In vitro antimicrobial assay of leaves, bark and fruits of Ficus auriculata collected from two different regions of Uttarakhand

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DOI: https://doi.org/10.22271/chemi.2021.v9.i1m.11347

Abstract

The present study is to evaluate the antimicrobial efficacy of Ficus auriculata fruits, leaves and bark collected from two different regions of Uttarakhand i.e, Almora and Haldwani. These parts of plants extracted in hexane, chloroform and methanol by successive soxhlet extraction technique. For antimicrobial assay, activity was done by agar well diffusion method against four human pathogenic bacteria (two gram positive and two gram negative bacteria). The results concluded that Almora leaves hexane extract possessed the higher antibacterial potential on increasing dose dependent manner at 800μg/ml (15.7±0.25 mm) Haldwani fruits hexane extract (15.5±0.25 mm) against E. coli. Almora bark chloroform extract inhibits the bacteria E. coli and B. subtilis with maximum zone (14±0.11 mm) and (12.4±0.15 mm). Haldwani leaves methanol extract showed the maximum inhibition zone (12±0.57 mm) against E. coli and Haldwani bark methanol extract showed the maximum inhibition zone (12.9±0.057 mm). These naturally occurring antimicrobials agents due to its safe and non toxic nature.

Keywords: Antimicrobial activity, Ficus auriculata, fruits, leaves and bark

1. Introduction

According to WHO 80% of world populations depends on medicinal plants and the rest of population health depends on commercial. About 21,000 species of plants used for their medicinal properties (Mahalakshmi M et al., 2014) [10]. India has the greatest resources of medicinal herbs endowed with a wide range of agroclimatic conditions and is known as the botanical backyard of the world. India is a biodiversity hotspot and a great variety of fruiting trees are indigenous to this region of the world as confirmed by various reports (Baliga MS et al., 2011) [2]. Over the centuries, Indian herbal drugs used as a major source of medicines for the treatment and prevention of many diseases. Ethnobotany embraces a complicated relationship between plants, people and way of life. This relationship between flora and human cultures is no longer confined to the use of vegetation for meals, clothing and shelters, but also includes their use for spiritual ceremonies, ornamentation and fitness care (Raghavendra MP et al., 2015) [19].

Ficus is a genus that consists of 750 species of medicinal plants primarily occurring in tropical and subtropical regions throughout the world. There is a large variation in the habitat of this species. Ficus genus belongs to the mulberry family (Moraceae) (Kunwar et al., 2006) [8]. Fig species are rich in nutrient, vitamins, mineral elements, water, and fats. Figs are rich source of calcium and fiber. According to USDA data for the Mission variety, dried figs are rich in fiber, vitamin K, copper, magnesium, manganese, calcium, potassium (Ahmad S et al., 2013) [11]. The literature survey reported that figs have been cultivated over 1100 years and these are one of the earliest cultivated plants for human use (Lansky EP et al., 2008) [9]. The genus can be generally reviewed by the very distinguishing syconium and factory latex and are collectively known as “figs”. Ficus plants are used by humans in different ways throughout the tropical and sub-tropical regions. Plants are origin of medication and nutrition and are used as decorative trees, devotional plants, lac hosts, fuel, fodder hedges or enclosures (Shi Y et al., 2018) [17].

Taxonomic classification: (Shilpakar A et al., 2009) [18]

Kingdom: Plantae (Plants)

Subkingdom: Tracheobionta (Vascular plants)
2. Materials and Methods

2.1. Collection of plant materials

We took three parts of plant fruits, leaves and bark of *Ficus auriculata* from two different altitudes of Uttarakhand. Places near to the Almora and Haldwani were selected for the collection of fruits, leaves and barks of *Ficus auriculata*. Washed off the sample to remove dust. After washed off cut the fruits, leaves and bark in to small pieces. They were kept in shade drying for the two weeks till then moisture content has been removed and then start the extraction process for the further progress.

2.2. Extraction

Extraction was done by successive soxhlet extraction technique.

