PHYTOCHEMICAL PROFILING OF PAKISTANI GARLIC VARIETIES; SPECIAL ATTENTION TO ANTIOXIDANT STATUS

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ABSTRACT: In the present study, locally grown garlic *i.e.* pink and white was evaluated for phytochemical profile. Pink garlic extracts from methanol, ethanol and water exhibited highest flavonols, flavanoids and DPPH by 10.20±0.50, 9.82±0.01 &9.20±0.03 mg/100g, 43.67±1.15, 42.50±1.59 &41.68±2.03 mg/100g and 53.53±1.78, 51.72±1.23 &51.19±1.03% respectively as compared to garlic white. The values for FRAP and ß-carotene were recorded maximumas9.47±0.47 mg TE/g and 69.44±0.54% in methanolic extracts. Similarly, highest ABTS value was exhibited by pink garlic methanolic extract (60.85±3.63 µmol TE/g). Maximum allicinwas4.63±0.02 mg/g in methanolic extract, however, white garlic extracts showed highest performance in ethanolic extract 0.961±0.002mg/g followed by methanolic 0.918±0.002mg/g.*In toto,* pink garlic extracts showed higher FRAP, DPPH, flavonoids, flavonols and ABTS values in comparison with garlic white. Moreover, amount of allicin was also higher in pink garlic.

Keywords: Garlic, antioxidant status, DPPH, FRAP, ABTS, allicin

INTRODUCTION

Functional/nutraceutical foods are the core element of diet based therapy owing to their health enhancing potential beyond the basic function of supplying nutrients. The use of such designer foods is an emerging trend among the health conscious consumers thereby captured the major share of the global nutrition market (Ares *et al*., 2009). Among the diet based interventional strategies, plants derived functional foods with rich phytochemistry are important that not only enhance wellness but also attenuate health risk factors (Tapsell*et al*., 2006; Shahidi, 2009).

Phytonutrients are endemic in the human diet from the ancient times providing natural shield against several metabolic syndromes. Amongst, garlic (*Allium sativum*L.) is an essential vegetable widely consumed as seasoning, flavoring and in culinary preparations. Alongside, it is utilized in folk medicines for curing various maladies (Rivlin, 2001). Garlic contains a range of phytochemicals that play an important role in the maintenance of human health and disease prevention (Butt and Sultan, 2009). Like other herbaceous plants, the composition of garlic varies withgeographical location, harvesting time, agronomic practices etc. Vegetables are rich source of phytonutrients such as carotenoids, anthocyanins and flavonoids etc. (Andersen and Jordheim, 2006). They have the ability to target at molecular level including enzyme kinetics, release of cytokines and signal transduction (Bárta*et al*., 2006).

According to estimation, about 95% of the sulfur from intact garlic cloves comes from two compounds namely S-allyl-cysteine sulfoxides and gamma-glutamyl-S-allylcysteine (Lawson *et al*., 2001). Nevertheless, the most abundant sulfur containing compound S-allyl-cysteine sulfoxide termed as allicin comprises 10 mg/g fresh or 30 mg/g dry weight of garlic (Lawson *et al*., 1995). During chopping and crushing, an odorless compound *i.e.*cysteinesulfoxides is readily converted to thiosulfinates, responsible for distinct freshly chopped garlic odor. However, allicin (diallylthiosulfinate) and S-allayl-cysteine are the main thiosulfinates out of which about 60-80% is allicin (Lawson *et al.,* 2001).

The concentration of allicin in fresh garlic varies depending upon variety, agronomical practices and region. The half-life of allicin at room temperature is 2-16 hr nonetheless, in crushed garlic or garlic juice around 2.4 days (Lawson, 1998).It has been deduced that most of the phytochemicals present in garlic and allied extract are derived from allicin thus helpful to improve human health. In aqueous garlic extract there is abundance of S-allayl cysteine and S-allylmercepto cysteine, involved in cholesterol lowering (Sterling and Eagling, 2001).

