

ESTIMATION OF TOTAL PHENOLIC CONTENT IN THREE VARIETIES OF ALLIUM CEPA L. AND ALLIUM SATIVUM L.

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Abstract : The present study was conducted to estimate and compare the total phenolic content in three varieties of *Allium cepa* L. and *Allium sativum* L. The materials under study include *Allium cepa* (Red onion), *Allium cepa* var. *aggregatum* (shallot), *Allium cepa* (white onion) and *Allium sativum* (garlic) in the family Liliaceae under monocotyledons. The amount of total phenolic content were calculated using Folin - Ciocalteu reagent at 765 nm wavelength in three different solvents and were compared.

IndexTerms – *Allium cepa* L., *Allium sativum* L.

I. INTRODUCTION

The present study investigates the presence of total phenolic content in three onion varieties and garlic. Onions are one of the edible species of large genus *Allium* consisting of more than 700 species (Burnie et al., 1999). The Genus *Allium* is very large and it possess so many wild edible species. It is widely distributed over temperate zones in the northern hemisphere (Hanelt, 1990; Gautam et al, 1997). The place of origin is assumed to be in central Asia and the Mediterranean regions are considered to be the secondary centres of origin. Majority species of onion grow in open sunny and dry land mainly in humid climates. Among the edible *Allium*, the onion (*Allium cepa* L.) stands in the first rank, in the warm-temperature hills of eastern Nepal, followed by garlic (*Allium sativum*) and shallot (*Allium cepa* *Aggregatum* group) (Gautam et. al.,1997). Liliaceae (commonly called lily family) comprises 250 genera and approximately 600 species of flowering plants with many of them been used as spices and vegetables. In India it is represented by 169 species. Onions, shallots and garlic come from the same genus *Allium*, and are commonly used as vegetables. Among them onions are most popular of these crops. These are a good source of vitamin B1. The onion (*Allium cepa*) plants are generally perennial herbs strong smelling when crushed (Anonymous 1999). The roots are adventitious fibrous (Rajitkar, 2003). Onions not only provide flavour but also provide health promoting phytochemicals. These are natural compounds found in onion which possess the capacity to promote health benefits in humans and offer protection from a variety of diseases. The organosulphur compounds show antimicrobial, antiallergenic, anti-inflammatory and antithrombotic properties (Block, 1997). The flavanols in onion like quercetin and kaempferol also possess different crucial biological roles for health maintenance like antiviral, antimicrobial anti-inflammatory, and anticancer activity, along with protection of the heart and brain (Alexander, 2006; Harwood et al.,2007; Utesch et al., 2008; Ansari et al., 2009). Garlic (*Allium sativum*) is generally used to flavour food. It is highly medicinal. The varieties differ in size and pungency. It is one of the best known herbs around the world. It is generally agreed that garlic evolved from the wild garlic *Allium longicuspis*. A single bulb of garlic possess an average of 15 to 20 cloves. It possesses more shelf life compared to onions. It is used for both culinary and medicinal purposes. Garlic repels vampires, repel mosquitoes, used to treat cold, cough and high blood pressure, fungal infections, cholesterol. Shallots (*Allium cepa* var. *aggregatum*) are very much similar to onions. A single plant of shallot produces a bunch of bulbs whereas the onions always occur as a single one. Onions and shallots possess sulphur-containing compounds, which makes eyes wet. It is also responsible for the strong odour. The underground stem modified into tunicate bulb consisting of reduced stem and axillary buds surrounded by inner fleshy scale leaves and outer membranous dry scales (Ranjitkar, 2003). Bulbs are uniform in shape, size and colour. Shapes ranges from spherical to nearly cylindrical and include flat and cone like bulbs. The bulb scale is the organ, which is responsible for the organ which is responsible for the food value in onion. Phenol or phenolic acid is an aromatic organic compound with the molecular formula C₆H₅OH. It is a white crystalline and volatile compound. Phenols are a class of compounds containing a hydroxyl group bonded to an aromatic hydrocarbon group. Phenols are chemically hydrobenzene. Phenols are estimated using Folin – Ciocalteu method. The Folin–Ciocalteu reagent (FCR) or Folin's phenol reagent or Folin–Denis reagent, also called the gallic acid equivalence method (GAE), is a mixture of phosphomolybdate and phosphotungstate used for the colorimetric in vitro assay of phenolic and polyphenolic antioxidants (Singleton, 1999). It is named after Otto Folin, Vintilă Ciocâlțeu, and Willey Glover Denis. The reagent does not measure only phenols, but will react with any reducing substance. It therefore measures the total reducing capacity of a sample, not just phenolic compounds. This reagent is part of the Lowry protein assay, and will also react with some nitrogen-containing compounds.

