EXTRACTION OF ORGANIC MICRONUTRIENTS, POLYPHENOLIC, SULPHIDE FROM GARLIC, SOYBEAN HUSK ECONOMICALLY & IT'S FIELD TRIAL ON COTTON CROP

Dr. Amrut Gunwantrao Gaddamwar*

Assistant Professor, Amolkachand Mahavidyalaya Yavatmal, Affiliated to Sant Gadge Baba Amravati University Amravati, Maharashtra, India,

Dr. Tukaram P.Chavan[1]

[1] Associate Professor, Amolkachand Mahavidyalaya Yavatmal, Affiliated to Sant Gadge Baba Amravati University Amravati, Maharashtra, India,

Mr. Sagar Narayan Dakhane[1]

[1]Assistant Professor, MES Abasaheb Garware College, Pune-04 Affiliated to Savitribai pune university Pune Maharashtra, India,,

Dr. Ramesh Tukaram Parihar[2]

[2]Assistant Professor, Department of Chemistry, Vidnyan Mahavidyalaya, Malkapur, District-Buldhana, Maharashtra State, Affiliated to Sant Gadge Baba Amravati University Amravati, Maharashtra, India,

Mr. G.Mallikarjun[2]

[2]Assistant Professor, Department of Chemistry, Govt. Degree College, Ibrahimpatnam, Ranga Reddy District, Telangana State, Affiliated to Osmania University Hyderabad, India,

ABSTRACT: *According to different reports, garlic husk contains different micronutrients in large quantity but it is very difficult to extract these micronutrients without losing its chemical nature in liquid states. India soil analysis reports, different states having deficient's in different micronutrients, it affects the crop yield. A micronutrients Mn, Fe, Ni, Cu, Zn, Co, Na, Mg, Si, Ca, and Cu was extracted chemically economical way and micronutrients were analyzed using atomic absorption spectrophotometer. Garlic husk has large potential to fulfill the micronutrient requirement of the field economically. Garlic husks can be used as an easily accessible source of natural bioactive compounds. It shows antioxidant activity, inhibited the growth of different pathogenic bacteria (Gram+) and showed ability to inhibit the growth of P. aeruginosa and K. pneumoniae these activities can probably be attributed to the major phenolic compounds, such as caffeic, p-coumaric ferulic and di-ferulic acids. The results obtained spoke in favor of the garlic husk as it is a promising source of natural antioxidants to be used for different biological, medicinal, food and agricultural applications.*

It provides an alternative to chemical micronutrients, antimicrobial, antioxidant and PGR, to increase the yield organically at low cost, to increase the fertility of the soil by retaining useful bacteria of the soil.

KEYWORDS: Soybean husk, Garlic husk, Extraction, the role of micronutrients, micronutrients comparisons.

I. INTRODUCTION

Garlic husk, soybean husk has gained importance due to the presence of these dissolved salts, micronutrients, macronutrients, essential elements, and beneficial elements in it. Some of these salts and metal ions are useful to the crops whereas some of these salts harmful to the human being and un-fit for domestic and industrial use. Garlic production occupies a leading position worldwide due to their wide usage in various sectors of the food industry. Husk is the main waste from garlic processing. The biological properties of garlic bulbs are well studied, but the husks are not well investigated to date. The total antioxidant capacities of aqueous and ethanol extracts of garlic husks and bulbs were determined by the ORAC method of the sample. The husks demonstrated the greatest total antioxidant capacities and significantly exceeded those of the bulbs. Garlic husk contains Allicin, alliin, ajoene, alliinase, peroxidase, miracynase, sucrose, glucose, minerals, vitamins, beta-carotene Actin, talin, paxillin, Antioxidant, Phenolic compound, minerals, vinyldithiins, and sulfides, Organosulfur compounds, Sulfur, Allyl disulfide, diallyl disulfide, allicin. Phenol, diallyl sulfur, furan carboxaldehyde S-allyl cysteine, S-allylmercaptocysteine, 1-methyl-1,2,3,4-tetrahydro-beta-carboline-3-carboxylic, Flavin, S-Allyl-L-cysteine, S-allyl cysteine sulfoxide, GPH-P (pepsin) y GPH-T (trypsin), Organic sulfides nitroso-sulfide, Organosulfur compounds, alk(en)yl-L-cysteine sulfoxides (ACSOs), (1R,3S)-1-methyl-1,2,3,4 tetrahydroβ-carboline-3-carboxylic acid, γ-glutamyl-S-allyl-L-cysteines (GSACs) and S-allyl-L-cysteine sulfoxide, diallyl sulfide (DAS), diallyl disulfide (DADS), diallyl trisulfide (DATS), and allyl methyl sulfide (AMS), ajoene, Vitamin C, total phenols, total flavonoids, free sugars content, and antioxidant activity therefore its extract-ant has potential as paste repellent nature, flower stimulant nature, plant growth regulator, root developer etc for the different crops.

i) Micronutrients are of two types which play a vital role in the increase of crop yield:

1**)** Metallic micronutrients (Cations) K+, NH_4^+, Na^+, Ca^{2+}, and Mg^{2+}.

