The Phytochemical Analysis and Amino Acid Profile of Allium *CEPA* (Onions) Extracts

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Abstract— *It has been established that plants which naturally synthesize and accumulate some secondary metabolites such as alkaloids, glycosides, tannins, volatile oil, minerals and vitamins have medicinal properties. Then phytochemical and amino acid profile of allium cepa extracts was carried out.*

The proximate values of the nutrients in 100g of raw onion were analyzed using qualitative and quantitative method. Amino acid profile of the onion extracts was done by extraction, digestion, distillation, hydrolysis, titration, and evaporation techniques and finally analysed using Technicon Seqential Multisample analyser (DNA model). The standard quantitative method of analysis was used to determine the phytochemicals (toxicants) in the onion extracts while qualitative method was used for the screening.. The result of proximate compositions proved that onions contain Water (86.5%), carbohydrate (11.1%), Protein (1.3%), Fats and oil (0.2%) and Ash (0.5%). The Amino acid profile revealed that Arginine (3.74g/100g) and Histidine (1.21g/100g) were present in the extracts. Others include; Isoleucine (2.22g/100g), Leucine (4.11g/100g), Lysine (3.60g/100g) Methionine (0.86g/100g), Phenylalanine (2.29g/100g), Threonine (1.25g/100g) and valine (2.01g/100g), in 100g of protein. There was a change in the values of amino acid profiles of the aqueous extract when compared with that of solvent extracts. The results of the phytochemical screening showed that; flavonoid, terpernoids, carbohydrate, cardiac glycosides, saponins, anthraquinone, phlobatannins, alkaloid and polyphenol were present. The amounts of toxicants in 100g raw onion were: Alkaloid (13.0%), Tannic acid (0.03g), Oxalate (1.15g), Cyanogenic glycosides (2.91mg) and phytic acid (6.05mg). likewise the solvent extracts. But at alpha – level ($p \geq 0.05$), there was no significant difference ($p = 0.222$) between the effects of aqueous and solvent extracts. The research has shown that; onions widely used as food, could serve as natural medicine, flavour enhancer and antimicrobial agent in food and pharmaceutical industries to improve Nigeria's ailing economy.

Keywords— Allium cepa, Amino acid, phytochemical screening.

I. INTRODUCTION

The history of herbal medicine dated back to 2800BC (Gerber, 2010). Medicinal herbs contain substances that can be used for therapeutic purposes and synthesis of useful drugs. Meanwhile, plants that possess therapeutic properties or exert beneficial pharmacological effects on the body of animals are generally termed "Medicinal plants" (Tapsell *et al*., 2006; Giovannimi *et al*., 2016). They have no specific morphological characteristic but possess some special qualities that make them medically important. It has been established that plants which naturally synthesize and accumulate some secondary metabolites such as alkaloids, glycosides, tannins, volatile oil, minerals and vitamins have medicinal properties (Edeoga and Erratodo, 2000; Benkeblia, 2004).

Allium cepa (Onions) is one of the edible species of a large genus (*Allium*) consisting of more than 700 species (Burnie *et al*., 1999). Among the edible *Allium*, the onion (*Allium cepa*) ranks first in the warm-temperate hills of eastern Nepal, followed by garlic (*Allium sativum*) and shallot (*Allium cepa* Aggregatum group) (Gautam *et al.*, 1997). *Allium cepa* belongs to the family of *Liliaceae.* It is also known as 'garden onion'or 'bulb'onion (Indu, 2017). It is one of the oldest cultivated

vegetables in history. *Allium cepa* is a tunicated bulb, round, and oblong in shape, invested with a shining, thin, dry membrane, of a reddish or white colour (Azu *et al*., 2007). It contains Carbohydrate, Protein, Fats, minerals and Vitamins (Vamshi et *al.,* 2010). Onion contains amino acids, which are very important elements for nearly all physiological functions. The "essential" amino acids are called "*essential"* because our bodies can't produce them; therefore, it's *essential* that they are included in our daily diet (Amino acid, 2017).

Onions are effective against common cold, heart disease, diabetes, osteoporosis (Wetli *et al.*, 2004), cough and sore throat (Ye, *et al*., 2013; Augusti, 1996). This could be due to certain chemical compounds believed to have anti; inflammatory, anti; cholesterol, anticancer and antioxidant properties present in onions (Ye, *et al*., 2013; Slimestad *et al.*, 2007).

Major active antibacterial components of onion in-vivo are the allicin derived organo-sulphur compounds (Tsao and Yin, 2001).

