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# A comparative study of antimicrobial and antioxidant activities of garlic (*Allium sativum* L.) extracts in various localities of Pakistan

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**The present study was conducted to analyze garlic extracts using compositional analysis and assessment of antibacterial and antioxidant properties against various microbes. It was observed that garlic consist of various bioactive compounds such as alkaloids, phenols, tannins and flavonoids with different concentration levels. Antimicrobial activity of different solvent extracts (n-hexane, chloroform, acetone, butanol and methanol) was tested against *Escherichia coli, Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Streptococcus epidermidis and Klebsella pneumonia* by using agar well diffusion assay. Results indicate that all the extracts were effective against the microorganism tested but n-hexane extract showed minimum activity against all microbes. Methanol extract was proven to be more effective against the *K. pneumonia*. MIC of these extracts was checked from 1-15 mg/mg concentration levels. Garlic plant extracts have rich contents of alkaloids, flavonoids, tannins and phenols. Various bioactive compounds are responsible for its medicinal properties.**

**Key words:** Antimicrobial activity, antioxidant activity, compositional analysis, flavonoids, alkaloids.

## INTRODUCTION

*Allium sativum* L. commonly known as garlic is among the oldest cultivated plant which is used for therapeutic purposes. Garlic has played one of the most important dietary and medicinal roles in human bodies for centuries and is used as a spice as well as medicinal herb. Garlic is a member of the lily (*Liliaceae*) family. It consists of more than 250 genera and 3700 species. These plants can tolerate unfavorable conditions, that is, winter and dryness due to their resistant structures: bulbs, tubers and rhizomes. The largest and most important representative genus of the *Alliaceae* family is *Allium*. It consists of 450 species, widely distributed in the northern hemisphere. There are over 300 varieties of garlic grown worldwide. In addition to the well known garlic and numerous other species are extensively grown for cooking purpose, such as leek (*Allium porrum* L.), scallion (*Allium fistulosum* L.), shallot (*Allium ascalonicum* Hort.), wild garlic (*Allium ursinum* L.), elephant garlic

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(*Allium ampeloprasum* L. var.*ampeloprasum*), chive (*Allium schoenoprasum* L.) and Chinese chive (*Allium tuberosum* L.) (Nuutila et al., 2002).

The biological and medical functions of members of *Alliaceae* family are mainly due to their high organo-sulphur compound contents. It also contains many other sulfur containing compounds such as alliin, ajoene, diallylsulfide, dithiin, S-allylcysteine, and enzymes, vitamin B, proteins, minerals, saponins, flavonoids, and maillard reaction products, which are non-sulfur containing compounds (Kojuri et al., 2007).

There is a wide range of reported therapeutic effects, such as hypolipidaemic, antiatherosclerotic, hypoglycaemic, anticoagulant, antihypertensive, antimicrobial, antidote (heavy metal poisoning) and hepatoprotective, preventing cold and flu symptoms through immune enhancement and exhibits anticancer and chemopreventive activities (Rivlin, 2001; Banerjee et al., 2003; Thomson and Ali, 2003; Amagase, 2006). The antimicrobial properties of crushed garlic have been known for a long time. A wide range of antimicrobial properties including antibacterial activity has been reported for crushed garlic (Bakri and Douglas, 2005). Recent chemical characterisation of sulphur compounds has shown that they are the main active antimicrobial agents (Rose et al., 2005).

The antioxidant properties of garlic and different garlic preparations are well documented (Cho and Xu, 2000; Nuutila et al., 2003; Saravanan and Prakash, 2004). The potential antioxidant properties of garlic are related to its phenolic and flavonoid fractions (Miller et al., 2000). Garlic has been used as an important flavouring agent, traditional medicine, and functional food to improve physical or mental health (Gedik et al., 2005; Pedraza-Chaverri et al., 2007).

Therefore, keeping in view the importance of garlic as an important medicinal food, the present study was conducted with the following aims and objectives: To evaluate proximate analysis of garlic samples, isolate phenolics, flavonoid, tannins and alkaloids from garlic and to determine antibacterial and antioxident activities of various extracts of garlic.

