

Antibacterial Effects of Extracts of Selected Medicinal Plants in Combination with Honey

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Abstract: In recent years drug resistance in human pathogenic bacterial strains has been commonly reported from all over the world. Sometimes use of antibiotics is associated with adverse effects on host like hypersensitivity and immune suppression. This has created various clinical problems in the treatments of infectious diseases. A large number of antimicrobial agents derived from traditional medicinal plants can be used as alternatives of antibiotics. In this study bacterial cultures were isolated from samples collected from different sites. The cultures were identified based on their colony morphology, gram stain reaction and different biochemical tests. Four bacterial cultures namely *E. coli*, *Klebsiella sp.*, *Staphylococcus sp.* and *Pseudomonas sp.* were identified and selected for the study. The effects of different antibiotics, honey and aqueous extracts of Ginger, Garlic, Turmeric, Basil and Clove on bacterial cultures were determined. The antibacterial effects of extracts of medicinal plants in combination with honey on different bacterial cultures were also studied. The results show that all the bacterial cultures were resistant to Penicillin G indicating that bacteria have developed antibiotic resistance. It was seen that the extracts of Ginger and Garlic have highest antimicrobial effect on these bacterial cultures followed by Turmeric and Basil. The aqueous extract of Clove did not show any antibacterial effects. Honey also showed antibacterial activity and it was interesting to see that when the extracts of medicinal plants were used in combination with honey then their antimicrobial effects significantly increased. In Ayurveda honey has been described as potent medicine for several uses and results of this study demonstrate that if extracts of these medicinal plants which are also used as spices in Indian food are used in combination with honey then their medicinal qualities may be enhanced.

Keywords: Drug resistance, Antibiotics, Medicinal plants, Honey, Antibacterial effects

I. Introduction

Infectious diseases are world's leading cause of death killing thousands of people every day. Infections due to different bacterial agents like pathogenic *E. coli*, *Salmonella sp.* and *Staphylococcus sp.* are most common (Alavijeh et al., 2012; Sarfaraz et al., 2018). In recent years drug resistance to human pathogenic bacterial strains has been commonly reported from all over the world which has resulted due to excessive use or misuse of antibiotics. Emerging multidrug resistant bacteria cause increase in mortality and increase the cost of treatment and care. The most problematic bacteria include *E. coli*, *Pseudomonas sp.*, *Staphylococcus sp.*, *Klebsiella sp.* etc. In addition to this problem sometimes antibiotics cause adverse effects on host like hypersensitivity and immune suppression. Hence, there is need to look for alternative treatments for infectious diseases (Alavijeh et al., 2012; Farzana, 2014). Scientists all over the world are focusing their attention toward natural and herbal products which can be used as antibacterial or antimicrobial agents. Many medicinal plants are rich in wide variety of secondary metabolites like tannins, terpenoids, alkaloids, flavonoids, phenol and quinines which can be used in herbal medicine to treat

infectious disease. Several studies have shown that medicinal plant extracts have antimicrobial activities (Rahman et al., 2015; Zeedan et al., 2016). Many medicinal plants are used as spices and are added during food preparation into almost every Indian food/ curry prepared daily like Turmeric (dried rhizome powder), Garlic (bulb), Ginger (fresh and dried rhizome) and Clove (bud). Basil leaves are routinely added for brewing tea in every Indian household. Studies have shown that Ginger, Garlic, Clove, Basil and Turmeric have antibacterial and antiviral activities against various pathogens (Abdulzahra and Mohammed, 2014; Marasini et al., 2015; Nabrdalik and Grata, 2016; Zeedan et al., 2016; Tambekar and Dahikar, 2016; Ginovyam et al., 2017; Feghali et al., 2018; Shah et al., 2019). Several studies have demonstrated the potent antifungal, antiviral and antibacterial effects of clove (Ibrahim and Salem, 2014; Marasini et al., 2015; Sakha et al., 2018). In addition, the antimicrobial activity of clove essential oil has been studied against a large number of multidrug resistant *Staphylococcus epidermidis*, *Staphylococcus aureus* and *E. coli* and some other bacterial pathogens (Mishra and Sharma, 2014; Packyanathan et al., 2017; Gonelimali et al., 2018). Gonelimali et al., (2018) experimentally proved that ethanolic extracts of Clove decrease the pH and hyperpolarization of plasma membrane of Gram positive and Gram negative bacteria, so they concluded that plant extracts may be used as natural antimicrobials and food preservatives (Saikumari et al., 2016; Gonelimali et al., 2018; Wongsawan et al., 2019).