2.2.1. Soxhlet extraction

This technique was carried out to obtain extracts for the phytochemical screening, biological and pharmacological activity. Fruit, leaves and Bark were meshed in grinder to get the powder of uniform size. Powder was packed in a thimble as filter paper. The thimble was then inserted into the Soxhlet apparatus and extraction was done by using hexane, chloroform and methanol as a solvent in a successive manner from non polar to polar solvent and extraction was continued for 9-10 hrs (Murugan. R et al., 2014) [[13]. Then hexane extract was collected and powder from the thimble was used for the next successive extraction of chloroform after drying it again yield was calculated. The same procedure has been followed by drying and powder in thimble was dried used for methanol extraction. Finally methanol, hexane and chloroform soluble fractions were obtained. All extracts obtained from two regions of uttarakhand were then evaporate in water bath and dried in a vacuum oven at 40-45 °C, and yield value for each extract was calculated. At last the extract were put on rotary evaporator or distillate to evaporate or collect solvent fo further use at 50° C to get the crude extract (Manandhar et al., 2019) [[11].

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**Diagram:**

![Flow chart of successive soxhlet extraction of fruits, leaves and bark of *Ficus auriculata*](image)

Labelled as:
- Plant parts (fruits, leaves and bark)
- Powder sample
- Successive soxhlet extraction
- Hexane
- Dried sample in rotary evaporator
- Soxhlet extraction
- Chloroform
- Dried sample in rotary evaporator
- Soxhlet extraction
- Methanol
- ME
- CE
- ME

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2.3. Determination of antibacterial activity FLB extracts of *F. auriculata*

2.3.1. Test organism collection: For antibacterial screening we collected human pathogenic bacterial strains i.e. *Escherichia coli*, *Salmonella typhi*, *Bacillus subtilis* and *Staphylococcus aureus* For the antibacterial study. We isolate these bacterial strains from Department of Public Health and Epidemiology, College of Veterinary and Animal Sciences, GBPUA&T pantnagar (INDIA). FLB extract of hexane, chloroform and from two different regions i.e., Almora and Haldwani. These sample gives the antibacterial screening against the bacterial strain and shows the effect against these bacteria. Two of them are gram positive strain (*Bacillus subtilis, Staphylococcus aureus*) and two of them are gram negative strain (*Escherichia coli, Salmonella typhi*). Antibacterial screening activity was done by disc-diffusion method (Singh R et al., 2005)\(^1\).

2.3.2. Preparation of bacterial inoculum: Bacterial inoculation prepared by the Luria bertani broth (HIMEDIA) for *Escherichia coli, Salmonella typhi, Bacillus subtilis* and *Staphylococcus aureus*. Nutrient agar was well dissolved in distilled water and then autoclaved for 30 minute about 120°C and 15 to 20 lbs pressure for bacterial colonies inoculation.

2.3.3. Preparation of agar plates: We prepared nutrient agar medium for bacterial culture and then poured in to sterile plates. Which should be done under laminar flow in undisturbed manner that kept the medium contamination free. Agar media uniformly spread over the sterile plates. Now agar nutrient plates was put in the incubator at 37°C for overnight until solidification of agar nutrient in sterile plates.

2.3.4. Placements of plates: After uniformly spread of agar in sterile plates, sterilized paper disc of 5 mm was dipped in FLB extract of hexane, chloroform and methanol of different concentration (200-1000 µL). Now these plates again put in to the incubator for overnight at 37°C for growing bacterial cultures. Next day measure the inhibition zone of different concentration and compared the zone of inhibition with standard Gentamicin (10µg/disc), amicacin (10µg/disc) and ofloxacin. Diameter of inhibition zone measured by scale (mm).

3. Statistical analysis

Each experiment was performed in triplicates and the mean values with standard deviation for the inhibition zones. The triplicate results were applied to represent the antibacterial activity of the leaves, bark and fruits in three different solvents.