Generally, chromatographic methods are utilized to investigate garlic volatiles that may affect the overall acceptability of the product. Several sulfur components were investigated including allylmethylsulfide, allylmercaptan, 3,3'-thiobis-1-propene and diallyl disulfide. The diallyl disulfides are the most prominent component for all garlic preparations. Keeping in view the health claims of garlic, present project was designed to characterize locally grown promising garlic varieties/line lehsangulabi (garlic pink) and lehsan white VRIG-11 (garlic white) with special reference to their active ingredients. Optimization and extraction of garlic extracts using different solvents were carried out and quantified with special reference to allicin through

HPLC. Alongside, the antioxidant activity was assed using various phytochemical assays.

MATERIALS AND METHODS

The present research was carried out in the Functional and Nutraceutical Research Section at the National Institute of Food Science and Technology (NIFSAT), University of Agriculture, Faisalabad. The locally grown garlic was used for respective preparations.

Chemical analysis of garlic varieties: Two promising indigenous garlic varieties/line *i.e*lehsangulabi (garlic pink) and lehsan white VRIG-11 (garlic white) wereselected. After cleaning, cloves of each variety/line were peeled for the preparation of aqueous garlic extract. Likewise, various analytical and HPLC grade reagents and standards were purchased from Merck (Merck KGaA, Darmstadt, Germany) and Sigma-Aldrich (Sigma-Aldrich Tokyo, Japan). The garlic varieties were analyzed for moisture content, crude protein, crude fat, crude fiber, total ash content and nitrogen free extract according to their respective protocols mentioned in AACC (2000). The sodium (Na) and potassium (K) were assessed by Flame Photometer-410 (Sherwood Scientific Ltd., Cambridge) whilst, calcium (Ca), iron (Fe) and manganese (Mn) were determined through Atomic Absorption Spectrophotometer (Varian AA240, Australia) (AOAC, 2006)

Extraction and analysis of garlic extracts: Garlic extracts from respective varieties/line were prepared and collected at 60 min using three solvents *i.e.* 50% aqueous methanol, 50% aqueous ethanol and water at constant temperature *i.e.*60^{o}C (Table 1). Afterwards, resultant extracts except water extract were filtered and subjected to Rotary Evaporator (Eyela, Japan).

The solvent extracts of both garlic varieties/line were estimated for their physicochemical, antioxidant potential and phytochemicals. To assess the antioxidative perspectives, total phenols (TP), DPPH radical scavenging activity (1,1-diphenyl-2-picrylhydrazyl), antioxidant activity by β-carotene assay, ABTS (2,2'-azino-bis, 3-ethylbenzothiazoline-6-sulfonic acid) test and FRAP (Ferric reducing antioxidant power) assay were performed. Likewise, flavonoids, flavonols and tannin were also determined.

Antioxidative and phytochemical profiling of garlic extracts: Different antioxidant assay and phytochemical indicators were performed by following their respective procedures.

Total polyphenols: Total polyphenols (TP) were measured by using Folin-Ciocalteu method following the protocol of Singleton *et al*. (1999)at 765 nm with UV/Visible Spectrophotometer (CECIL-CE7200) against control. Total polyphenols was estimated as gallic acid equivalent (mg gallic acid/g) by following expression;

Table 1. Garlic extracts prepared from different solvents

Treatments	Solvent extracts
T_1	Garlic pink methanolic extract
T_2	Garlic pink ethanolic extract
T_3	Garlic pink water extract
T_4	Garlic white methanolic extract
T_5	Garlic white ethanolic extract
T_6	Garlic white water extract

Methanol = (50% methanol + 50% water)
Ethanol = (50% ethanol + 50% water)

$C=c\times V/ m$

C = Total phenolic contents (mg/g plant extract, in GAE)
c = Concentration of gallic acid (mg/mL)
V = Volume of extract (mL)
M = Weight of garlic methanolic extract (g)