Onion has been used in many societies against bacterial and fungal infections in folk medicine. It could be used as natural preservative in foods to control microbial growth due to the antimicrobial activities onion possesses (Pszczola, 2002). Different Onions extracts differ in their antimicrobial activities, and micro-organisms vary in their responsiveness to the bioactive compounds of the extracts (Gorinstain *et al.,* 2010). The development of antibiotic resistance has become a global public health challenge which is causing ineffectiveness of antibacterial agents leading to increase in rate of morbidity and mortality. Therefore this research intends to assess the phytochemicals and amino acids present in *allium cepa* (onions*)* extracts to justify the claims on its use in folk medicine.

II. MATERIALS AND METHODS

2.1 Collection of Plant Material

The Onion bulbs (*Allium cepa*) were obtained from a Fruit and Vegetable Market in Owerri and transported to the applied microbiology laboratory of Nnamdi Azikiwe University Awka for analysis according to W. H. O. (2003) guidelines on good agricultural and collection practices (GACP) for medicinal plants.

2.2 Cold Water Extraction

By using Hander, (2005) method of extraction, the moisture of the onion bulb was removed by drying sliced onions inside an air-oven at 40^0C.

It was pulverized using electronic blender, and ten (10.0g) of the pulverized sample dissolved in 100mls distilled water.

It was sterilized at 121^0C for 15 minutes, left to soak for 2 days after which it was filtered aseptically, concentrated and stored at 4^oC for analysis.

2.3 Hot Water Extraction

By using Hander, (2005) method of extraction, the moisture content of onions was removed by drying sliced onions in oven at 40^0C. The sample was pulverized using electronic blender, mixed with sterile distilled water in a ratio of 1:20 (100 g in 1 L solvent).
It was stirred and heated for 4 hours.

After cooling, the extract was filtered aseptically, concentrated and stored at 4^oC for analysis.

2.4 Soxhlet Extraction

The peeled, sliced onions were dried in an oven at 40^oC, and then pulverized using electronic blender to a fine powder, stored at 4^oC for use.

By adopting Hander, (2005) method of extraction. The onion sample was extracted using ethanol solvent at a solute – solvent ratio of 1:10 for 6 hours in a Soxhlet extractor.

The Crude extract was kept in a sample bottle and stored at 4^oC prior to analysis.

The process was also repeated for petroleum ether solvent.

2.5 Proximate Compositions of the Onions

The Nutrient composition of the aqueous extracst were determined using the modified method described by Onwuka, (2005).

2.6 Amino Acid Profile

The Amino acid profile of Onions extracts was determined by using the methods described by Benitez (1989), Hander, (2005) and AOAC (2006).

2.7 Quantitative Determination of Phytochemicals

The phytochemicals were quantitatively determined by adopting Olajide *et al.*, (2011) and Onwuka (2005) methods of analysis.

2.8 Phytochemical Screening

The phytochemical screening of the extracts was done using standard qualitative procedure according to Harbone (1995).

III. RESULTS AND DISCUSSIONS

The results of the Proximate analysis of the onion, Amino acid profile of the onions extracts, quantitative analysis of the toxicants, Phytochemical Screening of the Onions extracts were presented in the Figures and Tables below.

TABLE 1
THE PROXIMATE COMPOSITIONS OF ONIONS EXTRACTS PER 100G OF EDIBLE PORTION

Food Nutrients	Percentage Compositions
Carbohydrate	11.1%
Moisture	86.5%
Protein	1.3%
Fat	0.2%
Ash	0.5%
Crude Fibre	0.4%