Gul and Bakhat, (2015) examined the antimicrobial activity and preservation potential of turmeric extract prepared in water, chloroform, ethanol and n-hexane against *E. coli*, *Staphylococcus aureus*, *Salmonella typhi* and *Candida albicans* by using disc diffusion method. Their results showed that aqueous extract of turmeric reduces the growth of *E. coli* and *Staphylococcus aureus*. Methanol extract of turmeric was effective for all the microbial cultures tested. Chloroform and n-hexane extracts of turmeric show weak activity against all the entire tested microbial cultures. Feghali et al., (2018) reported that curcumin extracted from turmeric show inhibitory effect on methicillin resistant *Staphylococcus aureus* (MRSA) (Feghali et al., 2018; Sakha et al., 2018).

Garlic exhibits powerful antimicrobial activities so Garlic can provide proper management for bacterial growth ranging from disinfectant, antiseptic, bacteriostatic and even bactericidal characteristics. Besides that, Garlic may have the ability to prevent and manage viral, fungal and even helminthes infections. Fresh Garlic has been found to have a significant role in managing food poisoning through killing the causative agents such as *Escherichia coli* (Abdulzahra and Mohammed, 2014; Wolde et al., 2018). Kshirsagar et al., (2020) reported that aqueous extracts of Garlic inhibit the activity of *Lactobacillus acidophilus* and *Streptococcus mutant* bacteria in dental caries and other oral infections.

Ginger has been valued for its antibacterial properties for thousands of years in Asian cultures (Ginovyam et al., 2017). Ginger (*Zingiber officinale*) is one of those plants which were seriously investigated over the years (Sebiomo et al., 2011; Patil and Pwar, 2016; Rahman et al., 2020). Rahman et al., (2020) determined the antibacterial effect of crude and ethanolic Ginger extracts against *Staphylococcus aureus*. The results indicate that there is strong antibacterial effect of crude and ethanolic ginger extracts against *Staphylococcus aureus*. It was also seen that the active ingredients of Ginger inhibits multiplication of bacteria by membrane disruption (Rahman et al., 2020). Ginger was found to have free radical removing activity (prevent oxidant formation), so prevents lipid oxidation. This finding can explain the well documented gastro-protective effects (Abdulzahra and Mohammed, 2014).

Nas and Ahmad, (2018) studied the phytochemical constituents and antibacterial activity of Ginger extracts against six pathogenic bacteria namely *Klebsiella pneumoniae*, *Salmonella typhi*, *Shigella sp.*, *Pseudomonas aeruginosa*, *E. coli* and *Staphylococcus aureus*. The results of phytochemical screening of the extracts showed that the Ginger contain alkaloid, anthraquinone, saponin, phenol, flavonoid, terpenoid, terpenoid and glycoside, steroid and reducing sugar while resin is absent. Their antibacterial assay showed that ethanolic extract has highest antibacterial activity followed by aqueous extract. The results of various studies justify that Ginger has therapeutic potential and can also be used as dietary supplement for food preservation (Dostalova et al., 2014; Rai et al., 2017; Nas and Ahmad, 2018).

The medicinal properties of basil have been studied in hundreds of scientific studies including *in vitro*, animal and human experiments. These studies reveal that basil has a unique combination of actions that include: antimicrobial, anti-diarrheal, anti-oxidant, anti-inflammatory, radio protective, anti-allergic, immunomodulatory and anti-coagulant activities. Basil's activity against water-borne and food-borne pathogens further suggests that it can be used in the preservation of food stuffs and herbal raw materials as well as for water purification and as a hand sanitizer. Antibacterial activity of aqueous, alcoholic and chloroform extracts of *Ocimum sanctum* were studied against different bacterial strains namely *Klebsiella sp.*, *Pseudomonas sp.*, *Staphylococcus aureus* and *Staphylococcus typhimurium* which shows broad spectrum activity of *Ocimum sactum* (Cohen, 2014; Nabradalik et al., 2016; Kulkarni and Adavirao, 2018; Shah et al., 2019).

Honey is a natural product which is used since ancient time in India as therapeutic agent. Honey has been shown to have antimicrobial, antibacterial (bactericidal and bacteriostatic), antioxidant and anti-inflammatory activities (Ibrahim and Salem, 2014; Wasihun and Kasa, 2016; Sarfaraz et al., 2018; Piotr et al., 2018; Mama et al., 2019). Mahandran and Kumarasamy, (2015) investigated the antibacterial activity of honey against *Staphylococcus aureus*, *Streptococcus pyogenes*, *E. coli* and *Proteus mirabilis* which showed that among the all bacterial cultures *Staphylococcus aureus* was most sensitive microbe and show maximum inhibitory zone. It was seen that when natural unheated honey was tested against pathogenic bacteria, oral bacteria, gastrointestinal tract pathogenic bacteria, as well as food spoilage bacteria then it shows some broad spectrum antibacterial activity (Mahandran and Kumarasamy, 2015). There are very few researchers who have studied the combined effects of honey with aqueous extracts of medicinal plants. Zeedan et al., (2016) studied the antibacterial effect of honey and herbal extracts of Ginger, Clove and Black cumin on different bacterial cultures. Their results showed that combination of honey with herbal extracts may have synergistic effects (Zeedan et al., 2016). Although a large number of studies have been done to demonstrate antibacterial effect of herbal extracts against different bacteria. However, very few studies support the effects of these extracts on drug resistance bacteria. In this study we have tested the effect of antibiotics, honey and different aqueous herbal plant extracts which are routinely used in Indian food as spices (Basil, Clove, Garlic, Ginger and Turmeric) against different food borne pathogens. We have also tested the effect of aqueous herbal extracts in combination with honey against the bacterial cultures. The objective of the study was to determine if combination of honey with aqueous herbal extracts increases the potency of these aqueous extracts in eliminating food borne pathogens.