4. Results and Discussion

Fruits, leaves and bark extract of *F. auriculata* which is collected from two different regions Almora and Haldwani (Nainital) were extracted in three different solvents i.e, hexane, chloroform and methanol. Different concentration of these extracts were used to antimicrobial assays. The *in vitro* antimicrobial activity were evaluated against two gram positive (*B. subtilis* and *S. aureus*) and two gram negative (*E. coli* and *S. typhi*) human pathogenic bacteria. Hexane extract of AL and HF shows the maximum zone of inhibition in increasing dose dependent manner from 200-800 µg/mL (9.2±0.15 mm to 15.7±0.25 mm) and (8±0.40 mm to 15.5±0.25 mm) against *E. coli*, AF and HL shows the maximum zone of inhibition (7±0 mm to 13.2±0.25 mm) and (7.4±0.42 mm to 11.1±0.28 mm) against *B. subtilis*, AB and HF shows the maximum zone of inhibition (7.1±0.1 mm to 9.5±0 mm) and (5.5±0.5 mm to 7.1±0.28 mm) against *S. typhi*. AF and HF shows the maximum zone of inhibition (8.1±0.15 mm to 11±0.05 mm) and (7±0 mm to 11.4±0.38 mm) against *S. aureus* in (Table 1 for Haldwani species, Table 2 for Almora species and Figure 2). Chloroform extract of AL and HL shows the maximum zone of inhibition from 200-800 µg/mL (7±0 mm to 11.9±0.17 mm) and (7.16±0.28 mm to 10.7±0.25 mm) against *S. typhi*, AB and HF shows the maximum zone of inhibition (10.2±0.20 mm to 14±0.11 mm) and (5±0 mm to 9.8±0.28 mm) against *E. coli*, AF and HL shows the maximum zone of inhibition (5±0 mm to 12.3±0.32 mm) and (7.10±0.28 mm to 10.8±0.104 mm) against *B. subtilis*, AB and HL shows the maximum inhibition zone (9±0 mm to 12.4±0.15 mm) and (7.36±0.57 mm to 12.1±0.23 mm) against *S. aureus* in (Table 3 for Haldwani species, Table 4 for Almora species and Figure 3). Methanol extract of AB and HB shows the maximum inhibition zone from 200-800 µg/mL (7.52±0.25 mm to 12.1±0.28 mm) and (8.20±0.34 mm to 12.9±0.057 mm) against *S. typhi*, AF and HL shows the maximum inhibition zone (6±0 mm to 8.2±0.2 mm) and (6±0.28 mm to 13±0.57 mm) against *E. coli*, AF and HL shows the maximum inhibition zone (7.13±0.15 mm to 11.16±0.15 mm) and 7.45±0.28 mm to 11.1±0.5 mm) against *B. subtilis*, AB and HF shows the maximum inhibition zone (7.1±0.1 mm to 12±0 mm) and (9.6±0.36 mm to 12.5±0.28 mm) against *S. aureus* in (Table 5 for Haldwani species, Table 6 for Almora species and Figure 4). Methanolic extract of stem bark of *F. auriculata* shown the maximum zone of inhibition against *E. coli* and hexane leaf extract shown the maximum inhibition zone against *S. aureus* (Gaire et al., 2011)\(^4\). Leaf extract of methanol and chloroform showed the maximum inhibition zone against *E. coli* and *S. typhi* (Kumari A et al., 2018).\(^7\) Ethanolic fruit extract showed the higher antibacterial potential against food poisoning bacteria (*Escherichia coli, Shigella flexneri* and *Staphylococcus epidermis*) and fruits showed the maximum potential against *Shigella flexneri* (Saklani et al., 2012).\(^10\) Alcoholic extract of leaves and fruits of *F. auriculata* showed the effective antibacterial potential against *S. aureus, B. subtilis* and *S. typhi*. Leaves shown the effective antibacterial potential than fruits, while fruits shown the effective antibacterial potential against *B. subtilis* (Fishawy et al., 2011).\(^3\)

| Haldwani | Concen.(µg/ml) | S. typhi | E. coli | B. subtilis | S. aureus |
|----------|---------------|----------|--------|-------------|----------|
| HFHE     |               |          |        |             |          |
| 200      | 5.4±0.5       | 8±0.40   | 7±0    | 9.16±0.28   |
| 400      | 5.85±0.57     | 10±0.28  | 8.25±0.43 |
| 600      | 6.16±0.28     | 15±0.57  | 9.16±0.28 |
| 800      | 7.1±0.28      | 15.5±0.25| 11.4±0.38|
| HLHE     |               |          |        |             |          |
| 200      | 6±0.11        | 6.8±0.0  | 7.45±0.42 |
| 400      | 7.2±0.25      | 7±0.11   | 8.23±0.40 |
| 600      | 7.8±0.28      | 10.9±0.11| 9.9±0.14  | 7.4±0.40  |
Table 2: Zone of inhibition (mm) of Almora leaves, bark and fruits hexane extract