Extraction of phenolic compounds and evaluation of the antioxidant and antimicrobial capacity of red onion skin (*Allium cepa* L.) was done by Viera, (2017). This study evaluated the content of phenolic compounds, total flavonoids and anthocyanins, as well as the antioxidant and antimicrobial activity, of red onion skin extract (*Allium cepa* L.) that was obtained by conventional

extraction at different times (30, 60, 120 and 240 minutes) and using different concentrations of ethanol (20, 40, 60 and 80%). The antioxidant activity of the extracts was evaluated using the following methods: DPPH radical sequestration, the inhibition of the auto-oxidation of the β -carotene system, FRAP and IC50. The antimicrobial activity was evaluated by the disk diffusion method. The best results for phenolics, flavonoids and anthocyanins were found using extraction with 80% solvent at 165, 60 and 120 minutes respectively. In relation to the antioxidant activity using the FRAP, DPPH and IC50 methods, the best values were found using 80% solvent at 120, 30 and 30 minutes of extraction respectively. The best value for antioxidant activity determined by the β -carotene method was with 59.32% solvent at 240 minutes of extraction. No antimicrobial activity of the extracts in relation to the tested microorganisms was detected. The results of this study showed that the concentration of 80% ethanol favoured the extraction of phenolic compounds, flavonoids and total anthocyanins, as well as producing the highest antioxidant activities found by the different methods. The best extracts in terms of antioxidant activity by the DPPH and IC50 methods were obtained with the lowest extraction time of 30 minutes, in contrast to the extraction time of 240 minutes, which produced the highest antioxidant activity by the β -carotene methods.

Comparison of Phenolic Content and Antioxidant Capacity of Red and Yellow Onions, were done by Anwei Cheng (2013). The total polyphenol (TP), flavonoid, proanthocyanidin (PAC) content, and antioxidant capacity of both onion varieties (red and yellow) were compared. The content of TP, flavonoids, and PAC was determined by Folin-Ciocalteu colorimetric method, AlCl₃, and by DMAC colorimetric method, respectively. The results showed that the contents of TP and flavonoids decreased from the outer to the inner layers in both onions, but there was no significant difference in PAC content. The outer layers had the highest antioxidant activity of extracts followed by a continuous decrease towards the inner layers in both varieties. The contents of phenolic acids and flavonoids were quantified by HPLC. Gallic acid, ferulic acid, and quercetin, as the main compounds in polyphenols, were detected in each layer of both onions. The red variety showed better antioxidant activity than yellow onion according to the linoleic acid system and DPPH assay. The higher contents of TP and flavonoids were associated with higher antioxidant activity.

Determination of Total Phenolic Content and Antioxidant Activity of Garlic (*Allium sativum*) and Elephant Garlic (*Allium ampeloprasum*) by Attenuated Total Reflectance–Fourier Transformed Infrared Spectroscopy, were done by Lu (2011). The total phenolic contents and antioxidant activities of garlics from California, Oregon, Washington, and New York were determined by Fourier transform infrared (FT-IR) spectroscopy (400–4000 cm⁻¹). The total phenolic content was quantified [Folin–Ciocalteu assay (FC)] and three antioxidant activity assays, 2,2-diphenyl-picrylhydrazyl (DPPH) assay, Trolox equivalent antioxidant capacity (TEAC) assay, and ferric reducing antioxidant power (FRAP), were employed for reference measurements. Four independent partial least-squares regression (PLSR) models were constructed with spectra from 25 extracts and their corresponding FC, DPPH, TEAC, and FRAP with values for 20 additional extracts predicted ($R > 0.95$). The standard errors of calibration and standard error of cross-validation were <1.45 (TEAC), 0.36 (FRAP), and 0.33 μ mol Trolox/g FW (DPPH) and 0.55mg gallic acid/g FW (FC). Cluster and dendrogram analyses could segregate garlic grown at different locations. Hydroxyl and phenolic functional groups most closely correlated with garlic antioxidant activity.

MATERIALS AND METHOD

The present work deals with the determination of phenolic contents in three vary of onion and garlic. Onion red (*Allium cepa* and a variety from Japan), small onion – shallot (Varieties of *Allium cepa* var. *aggregatum*) and garlic (*Allium sativum*) were collected from authorized vegetable shops of Ernakulam District during the months of December 2017 to April 2018 and were subjected to spectrophotometric studies.

