Peruvian shamans, to serve as a powerful botanic teacher of medicine. The genus *Ficus carica* consists of over 800 species and is one of about 40 genera of the mulberry family, Moraceae. *Ficus carica* consists of numerous varieties, significant genetic diversity, outstanding pharmacological activities and these are of remarkable commercial importance. Fig is distributed in Southwest Asia and the Eastern Mediterranean region, from the Turkey in the East to Spain and Portugal in the West; it is also grown commercially in parts of U.S.A. and Chile and to small extent, in Arabia, Persia, India, China and Japan. It is cultivated in India commercially few centers near Pune and Bellary and Anantpur districts. In Punjab, Uttar Pradesh and Mysore it is mostly grown scattered in gardens or in home yards (Bakshi *et al*., 1999) [6]. *Ficus* carica is studied in modern pharmacognosy from many centuries, however, figs have been used in medicines and this use was recorded in classical middle eastern and European medical writings. The placement of poultices of figs on tumors as treatment for abnormal swellings. Such swellings, according to reports of experts could have been due to infection or cancer (Ben *et al.,* 2003) [2].

Materials and method

The present research work was carried out in the Department of Botany, Islamia College University Peshawar, Pakistan. Brief accounts of materials as well as procedures used in it are described below.

Samples collection

Ficus carica is found in many regions Pakistan, the wild and cultivated plants were collected from district Mardan, Khyber Pakhtunkhwa, Pakistan in May 2016. The plants were identified by Dr. Naveed Akhter taxonomist at Department of Botany, Islamia College University Peshawar and voucher specimens were deposited in Herbarium of Botany Department of Islamia College University Peshawar, Pakistan.

Extraction

The fresh parts of the wild and cultivated varieties of *Ficus carica* leaves, were taken, rinsed with distilled water and kept under shade till drying. Extraction from plants parts were carried out by simple maceration process. The plants parts were taken and grounded in kitchen blender and soak in methanol, chloroform, n-hexane and ethyl acetate. These mixtures were kept for two weeks at room temperature 25^0 C in extraction bottles. After two weeks' mixtures were filtered twice, using Whatman-41 filter paper. Solvents were completely evaporated by rotary evaporator to obtain the extract (Ullah *et al.,* 2018) [15].

Requirements

Test samples, Nutrient agar (20g/l), Mcfarl and Barium sulphate turbidity standard, Cultures of bacterial strains, sterile normal saline solution (0.9 % NaCl w/v), bacterial slants, sterile cork borer, micropipette, petri plates, organic solvent (DMSO), incubator, standard antibiotic DOX (Doxycycline), spirit lamp, DPPH and Spectrophotometer were used in the experiment.

Sample preparation

The extracts (15mg) were dissolved in 10ml of DMSO. This stock solution 15 mg/10ml was again diluted, thus 8 concentrations of the extract were prepared i.e. 15 mg/ml, 12.5mg/ml, 10mg/ml, 7.5 mg/ml, 5mg/ml, 3mg/ml, 2mg/ml, 1mg/ml. Along with these solutions of Standard antibiotic (2mg/ml of the DOX) was also prepared. The solutions of extracts were used for test. Standard antibiotics and pure DMSO were used for positive and negative control. Dilutions with DMSO are presented in the table.

Table 1: Dilutions with DMSO (Dimethyl sulphoxide)

No	Conc.(mg/ml)	Stock sol (ml)	DMSO (ml)	Final Vol. (ml)
1	15	1.00	0.00	1
2	12.5	0.833	0.167	1
3	10.00	0.666	0.334	1
4	7.50	0.500	0.500	1
5	5.00	0.334	0.666	1
6	3.00	0.200	0.800	1
7	2.00	0.133	0.867	1
8	1.00	0.100	0.900	1

Preparation of nutrient broth media

Nutrient broth medium was used to grow bacteria for inoculums preparation. Its composition was;

a. Peptone form meat = 5g/l
b. Meat extract = 3g/l

Nutrient agar medium was used to perform antibacterial assay. Its composition was

1. Peptone form meat = 5g/l
2. Meat extract = 3g/l
3. Agar-agar = 12g/l

Nutrient broth media was prepared by dissolving 0.4 g/50ml of distilled water for the growth of bacterial inoculums; PH was adjusted at 7.0 and was autoclaved. Nutrient agar medium was prepared by dissolving 2.3g/100ml of distilled water for the growth of bacterial inoculum; PH was adjusted at 7.0 and was autoclaved.

McFarland 0.5 Barium Sulphate Turbidity Standard

The standard was prepared by adding 0.5 ml 0.048 M

Barium Chloride to 99.5 ml 0.36 N sulphuric acid. Barium sulphate turbidity standard (4-6 ml) was taken in screw capped test tube and was used and poured to inoculum give the same color as that of turbidity standard (Konemen, 1988).