## MATERIALS AND METHODS

### Collection and preparation of sample

Garlic samples were collected from local markets of Raja Bazar Rawalpindi and Sihala Bazar, Islamabad in fine plastic bags duly labeled with date of collection and stored at -20°C for further process. Garlic samples were oven dried at 60°C overnight. Samples were crushed into powder form using electric blender and were saved at temperature of 20°C for further use.

### Biochemical analysis of *Allium sativum* L.

#### *Determination of moisture contents and protein analysis*

Moisture contents of plant samples were analyzed by AOAC (1990) method. Protein content (nitrogen × 6.25) was determined by micro-Kjeldahl nitrogen analysis by using AOAC 979.09 and 920.87 methods (AOAC, 1990) and by Lowry's method (Lowry et al., 1951).

#### *Determination of oil contents*

The oil contents were analyzed by AOAC method, 920.85 with Soxhlet apparatus. In the Soxhlet extraction procedure, 5 g of the powdered sample were packed in a thimble and the oil was extracted with diethyl ether for 18 h (AOAC, 1990).

#### *Determination of total minerals (ash)*

For total ash contents, 4 g dried plant samples were taken in a pre weighed crucible and then shifted to furnace at 800°C for 1 h. Then samples were cooled in desiccators and weight was taken after cooling. Analysis of total ash was done by the method of AOAC (1990).

#### *Carbohydrate analysis*

Carbohydrate contents (%) of the samples were determined by subtracting the total percentages of crude protein, crude lipid, ash and moisture from 100 as outlined by Al-Khalifa (1996).

#### *Determination of metal ions*

Garlic samples underwent dry digestion as described by Solvak et al. *(*2006). One gram was placed in porcelain crucible and turned into ashes at 450°C for 18-20 h. The ash was then dissolved in 1 ml concentrated nitric acid ($HNO_3$) and was evaporated to dryness. Then it was heated again at 450°C for 4 h, treated with 1 ml concentrated $H_2SO_4$, 1 ml $HNO_3$ and 1 ml $H_2O_2$ and finally diluted with deionized water up to volume of 10 ml. Blank sample was also treated in the same way. Metal ions including Mg, Zn and Cu in the sample were determined by using flame atomic absorption spectrophotometry.

### Analysis of phytochemicals

#### *Determination of flavonoid*

Flavonoid contents were determined by dissolving 5 g of sample in 50 ml of 80% aqueous ethanol and the whole mixture was left in shaker incubator for 24 hThe extract was then centrifuged at 10,000 rpm, at 25°C for 15 min. Pellet was discarded and supernatant containing flavonoid was stored at 4°C. Estimation was carried out by a calorimetric assay as described by Lillian et al. (2007a). The flavonoid extract (250 µl) was mixed with 1.25 ml of distilled water and 75 µl of 5% $NaNO_2$ solution. After 5 min, 150 µl of a 10% $AlCl_3.H_2O$ was added and after 6 min 500 µl of 1 M NaOH and 275 µl of distilled water were also added to the mixture. The solution was mixed well and absorbance was measured at 415 nm. Different concentrations of quercetin (50 to 250 µg) were used as standard to draw the standard curve (Lillian et al., 2007a).

### Determination of phenolic compounds

Total phenols were extracted by boiling 2 g of defatted sample with 50 ml of diethyl ether in water bath for 15 min. The estimation was carried out by a calorimetric assay as described by Lillian et al. (2007b). 1 ml of sample was mixed with 1 ml of Folin Ciocalteu

reagent and after 3 min, 1 ml of saturated sodium carbonate solution was added to the mixture and volume was adjusted to 10 ml with distilled water. The reaction was kept in the dark for 90 min, after which the absorbance was measured at 725 nm. Different concentrations of phenol (50 to 500 mg) were used as standard to generate the standard curve and concentrations of garlic phenolics were measured (Lillian et al., 2007b).

#### Determination of tannins

Tannins were extracted by dissolving 0.5 g of sample in 100 ml of 70% acetone. Estimation of tannins was carried out by a calorimetric assay as described by Akindahunsi and Oyetayo (2006). Different concentrations of tannic acid (6.25 to 50 mg) were prepared by serial dilution from stock solution (50 mg/100 ml of 70% acetone). The absorbance was measured at 725 nm after the addition of 0.5 ml of folin phenol reagent and 2.5 ml of $Na_2CO_3$ (Akindahunsi and Oyetayo, 2006).