2) Non-Metallic micronutrients (Anions) NO_3^-, Cl^-, CO_3^-, SO_4^{2-}, and PO_4^{2-}.

The physiological role of essential plant micronutrients

Nitrogen: It makes plant dark green and succulent, promotes vegetative growth, and increases the cation exchange capacity of roots, and also makes it more efficient in absorbing other nutrients like P, K, and Ca.

Potassium: It increases the efficiency of the leaf in manufacturing sugar and starch and hence essential for potato and sugar beet crops, it helps to produce stiff straw in cereal and reduces lodging.

Calcium: It increases the stiffness of straw, promotes early root development and growth, provides basic materials for neutralization of organic acids, and encourages the speed of production by increasing the Ca/K ratio in soil solution. Magnesium, sulfur, iron, manganese, zinc, copper, molybdenum, and chlorine are micronutrients; each one plays its role in the development of plants. Most of these micronutrients are present in the water in different proportions.

Role of Micronutrients of sample: Manganese (Mn), Iron (Fe), Nickel (Ni), Copper (Cu), and Zinc (Zn) are the micronutrients which are under investigation. These metals are present in the sample. It plays a vital role in the growth as well as the yield of the crop.

Manganese (Mn): It acts as a catalyst in oxidation-reduction in the plant. It helps in chlorophyll formation and acts as an activator of enzymes. They are usually present in small amounts in natural soybean but many of them are toxic even at very low concentrations.

Iron (Fe): it is not a constituent of chlorophyll, but it is essential for its formation and also for the synthesis of protein and several metabolic reactions in the plant.

Nickel (Ni): Nickel is also required for the plant in a smaller amount for a better yield. It is a component of some plant enzymes; it metabolizes urea nitrogen into useable ammonia within the plant.

Copper (Cu): It activates a group of oxidizing enzymes and is a constituent of certain proteins. It is known to act as an “electron carrier” in enzymes which brings about oxidation-reduction and regulates the respiratory activities of plants.

Zinc (Zn): It is the constituent of various enzymes, therefore essential for several enzymes' reactions it also helps in the formation of growth hormones and chlorophyll of plants.

Cobalt (Co): Cobalt is an essential element and plays a critical role in the overall growth process of plants. Cobalt is necessary for the processes of stem growth, elongating the leaf coleoptiles, and expanding leaf discs. It is a critical element needed for a plant to reach maturity and for healthy bud development.

Sodium (Na): Higher *plants* require *sodium* to be able to grow to their full potential. Increased growth rates resulting from *sodium* is the result of improved water balance within the *plant* and resulting in cell expansion.

Potassium (K): Potassium acts as an activator for enzymes involved in protein synthesis and also for several enzymes involved in carbohydrates and nucleic acid metabolism. It increases the efficiency of the leaf in manufacturing sugar and starch hence it is essential.

II. MATERIALS AND METHODS:

Garlic husk (white and pink) were obtained locally in the region of Vidharbha, India. Freshly harvested garlic husk were randomly collected from different farms. Similar with local practice, the husk were dried by spreading them in a thin-layer inside a darkroom with open windows near the farms for 20 days. Samples were stored in a cabinet at 25oC. The sample husk were randomly selected from the husk sample and their outer covering manually removed and crushed in to powder form, it is used for the extraction of micronutrients solution, similarly soybean husk.

2.0 Preparation of Micronutrients solution from Garlic husk, Soybean husk: Garlic husk has been collected from the field, this garlic husk is further ground into powder of Garlic husk, this garlic husk has been transferred into a round bottom flask, it was treated with 90% hydrochloric acid, then to this reactant was treated with 80% sulphuric acid, after 24 hours stir it continuously, we got precipitate and filterant, separate the filterant with the help of watchman filter paper, this filterant was diluted with double distilled water. Micronutrients of this filterant were analyzed using atomic absorption spectrophotometer.

2.1 Investigation of Micronutrients: Atomic absorption spectrometer Model-AAS-280 were used to test the ppm of Manganese (Mn), Iron(Fe), Nickel (Ni), Copper(Cu), Zinc(Zn), Cobalt(Co), Sodium(Na), Potassium(K), present in samples 1,2,3,4 and 5 by using the following formula (Perkin-Elmer method)

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.1 Manganese (Mn):

Instrumentation - Atomic absorption spectrometer

Operating parameters:

1) Instrument: Atomic absorption spectrometer
i. Make - Perkin Elmer
ii. Model - AAS-280
2) Light source: It consist of Hallow Cathode lamp
3) Flame: Air Acetylene Flame is used
4) Wave length: 279.5 nm.

Materials and equipments:-

Calibrated Class ‘A’ glassware, double distilled water, Reagents.

Procedure:

Step -1: Preparation of standard stock solution:

Manganese, 1000 mg/L. Dissolve 1.000 gm of manganese metal in a minimum volume of (1+1) HNO_3. Dilute to 1 liter with 1% (v/v) HCl. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.