ESTIMATION OF TOTAL PHENOLIC CONTENT

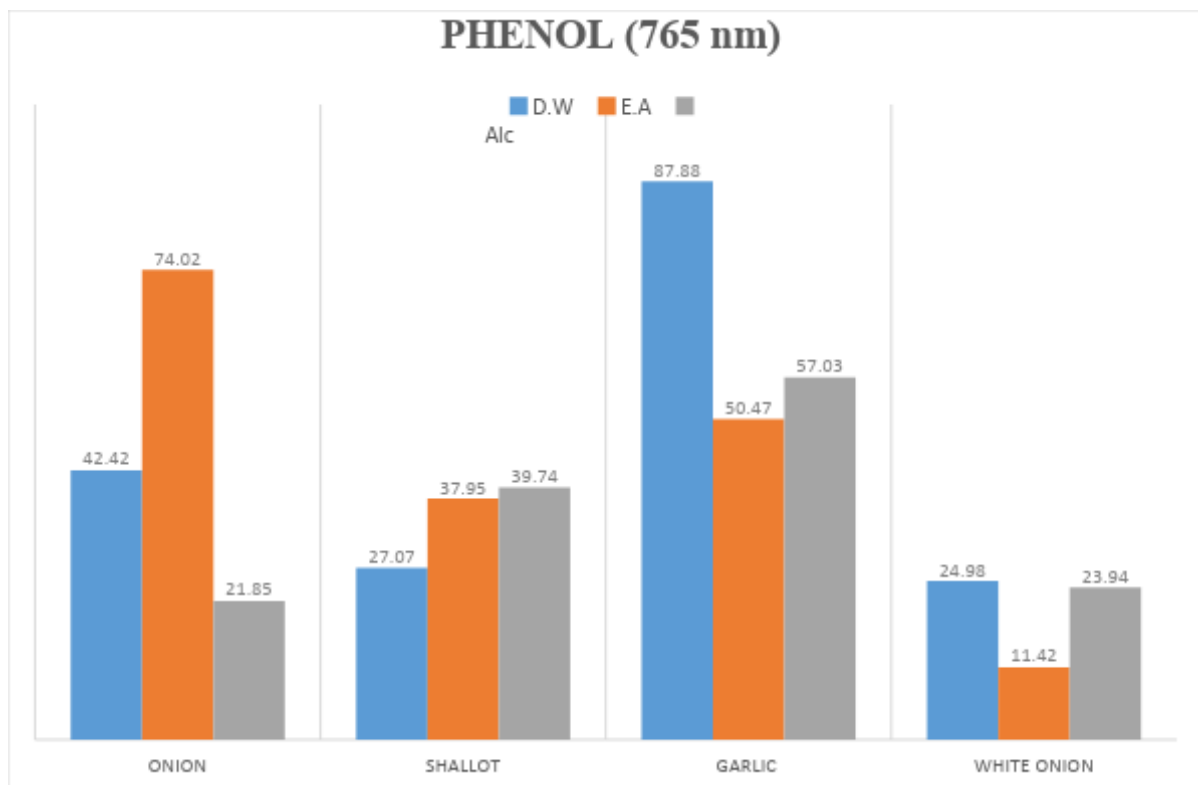
The amount of phenol was determined using Folin -Ciocalteu reagent at 765 nm wavelength in 3 different solvents (Distilled water, Alcohol and Ethyl acetate).

A volume of 0.5 ml of the plant extract (100 μ g/ml) was mixed with 2 ml of Folin-Ciocalteu reagent (diluted 1:10) were neutralized with 4 ml of sodium carbonate solution (7.5% w/v) The reaction mixture was incubated at room temperature for 30 minutes with intermittent shaking for colour development. The absorbance of the resulting blue colour was measured at 765 nm using spectrophotometer.

The total phenolic contents of the samples using Folin – Ciocalteu reagent are expressed in terms of Gallic acid equivalent. The reagent which is a mixture of phosphotungstic acid and phosphomolybdic acid which after oxidation of phenol, is reduced to a mixture of blue oxides of tungsten and molybdenum. The blue coloration produced has a maximum absorption in the region of 765nm and is proportional to the total quantity of phenolic compounds.

RESULTS

Fig 1: ESTIMATION OF TOTAL PHENOLIC CONTENT



DISCUSSIONS AND CONCLUSION

The amount of phenol was determined for all the samples using Folin - Ciocalteu reagent at 765nm wavelength in 3 different solvents (Distilled water, Alcohol and Ethyl acetate). The amount of phenol can be correlated to its antioxidant property. From the study it can be observed that garlic showed high amount of phenol among all the samples under study and lowest in white onion. Among the three solvents, distilled water and ethyl acetate showed high amount of phenol in garlic and lowest in white onion. In alcohol, garlic showed high amount of phenol but a low value was obtained in onion. The highest amount of phenol in onion was observed in ethyl acetate. The highest amount of phenol in White onion was observed in distilled water. The highest amount of phenol in garlic was observed in distilled water whereas the highest amount of phenol in Shallot was observed in alcohol.

In the present study, different extracting solvents were used and it was found out that they differed considerably in their capability to extract antioxidants. Conventionally, solvents such as methanol, ethanol, and acetone have been routinely used to extract phenolic/antioxidant compounds from fresh plants at different concentrations, which can allow them to diffuse without any trouble into the pores of the plant materials to leach out the active constituents. From the toxicological point of view water is considered to be safer than methanol. However, in general, the least polar solvents are considered to be most suitable for the extraction of antioxidant compounds, unless very high pressure is employed.

The non-lethal concentration of phenol provide enormous beneficial properties to human health. Sufficient amount of phenol is non-toxic to health and provide protection against various ailments, such as they act as cancer chemopreventives, antioxidants and also helps to promote healthy aging by minimizing DNA damage. The consumption of phenol rich food have resulted in an increase in total antioxidant capacity in the blood plasma of people. However, exposure to higher concentration of phenol may exhibit roles in genotoxicity, thyroid toxicity, respiratory disorders and pneumonia.

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