#### Determination of alkaloids

According to the method, dried sample was dissolved in ethanol (1:10) and was left on shaking for 24 h. Extract was concentrated near to dryness in oven and was re-dissolved in ethanol with addition of 1% HCl. The mixture was placed in refrigerator for three days. The solution was filtered and pH was maintained 8-10 and was extracted with chloroform by using separating funnel. Chloroform layer was recovered and ethanol layer was discarded whereas the solution was heated in hot water bath for evaporation. After that the sample was dried in oven to constant weight. Alkaloid contents were calculated on the basis of weight obtained and weight used (Gulfraz et al., 2011).

#### Bioassays

##### *Preparation of extract for antibacterial activity*

The powder form (80 mashes) of sample was extracted with different solvents (n-hexane, acetone, ethyl acetate, chloroform, butanol, ethanol and methanol) on the basis of their polarity. Initially the sample was extracted with n-hexane (1:10) by shaking for 24 h followed by centrifugation at 10,000 rpm for 15 min. Supernatant was then transferred to a pre-weighed falcon tube and residue was re-extracted with next solvent which was slightly polar then n-hexane. The same procedure was repeated with all solvents and all extracts were allowed to dryness in incubator. The dried extracts were dissolved in dimethylsulfoxide (DMSO) for antimicrobial assay.

##### *Microorganisms tested*

Antibacterial activity was tested against *Escherichia coli, Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Streptococcus epidermidis and Klebsella pneumonia* by using agar well diffusion method. Inoculums of all microbes were prepared in sterilized Lauria-Bertini media $gL^{-1}$ (10 g tryptophan, 10 g NaCl, 5 g yeast extract and distilled water) in separate test tubes which were then placed in shaker incubator at 37°C for 24 h to contain approximately $10^8$ cfu/ml.

#### Antibacterial activity

Antibacterial activity was tested by agar well diffusion method. Lauria-Bertini agar media was prepared and autoclaved at 121°C for 15 min which was then cooled and poured in Petri plates under sterilized condition of laminar flow hood. The wells of 6 mm were bored in each plate and the plates were inoculated with 30 µl of inoculum. Then 1000 µg/75 µl of each of sample was pippetted in each well and plates were incubated at 37°C for 24 h. After 24 h, zones of inhibition were measured in millimeter (mm).

#### Minimum inhibitory concentration (MIC)

Various concentrations of the garlic extracts were prepared: 1, 5, 10 and 15 mg/ml. The cultured plates were again seeded with test bacterial organism and allowed to solidify and thereafter punched with a sterile cork borer (5.0 mm diameter) to cut uniform wells. The open wells were filled with 0.05 ml of the extract. The plates then incubated at 37°C for 24 h. The lowest concentration of extract that showed inhibition of growth of the test organism was taken as minimum inhibitory concentration (MIC) as described by Ettebong and Nwafor (2009).

#### Antioxidant activity

The extracts (750 µl) of each sample were mixed with an equal amount of phosphate buffer (0.2 M, pH 6.6) and 1% potassium ferricyanide (a source of ferric ions). The mixture was incubated at 50°C for 20 min followed by addition of an equal amount of trichloroacetic acid (10%) to stop the reaction and was then centrifuged at 3000 rpm for 10 min. Upper layer (1.5 ml) was separated and mixed with an equal amount of distilled water and 0.1 ml $FeCl_3$ solution (0.1%). A blank was also prepared by using same procedure and the absorbance was measured at 700 nm as the reducing power (Gulfraz et al., 2011).

#### Statistical analysis

All experimental data was given as mean ± SD (Standard Deviation). Statistical analysis was conducted by using *t*-test.

## RESULTS AND DISCUSSION

Garlic samples were analyzed for chemical composition like total protein, oil content, and total carbohydrate and for phytochemicals. Whereas antibacterial and antioxidant activities of different garlic extracts were also assessed and results are presented in Figure 1.