Free radical scavenging activity (DPPH assay): The free radical scavenging activity *i.e.*DPPH (1,1-diphenyl-2-picrylhydrazyl) of garlic extracts was measured using the protocol of Muller *et al.* (2011). The absorbance was measured at 520 nm using UV/Visible Spectrophotometer and free radical scavenging ability was calculated by the following equation;

Reduction of absorbance (%) = [(AB - AA) / AB] × 100
AB = absorbance of blank sample (t = 0 min)
AA = absorbance of tested extract solution (t = 15 min)

Antioxidant activity (AA): Antioxidant activity of garlic extracts was estimated based on coupled oxidations of β-carotene and linoleic acid through spectrophotometer at 470 nm (Taga*et al*., 1984).

In (a/b) × 1/t = degradation rate of sample
ln = natural log
a = initial absorbance (470 nm) at time zero
b = absorbance (470 nm) after 40 min
t = time (min)

Antioxidant activity (AA) was expressed as % inhibition relative to control

$$AA = \frac{\text{Degradation rate of control - degradation rate of sample}}{\text{Degradation rate of control}} \times 100$$

ABTS (2,2'-azino-bis, 3-ethylbenzothiazoline-6-sulfonic acid) assay: The ABTS assay of garlic extracts were carried out using the protocol of Böhm*et al.* (2002).The prepared sample mixture was transferred to micro-cuvettes, centrifuged for 30 sec at 1000 *g* to separate the layers. The absorbance of lower layer was determined at 734 nm.

Ferric reducing antioxidant power (FRAP): The ferric reducing power of garlic extracts was estimated according to the protocol of Yuan *et al.* (2003) by measuring absorbance at 700 nm. During analysis, an

increase in absorbance (A) of the reaction mixture indicated reducing power.

Determination & quantification of flavonoids: Total flavonoids were estimated using the method of Ordonez*et al*. (2006). Total flavonoid content was calculated as quercetin equivalent (mg/100g) using the equation obtained from the calibration curve.The quantification of flavonols was done by the method of Kumaran and Karunakaran (2007). The flavonols content was expressed in quercetin equivalent (mg/100g) using the equation obtained from the calibration curve.

Estimation of tannins: The tannins were estimated by adopting the method of Nwinuka*et al*. (2005). Standard tannic acid solution was prepared from which a standard curve was drawn (absorbance versus concentration in mg/cm^3). From the curve, concentrations for each sample was obtained and used for the tannin content calculation (Nwinuka*et al*., 2005).

$$TC\ (g/100g) = \frac{C\ (mg) \times V_{ex}}{A \times M_s}$$

where
C (mg) = concentration from standard curve
Vex = extract volume (mL)
A = aliquot (mL)
Ms = mass of sample (mg)

HPLC Quantification: Different preparation of garlic extracts (methonolic, ethanolic and water) were subjected to HPLC quantification to estimate the allicin concentration by following the procedure of Bocchini*et al*. (2001).To conduct the experiment, HPLC Shim-Pack CLC-ODS C_{18} column (15 cm x 4.6 mm, 5.0 µm particle size) with UV/Visible detector was used. An Autosampler (WISP Model 710) uploaded the 20 µL sample. The temperature of internal column was maintained at room temperature. Estimation of allicin from representative samples was carried out at 254 nm while using mobile phase consisting of $DDHO_2$: Methanol (1:1) at the flow rate of 1.0 mL/min.

RESULTS AND DISCUSSION

Present study was planned to explore the nutraceutical worth of locally grown garlic varieties. For the intention, selected garlic samples were subjected to proximate and mineral composition. In this context, whole garlic (pink and white) were subjected to proximate profiling including moisture, crude protein, crude fat, crude fiber, ash and nitrogen free extract (NFE). Beside, mineral quantification and antioxidant potential estimations were also carried out by HPLC. Collected data was subjected to different statistical analysis to estimate the level of significance. The results with discussion of examined attributes are discussed herein.