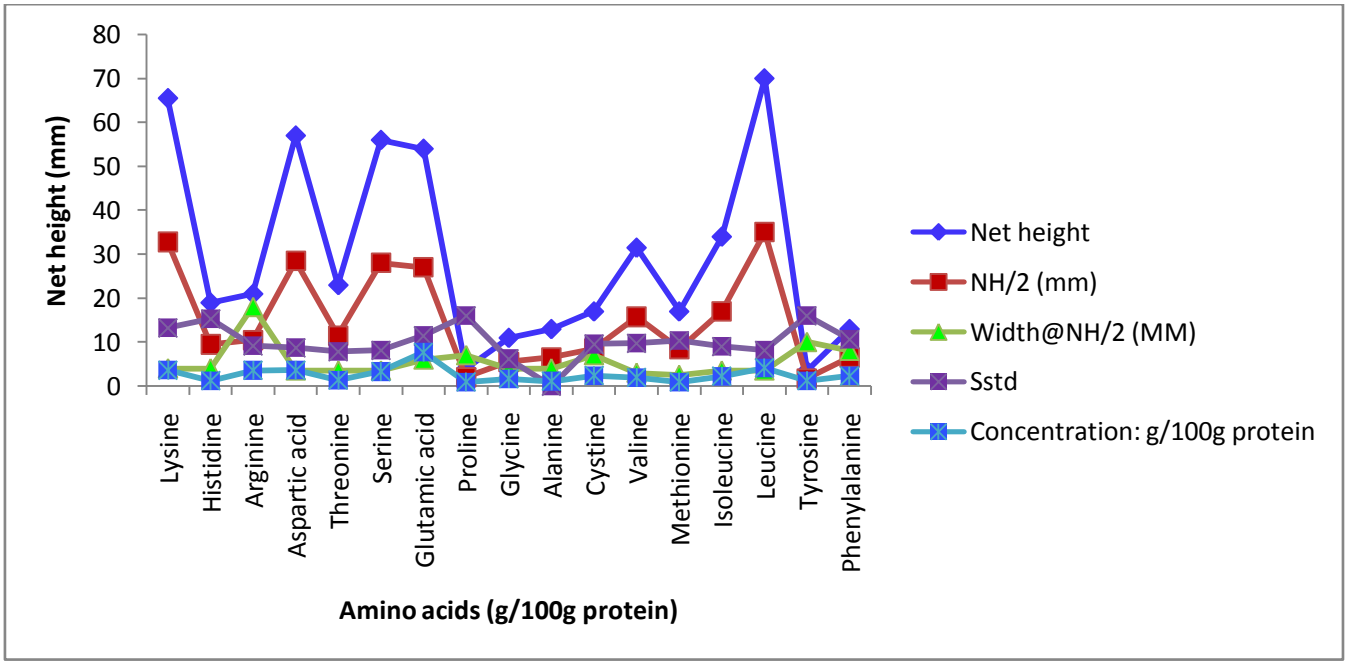

FIGURE 1: The amino acid profiles of *Allium cepa* (onion) cold water extract

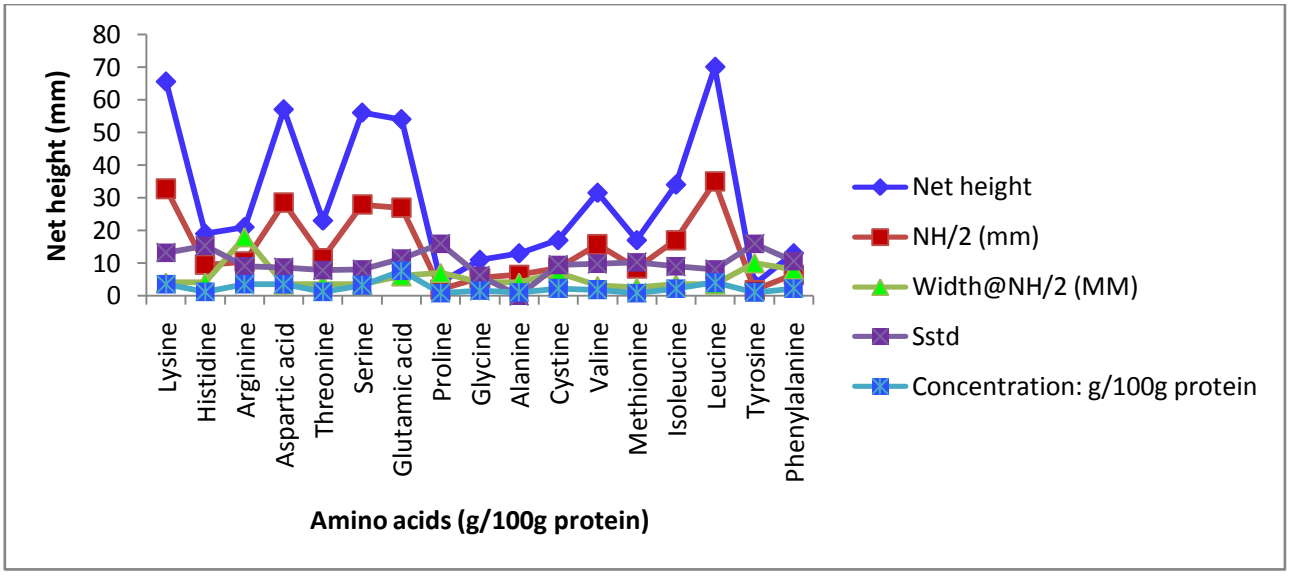

FIGURE 2: The amino acid profiles of *Allium cepa* (onion) hot water extract

