Pichia kudriavzevii – A potential soil yeast candidate for improving soil physical, chemical and biological properties

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Abstract

Soil yeasts exhibit an array of beneficial effects to plants *viz.*, plant growth promotion, phosphate solubilization, nitrogen and sulphur oxidation, etc. Yeasts remain as poorly investigated group of microorganisms that represent an abundant and dependable source of bioactive/chemically novel compounds and potential bioinoculants. Hence this study holds the key concept of assessing the performance of soil yeasts with potential plant growth promoting ability in soil quality improvement. Sixteen soil yeast isolates with plant growth promoting traits were assessed for biofilm forming potential and five potential soil yeast isolates were selected and identified through molecular technique. Soil incubation study was performed with these isolates to assess their impact on soil physical, chemical and biological properties. Due to inoculation of soil yeasts, notable changes were observed in soil physical, chemical and biological properties. Among the soil yeast isolates, *Pichia kudriavzevii* gave better results in soil incubation study.

Introduction

Environment safety has become the most important aspect of all the growing nations. Organic farming is one of the growing strategies all over the world to conserve the health of human beings and soil health too. Microbial inoculants are used for plant growth promotion and soil health improvement in organic farming. In recent days, microbial inoculants (biofertilizers) are popularly used as an alternative to chemical fertilizers due its merits like low cost, environment friendly, etc. The plants and microbes are benefited by each other in one way or other. The plant decides and selects its own microbes through secretion of root exudates and the microbe decides its plant host by its preferential selection process. The contribution of organic matter by microorganisms to soil through production of extracellular polymeric substances plays a pivotal role in improving the soil structure. Soil structure has the important influence on soil capacity to support the growth of plant, nutrient cycling and water holding capacity. Soil aggregation is an important parameter in soil structure maintenance and serves as a tool for evaluating the soil structure. The soil microorganisms play an inevitable role in formation of soil aggregates. The extracellular polymeric substances secreted by the soil microbes serve as cementing agent to form soil aggregates. Yeasts, the unicellular fungi are found in all ecosystems and also contribute significantly for microbial diversity. Certain yeasts are known to affect the soil physical property by producing high quantity of extracellular polymeric substances. Till now the soil aggregation studies are mainly focused with respect to organisms like earthworms, AM fungi, bacteria and very few studies reported on fungi. The occurrence and the stability of the aggregates play a key role in plant growth. Physical forces, chemical bonds and biological agents of the soils are considered as the causes of soil aggregation. With this background, the present study was formulated to study the role of soil yeasts in improving soil physical, chemical and biological properties.

Methods

Soil yeast isolates

The soil yeasts were isolated from different soil ecosystems and screened for various plant growth promoting traits (Ramya et al., 2019).

Biofilm formation and molecular characterization

The efficient soil yeast isolates with plant growth promoting traits were screened quantitatively for the biofilm forming capability under *in vitro* conditions using Crystal violet method as described by Au - O'Toole (2011). The biofilm forming capability of the cultures were assessed at different time intervals *viz.*, 5, 10 and 15 days. The high biofilm forming strain *Pseudomonas aeruginosa* was used as the reference strain.

Yeast DNA isolation was done by Lysis method and 18S rRNA was amplified in a thermal cycler (Harju et al., 2004). The 18S primers used for PCR were; ITS 1 as forward primer and ITS 4 as reverse primer.

Soil incubation study

The soil incubation study was conducted for all the five best biofilm producing soil yeast isolates along with the positive (*Aeromonas hydrophila*), negative (Baker's yeast- *Saccharomyces cerevisiae*) and uninoculated (without inoculums) sterile soil as control.

The soil was air dried and finely sieved by a

250 µm sieve. The soil was sterilized for three times by repeated cooling and heating.

The cultures were grown in broth and centrifuged to obtain the pellets which were then diluted with sterile water and population was also estimated by drop plate technique.

Then each soil yeast isolates were added accordingly that each gram of soil will have 10^8 CFU and mixed with soil separately in sterile trays under aseptic condition. Also sterile synthetic root exudates medium was also added for each pot at the rate of 30 mL per pot. Then the pots (11x10 cm) were filled with this soil mixture @ 400 g/pot and the moisture level is maintained at 60% throughout the incubation period. Three replications per isolate and also three technical replicates for two intervals (15 and 30) were maintained and incubated under laboratory conditions for 15 and 30 days.