Compositional Profiling: The proximate assay for pink and white garlic revealed moisture, crude protein, crude fat, crude fiber, ash and NFE as 67.05±5.20&66.85±4.19, 9.56±0.72 & 7.50±0.50, 0.75±0.05 & 0.56±0.03, 3.06±0.32 & 2.42±0.21, 3.53±0.31 & 2.95±0.23 and 16.05±1.08 &19.72±1.16%, respectively. Moreover, gross energy were recorded higher for pink garlic *i.e.*339.83±25.34kcal/100g than that of white 312.06±21.72 kcal/100g on dry weight basis (Table 2).

Table 2. Compositional analysis of garlic

Parameters (%)	Garlic pink	Garlic white
Moisture	67.05±5.20	66.85±4.19
Crude protein	9.56±0.72	7.50±0.50
Crude fat	0.75±0.05	0.56±0.03
Crude fiber	3.06±0.32	2.42±0.21
Ash	3.53±0.31	2.95±0.23
NFE	16.05±1.08	19.72±1.16
Energy (kcal/100g)	339.83±25.34	312.06±21.72

The results regarding proximate composition of present investigation are corroborated with the work of Bangash*et al.* (2011), probed the proximate composition of ten selected vegetables in Peshawar region for nutritional evolution. The reported values for moisture, crude fat, crudeprotein, fiber and ash of garlic were 66.80, 0.22, 4.01, 0.40 and 1.40%**,** respectively. Similarly, Odebunmi*et al.* (2010) examined African garlic for their proximate constituents and narrated values for moisture, crude fat, crude protein, crude fiber and ash as 76.86, 5.62, 8.75, 2.93 and 2.54%, respectively. Later, Otunola*et al.* (2010) observed 4.55, 4.08, 15.33, 0.72 and 2.10% of moisture, ash, crude protein, crude fat and crude fiber in the commonly consumed garlic cultivar of Nigeria. The differences among the white and pink garlic regarding their proximate profile may be attributed to varietal differences, genetic makeup, soil conditions and agronomic practices. The results regarding gross energy are in harmony with the findings of Nwinuka*et al.* (2005), observed 367.64 kcal/100g.

Table 3: Mineral profile of garlic

Minerals (mg/100g)	Garlic pink	Garlic white
Na	4.04±0.38	3.74±0.28
K	29.80±3.04	27.34±2.84
Ca	23.04±2.84	21.44±2.26
Fe	4.86±0.39	4.15±0.34
Mn	0.02±0.001	0.01±0.001

In the current investigation, minerals like sodium (Na), potassium (K), calcium (Ca) iron (Fe) and manganese (Mn) in pink and white garlic were

4.04±0.38& 3.74±0.28, 29.80±3.04 & 27.34±2.84, 23.04±2.84 &21.44±2.26, 4.86±0.39 & 4.15±0.34 and 0.02±0.001 & 0.01±0.001 mg/100g, respectively (Table 3).

The results for minerals are in resemblance with the earlier work of (Otunola*et al.*2010) who extensively examined the garlic for their mineral contents like Na, Ca, Fe, P, K, Zn, Mn and Mg; 4.10, 26.30, 5.29, 10.19, 54.00, 0.34, 0.001, 0.001 and 0.001mg/100g, respectively. Likewise, (Kumar*et al.*2010) illuminated that the garlic contained 30, 310 and 1.30mg/100g of Ca, P and Fe, respectively. The effect of climate, variety, soil and agronomic practices on mineral contents of garlic is evident from the early work of (Camargo*et al.* 2010), examined different garlic cultivar for their mineral profile and noticed that mineral profile also dependent on agro-climatic location and cultivar.

Antioxidant indices of garlic extract: Mean squares expounded significant differences due to treatments on total phenolic, flavonols, flavanoids, DPPH, FRAP, β-carotene and ABTS except for tannins. Mean values (Table 4) indicated highest TPC (51.65±3.25 mg GAE/100g) in T_1 (methanolic extract of pink garlic) followed by T_2 (ethanolic extract of pink garlic) 50.79±2.89 mg GAE/100g, T_3 (water extract of pink garlic) 45.83±1.56 mg GAE /100g, T_4 (methanolic extract of white garlic) 45.62± 2.51 mg GAE/100g and T_5 (ethanolic extract of white garlic) 45.14±2.12 mg GAE/100g whilst lowest in T_6 (water extract of white garlic) as 44.90±1.16 mg GAE/100g.