Bacterial strains used

Five strains of bacteria were used. Two were gram-positive i.e. *Bacillus subtilis* and *Staphylococcus aureas* while four were gram-negative; *Escherichia coli*, *Vibrio cholera* and *Enterobacter aerogenes* and Klebsiella pneumonia. The organisms were maintained on nutrient agar medium at 4^0 C.

Preparation of Inoculum

Centrifuged palates of bacteria from 24^0 hours old culture in nutrient both (SIGMA) of selected bacterial strains were mixed with physiological normal saline solution until a McFarland Turbidity Standard [10^{-6} colony forming unit (CFU) ml^{-1}] was obtained. Then this inoculum was used to seeding the nutrient agar.

Preparation of Seeded Agar Plates

Nutrient agar medium was prepared by suspending nutrient agar (MERCK) 2.8 g in 100 ml of distilled water; p H was adjusted at 7.0 and was autoclaved. It was allowed to cool up to 45^0 C. Then it was seeded with 10ml of prepared inoculum to have 10^{-6} CFU per ml. Petri plates were prepared by pouring 75ml of seeded nutrient agar and allowed to solidify. Ten wells per plate were made with sterile cork borer (5mm).

Pouring of test Solution; incubation and measurement of zone of inhibition

Using micropipette, 100 µl of test solutions was poured in respective wells. These plates were incubated at 37^0 C. After 24 hours of incubation; the diameter of clear zone of inhibitions were measured by a ruler. Antibacterial activity of 8 dilutions of each plant extract was determined against five bacterial strains.

Results and discussion

Methanolic leaves extract of cultivated *Ficus carica* L.

Methanolic leaves extract of cultivated variety of *Ficus carica* was tested against *Bacillus subtilis*, it exhibited the 16 mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 6mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 13mm, 12mm and 11mm inhibitory zone. At the concentration of 50mg/ml the zone of inhibition was 9mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 10mm and 7mm. At concentration of 1mg /ml showed 6mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of cultivated variety of *Ficus carica* was tested against *Staphylococcus aureas*, it exhibited the 25mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 12mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 25mm, 25mm and 20mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 15mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 14mm and 13mm. At concentration of 1mg /ml showed 12mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of cultivated variety of *Ficus carica* was tested against *Vibro cholera*, it exhibited the 15mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 4mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 15mm, 15mm and 12mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 10mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 9mm and 7mm. At concentration of 1mg /ml showed 4mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of cultivated variety of *Ficus carica* was tested against *Enterobacter aerogenes*, it exhibited the 16mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 10mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 14mm, 16mm and 15mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 16mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 12mm and 11mm. At concentration of 1mg /ml showed 6mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of cultivated variety of *Ficus carica* was tested against *Escherichia coli*, it exhibited the 16mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 6mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 15mm, 14mm and 13mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 10mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 10mm and 10mm. At concentration of 1mg /ml showed 6mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of cultivated variety of *Ficus carica* L. was tested against *Klebsiella pneumoniae*, it exhibited the 17mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 6mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 12mm inhibitory zone. At the concentration of 50mg/ml the zone of inhibition was 10mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 8mm. At concentration of 1mg /ml showed 6mm inhibitory zone. Standard DOX showed 20mm inhibitory zone (Table. 1: fig. 1).

Methanolic leaves extract of wild *Ficus carica* L.

When the methanolic leaves extract of wild variety of *Ficus carica* was tested against *Bacillus subtilis*, it exhibited the 26mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 10mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 26mm, 26mm and 18mm inhibitory zone. At the concentration of 50mg/ml the zone of inhibition was 15mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 13mm and 12mm. At concentration of 1mg /ml showed 10mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of wild variety of *Ficus carica* L. was tested against *Staphylococcus aureas*, it exhibited the 20mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 9mm. Further