Soil property analysis

Sl.No	Properties	Method
Soil physical properties		
1.	Mean weight diameter, geometric mean diameter, macro aggregate stability (4-0.25 mm), micro aggregate stability (0.25-0.05 mm), total water stable aggregates and dry aggregate ratio	Wet sieving procedure (Yoder, 1936).
Soil chemical properties		
2.	Soil pH	Jackson, (1973)
3.	Soil organic carbon	Walkley and Black, 1934
4.	Soil labile carbon	Blair *et al.*, 1995
5.	Soil protein index (SPI)	Wright and Upadhyaya, 1998; Clune, 2007
Soil biological properties		
6.	Soil microbial biomass carbon (MBC)	Jenkinson and Powlson, 1976
7.	Dehydrogenase activity	Casida Jr *et al.*, 1964

Soil colloidal polysaccharides

Colloidal exopolysaccharides was measured with 1g of air dried soil samples.

It was extracted by 10 mL of 100 mM EDTA which was added to1g dry soil and centrifuged at 3600 rpm for 15 min. The aliquot was transferred to a new pre-weighed centrifuge tube and precipitated by 5 mL dehydrated 70% cold ethanol in splits. Then the tube was kept in refrigerator overnight for precipitation and the centrifuged at 8000xg for 5 min and then oven dried at 60^{o}C until constant weight. Finally the weight of the precipitate along with the centrifuge tube was taken to measure the amount of colloidal exopolysaccharides.

Statistical analysis

All the data were subjected to statistical analysis using analysis of variance (ANOVA) at $p < 0.05$ levels. Further the treatment means were statistically differentiated by performing Duncan's Multiple Range Test (DMRT) at $p < 0.05$ levels. Statistically differentiated means were denoted by different alphabets.

Results

The results of the biofilm formation assay performed at particular time intervals *viz.*, 5th day, 10th day and 15th day indicated that the ratio of biofilm producing cells and planktonic cells of the yeast isolates decreased from 5th day 15th day. This indicates the stability of the yeast isolates to form biofilm over a period of time. The isolate OT3 8 showed increase in B/P ratio with increase in time interval (Fig. 1).

Molecular identification

Homology search of nucleotide sequence obtained from isolates OT3 5, OT3 2, OT3 8, OT3 12 and RT2 4 with other 18S rRNA gene sequences was carried out individually. The 18S rRNA sequence of the five

isolates OT3 5, OT3 2, OT3 8, OT3 12 and RT2 4 showed 98.5% homology with *Candida tropicalis*, *Pichia kudriavzevii*, *Pichia kudriavzevii*, *Candida tropicalis* and *Pichia kudriavzevii* respectively (Table 1).

Effect of soil yeasts on soil physical properties

The soil yeast isolate OT3 8 recorded geometric mean diameter (GMD) of 0.40 mm which is almost comparable with the positive biofilm former inoculated soil. On comparing with 15 DAI, GMD was found to be increased after 30 days after inoculation with soil yeasts (Fig. 2). The soil yeast isolates OT3 8 and OT3 12 positively influenced the mean weight diameter (MWD) of the soil over the period of incubation. The positive biofilm former recorded the highest MWD of 2.22 mm on 30 DAI (Fig. 3). In wet sieving method of soil aggregation analysis, the positive biofilm former showed more macro aggregates stability. The soil yeast isolates OT3 8 and RT2 4 showed high macro-aggregate stability on 30 DAI (Table 2). It was found that in all the treatments a gradual increase in macro aggregation of soil particles was observed with the incubation time.

Micro aggregation stability of OT3 2 inoculated soil was found and the results revealed that, the stability of micro aggregates also increased with the time (Table 2). The total water stable aggregates of the inoculated soil were found to be positively influenced by OT3 12 and OT3 8 on 15 DAI & 30 DAI. The soil yeast isolate OT3 2 recorded the highest dry aggregate ratio on 15 and 30 DAI (Table 3).