Table 4: Antioxidant indices of garlic extracts

Treatments	TPC (mg GAE/100g)	Flavanols (mg /100g)	Flavonoids (mg /100g)	DPPH (%)
T_1	51.65±3.25a	10.20±0.50a	43.67±1.15a	53.53±1.78a
T_2	50.79±2.89a	9.82±0.01b	42.50±1.59b	51.72±1.23b
T_3	45.83±1.56b	9.20±0.03c	41.68±2.03b	51.19±1.03bc
T_4	45.62±2.51b	9.15±0.05c	40.55±3.56c	50.84±1.45c
T_5	45.14±2.12b	8.87±0.01d	40.47±2.45c	50.51±1.89c
T_6	44.90±1.16b	8.66±0.08e	36.11±1.12d	49.71±1.77d

Likewise, pink garlic extracts *i.e.* methanol, ethanol and water exhibited highest flavonols, flavanoids and DPPH by 10.20±0.50, 9.82±0.01 &9.20±0.03 mg/100g, 43.67±1.15, 42.50±1.59 &41.68±2.03 mg/100g and 53.53±1.78, 51.72±1.23 &51.19±1.03% respectively as compared to garlic white *i.e.*9.15±0.05, 8.87±0.01 &8.66±0.08 mg/100g, 40.55±3.56, 40.47±2.45 &36.11±1.12 mg/100g and 50.84±1.45, 50.51±1.89 &49.71±1.77%, correspondingly.

The values for FRAP and ß-carotene were recorded maximum (9.47±0.47 mg TE/g and 69.44±0.54%) in methanolic extract of pink garlic as compared to white garlic methanolic extract (8.51±0.02 mg TE/g and 64.50±3.15%) trailed by ethanolic extract (9.33±0.21 mg TE/g&67.36±1.12% and 8.47±0.14 mg TE/g &62.35±2.89%, respectively) however, the least output was obtained for water extract of pink and white garlic as 8.52±0.23 mg TE/g &64.60±2.56% and 8.18±0.16 mg TE/g &61.50±1.89%, correspondingly. Similarly, highest ABTS value was exhibited by pink garlic methanolic extract (60.85±3.63 µmol TE/g) followed by white garlic methanolic extract (57.27±3.69 µmol TE/g). Similar pattern was observed in ethanolic extract; pink garlic showed higher ABTS value (58.59±2.48 µmol TE/g) as compared to garlic white (57.11±1.47 µmol TE/g) whilst in water extract the recorded values were 58.14±1.87 µmol TE/g and 55.81±1.29 µmol TE/g, respectively. The solvents imparted non-substantial differences in tannin contents. The maximum tannins were noticed in methanolic extract of pink garlic 0.93±0.01 mg/100g whilst lowest 0.81±0.02 mg/100g in water extract of white garlic (Table 5).

The results of instant investigation are comparable with the findings of Bozin*et al.* (2008), they illuminated that polar fractions of garlic contained appreciable antioxidant potential. The recorded values for TPC and flavonoid varied from 5 to 98.50mg/100g GAE and 4.16 to 6.91ug/QE, respectively. Moreover, the tested garlic extracts exhibited strong free radical scavenging activity ranged from 36 to 84.70%. They observed that the presence of different bioactive constituents like allicin and diallyl sulfide showed strong antioxidant activity by quenching free radicals, electron donation and metal chelating.