### Biochemical analysis of garlic

Chemical composition of garlic shows higher concentration of carbohydrate (67.5-68.5%) and low concentration of oil (3.6-3.8 %) in samples (Figure 1). It was assumed that prolonged heating affected the oil quantity and resulted in the degradation of polyunsaturated fatty acids (Yunusova et al., 1998). The concentration level of protein (17.5-17.6%), moisture (9.6 to 10.2%) and ash (0.81-0.9%) were found in garlic samples (Figure 1). The values of different parameters analyzed from garlic were almost similar and comparable to values reported by other researchers (Haciseferogullar et al., 2005;

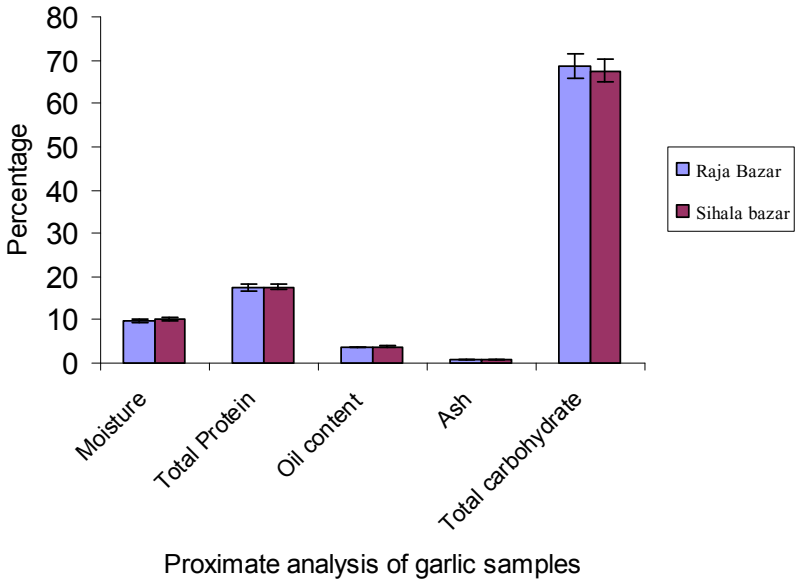


**Figure 1.** Comparison of proximate analysis (%) of garlic samples from Raja bazaar and Sihala bazaar.

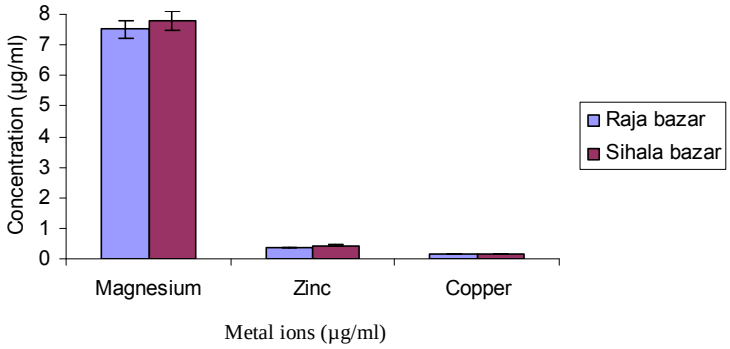


**Figure 2.** Comparison of metal ions (µg/ml) of garlic samples from different areas.

Nwinuka et al., 2005) especially the value for total protein which were similar to the study (17.5%) whereas literature showed that ash content in garlic is 4.06% but in the present study both garlic samples showed low ash contents (0.81-0.9%), whereas moisture contents (66.32%) obtained in literature was higher than that in this study (9.6-10.2%). Garlic samples were found to be good sources of carbohydrates but poor sources of ash, protein and fat. On the basis of protein and less oil content in garlic, it is a useful diet and could be used as a good medicinal food. Protein content was found to be considerably higher than concentrations in other vegetables such as bean and pea but crude oil contents were considerably lower in quantity.

Comparison of chemical composition of garlic from two localities is given in Figure 1. The results show that percentage of carbohydrate is higher in both samples whereas the concentration of moisture, protein, oil content and ash was found to be higher in sample collected from Sihala as compared to the other sample. The values obtained for moisture, oil, ash and total carbohydrate contents were significantly different with each other except the value for total protein which is non significant. The higher concentration of moisture, protein, oil contents and ash in Sihala samples might be due to better environmental conditions in this area.