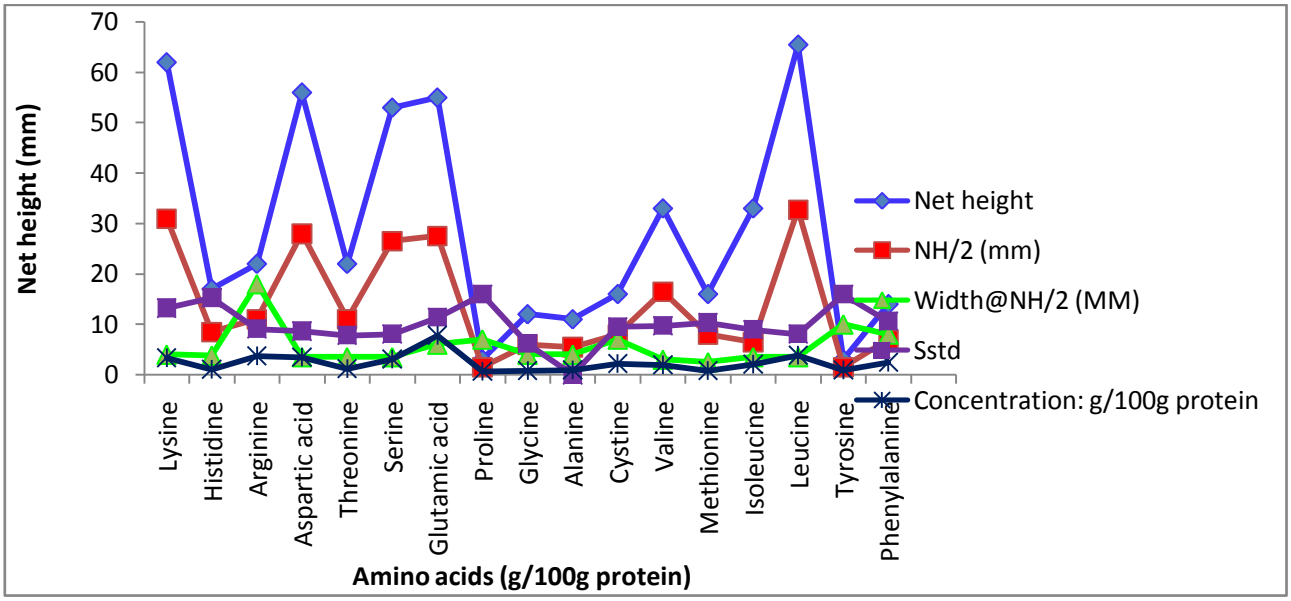

FIGURE 3: The amino acid profiles of *Allium cepa* (onion) ethanolic extract

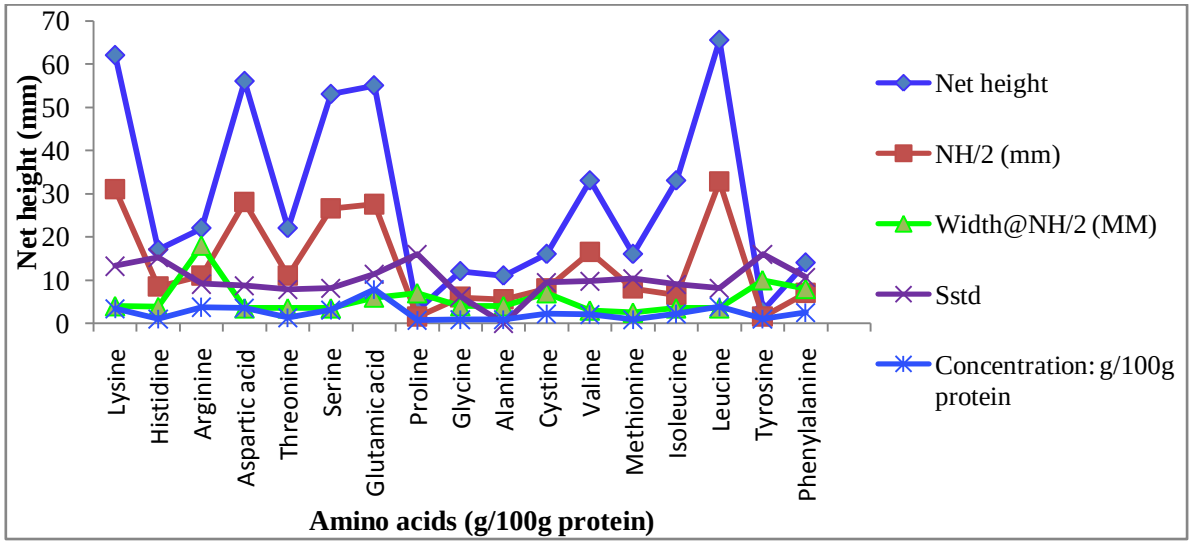

FIGURE 4: The amino acid profiles of *Allium cepa* (onion) Petroleum ether extract