II. Material And Methods

Isolation and identification of bacteria: Bacterial samples were collected from different sites including soil, sewage, kitchen sink, mouth, computer surface and skin of humans. Nutrient agar medium was used for the isolation of bacterial colonies from different samples. The cultures were isolated by dilution plating and were purified by streak plating method. Identification of the bacterial cultures were based on the results obtained during the study in the colony morphology, reaction with gram stain, growth on selective media (MacConkey agar media and EMB agar media) and the biochemical tests (motility test, indole production test, methyl red test, voges proskauer test, citrate utilization test, triple sugar iron agar test, urease test, oxidase test). Pure cultures of the bacteria were then maintained in on nutrient agar slants and stored at -20°C for further studies (Kowsalya, 2012).

Study of antibacterial susceptibility: Four bacterial cultures were used in the study which included three Gram negative bacteria namely *Klebsiella sp.*, *E. coli*, *Pseudomonas sp.* and one of Gram positive bacteria *Staphylococcus sp.* The study was done on Muller Hinton Agar using disc diffusion method. All the selected four bacterial cultures were swabbed on Petri plates. Antibiotics used in experiments were chloramphenicol (30 mcg per disc), tetracycline (10 mcg per disc), bacitracin (200 mcg per disc), streptomycin (30 mcg per disc), penicillin G (6000 mcg per disc) and nystatin (50 mcg per disc). In this experiment nystatin was used as a control because this is antifungal antibiotic. The inhibition zones formed around different antibiotics were measured in centimetre (cm) and recorded as results (Kowsalya, 2012).

Study of antibacterial effects of different medicinal plants: The medicinal plants used for studying the antimicrobial effects included Basil (*Ocimum sanctum*), Clove (*Syzygium aromaticum*), Garlic (*Allium sativum*), Ginger (*Zingiber officinale*) and Turmeric (*Curcuma longa*). The parts of these plants which were used in the study are the leaves of Basil, flower buds of Clove, rhizome of Garlic and Ginger and dried rhizome powder of Turmeric. Rhizome powder of Turmeric is called haldi and is used for curry making in India. The aqueous extracts of Clove, Basil, Garlic, Ginger and Turmeric were prepared by dissolving 0.5 gram (5%) and 1 gram (10%) of crushed plant part in 10 ml of sterilized distilled water. The plant extracts were vortexed vigorously and left for thirty minutes after which they were filtered and the filtrate was used for studying the antibacterial effects. All the selected four bacterial cultures were swabbed on Petri plates containing Muller Hinton Agar medium. The antibacterial effects of different plant parts were studied by disc diffusion method as well as agar well diffusion method. The plates were incubated at 37°C for 24 hours in upright position and after 24 hours the antibacterial activity was measured in terms of the diameter of the zone of inhibition (cm) (Mishra and Sharma, 2014; Gonelimaliet al., 2018; Suzan et al., 2018).

Study of antibacterial effects of honey and different medicinal plants in combination with honey: The pure honey purchased from market was used for studying the antibacterial effects on different isolated bacterial cultures by disc diffusion method as well as agar well diffusion method. In the disc diffusion method a sterilized filter paper disc of 6 mm was dipped in pure honey and used for study the antibacterial effect and in agar well diffusion method a hole was prepared in Petri plate containing Muller Hinton Agar medium and 1 ml pure honey was poured in the

well to study its antibacterial effects. For the study of effects of medicinal plants (Basil, Clove, Garlic, Ginger and Turmeric) in combination with honey 1 gm (10%) of plant part was crushed with 10 ml of pure honey and mixed well. The mixture was kept at room temperature and then used for studying the antibacterial effects by disc diffusion as well as agar well diffusion method (Mishra and Sharma, 2014; Gonelimali et al., 2018; Suzan et al., 2018).

