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Effect of Yeast as Biopromotor for Soil Amendment on the Growth of Sorghum Bicolor and Arachis Hypogea

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Abstract:- Yeast is used asbio-fertilizer in agriculture for the biological activity and protection for animal, human and the environment. Plant growth promoting by yeast enhanced the plant root and growth development. The variable nutritional and hormonal stability, producing plant growth regulators, solubilizing nutrients in soil by yeast can promote plant growth. In the current research the analysis of plant growth enhancement, phytochemical constituent and the antioxidant activity of Sorghum bicolor and Arachis hypogeal was done.

Keywords:- *Yeast, Biofertilizer, Plant growth.*

I. INTRODUCTION

Now a day's the use of synthetic fertilizers by farmers to obtainanim proved yield of various field crops. Chemical fertilizers and pesticides can reduce soil fertility and adverse health harms to the consumers. Owing to side effects of chemical fertilizers, interest has been stimulated for the make use of organic manures (Follet et al., 1981).Non-conventional sources of enormous organic matter status of soil are acquiring much attention due to their easy availability, prompt response and feasibility in using over large area in less time. Extreme use of chemical fertilizers will build up the environment related problems and situation can be improved through the use of bio-fertilizers (Saadatnia et al., 2009). Use of chemical fertilizers in the crop field contributes greatly to the deterioration of the environment, depletion in soil fertility, reduce in agricultural production and soil degradation (Inbar et al., 1993). Yeasts are unicellular fungi that multiply primarily through asexual condition and grow up quickly on carbohydrates which often through fermentative and respiratory pathways. Since a consequence of their nutritional preference, yeast populations are generally an order of magnitude higher in the rhizosp here as opposed to the bulk soil (Botha, 2011). Yeasts showed wide range of plant growth promoting individuals including pathogen inhibition (El-Tarabily and Sivasithamparam, 2006). Phytohormone production (Nassar et al., 2005); phosphate solubilisation (Mirabal Alonso et al., 2008);N and S oxidation (Falih and Wainwright, 1995); siderophore production (Sansone et al., 2005) and stimulation of my corrhizal root colonization (Mirabal Alonso et al., 2008). Yeasts show antimicrobial and helpful substance desirable for plant growth from amino acids and sugars concealed by bacteria, plant roots and organic matter (Boraste et al., 2009). Saccharomyces cerevisiaeis considered as a new promising plant growth promoting yeast for dissimilar crops. Recently, it became a positive substitute to artificialfertilizers carefully used for animals, human and environment (Omran, 2000).

II. MATERIALS AND METHODS

A. Preparation of yeast strains

Commercial yeast was used for whole study. Saccharomyces cerevisiaeis was grown on Yeast – Peptone Dextrose (YPD) agar. Single cell isolated colonies were inoculated in conical flask containing 50ml of yeast – peptone dextrose (YPD) media. Saccharomyces cerevisiaeis culture was incubated for 18 to 24 hours prior to inoculation

- *Identification of yeast*

The isolated yeast was found on the basis of Gram's staining and biochemical characteristics (MacFaddin, 1980) and results were found with the use of Bergey's Manual of systematic Bacteriology (Krieg and Holt, 1984).

- *Physiochemical analysis for Soil samples*

The calcium, chloride, magnesium, TDS, Iron, phosphate content were analysed in soil incorporate with different concentration of yeast

- *Analysis of plant growth promoting activity of yeast*

- *Qualitative assay of IAA*

Saccharomyces cerevisiaeis isolates was inoculated in YPD broth with L-Tryptophan 500mg/l at 28ºC for one week. Completely grown cultures were centrifuged at 3000 rpm for 30 min and then 2 ml of supernatant was mixed with two drops of orthophosphoric acid and 4ml of the Salkowski reagent. Development of pink colour indicates IAA production was determined.