TABLE 2
THE TOXICANTS (PHYTOCHEMICALS) IN ONIONS

Oxalate (g/100g)	Tannin (g/100g)	Phytic acid (mg/100g)	% Alkaloids	Cyanogenic glycosides (mg/100g)	Sample
1.78	0.06	7.33	15.11	3.79	A
1.27	0.05	7.01	15.32	3.51	B
1.15	0.03	6.05	13.03	2.91	C
1.20	0.04	6.08	13.06	3.01	D

Key: A=Ethanolic extract, B=Pet. Ether extract, C=Cold Water Extract, D= Hot water extract

TABLE 3
PHYTOCHEMICALS SCREENED IN *ALLIUM CEPA* (ONION) EXTRACTS

Class of Compounds	Cold Water Extract	Hot Water Extract	Ethanolic Extract	Pet. Ether Extract
Alkaloids	-	-	+	+
Flavonoids	+++	+++	+++	+++
Tannins	+	+	+	+
Saponin	+	+	+	+
Terpenoid	++	++	+	+
Glycosides	++	++	+	+
Steroids	+	++	+	+
Phlobatannin	+	++	+	+
Polyphenols	++	+++	++	++
Anthraquinone	-	-	+	+
Anthranoid	-	-	-	+
Glucose	++	++	++	++

Key: + = Presence; - = Absence

IV. DISCUSSIONS

The presence of fibre as shown in the proximate compositions (Table 1.0) which are non digestible ingredients fermented by bifidobacteria help in maintaining the health of the gut and colon. Table 1.0, also shows that Onion contains some minerals elements and vitamin C, and this makes it important in human nutrition (FAO, 2011; Robertson *et al.*, 1991).

The results of the amino acid profiles of the onions extracts (Figure 1.0 to 4.0), shows the presence of Histidine, Isoleucine, Leucine, Lysine, Methionine, Phenylalanine, Threonine and valine as the essential amino acids (Anonymous, 2007), Tryptophan was destroyed by 6N HCl during processing of the sample (hydrolysis). Arginine is regarded as an essential and non essential amino acid. Other non essential amino acids present in the onion extracts include: aspartic acid, serine, glutamic acid, proline, glycine, alanine, cystine and tyrosine (Anonymous, 2007).

Histidine is unique because it is both essential and non essential amino acid. The bodies of living organisms needs histidine to develope and maintain healthy tissues, especially myeline sheath of nervous cells to ensure the transmission of messages from the brain to organs throughout the body (Anonymous, 2007). Isoleucine is another branched-chain amino acid that cannot be produced in the body and must be obtained from the food we eat. It is essential for proper blood clotting and muscle repair. This implies that the result of the amino acid profiles of this research has been in agreement with the *antithrombotic effects of onion* (Bordia *et al.*, 1996).

The symptoms of isoleucine deficiency include; dizziness, fatique, headaches, confusion and depression. Isoleucine plays important role in the transport of oxygen from the lungs to the various parts of the body and the production of hemoglobin, which contains iron. Isoleucine is also important for the efficient metabolism of glucose and boosts the body's energy levels. The result of this research is therefore in line with the *antidiabetic effects of onion* (El-Demerdash, *et al*., 2005). When given orally, isoleucine reduces the level of sugar in the blood by 20 percent and increases sugar absorption in the muscles by 71 percent without actually increasing the level of insulin in the blood (Anonymous, 2007).

Lysine is a key component in hormones and enzymes and plays a vital role in the production of collagen, a substance that is critical in bone, muscle, cartilage, and skin formation. Lysine plays a key role in calcium absorption by reducing the amount of calcium being excreted in the urine. It promotes the growth of hair, nails, teeth, and bones. The result of the amino acid, Lysine therefore supports the work of Muhlbauer *et al.,* (2002), that onions have *bone strengthening properties*.

Phenylalanine present in the onion extracts (Figure 1.0 to 4.0) is regarded as a forerunner of tyrosine, the anti-depressant dopamine, norepinephrine, epinephrine and the skin pigment, melanin. Eating foods rich in phenylalanine will help to prevent mood swings which help one out of lethargy, sluggishness, feeling of low morale and anxiety (Anonymous, 2017).

The amino acid, Threonine found in the onion extracts is important in the formation of bones, cartilage, hair, teeth and nails. It is also responsible for the growth and development of liver muscles, skeletal muscles and the small intestines. Threonines also help in the production of antibodies to strengthen the immune system. It is mostly found in the nervous system and help to fight depression. It helps speed the healing of wounds and recovery from injury by helping build strong bones. These showed that the results of amino acid profile of the onion extracts have been in agreement with the anti-inflammatory and bone strengthening properties of onion (Sparg *et al.,* 2004; Matheson *et al.,* 2009). There were differences in the values of amino acid profiles of the aqueous extract and that of the solvent extracts.