Step -2: Preparation of Test solution: Aspirated directly

Calculation:

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.2 Iron (Fe):

Instrumentation - Atomic absorption spectrometer

Operating parameters:

1. Instrument: Atomic absorption spectrometer
2. Make - Perkin Elmer
3. Model - AAS-280
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Air Acetylene Flame is used.
6. Wavelength: 248.3 nm.

Materials and types of equipment:-
Calibrated Class ‘A’ glassware, double distilled water, Reagents.
Procedure:
Step -1: Preparation of standard stock solution:
Iron, 1000 µg/ml. The solution is prepared in 1 liter with 1% nitric acid. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.
Step -2: Preparation of Test solution: Aspirated directly

$$\mu g/ml = \frac{\text{(sample absorbance) X (Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.3 Nickel (Ni):
Instrumentation - Atomic absorption spectrometer
Operating parameters:
1. Instrument: Atomic absorption spectrometer
2. Make-Perkin Elmer
3. Model - AAS-280
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Acetylene Flame is used
6. Wavelength: 232.0 nm

Materials and types of equipment:-
Calibrated Class ‘A’ glassware, double distilled water, Reagents.
Procedure:
Step -1: Preparation of standard stock solution:
Nickel, 1000µg/ml, a solution is prepared in 1 liter with 1% nitric acid. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.
Step -2 : Preparation of Test solution: Aspirated directly
Calculation:

$$\mu g/ml = \frac{\text{(sample absorbance) X (Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.4 Copper (Cu):
Instrumentation - Atomic absorption spectrometer
Operating parameters:
1. Instrument: Atomic absorption spectrometer
2. Make - Perkin Elmer
3. Model - AAS-280
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Acetylene Flame is used.
6. Wavelength: 324.8 nm.

Materials and types of equipment:-
Calibrated Class ‘A’ glassware, double distilled water, Reagents.
Procedure:
Step -1: Preparation of standard stock solution:
Copper, 1000µg/ml solution is prepared in 1litre with 1% nitric acid. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.
Step -2: Preparation of Test solution: Aspirated directly
Calculation:

$$\mu g/ml = \frac{\text{(sample absorbance) X (Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.5 Zinc (Zn):
Instrumentation - Atomic absorption spectrometer

Operating parameters:

1. Instrument: Atomic absorption spectrometer
2. Make - Perkin Elmer
3. Model - AAS-280
4. Light source: Hallow Cathode lamp
5. Flame: Acetylene Flame
6. Wave length: 213.9 nm

Materials and types of equipment:-

Calibrated Class ‘A’ glassware, double distilled water, Reagents.

Procedure:

Step -1: Preparation of standard stock solution:

Zinc, 500µg/ml solution is prepared in 1litre with 1% Hydrochloric acid. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.

Step -2 : Preparation of Test solution: Aspirated directly

Calculation:

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.6 Cobalt (Co):

Instrumentation - Atomic absorption spectrometer

Operating parameters:

1. Instrument: Atomic absorption spectrometer
2. Make - Perkin Elmer
3. Model - AAS-280
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Air Acetylene Flame is used.
6. Wavelength: 240.7 nm.

Materials and types of equipment:-

Calibrated Class ‘A’ glassware, double distilled water, Reagents.

Procedure:

Step -1: Preparation of standard stock solution:

Cobalt, 1000 mg/L. Dissolve 1.000 gm of cobalt metal in a minimum volume of (1+1) HCl. Dilute to 1 liter with 1% (v/v) HCl, Prepare standard solution, having known concentration of the metal to be determined in the same solvent.

Step -2: Preparation of Test solution: Aspirated directly

Calculation:

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.7 Sodium (Na):

Instrumentation - Atomic absorption spectrometer

Operating parameters:

1. Instrument: Atomic absorption spectrometer.
2. Make - Perkin Elmer.
3. Model - AAS-280.
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Air Acetylene Flame is used.
6. Wavelength: 589.0 nm.

Materials and types of equipment:-

Calibrated Class ‘A’ glassware, double distilled water, Reagents.

Procedure:

Step -1: Preparation of standard stock solution:

Sodium, 1000 mg/L Dissolve 2.542 gm of sodium chloride (NaCl), in deionized water and dilute to 1 liter with deionized water. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.

Step -2: Preparation of Test solution: Aspirated directly

Calculation:

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

2.1.8 Potassium (K):

Instrumentation - Atomic absorption spectrometer

Operating parameters:

1. Instrument: Atomic absorption spectrometer
2. Make - Perkin Elmer
3. Model - AAS-280
4. Light source: It consists of a Hollow Cathode lamp.
5. Flame: Air Acetylene Flame is used.
6. Wavelength: 766.5 nm.

Materials and types of equipment:-

Calibrated Class ‘A’ glassware, double distilled water, Reagents.