- *Assay of Gibberelic acid production: (Borrow et al., 1955)*

A quantity of YPD broth was prepared for yeast isolates respectively and sterilized. Soon after than the culture was centrifuged for 8000 rpm for 10 min. 15 ml of supernatant, 2ml of zinc acetate reagent (21.9 g zinc acetate +1ml glacial acetic acid and the volume make equal to 100 ml with distilled water) was added. After 2 minutes, 2 ml of potassium

ferrocyanide (10.6% in distilled water) was added and was centrifuged at low speed (2000 rpm) for 15 minutes. To 5 ml of supernatant and 5ml of 30 percent HCL was added and combination was incubated for 20°C for 75 min. For blank of 5ml of 5 percent HCL was used. Absorbance was read at 254 nm concentration of gibberllins was calculated by preparing stand curve by using gibberellic acid (GA_3) as standard (100-1000 µg/ml).

- *Assay of Siderophore production (Ramyasmruthi et al., 2012)*

- *Tetrazolium test*

A pinch of tetrazolium and 2 drops of NaOH were added to 0.01 g of siderophore fractions of the saccharomyces cerevisiaeis. The appearance of deep red colour is indicative of siderophore.

- *Assay of Phosphate solublization production (Subba rao, 1999)*

Phosphate-solublization test was conducted qualitatively by plating the Saccharomyces cerevisiaeis isolates in agar containing precipitated tricalcium phosphate. The medium was a modification of pikovskaya medium. Saccharomyces cerevisiaeis isolates culture were streaked on the surface of agar plates and incubated at 28°c for 3 days. After 3 days, the colonies showing the clear zones around them were considered as positive for positive phosphate solubilisation.

- *Preparation of yeast for plant growth* (*Moustafa et al., 2008)*

Dry yeast was well dissolved firstly in slight sugar solutions and cultivation for 12hr. Before application, the solutions were diluted to the required concentrations using sterile distilled water. Seeds were soaked in defined yeast concentrations (1%, 2% and 5%) at overnight before sowing. Soaked seeds in pure water acted as control. For foliar spraying or soil inoculation treatments, seeds were sown without soaking.

- *Experimental code and design for pot study*

Pot 1 - Control (water)
Pot 2 - 1% S.cerevisiae (spraying)
Pot 3 - 2% S.cerevisiae (spraying)
Pot 4 - 5% S.cerevisiae (spraying)
Pot 5 - Control (seed coated with water)
Pot 6 - seed coated with 1% S.cerevisiae
Pot 7 - seed coated with 2% S.cerevisiae
Pot 8 - seed coated with 5% S.cerevisiae

- *Biometric analysis*

The plants were randomly selected for recording the shoot length and root length, wet weight, dry weight, Carbohydrate, protein, chlorophyll and carotenoids were analysed.

- *Qualitative phytochemical analysis (Harborne, 1973)*

Phytochemical analysis for major phyto constituents of the plant extract under taken using standard qualitative method as described by Harborne, 1973.

- *Antioxidant analysis Chelating activity (Harpreet Walia et al., 2011):*

The chelating activity of extracts was measured as given by (Harpreet Walia et al., 2011) with little modifications. 1ml of extract with different concentration was mixed with 3.5ml methonal and then the mixture was mixed with ferrous chloride (2Mm, 0.1ml) and ferrozine (1Mm, 0.2ml) for 10 min at room temperature. The absorbance was measured at 562 nm against a blank in which the extract was not added. The % of inhibition was calculated as:

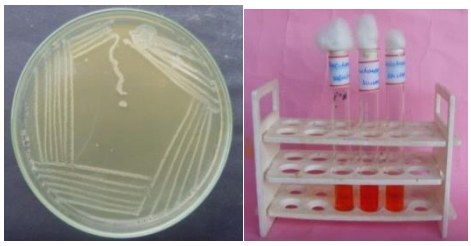
$$\% \text{ Inhibition} = B_O\text{-}B1/B_O*100$$

Where, B_O is the absorbance of control, B1 is the absorbance of reaction mixture.