## Metal ion detection

Metal ions such as magnesium, zinc and copper from garlic samples were analyzed by Flame Atomic Absorption Spectroscopy (FAAS) and results are given in Figure 2. Macro elements including Mg (7.504 ± 0.1 and

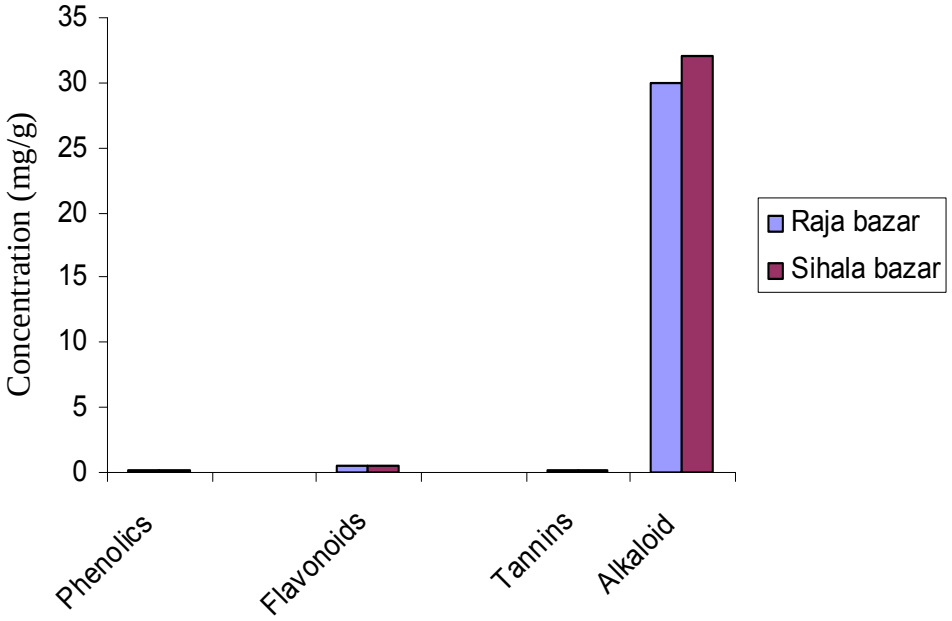


**Figure 3.** Comparison of phytochemical analysis of garlic samples from different areas.

7.77± 0.1 µg/ml) were found in high concentrations in samples from Raja bazaar Rawalpindi and Sihala bazaar Islamabad, respectively while other micro elements including Zn (0.370 ± 0.01 and 0.428 ± 0.07 µg/ml) and Cu (0.147 ± 0.01 and 0.16 ± 0.01 µg/ml) are found in lesser amounts. All the values obtained are significantly different from each other. Many of these metal ions act as cofactor for different enzymes. Higher concentration of Mg (7.504 ± 0.1 and 7.77± 0.1 µg/ml) are present in garlic whereas reported Mg, Cu and Zn concentrations in garlic were 1.056 ± 0.025 , 0.0912 ± 0.07 and 0.0274 ± 0.01 µg/ml, respectively (Haciseferogullar et al., 2005). Presence of all these metal ions increases the nutritional value of garlic (Metwally, 2009). The metal ion composition of garlic is mainly dependent on their ecosystem and soil composition. Furthermore garlic has higher capability to extract metal ions from other substrate. Therefore, metal ion composition of garlic is mainly dependent on their ecosystem and substrate composition (Richa et al., 2005). Zn and Mg activate DNA polymerase, which is necessary for synthesis and transcription of DNA. Magnesium is also cofactor for enzymes that play very important role in glycolysis to control sugar metabolism in the body by converting glucose to pyruvate. Comparatively, metal ions concentration increases for Sihala bazaar Islamabad as shown in Figure 2. These increases may be due to variations in soil condition of these areas.

## Analysis of phytochemicals

The results of organic compounds such as phenolics, flavonoid and tannin of two garlic samples collected from different areas are shown in Figure 3. The data shows that phenolics (0.152 ± 0.05 and 0.162 ± 0.01 mg/g), flavonoid (0.451 ± 0.03 and 0.498 ± 0.01 mg/g), tannins (0.167 ± 0.05 and 0.123± 0.048 mg/g) and alkaloid (30.2 ±0.5 and 32.1 ± 2.0 mg/g) were present in garlic samples (Figure 3). All the values obtained after analysis were significantly different from each other (Gulfraz et al., 2011).