Procedure:

Step -1: Preparation of standard stock solution:

Potassium, 1000 mg/L Dissolve 1.907 gm of potassium chloride (KCl), in deionized water and dilute to 1 liter with deionized water. Prepare standard solution, having known concentration of the metal to be determined in the same solvent.

Step -2: Preparation of Test solution: Aspirated directly

Calculation:

$$\mu g/ml = \frac{(\text{sample absorbance})\ X\ (\text{Standard concentration in } \mu g/ml)}{\text{Standard Absorbance}}$$

Similarly magnesium (Mg), Silicon (Si), Calcium (Ca) was analyzed.

Above extracted soybean, garlic husk samples field trials were conducted on cotton crop which gave extraordinary results on cotton crop. 150 ml of soybean based husk solution is mixed with 150 ml of garlic based liquid in 200 liters of water, it was foliar sprayed 3 to 4 times on cotton crop at every 20 days interval after 100 days from the date of sowing of cotton crop.

III. DISCUSSION:

To make farmers income more than double is very difficult task due to uncertain climatic condition, untimely rain but by reducing expenditure using novel organic micronutrients solutions extracted from soybean, garlic husk economically from soybean waste husk, as well as garlic husk which were generated in the field in large quantity which has large potential to fulfill the requirement of micronutrients. Due to presence of phenolic, organo-sulphure, nitoroso-sulfure it acts as paste repellent as well as plant growth regulator and flower stimulant and which Provide poison-free vegetables to society. It is the best alternative to uplift the farmer’s life economically.

A benefit to farmers: A farmer gets cost-effective micronutrients, paste control, flower stimulant as well as plant growth regulator in a single solution which is not only completely organic but also save the labor cost of the farmer & quality of micronutrients in a liquid state, due to the liquid state rate of uptake by the plant were more, which is beneficial to crops as compared to other micronutrients. It is more profitable for the farmers & safe for society.

Future Plan: It fulfills the need for 21st-century micronutrient requirements, paste control, PGR and flower stimulant together for the different agricultural crops. It is formulated in such way which contains large quantity of micronutrients as well as PGR and flower stimulant together, which is completely organic, extracted 100% from soybean, garlic husk economically.

Table 1 shows micronutrients present in Soybean husk based liquid Sample in ppm

Sr. No.	Name of element	Concentrations in ppm or mg/L	Pick area	Pick hit
1	Sodium (Na)	14.378	2.95	0.23
2	Potassium (K)	30.649	2.35	0.27
3	Magnesium (Mg)	31.132	2.76	0.39
4	Zinc (Zn)	12.517	5.18	0.27
5	Nickel (Ni)	0.840	5.40	0.35
6	Silicon (Si)	0.370	2.63	0.23
7	Calcium (Ca)	16.632	2.70	0.39
8	Iron (Fe)	17.830	1.95	0.28
9	Cobalt (Co)	1.229	3.74	0.34
10	Magnase (Mn)	10.055	2.34	0.54
11	Cupper (Cu)	1.486	4.29	0.76