III. RESULTS AND DISCUSSION

A. *Identification of Saccharomyces Cerevisiae*

The identification of Commercial Saccharomyces cerevisiaea test isolates under the study by performing Gram's staining procedure, biochemical tests as recommended in the Bergey's Manual. The production of acid with gas that indicated the presence of fermentation of glucose, sucrose and maltose. Colony morphology showed smooth white, creamy colour and ovoid shaped organism. The isolate was confirmed as Saccharomyces cerevisiaea



B. *Qualitative nutrient analysis*

To analysis the ability of yeast to produce IAA, Gibberllic acid, siderophore, carbohydrate and protein. The Production of pink to red colour point toward the occurrence of IAA. The production of Yellow colour indicated the presence of Gibberllic acid. The production of red colour indicated the presence of siderophore. Tian et al., 2009 reported that Siderophore are minute, high-affinity iron chelating compounds concealed by Bacteria, fungi. The production of siderophore by Saccharomyces cerevisiaea was done in YPD medium.

Phosphate solubilizing activity was checked for Saccharomyces cerevisiaea. In Present study the Pikovskaya agar media, Saccharomyces cerevisiaea was formed a clear halo zone at 9 mm. Similarly were found by Yasmin et al., 2009. He revealed that the Saccharomyces cerevisiaea

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degraded and solubilized the insoluble phosphate into soluble forms and produced a clear zone in Pikovskaya media.

The hydrolytic enzyme of Amylase in which starch converts into amylase enzyme it produced a clear halo zone. Punitha et al., 2010 reported that the Bacillus sp utilize starch as a substrate and converts into maltose by secreting amylase enzyme that can be complete by the formation of brick red colour for the period of enzymatic assay.

The hydrolytic enzyme of protease it produced a clear halo zone. Carl a biliniski et al., 1986 screened Saccharomyces for extracellular proteinase production by inoculation in skim milk agar media. Saccharomyces cerevisiaea strains indicated the production of extracellular acid proteinase by forming clear halo zones.

Lipase hydrolyzed to fatty acid it formed a clear halo zone. Punitha et al., 2010 reported the lipids are hydrolyzed to fatty acid and glycerol. The olive oil was creating out to be the nearly all equitable substrate for enzyme production.

The quantitative measurement assay for the ability of Saccharomyces cerevisiaea to construct IAA in the occurrence of the precursor L-Tryptophan (50, 100, 150, 200, 250 µg/ml) while Compared to all concentrations, the highest amount of IAA was obtained at 200 µg/ml. Yadava et al., 2010 reported IAA producing yeast was believed to improve root length and augmented root growth. In their study yeast also produced IAA at 150 µg/ml concentration.

The Gibberllic acid production from Saccharomyces cerevisiaea was estimated. The maximum amount of Gibberllic acid was observed at 30µg/ml. El-Tohamy and El-Greadly, 2007 studied that The highest plant growth and productivity of Sorghum bicolor and Arachis hypogea was observed in high levels of Gibberllic acid. However, the high level of yeast had also significant values evaluate with control plants. Improvement of growth parameter and yield in response to yeast application was proved in plants. Mishustin, 1963 reported that the Gibberllic acid were extracted and estimated by spectrophoto metric method which found to be Azobacter spp.(VMP21) 67 µg/ml, A.armeniacus (VMP22) 20 µg/ml, A. vinelandii (VMP23) and A. Chroococcum (VMP28) 60 µg/ml, A. vinelandii (VMP29) 50 µg/ml, A. paspali (VM30) 65 µg/ml and A. Chroococcum (VMP31) 70 µg/ml.

C. *Physiochemical Analysis Of Soil*

Physiochemical analysis of soil and different concentration of yeast. The pH of the soil was 7.0, Total Dissolved Solid (TDS) content was 980mg/g, the total calcium, magnesium, phosphate, iron, nitrogen and chloride were 220mg/g, 160 mg/g, 2.00 µg/g, 2.00µg/g, 9.6mg/g, 111.23 mg/g respectively. Mahadevi et al., 2016 reported the pH as 6.2, TDS as 1820mg/g, calcium as 280mg/g and Magnesium as 168mg/g for the soil.