III. Result and Discussion

Identification and selection of bacterial cultures: Twenty four bacterial cultures were isolated from samples collected from different places and were purified by streak plating are shown in Table 1. Gram staining of cultures was done and it was seen that sixteen cultures were Gram positive and eight were Gram negative. Three different looking Gram negative cultures and one Gram positive culture was selected and different biochemical tests were performed and the cultures were identified. The results are shown in Table 2 and Figure 1. The results show that culture number N5 was *E. coli* as it showed metallic green color colonies in the confirmatory test on EMB plate (Figure 1A). This culture was Gram negative, motile, lactose fermenting, MR test positive, VP test negative and Indole positive. Culture number N7 was identified as *Klebsiella sp.* as it showed mucoid pink colonies on MacConkey agar plate (Figure 1B). This culture was Gram negative, lactose fermenting, catalase positive, MR test negative, VP test positive and Indole negative. Culture number N14 was identified as *Staphylococcus sp.* and it showed spherical (cocci) shape in characteristic grape like cluster under microscope (Figure 1C). This culture was Gram positive, lactose fermenting, coagulase positive, MR positive, VP positive and Indole negative. Culture number N15 was identified as *Pseudomonas sp.* and it showed greenish blue pigment in the medium (Figure 1D). This culture was Gram negative, lactose non fermenting, MR negative, VP negative and Indole negative. These cultures were isolated from kitchen and other household places which frequently come in contact with human beings. These bacterial cultures are common food borne pathogens which may cause health hazards for persons coming in contact with these microbes due to contamination of food and water (Mostafa et al., 2018; Gabriel et al., 2018). Our study was focused to determine if spices (Clove, Garlic, Ginger, Basil and Turmeric) added during food preparation in Indian food may help in eliminating these pathogens and thus decreasing negative effects on health.

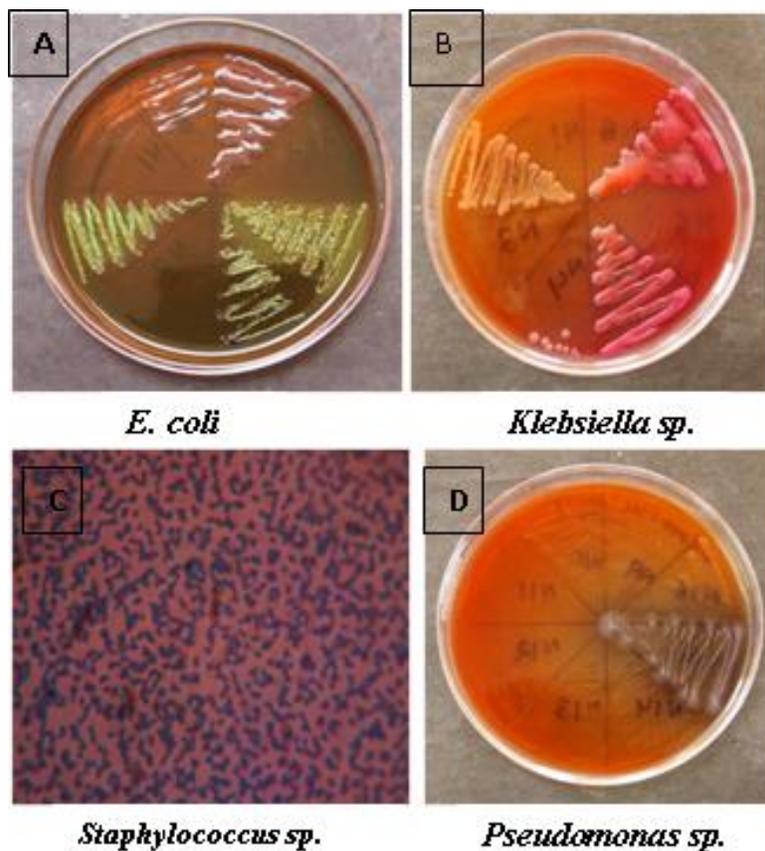


Figure no 1: Identification of selected bacterial cultures

Table no 1: List of bacterial cultures isolated from different places

S.No.	Culture number	Place of isolation
1.	N1, N2, N3, N4, N5, N6, N7, N8, N21	Sewage
2.	N9, N10, N13	Skin of human
3.	N11, N15, N23	Kitchen sink
4.	N12, N14, N20, N22	Mouth
5.	N16	Computer surface
6.	N17, N19, N24	Soil

Table no 2: Characteristics of bacterial cultures

S. No.	Culture number	Gram stain reaction	Lactose fermentation	MR test	VP test	Indole test
	N5	-	+	+	-	+
	N7	-	+	-	+	-
	N14	+	+	+	+	-
	N15	-	-	-	-	-