The results showed that phenolics (0.152 ± 0.05 mg/g), flavonoids (0.451 ± 0.03 mg/g), tannins (0.167 ± 0.048 mg/g) and alkaloid (30.2 ± 0.5 mg/g) were found in garlic sample from Raja bazaar Rawalpindi whereas values of phenolics (0.162 ± 0.01mg/g), flavonoids (0.498 ± 0.01 mg/g), tannins (0.123 ± 0.01 mg/g) and alkaloids (32.1 ± 2.0 mg/g) obtained from garlic sample Sihala bazaar Islamabad are significantly different from each other (Figure 3). Further, the figure showed that there is significant difference among all the samples with respect to concentrations of chemical compounds in the samples collected from different areas. This data also shows gradual increase in values for chemical compound composition of Sihala bazaar Islamabad. These increases may be due to variations in environmental conditions as well as soil of that area.

## Antibacterial activity of garlic extracts

Historically, garlic has been used worldwide against high bacterial infections. Garlic exhibits broad spectrum anti-bacterial activity against both Gram-positive and negative

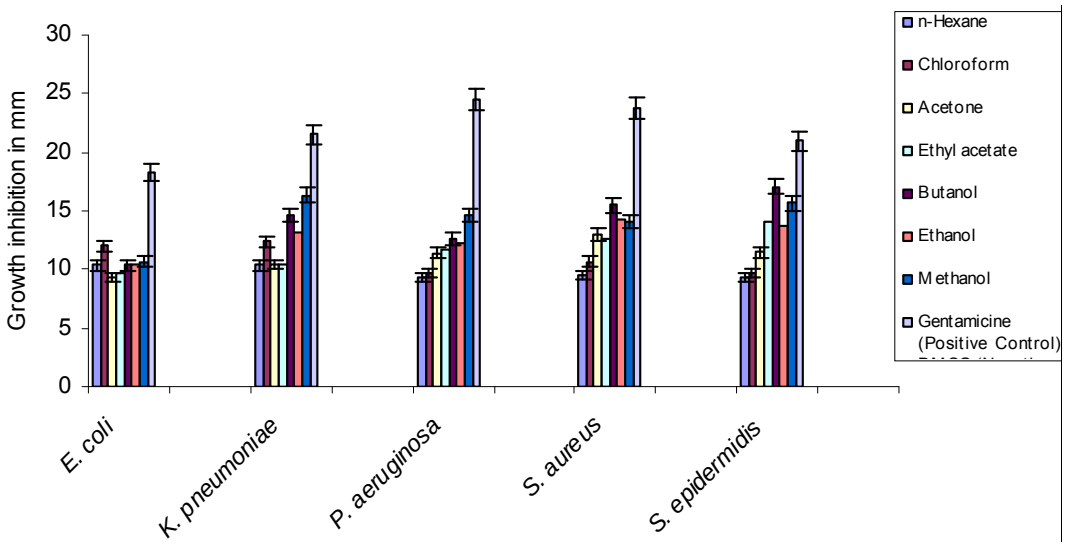


**Figure 4.** Antibacterial activity of garlic extracts.

**Figure 5.** Antimicrobial activity of different garlic extracts against *K. pneumoniae.*

**Figure 6.** Antimicrobial activity of different garlic extracts against *S. epidermidis.*

bacterial strains. The growing concern about food has just led to the progress of antimicrobial agents to manage food borne microorganisms (Nevas et al., 2004). Spices are the most frequently used normal remedial agents in foods and has been used traditionally for preserving foods and to enhance flavor of food (Souza et al., 2005).

Antibacterial activity of different extracts (n-hexane, chloroform, acetone, butanol, ethanol and methanol) of garlic samples were tested against *E. coli, S. aureus*, *P. aeruginosa*, *S. epidermidis and K. pneumonia* (Figure 4). According to our results, n-hexane, chloroform, acetone, butanol, ethanol and methanol medium have shown inhibitory effects against the tested microorganisms (Ali and Blunden, 2003). It was observed that n-hexane extract of garlic showed activity only in the the range of 9.3±0.5-10.3±0.5 mm against all microorganisms. Acetone extracts showed less activity against *E. coli* but was active against all other microbes. The ethanol extract was proved to be effective against *S. aureus, K. pneumineae* and *S. epidermidis* but was not much effect on *E. coli*. Methanol extracts were proven to be more effective against all the microbes (Gulfraz et al., 2011).