The observations of (Lu*et al.* 2011) confirmed the current findings for garlic antioxidant potential. They used different garlic cultivars for assessing their antioxidant capacity through FRAP, DPPH, TEAC and TPC assay. The observed values for the respective traits were 7.62 to 10.83 µmolTrolox/g, 7.60 to 9.47 µmolTrolox/g, 57.86 to 60.35 µmolTrolox/g and 15.61 to 17.35 mg gallic acid/g on fresh weight basis. They ascribed these variations due to varietal differences as diverse cultivation induce different amount of organosulfur compounds withpotent antioxidant activity.

The variations in the antioxidant potential of pink and white garlic in current study may be due to the difference between their genetic makeup, producing more organosulfur compounds in pink garlic as compared to white garlic. The work of Temitope*et al.* (2010) elucidated that the total phenolic contents of garlic ranged from 8.29 to 14.5 mg/g GAE, affected by garlic variety, extraction medium and method.

Table 5: FRAP, β-carotene, ABTS and tannins contents of garlic

Treatments	FRAP (mg TE/g)	β-carotene (%)	ABTS (µmol TE/g)	Tannins (mg /100g)
T_1	9.47±0.47a	69.44±0.54a	60.85±3.63a	0.93±0.01
T_2	9.33±0.21a	67.36±1.12b	58.59±2.48b	0.93±0.03
T_3	8.52±0.23b	64.60±2.56c	58.14±1.87b	0.90±0.02
T_4	8.51±0.02b	64.50±3.15c	57.27±3.69c	0.85±0.01
T_5	8.47±0.14b	62.35±2.89d	57.11±1.47c	0.84±0.03
T_6	8.18±0.16b	61.50±1.89e	55.81±1.29d	0.81±0.02

T_1 = Garlic pink methanolic extract T_2 = Garlic pink ethanolic extract T_3 = Garlic pink water extract
T_4 = Garlic white methanolic extract T_5 =Garlic white ethanolic extract T_6 = Garlic white water extract

Wangcharoen and Morasuk(2007) probed the effect of heating on the antioxidant capacity of garlic extracts. Purposely, they formulated a model system comprised of four tests *i.e.*ABTS, DPPH, FRAP and TPC for the assessment of garlic antioxidant potential. The observed values for the respective assay were 5.17, 1.13 & 0.57 Trolox and 1.29 GAE. They inferred that the antioxidant activity of garlic was directly proportional to allicin, organosulfur volatile compounds and flavanoids. Likewise, Gorinstein*et al.*(2006) narrated 26.1µmol Trolox equivalent and 11.42 mg GAE/g for ABTS and TPC of garlic, respectively. Nonetheless, research work of Jastrzebski*et al.* (2007) exhibited lower amount of total phenolic *i.e.*49.3 mg GAE/100g. One of their peers, Wangcharoen and Morasuk (2007) tested garlic for antioxidant capacity through TPC, FRAP, ABTS and DPPH and reported values 0.41 GAE/g, 0.14 , 1.06 and 0.16 mg vitamin C equivalent/g.

Similarly, Leelarungrayub*et al.* (2006) delineated that the free radical scavenging activity of garlic varied from 43.02 to 65.32%. The variations in the values of ABTS, FRAP, TPC and DPPH are due to the adaptation of different procedures and reference standards however, there are conclusive evidences advocating strong antioxidant ability of garlic.

Extraction conditions require core attention owing to their effect on overall experimental efficiency and in case of garlic bioactive compounds extraction the aqueous methanol and ethanol performed better as compared to water. The higher antioxidant activity of methanolic extract in current experiment is in accordance with the work of (Park and Chin 2010),who noticed higher TPC, DPPH, ABTS and FRAP values for methanolic extract than that of water. Similarly, Strail*et al.* (2006) examined the antioxidant activity of methanolic, ethanolic and water extracts of garlic. The results showed higher antioxidant activity of methanolic extract than rest of the extracts. They deduced that the polarity of the solvents was the primary factor for optimum recovery of phenolics. Different groups of scientists including (Bozin*et al.* 2008) and (Frankel and Meyer 2000) elucidated that the solvent to material ratio and polarity of solvent were the considerate factors for maximum phenolic recovery. In this context, methanol with 1:1 ratio with water was most effective than ethanol and water.