Study of antibiotic susceptibility: Antibiotic susceptibility pattern of selected four bacteria was studied to determine their drug resistance pattern. Five different antibiotics namely chloramphenicol, tetracycline, bacitracin, streptomycin and penicillin G were used in the study and nystatin was used as control. The zone of inhibition (cm) formed around the antibiotic discs in Petri dishes having different bacterial cultures were measured and the results are shown in Figure 2. It was seen that all the four bacterial culture were sensitive to chloramphenicol and streptomycin. The sensitivity of chloramphenicol was highest. The bacteria *E. coli*, *Klebsiella sp.* and *Staphylococcus sp.* were sensitive to tetracycline but penicillin resistant. Bacteria *E. coli*, *Klebsiella sp.* and *Pseudomonas sp.* were resistant to bacitracin but *Staphylococcus sp.* was sensitive to bacitracin. Interestingly in the study all the four bacterial cultures were resistant to penicillin G and three bacterial cultures were resistant to bacitracin. These results strongly indicate that commonly isolated bacterial cultures have also developed resistance to multiple drugs. Multidrug resistant bacteria are taken up along with contaminated food and water and may result in food borne infections.

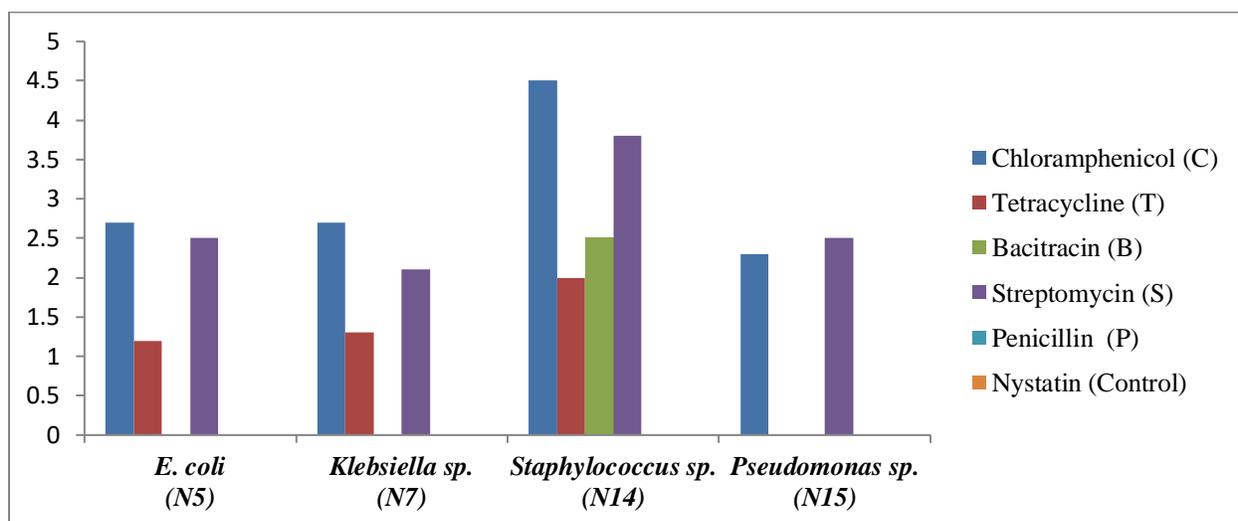


Figure no 2: Effects of different antibiotics on bacterial cultures

Antibacterial effect of honey and aqueous extracts of medicinal plant on growth of bacterial cultures: The antibacterial effects of pure honey and 5% and 10% extracts of Basil, Clove, Ginger, Garlic and Turmeric were determined. It was seen that pure honey showed substantial antimicrobial effects against all the bacterial cultures results are shown in Figure 3. The 5% aqueous extracts of Garlic and Ginger showed significant antibacterial effect against all the four bacterial cultures. However, the 5% aqueous extracts of Clove, Turmeric and Basil did not show antibacterial effect against these bacterial cultures. Further, it was seen that 10% aqueous extracts of Ginger and