The result of other phytochemicals in Table 3.0 shows that Flavonoids and Organosulfur compounds are the two major classes of secondary metabolites in Onion. Alkaloid was absent in the aqueous extract of the Onion sample. This could be due to the insolubility of alkaloid in water.

The major flavonoid found in onion is quercetin, present in conjugated form, as quercetin 40-O-*β*-glycopyranoside, quercetin 3,40-O-*β*-diglycopyranoside, and quercetin 3,7,40-O-*β*- triglycopyranoside (Sellappan and Akoh, 2002; Koncic and Jug, 2011). The quercetrin, act as anti-bowel disease (Yoshida *et al.,* 1998a), enhances immune system activity like cold and fevers, mouth and pharynx inflammation, infections, bronchitis/cough, hypertension, dyspepsia and arteriosclerosis (Dorant *et al.,* 1996). The extract could be applied on wounds and stings, on the skin, to remove warts and even unwanted skin blemishes (Lai and Roy, 2004). This ability could be due to the presence of flavonoid and other organosulfur compounds in Onion which act to scavenge oxidizing agents that inhibit the oxidation of fatty acids, thereby preventing the formation of pro-inflammatory messengers and also inhibit bacterial growth (Zohri *et al.,* 1995; Kim, 1997).

Phenolic compounds present in the onion extract (Table 3.0) are potentially toxic to the growth and development of pathogens (Edeoga *et al.,* 2000). The oxidation inhibiting activity of tannins in onions could be attributed to their Gallic and digallic acids (Edeoga *et al.,* 2000).

Saponins present in the extract (Table 3.0), justify the use of onion in treatment of gastrointestinal tract disturbances by traditional healers (Sampath *et al.,* 2010). Saponins also have anti-inflammatory activity and this could account for the application of onion extract on wounds and stings on the skin by traditional medicine practitioners (Sparge *et al.,* 2004). It fights against cancer by helping the cell DNA to stop producing bad cells and make only good ones (Yuan, et al., 2008).

Steroids present in the onion extracts (Table 3.0), are used as starting materials in the synthesis of hormonal drugs, and this could support the reason for its use (Onion) to increase libido and strengthening of reproductive organs (Sampath *et al.,* 2010).

Onion contains toxicants (phytocemicals) although in small quantity as shown in Table 2.0, which implies that; individuals are not allowed to consume more than 10 bulbs (approximately105g) of raw onion per day due to presence of cyanogenic glycoside, phytic acids and Tannic acid in them to avoid mouth and body odour (Banerjee *et al.,* 2003).

V. CONCLUSION

This study has shown that the efficacy of the extracts might be due to presence of bioactive constituents in Onion. These bioactive molecules are potential sources of antimicrobial drugs that could tackle the problem of drugs resistance that is currently a challenge to our country's Food and Medical industries.

REFERENCES

[1] AOAC (Association of Official Analytical Chemists)(2006). Official Method of Analysis of the AOAC (W. Horwitz Editor) Eighteenth Edition. Washington D.C, pp. 899–911

[2] Augusti, K. (1996). Therapeutic values of onion and garlic. *Indian Journal of Experimental Biology* **34**:634–640.

[3] Banerjee, S. K., Mukherjee, P. K. andMaulik, S. K. (2003). Garlic as an antioxidant: The good, the bad and the ugly. *Phytotherapy. Research*. **17**:97–106.

[4] Ben´ıtez, V., Moll´a, E., Mar´ıa, A., Mart´ın-Cabrejas, M. A., Aguilera, Y., Francisco, J., L´opez-Andr´eu, F. J. and Esteban, R.M. (2011). Effect of sterilisation on dietary fibre and physicochemical properties of onion by-products. *Journal of Food Chemistry*. **127**:501–507.

[5] Benitez, L. V. (1989). Amino Acid and fatty acid profiles in aquaculture nutrition studies, p. 23- 35. In S.S. De Silva (ed.) Fish Nutrition Research in Asia. Proceedings of the Third Asian Fish Nutrition Network Meeting. Asian fish. Society Special Publication. **4**, p. 166.

[6] Bordia, T., Mohammed, N., Thomson, M. and Ali, M. (1996). An evaluation of garlic and onion as antithrombotic agents. *Prostaglandins Leukot Essential Fatty Acids*. **54**:183–186.

[7] Burnie, G., Forrester, S., Greig, D., Guest, S., Harmony, M., Hobley, S., Jackson, G., Lavarack, P., Melanie, L., Donald, R.M., Macoboy, S., Molynenx, B., Moodie, D., Moore, J., North, T., Newan, D., Pienaar, K., Purdy, G., Silk, J., Ryan, S., Schien, G. (1999). *Botanica:* The illustrated A-Z of over 10,000 garden plants, (3rd Ed.), Random House Australia pty Ltd, New South Wales, pp 74.