Figure 1 shows micronutrients present in Soybean husk based liquid Sample in ppm

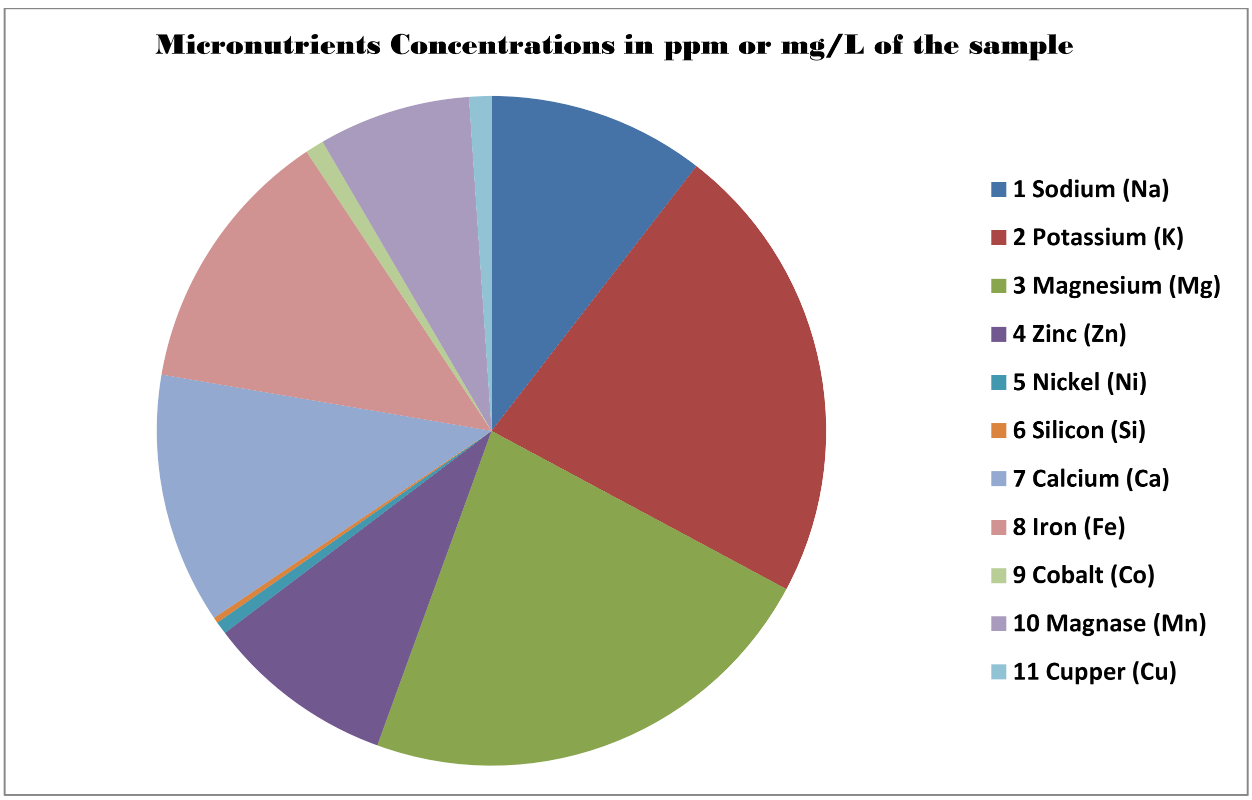

Table 2 shows that micronutrients present in garlic base husk

Sr. No.	Name of element	Concentrations in ppm or mg/L	Pick area	Pick hit
1	Sodium (Na)	12.289	3.92	0.67
2	Potassium (K)	23.507	3.15	0.28
3	Magnesium (Mg)	10.076	4.18	0.39
4	Zinc (Zn)	12.324	2.62	0.23
5	Nickel (Ni)	0.959	2.35	0.32
6	Silicon (Si)	0.471	3.87	0.33
7	Calcium (Ca)	15.265	4.18	0.28
8	Iron (Fe)	12.43	2.76	0.416
9	Cobalt (Co)	1.741	2.15	0.496
10	Magnase (Mn)	11.146	2.34	0.77
11	Cupper (Cu)	1.639	2.76	0.22

Figure 2 shows micronutrients present in Garlic turpale based liquid Sample in ppm