Garlic showed antibacterial activities against all the four bacteria and 10% aqueous extracts of Turmeric and Basil showed antibacterial effects against *E. coli*, *Staphylococcus* sp. and *Pseudomonas* sp. whereas there was no effect of these extracts on the growth of *Klebsiella* sp. The 10% aqueous extract of Clove did not show any antibacterial effect against these four cultures. So it was interesting to see that Ginger and Garlic are highly potent in killing bacteria and they show high antibacterial effects against all bacterial cultures even at 5% concentration. Turmeric and Basil are less potent and they show antibacterial effects against some bacterial cultures only at high concentration. Earlier studies have shown that the ethanolic extract of Clove and Ginger show antimicrobial activity at 10 mg/ml concentration against some food poisoning bacteria. It had inhibitory effect against four of the pathogenic strains (*Bacillus cereus*, *Staphylococcus aureus*, *E. coli* and *Pseudomonas aeruginosa*) while Ginger was effective against three of them (*Bacillus cereus*, *Staphylococcus aureus* and *Pseudomonas aeruginosa*) and did not have any effect on *E. coli* (Mostafa et al., 2018). Researchers also reported that aqueous extract of clove did not show inhibitory effect against all the pathogenic bacteria. However, the methanolic extract of Clove was reported to be potentially effective against *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E. coli* with minimum inhibitory concentration (MIC) ranges from 0.1 to 2.31 mg/ml (Pandey and Singh, 2011). Gonalimali et al., (2018) determined the antibacterial potential of ethanolic and aqueous extract of clove at 20% concentration on Gram positive, Gram negative bacteria and antifungal activity of one fungus *Candida albicans*. Both extracts inhibited the growth of microorganisms with different potency. Ethanolic extract of Clove showed antifungal activity against *Candida albicans* with zone of inhibition 25.2 mm. Ethanolic and aqueous extracts of Clove showed highest activity against *Bacillus cereus* (18.2 mm and 15.1 mm) but the minimum inhibitory concentration of aqueous extract is 0.315% and minimum inhibitory concentration of ethanolic extract was 0.625%. Mostafa et al., (2018) showed that the differences in MIC of plant extracts are due to variation in their chemical constituents and volatile nature of their components. The ethanolic extract has lower MIC values than aqueous extracts which support that alcoholic extracts have higher antimicrobial activities than aqueous extracts. So our results disagree with Mostafa et al., (2018) findings who have studied the antibacterial potential of Clove in aqueous extract it may be due to different levels of chemical constituent of plants. Zeedan et al., (2016) showed that the ethanolic extract of Clove, Ginger and Black cumin at 10%, 20% and 50% concentration showed significant antimicrobial activity against all the tested bacterial cultures and the least antibacterial effect was shown by aqueous and ethanolic extract of Ginger. We also found that 5% and 10% aqueous extract of Ginger inhibited the growth of *E. coli*, *Staphylococcus aureus*, *Klebsiella* sp. and *Pseudomonas* sp. Rahman et al., (2020) determined the antibacterial activity of crude Ginger extracts at different concentrations on *Staphylococcus aureus*. The results indicate that there was no inhibition of *Staphylococcus aureus* from 5% to 60% crude Ginger extract which differ with our studies. The inhibition of growth is started from 70% crude Ginger extract and complete growth inhibited at 100% concentration in the study of Rahman et al., (2020). Nas and Ahmad, (2018) determined the antibacterial activity of aqueous, ethanol and n-hexane extracts of Ginger at 10 microgram/ml concentration. Their antibacterial assay showed that ethanolic extract has highest antibacterial activity followed by aqueous and n-hexane extract. This result clearly show that ethanol extract has maximum number of phytochemical components compared to aqueous and n-hexane and better solubility of the bioactive constituents in ethanol than in aqueous and n-hexane extracts. Gul and Bakhat, (2015) determined the antimicrobial activity and preservation potential of turmeric extract prepared in water, chloroform, ethanol and n-hexane with 1% concentration against *E. coli*, *Staphylococcus aureus*, *Salmonella typhi* and *Candida albicans* by using disc diffusion method. Their results showed that aqueous extracts of turmeric reduces the growth of *E. coli* and *Staphylococcus aureus*. Methanol extract of turmeric was effective for all the microbial cultures. Chloroform and n-hexane extracts of turmeric show weak activity against all the entire tested microbial cultures. So their findings disagree with our results as we did not found any antibacterial activity of aqueous extract of Turmeric at 5% concentration whereas inhibition of the growth of bacterial cultures was seen at 10% concentration. Gabriel et al., (2018) examined the antibacterial activity of crude, ethanolic and methanolic extracts of Basil against one human pathogen, *Vibrio* sp. which cause diarrhea. Their results showed that the ethanolic extract was more effective than methanolic and crude extract. So the result of their study showed that Basil can be used for controlling diarrheal illness due to contamination of drinking water. Semeniuc et al., (2016) also demonstrated that essential oil of Basil plant had mild antibacterial effect against *E. coli* and *Bacillus cereus* but did not inhibit *Staphylococcus aureus*, *Salmonella typhimurium* and *Pseudomonas aeruginosa* (Semeniuc et al., 2016). On the other hand our results showed that 10% aqueous extracts of Basil showed antibacterial effects against *E. coli*, *Staphylococcus* sp. and *Pseudomonas* sp. whereas there was no effect of these extracts on the growth of *Klebsiella* sp. disagree with who found that Basil oil inhibit the growth of *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *E. coli* at 2.25% and 4.5% concentration (Yamini et al., 2016). In view of the above it appears that the antibacterial effects of extracts of

medicinal plants show slight variation in results reported by different authors, which may be presented in different plant varieties due to variation in chemical constituents of plants.