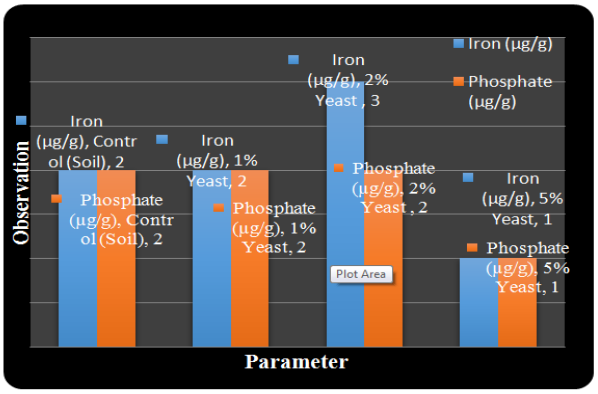

Fig 1:- Parameter

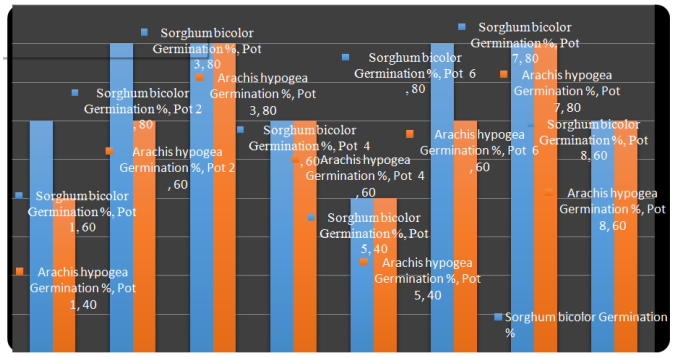

Fig 2:- Seed germination on sorghum bicolor and Arachis hypogea

D. *Biometric Analysis*

The significant difference in all parameters like plant height, root length, number of leaves, plant dry weight and plant fresh weight. The biometric analysis of Sorghum bicolor and Arachis hypogea. It was observed that the maximum height, shoot length, root length was achieved in pot-2 by spraying1% S.cerevisiae to Sorghum bicolor (51.0cm, 6.0cm, 45.0cm) while compared to pot 1/control as Soil were (23.50cm, 8.00cm, 15.50cm). Hence this result concluded that the maximum plant growth was observed in pot 2.

The fresh and dry weight, branch of treatment of pot-2 by spraying 1% S. cerevisiaeto Sorghum bicolor (0.89g, 0.21g, 4 counting) while compare to the pot 1 control as soil (0.33g,0.14g, 3 counting). The biometric analysis of Arachis hypogea. It was observed that the maximum height, shoot length, root length was achieved in pot-2 by spraying 1% S.cerevisiae to Arachis hypogea (30.80cm, 24.0cm, 6.80cm) while compare to pot 1 controlas soil (29.50cm, 6.00cm, 23.50cm).

The fresh and dry weight, number of leaves, branch of treatment of pot-2 by spraying 1% S.cerevisiae to Arachis hypogea in (4.10g,1.25g, 49 no's, 8 (counting) while compare to the pot 1 control as soil were (3.24g, 0.87g,24 no's, 5counting). The maximum growth was observed in pot 2. Moustafa El- sayed shalaby et al., 2008 reported that the concerning root parameters, their length, diameter and fresh

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weight were consider ablyim proved during the second season compared to the first one, representing a quite organization with the experimental conditions. The highest percentage of shoot dry weight of 51.17% was attaining in the second season due to seed treatment with 5 g L-1 of yeast solution. The chlorophyll and carotenoid content of Sorghum bicolor. The maximum chlorophyll content were found in treatment of pot-2 (1% S.cerevisiae (spraying) of Sorghum bicolor (chlorophyll a - 0.80, chlorophyll b-0.38, carotenoid-1.85) when compared to other treatments. Table 11 and figure 9 shows the chlorophyll and carotenoid content of Arachis hypogea. The highest amount of chlorophyll content in an treatment of pot-2 (1% S.cerevisiae (spraying) of Arachis hypogea (chlorophyll a -0.61, chlorophyll b-0.46, carotenoid-1.07). Amprayn et al., 2012 stated that the use of the three yeasts bring on the arrangement of photosynthetic pigments (chlorophyll a and b and carotenoid). However, K. walti (100 ml plant-1) occupied in the highest increase in the pigments contents (0.86 and 0.22 and 1.02mg g-1 fresh leaves, respectively). The differentiation between yeast strains competence to improve the growth of plants.