Higher inhibition zone of butanol extract of garlic have been shown against *S. epidermidis* (16±1.0 mm) followed by methanol extract against *K*. *pneumineae* (15±1.0 mm). The results are almost similar to those reported by Pundir et al. (2010) when they studied the antibacterial activity of garlic extracts (Figures 5 to 6). In the present study, the ethanolic extracts of garlic showed inhibitory activity against all the five bacterial strains in which the diameter of zone of inhibition was 10.3±0.5 mm to 14.3±0.5 (Figure 4). The butanol extract revealed maximum zone of inhibition (16±1.0 mm) against *S. epidermidis* followed by *S. aureus* (15±1.0 mm) and *K. pneumineae* (14.6±0.5 mm) (Sulieman et al., 2007).

**Table 1.** Minimum inhibitory concentration (mg/ml) of various extracts of garlic sample (diameter of zone inhibition in mm).

| Microorganism | Diameter of zone of inhibition | | | |
|---|---|---|---|---|
| | Extract concentration (mg/ml) | Growth inhibition zone diameter | | |
| | | Butanol | Ethanol | Methanol |
| *E. coli* | 15 | - | 6±0.5 | 8±1.0 |
| | 10 | - | 5±0.5 | 8±1.0 |
| | 5 | - | - | - |
| | 1 | - | - | - |
| *K. pneumoniae* | 15 | 8±1.0 | 10±1.0 | 17±1.0 |
| | 10 | 5±0.8 | 4±1.0 | 14±1.0 |
| | 5 | - | - | 11±0.5 |
| | 1 | - | - | - |
| *S. aureus* | 15 | 10±1.0 | 18±1.7 | 9±1.0 |
| | 10 | 4±0.5 | 12±1.0 | 9±1.0 |
| | 5 | - | - | - |
| | 1 | - | - | - |
| *P. aeruginosa* | 15 | 11±1.0 | 16±1.0 | 13±1.0 |
| | 10 | 8±1.0 | 8±1.3 | 7±1.0 |
| | 5 | - | - | - |
| | 1 | - | - | - |
| *S. epidermidis* | 15 | 14±1.0 | 5±0.9 | 11±1.0 |
| | 10 | 6±1.0 | - | 8±1.0 |
| | 5 | 3±0.5 | - | 8±1.0 |
| | 1 | - | - | - |

- No inhibition. Values are Mean ± SD after triplicate analysis.

Values in terms of garlic extracts against *E. coli*, *S. aureus* revealed the highest antibacterial activity was observed against *E. coli*. Antimicrobial activity of all extracts for *E. coli* was in the range of (9.3±0.5-11±1.0 mm), *K. pneumineae* (10.3±0.5-15±1.0 mm), *P. aeruginosa* (9.3±0.5 -14.6±0.5 mm), *S. aureus* (9.3±0.5-15±0.5 mm) and *S. epidermidis* (9.3±0.5 -16±1.0 mm) (Figure 4).

The antibacterial activity of garlic has been reported by many workers against *S. aureus, E. coli* and *K. pneumoniae* (Jabar and Mossani, 2007) and *E. coli* and *S. aureus* (Vuddhakul et al., 2007). Shelef (1983) reported that allicin (an essential oil) isolated from garlic inhibited bacterial growth and it was shown that most of the antimicrobial activity was due to phenolic compounds like eugenol, thymol and carvacol found in garlic (Gulfraz et al., 2011).

## Minimum inhibitory concentration (MIC) of garlic extracts

Garlic extracts in methanol, ethanol and butanol showed better inhibition against the bacterial strains used, as compared to other extracts, so different concentrations of these extracts were subjected to determination of MIC. The concentrations of the extracts used for MIC determination were 15, 10, 5 and 1 mg/ml and the results are given in the Table 1. It was observed that the most susceptible bacterial strain was *S. epidermidis* against butanol extract (3±0.5 mm) at concentration of 5 mg/ ml and it was followed by *K. pneumonea* against methanolic garlic extract (11±0.5 mm) at concentration of 5 mg/ml. Whereas *S. aureus* showed (12±1.0 mm) inhibition when 5 mg/ml of ethanol extract was applied. All bacterial strains showed higher susceptibility against methanol extracts, moderate activity against ethanol and less activity against butanol extract. The large size of zones of inhibitions indicates the increase in the concentration of extracts and the inhibition activity of extracts. Furthermore, gentamicine a standard antimicrobial agent had shown zones of inhibition ranging from 18±0.5 - 24±0.5 mm, and highest value for *P. aeruginosa* (24±0.5 mm). The zones of inhibition of effective extracts were close to that of the drug. It seems that the organisms may