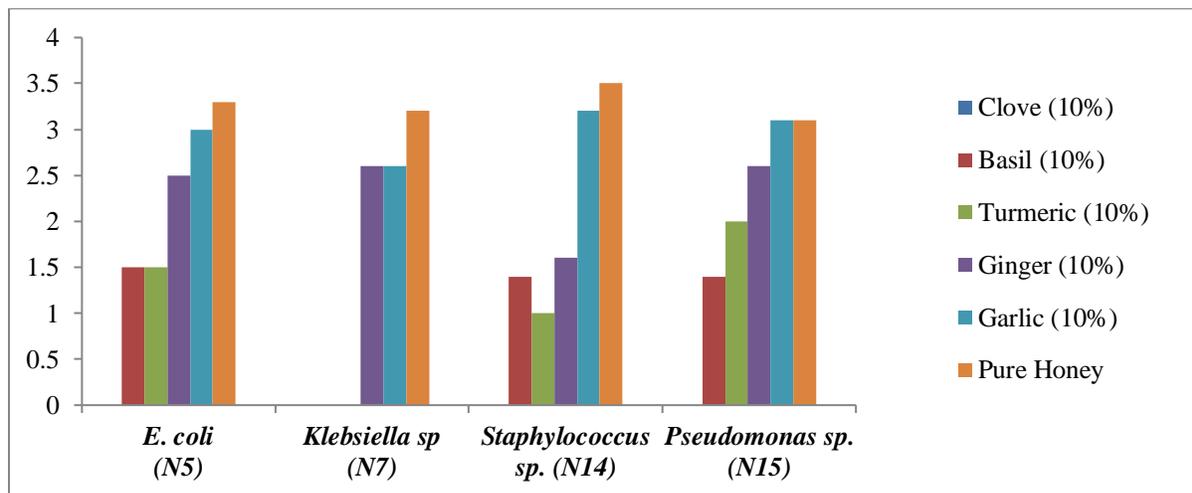


Figure no 3: Effects of aqueous extracts of medicinal plants and honey on bacterial growth

Antibacterial effects of medicinal plant extracts in combination with honey on growth of bacterial cultures:

The 10% extracts of medicinal plants were prepared in honey and they were used to study the antibacterial activity. The results are shown in Figure 4. It was seen that the extracts of medicinal plants in combination with honey cause little synergistic effect and lead to increase in antibacterial activity of these extracts. The effect was more pronounced against *Pseudomonas sp.* and *Staphylococcus sp.* in comparison to *E. coli* and *Klebsiella sp.* There was no antibacterial effect of Clove even in combination with honey. Moreover, Turmeric and Basil also did not show any antibacterial effects against *Klebsiella sp.* when combined with honey. These results suggest that the mixing of medicinal plant extracts with honey may enhance their antibacterial effects. Such results have also been obtained by Zeedan et al., (2016) who studied that the antibacterial effect of honey and herbal extracts of Ginger, Clove and Black cumin at 10%, 20% and 50% concentration on different bacterial cultures. Their results showed that combination of honey with herbal extracts may have synergistic effects (Zeedan et al., 2016). These results are also agree with earlier studies which show that there is synergistic effect on antimicrobial activity from the combination of Ginger and honey against oral isolates (Eja et al., 2011; Patel et al., 2011). Thus, it appears that combination of honey with herbal extracts may increase their potency.