| Almora | Concen.(µg/ml) | S. typhi | E. coli | B. subtilis | S. aureus |
|--------|----------------|----------|---------|-------------|-----------|
|        | 200            |          |         |             |           |
|        | 400            |          |         |             |           |
|        | 600            |          |         |             |           |
|        | 800            |          |         |             |           |
| AFHE   | 200            | 5.2±0.25 | 0±0     | 8.2±0.25   | 8.1±0.15  |
|        | 400            | 7.3±0.30 | 6.3±0.15| 9.3±0.35   | 9.1±0.28  |
|        | 600            | 8±0.05  | 7±0     | 10±0       | 10±0      |
|        | 800            | 9.16±0.2 | 8±0.05  | 13.2±0.25  | 11±0.05   |
| ABHE   | 200            | 7.1±0.10 | 5±0     | 7±0.05     | 7.2±0.3   |
|        | 400            | 8±0.05  | 6.3±0.15| 8.18±0.16  | 8±0.0     |
|        | 600            | 9.1±0.23| 7.14±0.2| 9.3±0.20   | 9.1±0.10  |
|        | 800            | 9.5±0.0 | 7.4±0.1 | 10±0       | 10.8±0.28 |
| ALHE   | 200            | 11.1±0.0| 9.2±0   | 8.2±0.34   | 6.16±0.15 |
|        | 400            | 6±0.11  | 10±0.2  | 9.2±0.20   | 8.18±0.31 |
|        | 600            | 6.2±0.25| 12±0.0  | 11.1±0.23  | 8.9±0.11  |
|        | 800            | 7±0.0   | 15.7±0.25| 12.2±0.25  | 10±0.05   |
| Amikacin | (10 µg/ml) | 23       | 20      | 20          | 22        |
| Control |                | 0        | 0       | 0           | 0         |

*Mean of three replications with ± standard deviation

Fig 2: Zone of inhibition of Haldwani and Almora leaves, bark and fruits in hexane extract

Table 3: Zone of inhibition (mm) of Haldwani leaves, bark and fruits chloroform extract

| Haldwani | Concen. (µg/ml) | S. typhi | E. coli | B. subtilis | S. aureus |
|----------|-----------------|----------|---------|-------------|-----------|
|          | 200             |          |         |             |           |
|          | 400             |          |         |             |           |
|          | 600             |          |         |             |           |
|          | 800             |          |         |             |           |
| HFCE     | 200             | 5.1±0.28 | 5±0     | 7±0.0      | 9.8±0.28  |
|          | 400             | 5.8±0.28 | 6.16±0.28| 7.6±0.57  | 9.8±0.28  |
|          | 600             | 7.5±0.5 | 7.16±0.28| 8.6±0.0   | 10.6±0.57 |
|          | 800             | 9.5±0.5 | 9.8±0.28 | 10.16±0.28| 11.58±0.14|
| HLCE     | 200             | 7.16±0.28| 5.25±0.43| 7.1±0.28  | 7.36±0.32 |
|          | 400             | 8.25±0.43| 6.25±0.25| 8.25±0.25 | 10.26±0.25|
|          | 600             | 8.75±0.43| 7.08±0.14| 9.1±0.17  | 11.4±0.41 |
|          | 800             | 10.78±0.2| 8.25±0.43| 10.8±0.104| 12.1±0.23 |
| HBCE     | 200             | 0±0     | 5±0     | 7±0.0      | 6.9±0.05  |
|          | 400             | 0±0     | 5.3±0.28| 8±0.15    | 7.5±0.05  |
|          | 600             | 8±0.17  | 6.1±0.17| 8.5±0.05  | 8.36±0.32 |
|          | 800             | 9.2±0.2| 7.16±0.15| 9.1±0.17  | 9.2±0.20  |
| Amikacin | (10 µg/ml)     | 23       | 20      | 20          | 22        |
| Control  |                | 0        | 0       | 0           | 0         |

*Mean of three replications with ± standard deviation

Table 4: Zone of inhibition (mm) of Almora leaves, bark and fruits chloroform extract

| Almora | Concen.(µg/ml) | S. typhi | E. coli | B. subtilis | S. aureus |
|--------|----------------|----------|---------|-------------|-----------|
|        | 200            |          |         |             |           |
|        | 400            |          |         |             |           |
|        | 600            |          |         |             |           |
|        | 800            |          |         |             |           |
| AFCE   | 200            | 7.1±0.15 | 6±0     | 5±0.0      | 5±0.0     |
|        | 400            | 8±0.11  | 7±0.11  | 19±0.15   | 8±0.20    |
|        | 600            | 9±0.0   | 7.6±0.1 | 11±0.0    | 8.4±0.11  |
|        | 800            | 10.2±0.2| 8.16±0.15| 12.3±0.32 | 9.1±0.17  |