[8] Dorant, E., Van Den Brandt, P. and Goldbohm, R. A. (1996). A prospective cohort study on the relationship between onion and leek consumption, garlic supplement use and the risk of colorectal carcinoma in The Netherlands. *Carcinogenesis*. **17**:477–484.

[9] Edeoga, H.O., Erratado (2000). Alkaloid, Tannin, and Saponin Contents of some Nigeria Medical plants. *Journal of medicinal Aromatic plant science.* **2:** 1010-1015.

[10] El-Demerdash, F.M.,Yousef, M. I. and Abou El-Naga, N. I. (2005).Biochemical study on the hypoglycemic effects of onion and garlic in alloxan-induced diabetic rats. *Food and Chemical and Toxicology*. **43**:57–63.

[11] Food and Agricultural Organization (2011). Onions: Post-Harvest Operation. Food and Agriculture Organization of the United Nations. FAO Statistics. (2011). Productions, Crops. 1075.

[12] Gautam, S.R., Neupane, G., Baral, B.H., Rood, P.G. and Pun, L.(1997). Prospects of onion cultivation in the warm-temperate hills of eastern Nepal and its research and development strategies for commercial production. *ISHS* Acta Horticulturae. **433,** 83-94.

[13] Gerber, C., (2010). History of herbal medicine. Available from: **http:**//herbs.lovetoknow.com/History_Herbal Medicine.

[14] Giovannini, P., Howes, M.R., Edward, S.E. (2016)."Medicinal plants used in the traditional management of diabetes and its sequelae in central America: A review". *Journal of Ethnoparmacology*. **184:** 58-71.

[15] Gorinstain, S., Leontowicz, H., Leontowicz, M., Najman, K., Bieecki, W., Ham, K.-S., Kang, S.-G., Paredes-Lopez, O., Leticia Martinez-Ayala, A. and Trakhtenberg, S. (2010). Aorta and liver changes in rats fed cholesterolcontaining and raw vegetable-supplemented diets: Experiments in vitro and in vivo. *Journal of Agriculture and Food Chemistry.* **59**:7441–7451.

[16] Griffiths, G., Trueman, L., Crowther, T., Thomas, B. and Smith, B. (2002). Onions-a global benefit to health. *Phytotherapy Research*. **16**:603–615.

[17] Harbone, J.B (1995). Phytochemical methods, 1st Edition Chapman Halls, London. Pp 20-22.

[18] Hander, S.S., (2005). Tradition and modern method of extractions of essential oils from aromatic plants. Presentation at the training course on cultivation, post-harvesting and processing technologies of medicinal and aromatic plants in developing countries. ICS-UNIDO organized at Bomako, Mali (West Africa). In: extractions technologies of medicinal and aromatic plants. Pp. 22-23.

[19] IBM (2011). ® SPSS®Statistics. Chicago, Illionis: SPSS Inc: version 19.0

[20] Indu, M. (2017). Origin and History of Onions. *IOSR Journal of Humanities and Social Science (IOSR-JHSS*), vol.22, issue 9, verse 13, pp 7-10.

[21] Koncic, M. Z. and Jug, M. (2011). Antioxidant and bioadhesive properties of onions (*Allium cepa*) processed under acidic conditions. *International Journal of Food Properties*. **14**:92–101.

[22] Lai, P. K. and Roy, J. (2004). Antimicrobial and chemopreventive properties of herbs and spices. *Curr. Med. Chem.* **11**:1451–1460.

[23] Matheson, E. M., MAinous, A. G. and Carnemolla, M. A. (2009). The association between onion consumption and bone density in perimenopausal and postmenopausal non-Hispanic white women 50 years and older. *Menopause.* **16**:756–759.

[24] Muhlbauer, R. C., Lozano, A. and Reinli, A. (2002). Onion and a mixture of vegetables, salads, and herbs affect bone resorption in the rat by a mechanism independent of their base excess. *Journal of Bone Mineral Research.* **17**:1230–1236.

[25] Pszczola, D.E. (2002). Antimicrobials: setting up additional hurdles to ensure food safety. *Food and Technology*, **56,** 99-107.

[26] Olajide, R, Akinsoyinu, A.O, Babayemi,O.J, Omojola, A.B, Abu, A.O and Afolabi, K.D.(2011). Effect of processing on Energy values, Nutrient and Anti-nutrient components of wild Cocoyam. (*Colocasia esculenta*) corn.Pakistan. *Journal of Nutrition*, **10**(1):29-34.