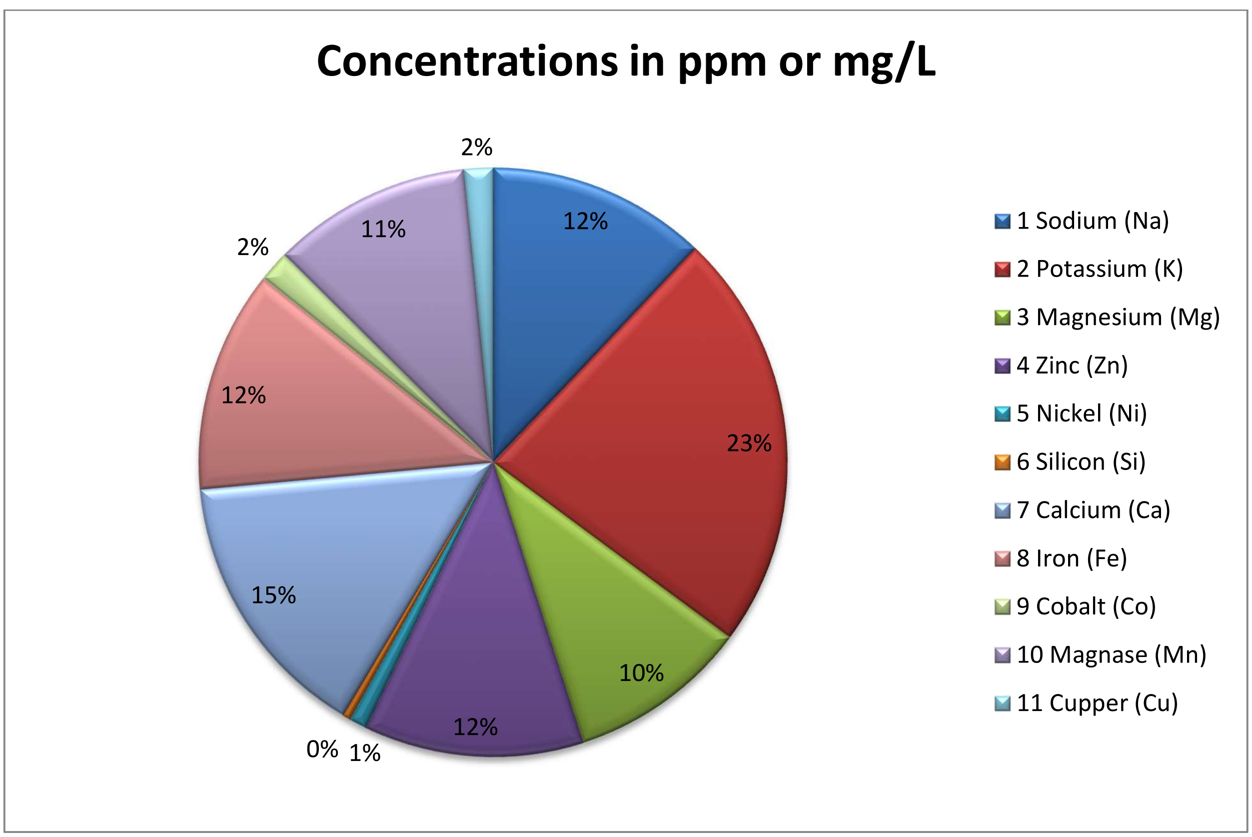

Figure 3 shows micronutrients pick area & hit of Soybean in AAS

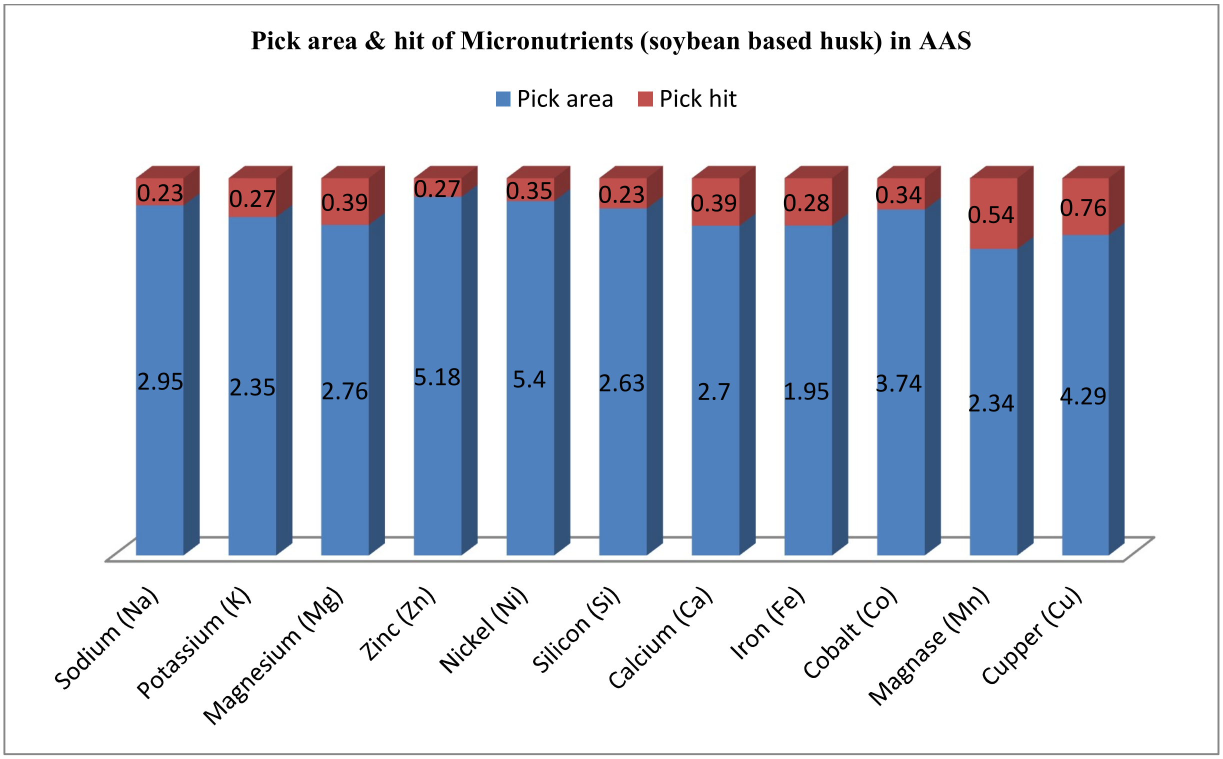

Figure 4 shows micronutrients pick area & hit of Garlic Turpale in AAS

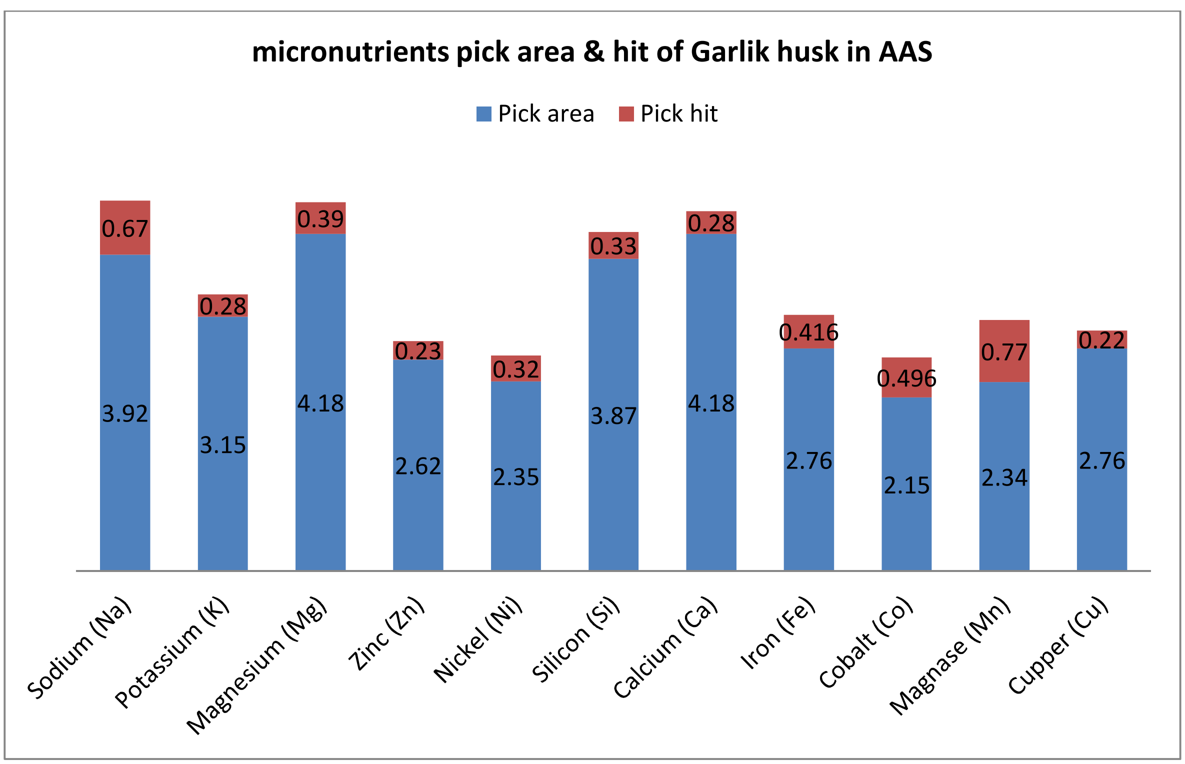

Figure 5 shows that images of soybean and garlic husk

Figure 6 shows that images of cotton crop cultivated using samples

IV. RESULTS:

From table 1, 2, and figure 1, 2, 3 & 4 it is clear that the concentration of micronutrients (beneficial elements) in the given soybean, Garlic samples was found to be Sodium (Na)-14.378, 12.289, Potassium (K)-30.649, 23.507, Magnesium (Mg)-31.132, 10.076, Zinc (Zn)-12.517, 12.324, Calcium (Ca)-16.632, 15.265, Iron (Fe)-17.830, 12.43, Magnase (Mn)-10.055, 11.146 and essential elements are found to be Nickel (Ni)-0.840, 0.959, Silicon (Si)-0.370, 0.471, Cobalt (Co)-1.229, 1.741, Copper (Cu)-1.486, 1.639 in ppm or mg/L respectively. These organic beneficial and essential elements are found in a requisite amount as per the short and long-duration crops requirements which are cost-effective as compared to available chemical micronutrients but in garlic extract not only above micronutrients but also phenolic, Organic sulfides nitroso-sulfide, Organo-sulfur compounds are present therefore it shows paste repellent nature and acts as flower stimulant as well as PGR. As compared to soybean husk extract Garlic husk extract is best for the different crops in agriculture. Field trials of above samples on cotton crop were conducted which graves extraordinary results.

ACKNOWLEDGMENTS

Great support from Dr. Rammanohar Mishra, principal of the Amolkachand Science College Yavatmal, and Dr. P.R.Rajput Principal of SSSKR Innani Science College, Karanja lad District Washim, Maharashtra, India.

REFERENCES:

1. Aala, F.; Kalsom, U.; Nulit, R.; Rezaie, S. 2014. Inhibitory effect of allicin and garlic extracts on growth of cultured hyphae. Iran J Basic Med Sci 17: 150-154.