*Mean of three replications with ± standard deviation
### Table 5: Zone of inhibition (mm) of Haldwani leaves, bark and fruits methanol extract

| Haldwani | Concen.(µg/ml) | S.typhi   | E.coli    | B.subtilis | S.aureus |
|----------|----------------|-----------|-----------|------------|----------|
| HFME     | 200            | ±0±0.57   | 7.1±0.28  | 6.1±0.28   | 9.3±0.76 |
|          | 400            | 7.1±0.28  | 7.5±0.5  | 7.3±0.57   | 10.8±0.28|
|          | 600            | 7.5±0.5   | 9.5±0.5  | 8.3±0.28   | 11.5±0.5 |
|          | 800            | 8.5±0.5   | 10.3±0.57| 9.3±0.57   | 12.5±0.28|
| HLME     | 200            | 6.0±0.57  | 6±0.28   | 7.45±0.28  | 6±0.76   |
|          | 400            | 7.2±0.28  | 7.06±0.5 | 8.23±0.57  | 7.16±0.28|
|          | 600            | 7.8±0.5   | 10.9±0.5 | 9.9±0.28   | 7.4±0.5 |
|          | 800            | 8.6±0.5   | 13±0.57  | 11.1±0.57  | 10.2±0.86|
| HBME     | 200            | 8.2±0.34  | 5.1±0.28 | 6.8±0.15   | 9.6±0.36 |
|          | 400            | 9.06±0.11 | 6.1±0.10 | 7.3±0.28   | 10.1±0.36|
|          | 600            | 10±0.0    | 7±0.11   | 8±0.50     | 11±0     |
|          | 800            | 12.9±0.05 | 8±0.00   | 9.1±0.28   | 12.1±0.23|
| Amikacin | (10 µg/ml)     | 23        | 20        | 20         | 22       |
| Control  |                | 0         | 0         | 0          | 0        |

*Mean of three replications with ± standard deviation

### Table 6: Zone of inhibition (mm) of Almora leaves, bark and fruits methanol extract

| Almora | Concen.(µg/ml) | S.typhi   | E.coli    | B.subtilis | S.aureus |
|--------|----------------|-----------|-----------|------------|----------|
| AFME   | 200            | 5.26±0.25 | 6±0       | 7.13±0.15  | 9±0.10   |
|        | 400            | 6±0.0     | 6.3±0.2   | 8.1±0.17   | 9.8±0.28 |
|        | 600            | 8±0.11    | 7.2±0.05  | 9±0.0      | 11±0     |
|        | 800            | 10.1±0.17 | 8.2±0.2   | 11.2±0.15  | 11.4±0.11|
| ABME   | 200            | 7.2±0.25  | 6±0.0     | 8±0.0      | 7.1±0.1 |
|        | 400            | 8±0.0     | 7.2±0.05  | 9.1±0.15   | 8±0.0   |
|        | 600            | 9.26±0.25 | 7.3±0.15  | 9.2±0.30   | 9±0.10  |
|        | 800            | 12.1±0.28 | 8.1±0.15  | 9.8±0.62   | 12±0.0  |
| ALME   | 200            | 0±0.0     | 0±0.0     | 7±0.11     | 0±0.0   |
|        | 400            | 5.1±0.28  | 0±0.0     | 8.1±0.23   | 6.1±0.15|
|        | 600            | 6.2±0.25  | 6.3±0.26  | 9.1±0.1    | 7.2±0.2 |
|        | 800            | 7.3±0.26  | 7.2±0.20  | 10±0.11    | 8.1±0.17|
| Amikacin | (10 µg/ml)     | 23        | 20        | 20         | 22       |
| Control |                 | 0         | 0         | 0          | 0        |

*Mean of three replications with ± standard deviation
5. Conclusion
In present study hexane extract and chloroform extract of Almora species shows the maximum zone of inhibition against human pathogenic bacteria while methanol extract of Haldwani (Nainital) species shows the maximum zone of inhibition against human pathogenic bacteria. A number of antibiotics are becoming less effective due to development of resistance and this has caused serious clinical problems in the treatment of infectious diseases. So, it also confirming the more possibility of bioactive compounds that we will isolate for future aspects and are useful for rationalizing the use of this plant in primary health care.