[27] Onwuka, G.I (2005). Food analysis and instrumentation: food Toxicants, p. 128

[28] Robertson, J. M., Donner, A. P. and Trevithick, J. R. (1991). A possible role for vitamin C and E in cataract prevention. *American Journal of Clinical Nutrition.* **53**:346S–351S.

[29] Sampath, K.P., Debjit B., Chranjib, Biswajit and Pankaj, T. (2010). *Allium cepa*: A traditional medicinal herb and its health benefits, *Journal of Chemical and pharmaceutical Research*, **2** (1): 283-291.

[30] Santas, J., Almajano, M. P. and Carbo, R. (2010). Antimicrobial and antioxidant activity of crude onion (*Allium cepa*) extracts. *Int. Journal of Food Science and Technology.* **45**:403–409. 1375

[31] Saulis, A.S., Mogford, J.H., and Mustoe, T. A, (2002). Effect of mederma on hypertrophic scrring in the rabbit ear model, Plastic Reconstructive Surgery, **110**(1): 177 – 183.

[32] Sellappan, S. and Akoh, C. (2002). Flavonoids and antioxidant capacity of Georgia-grown Vidalia onions. *Journal of Agriculture and Food Chemistry.* **50**:5338–5342

[33] Slimestad, R., Fossen T., and Vagen I.M. (2007). Onion a source of unique dietary flavonoids. *Journal of Agriculture and Food Chemistry*. **55** (25): 10067-10080.

[34] Sparge, S. G., Light, M. E. and van Staden, J. (2004). Biological activities and distribution of plant saponins. *Journal of Ethnopharmacology*. **94**:219–243.

[35] Ross, I.A. (2001). Medicinal plants of the world: chemical constituents, Traditional and Modern Medicinal Uses, Human press, Totowa, Volume 2, pp 1-9.

[36] Tapsell, L.C., Hemphill, I., and Cobiac, L. (2006). "Health benefits of herbs and spices: the past, the present, the future". *Medical journal of Austra*. **185 (**4): 14-24.

[37] Tsao, S.M. and Yin, M.C. (2001). In-vitro antimicrobial activities of four diallyl sulphides occurring naturally in garlic and Chiness leek oils. *Journal of Medical Microbiology*,***50***, 646-649.

[38] Vamsh,i K., Sharath, N., Rao, K.N.V., David, B., Sandhya, S., Sudhakar, K., Saikumar, P., Sudha, P., and Chaitanya, R.K (2010). A comprehensive review on *Allium cepa. Journal of Addvanced Pharmaceutical Research **1**:2, 94 – 100*

[39] Wang, H. X. and Ng, T. B. (2001). Purification of allivin, a novel antifungal Protein from bulbs of the round-cloved Garlic. *Life Science*. **70**:357–365.

[40] Wang, T. J., Gona, P., Larson, M. G., Tofler, G. H., Levy, D., Newton-Cheh, C., Jacques, P. F., Rifai, N., Selhub, J., Robins, S. J., Benjamin, E. J., D'Agostino, R. B. and Vasan, R. S. (2006). Multiple biomarkers for the prediction of first major cardiovascular events and death. *New English Journal of Medicine.* **355**:2631–2639.

[41] Wetli, H. A., Brenneisen, R., Tschudi, I., Bigler, P., Sprang, T., Schurch, S. and Muhlbauer, R. C. (2004). Gamma-glutamyl-peptide isolated from onion by bioassay guided fractionation inhibits resorption activity of osteoclast.**In**: 26th Annual Meeting of the American Society for Bone and Mineral Research, pp. S314–S314.

[42] World Health Organization (2003). WHO guidelines on good agricultural and collection practices (GACP) for medicinal plants. P. 55-60.

[43] Ye, C.L., Dai, D.H., and Hu, W.L. (2013). Antimicrobial and antioxidant activities of the essential oil from Onion (*Allium cepa*). *Food control* **30:** 48-53.

[44] Yoshida, H., Katsuzaki, M., Ohta, R., Ishikawa, K., Fukuda, H., Fujino, T. and Suzuki, A. (1998a). Antimicrobial activity of the thiosulfinates isolated from oil-macerated garlic extract. *Biosci. Biotechnol. Biochem.* **63**:591–594.

[45] Yuan, L., Ji, T. F., Wang, A. G., Yang, J. B. and Su, Y. L. (2008). Two new furostanol saponins from the seeds of *Allium cepa. Chinese Chem. Lett.* **19**:461–464.

[46] Zohri, A. N., Abdel-Gawad, K. and Saber, S. (1995).Antibacterial, antidermatophytic and antitoxigenic activities of Onion (*Allium cepa*) oil. *Microbiological Research.* **150**:167–172.