2. Abdel-Daim, M.; Abdelkhalek, N.; Hassan, A. 2015. Antagonistic activity of dietary allicin against deltamethrin-induced oxidative damage in freshwater Nile tilapia; Oreochromis niloticus. Ecotoxicology and Environmental Safety 111: 146-152.
3. Abdel-Hafeez, E.; Ahmad, A.; Kamal, A.; Abdellatif, M.; Abdelgelil, N. 2015. In vivo antiprotozoan effects of garlic (*Allium sativum*) and ginger (*Zingiber officinale*) extracts on experimentally infected mice with Blastocystis spp. Parasitology Research 114: 3439-3444.
4. Abid-Essefi, S.; Zaied, C.; Bouaziz, C.; Ben Salem, I.; Kaderi, R.; Bacha, H. 2012. Protective effect of aqueous extract of Allium sativum against zearalenone toxicity mediated by oxidative stress. Experimental and Toxicologic Pathology 64: 689-695.
5. Addis, W.; Abebaw, A. 2018. Determination of heavy metal concentration in soils used for cultivation of Allium sativum L. (garlic) in East Gojjam Zone, Amhara Region, Ethiopia. Cogent Chemistry 3.
6. Adwan, G., & Mhanna, M. (2008). Synergistic effect of plant extracts and antibiotics on Staphylococcus aureus strains isolated from clinical specimen. Middle-East Journal of Scientific Research, 3, pp 134-139.
7. Ahmed, Z., Khan, S., Khan, M., Tanveer, A., & Lone, Z. (2010). Synergistic effect of Salvadora persica extracts, tetracycline and penicillin against Staphylococcus aureus. African Journal of Basic & Applied Sciences, 2(1-2), pp 25-29.
8. Aiyegoro, O., & Okoh, A. (2009). Use of bioactive plant products in combination with standard Antibiotics: Implications in antimicrobial chemotherapy. Journal of Medicinal Plants Research, 3(13), pp 1147-1152.
9. Almeida, A., Farah, A., Silva, D., Nunan, E., & Gl_oria, M. (2006). Antibacterial activity of coffee extracts and selected coffee chemical compounds against enterobacteria. Journal of Agricultural and Food Chemistry, 54(23), pp 8738-8743.
10. Alotaibi, A., Alodeani, E., & Izhari, M. (2014). Assessment of cooperative repression of uropathogens by certain antimicrobics and essential oils. International Journal of Research Studies in Biosciences, 2(10), pp 1-6.