it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 21mm, 20mm and 15mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 15mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 12mm and 11mm. At concentration of 1mg /ml showed 9mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of wild variety of *Ficus carica* L. was tested against *Vibrio cholera*, it exhibited the 13mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 4mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 13mm, 13mm and 11mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 11mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 11mm and 8mm. At concentration of 1mg /ml showed 8mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of wild variety of *Ficus carica* L. was tested against *Enterobacter aerogenes*, it exhibited the 20mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 10mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 18mm, 17mm and 15mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 13mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 12mm and 10mm. At concentration of 1mg /ml showed 5mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of wild variety of *Ficus carica* was tested against *Escherichia coli*, it exhibited the 13mm inhibition zone at concentration of 15mg/ml. While minimum inhibition concentration value was 6mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 12mm, 12mm and 1mm inhibitory zone. At the concentration of 5.0mg/ml the zone of inhibition was 10mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 8mm and 8mm. At concentration of 1mg /ml showed 6mm inhibitory zone. Standard DOX showed 20mm inhibitory zone. When the methanolic leaves extract of wild variety of *Ficus carica* was tested against *Klebsiella pneumoniae*, it exhibited the 14mm inhibition zone of inhibition at concentration of 15mg/ml. While minimum inhibition concentration value was 5mm. Further it was found that at 12.50mg/ml, 10mg/ml and 7.50mg/ml concentrations showed 13mm, 10mm and 10mm inhibitory zone. At the concentration of 50mg/ml the zone of inhibition was 10mm. While at the concentration of 3mg/ml and 2mg/ml inhibitory zone were 10mm and 7mm. At concentration of 1mg /ml showed 5mm inhibitory zone. Standard DOX showed 20mm inhibitory zone (Table 2: fig. 1)

Table 2: Zone of inhibition (mm) after 24 hours showing Antibacterial activity of cultivated Ficus carica methanolic leaves extract against Bacillus subtilis, Staphylococcus aureas, Vibro cholera, Enterobacter aerogenes, Escherichia coli, Klebsiella pneumoniae.

Concentration mg/ml	Methanolic extract of cultivated sp against B. subtilus (mm)	Methanolic extract of cultivated spp against S. aureas (mm)	Methanolic extract of cultivated sp against V. cholera (mm)	Methanolic extract of cultivated sp against E. aerogenes (mm)	Methanolic extract of cultivated sp against E. coli (mm)	Methanolic extract of cultivated sp Against K. pneumonia (mm)
15	16	25	15	16	16	17
12.5	13	25	15	14	15	12
10	12	25	15	16	14	12
7.5	11	20	12	15	13	12
5	10	15	10	16	10	10
3	9	14	9	12	10	8
2	7	13	7	11	10	8
1	6	12	4	10	10	8

Table 3: Zone of inhibition (mm) after 24 hours showing Antibacterial activity of wild *Ficus carica* L. methanolic leaves extract against *Bacillus subtilis*, *Staphylococcus aureas*, *Vibro cholera*, *Enterobacter aerogenes*, *Escherichia coli*, *Klebsiella pneumoniae*.

Concentration mg/ml	Methanolic extract of wild sp against B.subtilus (mm)	Methanolic extract of wild Spp against S.aureas (mm)	Methanolic extract of wild Spp against V.cholera (mm)	Methanolic extract of wild Spp against E.aerogenes (mm)	Methanolic extract wild spp against E.coli (mm)	Methanolic extract of wild spp against K.pneumoniae (mm)
15	26	20	13	20	13	14
12.5	26	21	13	18	12	13
10	26	20	13	17	12	10
7.5	18	15	11	15	12	10
5	15	15	11	13	10	10
3	13	12	11	12	8	10
2	12	11	8	10	8	7
1	10	9	8	5	8	5