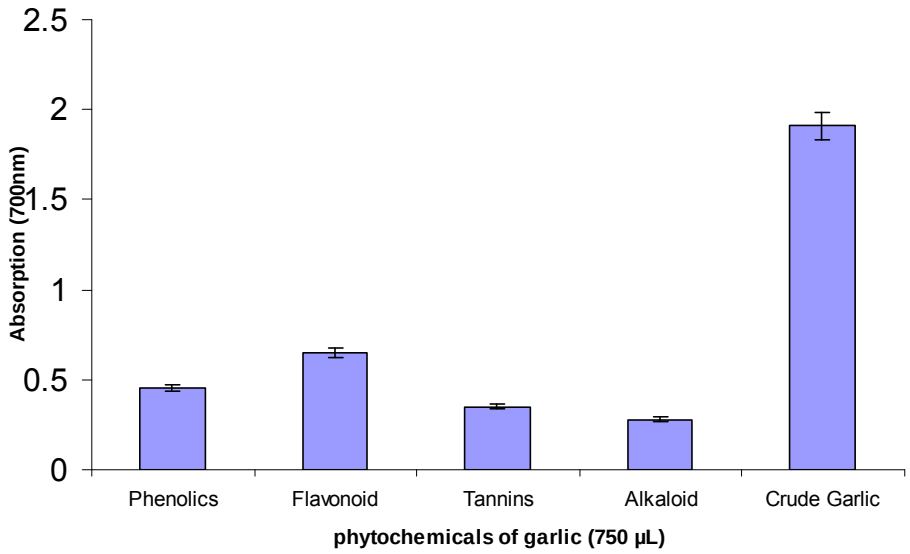


**Figure 7.** Antioxidant activity of phytochemicals found in garlic samples. Mean ± SD after triplicate analysis.

need higher concentrations of extracts to kill them, due to their cell wall components (Gulfraz et al., 2011). Antimicrobial activities for butanol extract values ranges from (3±0.5 - 14±1.0 mm), ethanol extract (4±1.0 - 18±1.7 mm) and methanol extract (6±1.0 - 17±1.0 mm) for all microorganism tested (Table 1). The results showed that the MIC values range between 3±0.5 - 18±1.7 mm.

## Antioxidant activity of phytochemicals

Antioxidant activity of different phytochemicals, that is, phenols and tannins were determined and it was found that antioxidant activities of phenolics (0.45 ± 0.01 mg/g), flavonoid (0.65 ± 0.01 mg/g), tannins (0.35 ± 0.01 mg/g), alkaloid (0.28 ±0.01 mg/g) and crude garlic (1.91 ± 0.12 mg/g) were present. Results of antioxidant activity are shown in Figure 7. It shows that crude garlic (1.91 ± 0.12 mg/g) has higher antioxidant activity than all other phytochemicals and alkaloid has less antioxidant activity than other phytochemicals. Among all phytochemicals, total phenols, flavonoids and tannins had shown antioxidant activity (Figure 7). Flavonoids and alkaloids of garlic extract which was evaluated using ascorbic acid as a standard. Assay involved the use of $FeCl_3$/ $K_3Fe(CN)_6$ complex as a source of ferric ions which may reduce to ferrous ion in the presence of extracts containing active flavonoid and alkaloids and was confirmed by production of green colour complex which intensity was measured spectrophotometrically. Thus, increase in absorbance of experiment was due to increased antioxidant activity (Nethravathi et al., 2006). Flavonoid and tannins are polyphenolic compounds which donate their hydrogen atom to ferric ions and convert them to their reduced form resulting in the production of intense green colour and greater absorbance. These polyphenolic compounds act as antioxidant agent and may protect the body from oxidative damage.

The present study indicates the presence of different concentrations of polyphenolic compounds in garlic which may protect the human body from its damaging effects. Therefore, utilization of garlic in diet may become useful in preventing number of diseases like carcinogenesis and cardiovascular diseases in human as reported by Gezer et al. (2005).

## Conflict of Interests

The author(s) have not declared any conflict of interests.

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