11. Adeyi, O., 2010. Proximate composition of some agricultural wastes in Nigeria andtheir potential use in activated carbon production. J. Appl. Sci. Environ. Manag.14, 55–58.
12. AFNOR, 1986. Association Franc¸ aise de Normalisation. Céréales et produitscéréaliers. Détermination de la teneur en matières grasses totales. NF V 03-713.,pp. 39–155.
13. Amagase, H., 2006. Clarifying the real bioactive constituents of garlic. J. Nutr. 136,716–725.
14. Amagase, H., Petesch, B.L., Matsuura, H., Kasuga, S., Itakura, Y., 2001. Intake of garlicand its bioactive components. J. Nutr. 131, 955–962.
15. AOAC, 1997. Association of Official Analytical Chemists. Official methods of Analysis.Assoc. of Analytical Chemists, Washington, DC, USA.Asamarai, A.M., Addis, P.B., Epley, R.J., Krick, T.P., 1996. Wild rice hull antioxidants.J. Agric. Food Chem. 44, 126–130.
16. Sokamte T A, Mbougueng P D, Tatsadjieu N L and Sachindra N M 2019 Phenolic compounds characterization and antioxidant activities of selected spices from Cameroon *S. Afr. J. Bot.* **121** 7–15
17. Embuscado M E 2015 Spices and herbs: Natural sources of antioxidants–a mini review *J. Funct. Foods* **18** 811–9
18. Nychas G-J E and Tassou C C 2014 Traditional Preservatives – Oils and Spices *Encyclopedia of Food Microbiology (Second edition)* ed C A Batt and M L Tortorello (Amsterdam : Academic Press, Elsevier, Ltd.) pp 113–8
19. Cortes-Rojas D F de Souza C R F and Oliveira W P 2014 Clove (Syzygium aromaticum): a precious spice *Asian Pac. J. Trop. Biomed.* **pp** 90–6
20. Trivedy R. K. and P. K. Goel (1984): Hand book of chemical and Biological method for water pollution studies Environmental publications, Karad: 1-247
21. A text book of Environmental studies by Dr P.R.Rajput et al 2013, Lambert publication Private Limited, Germany.
22. Department of Agriculture and Cooperation, Min. of Agriculture, Government of India Report-2012
23. Snober H. Bhat et al (2011) “Correlation of Soil Physico-Chemical Factors With Vam Fungi Distribution Under Different Agro-ecological Conditions” International Journal of Pharma and Bio Sciences, vol-2, issue-2, pp B-98-107.
24. Department of agriculture, Government of Maharashtra-2010.
25. Department of Agriculture and Cooperation, Min. of Agriculture, Government of India Report-2012.
26. Krutuja Bhosale Govt. of Maharashtra Regional imbalance committee report-2012
27. Gaddamwar et-al (2013), “Influence of constituents of coconut water on fenugreek plant” International journal of herbal medicine, Vol-1, Issue-2, pp-162-168.
28. E A Kotenkova1 and N V Kupaeva (2019), “Comparative antioxidant study of onion and garlic waste and bulbs”, IOP Conf. Series: Earth and Environmental Science 333, pp 1-5.
29. Fatma Kallel et.al (2014), “Garlic (Allium sativum L.) husk waste as a potential source of phenoliccompounds: Influence of extracting solvents on its antimicrobial andantioxidant properties”, Industrial Crops and Products 62, pp 34–41.
30. Teofilo Macario Espinoza Tellez et.at (2020), “Garlic (Allium sativum L) and its beneficial properties for health: A Review El ajo (Allium sativum L) y sus propiedades beneficiosas para la salud: Una revision”, Agroind. sci., Vol 10, issue 1, pp 103-115