Fig 3

The carbohydrate content of Sorghum bicolor and Arachis hypogea. It was recognized that the level of total carbohydrate content of the plant Sorghum bicolor and Arachis hypogea were highly enhanced by S.cerevisiae. Among this treatment pot-2 (1% S.cerevisiae (spraying) of Sorghum bicolor was recorded highest carbohydrate content in Sorghum bicolor (233.20 µg) and Arachis hypogea (396.30 µg). Ramadan agamy et al., 1989 reported as therefore, the content of total sugars and total protein in leaves improved significantly as of the use of the yeasts apart from in one case (P. transvaalensis, 50 ml pot1). The data point to the three yeasts induced sucrose formation in the beet roots significantly as compared with the control. The maximum dose (100 ml pot-1) was the best inducer among all cases. However, K. walti (100 ml/plant) caused the utmost increase in the sucrose content. It increased the sucrose content by 42.45% of the give up of the control.

The protein content of Sorghum bicolor and Arachis hypogea. The highest level of protein content were obtained in pot-2 (1% S.cerevisiae (spraying) of Sorghum bicolor (3680 µg) and Arachis hypogea (2450 µg). Gaballah et al., 2012 reported the increase in total soluble protein content could be attributed to the growth hormone produced by yeast and the direct stimulation of protein synthesis. Stino et al., 2009 providing plants with essential nutrient elements required for protein formation.

The different types of phytochemical analysis of carried out two different plants of Sorghum bicolor and Arachis hypogea. The Alkaloids, Flavonoids, Steroids, Protein, and Carbohydrate present in leaves of Sorghum bicolor whereas Saponins, Tannins, Terpenoids, Quinine and Glycosides absent. Omoyeni et al., 2011 reported the screening showed the presence of alkaloids, flavonoid, steroid, protein and carbohydrate whereas Saponins, Tannins, Terpenoids, Quinine and Glycosides are absent.

The leaves of Arachis hypogea showed the presence of Alkaloids, Glycosides, Steroids, Protein, and Carbohydrate whereas Tannins, Terpenoids, Flavonoids, Quinones and Saponins were completely absent. Rajinikanth Marka et al., 2013 reported as Phytochemical analysis of various solvent extracts of leaf of A. hypogaea revealed the presence of alkaloids, glycosides, fats, oils, phenols, lignins whereas tannins, flavonoids, quinones and saponins were completely absent.

E. *Antioxidant activity*

The antioxidant was tested for selected plants. The current work was experiential that the chelating activity of Sorghum bicolor and Arachis hypogea showed the good results. The result indicated the plant extract have iron binding capacity which may be due to the presence of polyphenol that turn aside the cell beginning free radical damaged by reducing of transition metal ions. Halliwell et al., 1995 reported the results the plant were interfered with the formation of ferrous – ferrous zine complex. Indicating, that it have marked iron chelating activities and capture ferrous iron before the formation ferrozine. Transistion mental chelating to from low redox potential complex is an important effect and determines the chelating of iron (II) is one method for evaluate this property. Since the reaction is dependent on the affinity of the an antioxidant towards iron (II) in relation to ferrozine, the assay is affective by both binding constant and concentration of antioxidant and thus only strong iron antioxidant chelator is founded.

IV. CONCLUSION

The results are promising that the yeast one of the bio fertilizer in agriculture. Application of yeast increases the overall growth of the treated plant. In addition they could enhance the physical and chemical properties of soil, the water holding capacity and more nutrients to the soil. To put off the environmental pollution from widespread application of artificial fertilizer it should be referred to farmers to assure the community physical condition and a manageable agriculture.

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