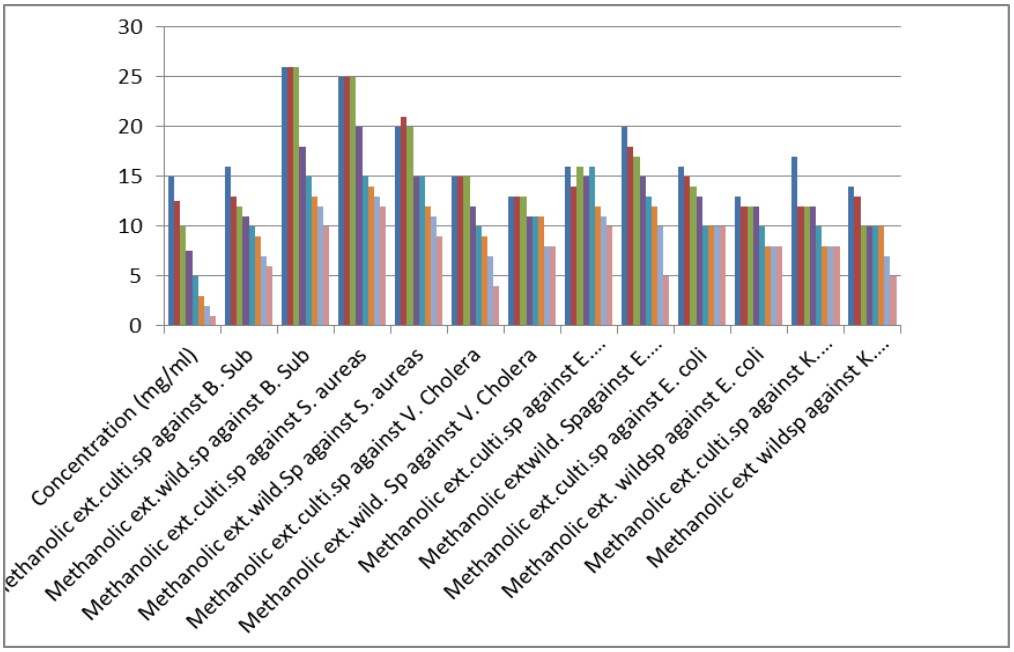

Fig 1: Comparison of Zone of inhibition (mm) of cultivated and wild species of *Ficus carica* L. methanolic leave extract against *Bacillus subtilis*, *Staphylococcus aureas*, *Vibrio cholera*, *Enterobacter aerogenes*, *Escherichia coli* and *Klebsiella pneumoniae*.

Conclusion

The study concluded that wild and cultivated species of *Ficus carica* L. leaves are considered as medicine in the health care system. The results concluded that leaves of *Ficus carica* L. are good antibacterial agents. Methanolic extracts of cultivated and wild species of *Ficus carica* can be used as strong antibacterial agents. Maximum antibacterial percentage inhibition was shown by wild *Ficus carica* L. leaves against *Bacillus subtilis* in methanolic extract followed by cultivated type in methanolic extract.

References

1. Ali A, Rashid M, Sultan A, Irfan M. Anisochilus carnosus (L. f.) Wall. ex Benth. (Lamiaceae) a new generic record for Pakistan. Plant Science Today. 2017; 4(3):102-105. http://dx.doi.org/10.14719/pst. 2017.4.3.316.
2. Ben-Noun LL. Figs—the earliest known ancient drug for cutaneous anthrax. Annals of Pharmacotherapy. 2003; 37(2):297-300.
3. Bhattarai S, Chaudhary RP, Taylor RSL. Screening of selected ethnomedicinal plants of Manang district, Central Nepal for antibacterial activity. Ethnobot. 2008; (20):9-15.
4. Devi K, Karthikai GD, Thirumaran G, Arumugam R, Anantharaman P. Antibacterial activity of selected medicinal plants from Parangipettai coastal regions; Southeast coast of India. World Appl Sci J. 2009; 7(9):1212-1215.
5. Dzhambazov B, Daskalova S, Monteva A, Popov N. In vitro screening for antitumour activity of Clinopodium vulgare L. (Lamiaceae) extracts. Biological and Pharmaceutical Bulletin. 2002; 25(4):499-504.
6. Guha Bakshi DN, Sensarma P, Pal DC. lexicon of medicinal plants in India. Naya Prokash, 1999.
7. Hunter D. Life in the fast lane: High- throughput chemistry for lead generation and optimisation. Journal of Cellular Biochemistry. 2001; 84(37):22-27.
8. Irfan M, Ahmad I, Saeed SH. Traditional medicinal plant knowledge of some spermatophytes of Samar Bagh Valley, Lower Dir district, Pakistan. Plant Science Today 2017; 4(4):151-153. doi: 10.14719/pst.2017.4.4.334
9. Irfan M, Ali I, Kashf RA. Ethnobotanical survey of the flora of Maidan Valley, Lower Dir District, Khyber Pakhtunkhwa Province, Pakistan. Plant Science Today. 2018; 5(2):68-71. https://dx.doi.org/10.14719/ pst.2018.5.2.379
10. Irfan M, Khan I, Ali A, Khan R, Ali A, Jan G. Ethnomedicinal Uses of the Plants of Tehsil Laalqilla, District Lower Dir, Khyber Pakhtunkhwa, Pakistan. Journal of Applied Environmental and Biological Sciences. 2018; 8(6):61-66.
11. Khan MA, Khan MA, Mujtaba G, Hussain M. Ethnobotanical study about medicinal plants of Poonch valley Azad Kashmir. J animal plant Sci. 2012; 22:493-500.
12. Liu Y, Wang MW. Botanical drugs: challenges and opportunities: contribution to Linnaeus Memorial Symposium 2007. Life Sciences. 2008; 82(9-10): 445-449.
13. Luna LE. The concept of plants as teachers among four mestizo shamans of Iquitos, northeastern Peru. Journal of Ethnopharmacology. 1984; 11(2):135-156.
14. Mir MA, Sawhney SS, Jassal MMS. Qualitative and quantitative analysis of phytochemicals of Taraxacum officinale. Wudpecker Journal of Pharmacy and Pharmocology. 2013; 2(1):1-5.
15. Ullah SW, Khan Ali W, Khan MS, Sajad MA, Nabeela Irfan M. International Journal of Biosciences IJB. 2018; 13(5):438-447.