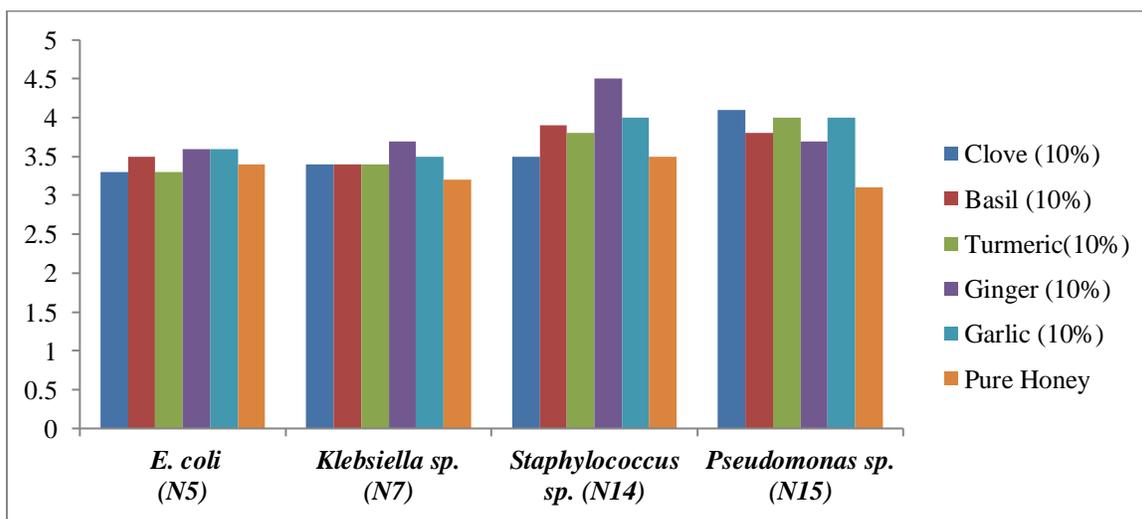
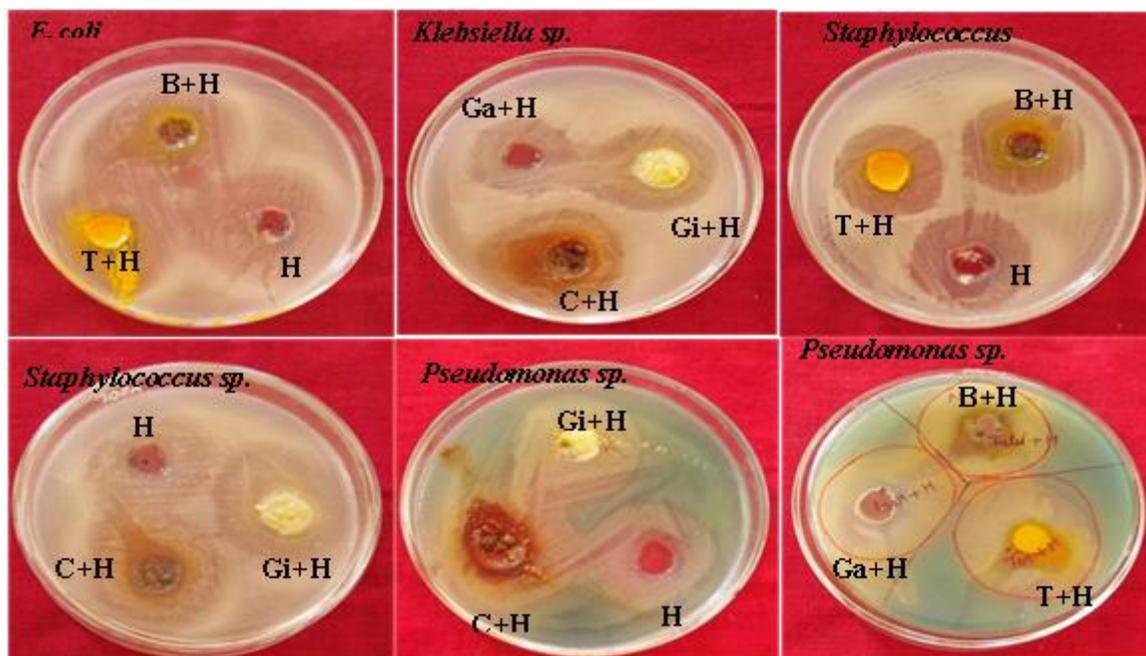


Figure no 4: Effects of extracts of medicinal plants in combination with honey and only honey on bacterial cultures



B+H = Basil+ Honey, T + H = Turmeric + Honey, Ga + H = Garlic + Honey, C + H = Clove + Honey, Gi + H = Ginger + Honey, H = Honey.

Figure no 5: Effects of extracts of medicinal plants in combination with honey and only honey on bacterial cultures

IV. Conclusion

From our study it can be concluded that aqueous extracts of Ginger, Garlic, Turmeric and Basil have significant antimicrobial effects on commonly encountered food borne pathogens namely *E. coli*, *Klebsiella sp.*, *Pseudomonas sp.* and *Staphylococcus sp.* The effects of these aqueous extracts are strong enough to inhibit the growth of these bacterial cultures which cannot be killed by Penicillin and Bacitracin antibiotics as they have become resistant to these antibiotics. Hence, the use of these spices as food ingredients may help in eliminating these food borne pathogens. These may help in improving the overall health of human who consume spices as food. Further, it was interesting to find that combination of honey with aqueous extracts of Basil, Garlic, Ginger and Turmeric have increased antibacterial effects. Thus, if these spices are mixed with honey and consumed then they may eliminate pathogens in mouth and help in improving oral hygiene and reducing food borne infections. The marked effects of mixture of Ginger, Turmeric, Garlic and Basil extracts with honey have a broad spectrum antimicrobial activity means it enhances the effect of antibacterial activity of plants. The new formula can be used as antibacterial agents as natural alternative to antibiotics at low concentration but still further extensive studies should done to explore the hidden properties of these plant extracts and honey.

In future research is required to study the mechanism of interaction of honey with plant extracts. Moreover, the effect of more medicinal plants and their different concentration with honey can be tested on other bacterial cultures.

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