The results regarding total antioxidant activity (β-carotene) are in harmony with the research work of (Queiroz*et al.* 2009),who conducted a study to evaluate the antioxidant activity of garlic and its products like fresh garlic, chopped with salt, chopped without salt and fried garlic. They conducted DPPH and β-carotene activity and observed that the fried garlic exhibited the highest values for respective tests as 70.05 and 66.36%. Mechanistically they described that higher antioxidant activity of the garlic was due to the presence of polyphenols and organosulfur compounds.

In the nutshell, garlic is one of the rich sources for efficient antioxidants recovery thereby providing protection against various infirmities. Among the different solvents, methanol exhibited the highest yield due to its polarity differences. The garlic showed higher antioxidant activity owing to the presence of allicin, S-allylcysteine, diallylsulfide and diallyldisulfide that quench free radicals, chelate metal ions and prevent lipid peroxidation. The functional properties of garlic proved its therapeutic worth against life threatening maladies.

Allicinquantification: Mean squares showed significant effect of solvents on allicin content of different extracts. Means for the effect of solvents (Table 6) elucidated maximum allicin (4.63±0.02mg/g) in methanolic extract followed by ethanolic extract (2.32±0.01mg/g) whereas, water extract showed minimum value (0.966±0.003mg/g) in pink garlic. However, white garlic extracts presented highest allicin in ethanolic extract 0.961±0.002mg/g

followed by methanolic 0.918±0.002mg/g whilst water extract exhibited least value 0.774±0.001mg/g.

Table 6: Quantification of allicin in different garlic extracts

Treatments	Concentration (mg/g)
T_1	4.63±0.02
T_2	2.32±0.01
T_3	0.966±0.003
T_4	0.961±0.002
T_5	0.918±0.002
T_6	0.774±0.001

The results of present research are in harmony with the earlier work of (Baghalian*et al.* 2005), scrutinized the allicin contents of different Iranian garlic through HPLC. The results indicated that allicin in different cultivars varied from 17.5 to 134.5mg/g. The differences in the allicin values were due to origin, agronomic practices and genetic make up. Likewise, (Arzanlou and Bohlooli 2010) evaluated allicin value in aqueous extracts from different green garlic parts like leaf, shoot and young bulbs. The highest allicin contents were noticed in whole green garlic 0.48 mg/mL trailed by shoot and leaf extracts 0.44 and 0.26 mg/mL, respectively.

The effect of solvents on the allicin contents is evident from the findings of Li and Shi-ying (2007); noticed varying content of allicin using different solvents. The methanol, ethanol and ethyl acetate were performed better in comparison with water. The core reason was the polarity differences among the solvents.

Baghalian*et al.* (2006) conducted a trial to evaluate the effect of genetic differences, morphological traits and agro-climatic conditions on 24 garlic cultivars. The recorded allicin contents ranged from 3.84 to 18.25 mg/g in all tested samples. The results of instant study are in corroboration with the view of British Pharmacopoeia (1998) that a good garlic variety should provide 4.5 mg/g allicin.

The variations in the pink and white garlic are supported by the work of Schulz *et al.* (1998) and Baghalian*et al.* (2005), they observed variations in allicin contents owing to the source and genetic variations.

Conclusions: Garlic contains array of phytochemicals that play an important role in improving human health and disease prevention. Health promoting aspects of garlic are mainly accredited to its sulfur containing compounds mainly allicin and S-allayl-L-cysteine. For the intention, garlic samples were subjected to physico-chemical characterization, allicin quantification and antioxidant potential. For the extraction of bioactive molecules different solvents *i.e.* aqueous methanol, aqueous ethanol and water were used at constant temperature and time. Regarding bioactive molecules extraction, methanol exhibited highest polyphenols than that of ethanol and water. The pink garlic extracts showed higher FRAP, DPPH, flavonoids, flavonols, ABTS and tannins values in comparison with garlic white. Moreover, amount of allicin was also higher in pink garlic.

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