6. Acknowledgement
I would like to express my sincere thanks and gratitude to my advisor Dr. Shirish Tandon, advisory committee and G.B.P.U.A&T advisor Dr. Shishir Tandon, advisory committee and I would like to express my sincere thanks and gratitude to my

7. References
1. Ahmad S, Bhatti FR, Khaliq FH, Irshad S, Madni A. A review on the prosperous phytochemical and pharmacological effects of Ficus carica. International Journal of Bioassays 2013;2:843-849.
2. Baliga MS, Pai RJ, Bhat HP, Palatty PL, Boloor R. Chemistry and medicinal properties of the Bakul (Mimusops elengi Linn): A review. Food Research International 2011;44(7):1823-9.
3. El-Fishawy A, Zayed AF, Afifi S. Phytochemical and pharmacological studies of Ficus auriculata Lour. Journal of Natural Products 2011;4:184-195.
4. Gaire BP, Lamichhane R, Sunar CB, Shilpakar A, Neupane S, Panta S. Phytochemical screening and analysis of antibacterial and antioxidant activity of Ficus auriculata (Lour.) stem bark. Pharmacognosy journal 2011;3(21):49-55.
5. George M, Joseph L, Paul MN. Ficus auriculata: a pharmacological update. International Journal of Current Research and Academy Review 2016;4:26-31.
6. Khatun MJM, Rahman MM, Rahim MA, Jakariya M, Mirdah MH. Study on the ethnomedicine and nutritional status of three edible Ficus species in hill district of Bangladesh. International Journal of Minor Fruits, Medicinal and Aromatic Plants 2016;2(1):35-40.
7. Kumari A, Verma R, Sharma M, Chauhan P, Kumar A. Evaluation of Phytochemical, antioxidant, antibacterial and anti-cancerous activity of Ficus auriculata Lour. And Osyris wightiana Wall. Ex Wight. Bulletin of Environment, Pharmacology and Life Sciences 2018;7(8);64-70.
8. Kunwar RM, Bussmann RW. Ficus (Fig) species in Nepal: A review of diversity and indigenous uses. Lyonia 2006;11(1):85-97.
9. Lansky EP, Paavilainen HM, Pawlus AD, Newman RA. Ficus spp. (fig): Ethnomedicine and potential as anticancer and anti-inflammatory agents. Journal of Ethnopharmacology 2008;119(2):195-213.
10. Mahalakshmi M, Parimala M, Shoba FG. Evaluation of anti-diarrhoeal potential pf methanol extract of Ficus bengalensis Linn. Stem bark and root bark. International Journal of Pharmacognosy and Phytochemical Research 2014;6(3):454-458.
11. Manandhar S, Luitel S, Dalal RK. In vitro antimicrobial activity of some medicinal plants against human pathogenic bacteria. Journal of tropical medicine 2019.
12. Murti K, Kumar U. Anti-bacterial activity of Ficus bengalensis and Ficus racemosa roots L. American Journal of Microbiology 2011;2(1):21-4.
13. Murugan R, Parimalazhagan T. Comparative evaluation of different extraction methods for antioxidant and anti-inflammatory properties from Osbeckia parovifolia Am.– An in vitro approach. Journal of King Saud University -Science 2014;26(4):267-275.
14. Parekh J, Chanda S. In vitro antimicrobial activity and phytochemical analysis of some Indian medicinal plants. Turkish Journal of Biology 2007;31(1):53-8.
15. Raghavendra MP, Prasad AD, Shyma TB. Investigations on anti-diabetic medicinal plants used by tribes of Wayanad district, Kerala. International Journal of Pharmaceutical Sciences and Research 2015;6(8):3617.
16. Saklani S, Chandra S. In vitro antimicrobial activity, nutritional profile and phytochemical screening of wild edible fruit of Garhwal Himalaya (Ficus auriculata). International Journal of Pharm Sci Rev Res 2012;12(2):61-64.
17. Shiyi M, Mon AM, Fu Y, Zhang Y, Wang C, Yang X et al. The genus Ficus (Moraceae) used in diet: Its plant diversity, distribution, traditional uses and ethnopharmacological importance. Journal of Ethnopharmacology 2018;226:185-196.
18. Shilpakar A, Gaire BP, Bahadur SC, Lamichhane R, Neupane S. Phytochemical Screening and analysis of antibacterial and antioxidant activity of Ficus auriculata, Lour. Stem bark. Ph.D. Thesis, Pokhara University Nepal 2009.
19. Singh R, Jain A, Panwar S, Gupta D, Khare SK. Antimicrobial activity of some natural dyes. Dyes and pigments 2005;66(2):99-102.

Fig 4: Zone of inhibition of Haldwani and Almora leaves, bark and fruits in methanol extract