Effect of soil yeasts on soil chemical properties

Studies on the effect of soil yeasts on soil pH revealed that the pH of the soil remained neutral with due course of incubation after inoculation with the soil yeast isolates (Fig. 4). Inoculation of soil yeast isolate OT3 recorded higher soil organic carbon content of 7722.77µg g^{-1} and 8356.81 µg g^{-1} of soil after 15 and 30 DAI respectively (Fig. 5). Soil labile carbon status was found to be positively influenced due to the inoculation of soil yeast isolates (Table 4). The soil protein index due to the inoculation of soil yeast isolates ranged from 3.78 to 8.29 µg/g of soil on 15 DAI (Table 4).

Effect of soil yeasts on biological properties

The soil microbial biomass carbon content of the inoculated soil ranged from 472.59 to 977.78 µg/g of soil and 462.59 to 906.78 µg/g of soil on 15 and 30 DAI respectively. The isolate OT3 12 recorded the maximum value of 977.78 µg/g of soil and 906.78 µg/g of soil on 15 and 30 days after inoculation (Table 5). Higher dehydrogenase activity (0.64 µg TDF $g^{-1}day^{-1}$) was observed in OT3 8 inoculated soil on 15 DAI (Table 5). Soil colloidal polysaccharide content of the treated soils was found to be positively influenced by the microbial inoculation (Fig. 6).

Discussion

Plants are closely associated with soil, hence plant microbe interactions are not only essential for the plant in terms of their nutrition, growth promotion, bio-control, stress alleviation etc., but they also influence soil physical, chemical and biological properties through biogeochemical cycles

(Velmourougane et al., 2017). Several reports related to agriculturally important microbial biofilms are available from India and Srilanka (Velmourougane et al., 2017). Extensive studies were performed with cyanobacteria based bacterial and fungal biofilms and their phytopathogenic activity (Prasanna et al., 2013). The agronomic potential of leguminous crops was improved due to application of biofilmed preparations of *Anabaena/Trichoderma* with different agriculturally useful bacteria/fungi (Prasanna et al., 2014). Swarnalakshmi et al. (2013) reported that the soil biological and chemical properties were enhanced due to application of cyanobacterial-based biofilms in the wheat rhizosphere. According to Prasanna et al. (2015) microbial biofilm inoculation in flooded and SRI (System of Rice Intensification) rice recorded differential effect on plant growth and soil nutrient dynamics. The results of our study revealed that, all the sixteen yeast isolates possessed the ability to produce biofilm. Four isolates produced biofilm in higher amounts than the reference strains *Pseudomonas* sp. and one isolate produced in equal amounts with that of the reference strain. From the results of our study it has been concluded that a better understanding of yeast biofilm formation in crop rhizosphere and development of more effective biofilmed biofertilizers are potential areas for future research to sustain the agricultural productivity.

The regular application of organic manure increases the soil microbial count, improves soil characteristics and involved in mineral transformation (Fernández *et al.*, 1997; Bastida et al., 2008). The overall outcome of the experimental results showed some improvement in physical properties of soil like mean diameter, macro and micro-aggregates, dry aggregation ratio and water stable aggregates with the application of yeast isolates than the control (uninoculated) soil. Investigation of results revealed that the application of yeast inoculants contribute more to micro-aggregate formation. The possible mechanism underlying for this may be cementation of soil micro-aggregates due to EPS production by the inoculated yeast isolate which led to increase in micro-aggregate formation. Garcia-Franco et al. (2015) have stated that formation of micro-aggregates within macro-aggregates may increase soil stability. Microaggregates (< 250 µm) are formed from organic molecules *i.e.*, polysaccharides attached to clay (Cl) and polyvalent cations (P) to form compound particles (Cl–P–OM) (Tisdall, 1996). According to Le Bissonnais, (1996) unstable aggregations formed in soil led to low infiltration rate of soil. He also stated that the Mean Weight Diameter (MWD) of the soil aggregate at the range of 0.7 mm was considered as very unstable aggregates but 3.4 mm indicates very stable aggregates. In the present study, inoculation of OT3 8 and RT2 4 yeast isolates increased the MWD upto1.8 mm at 30 DAI and it is expected that it will be further improved with increase in incubation period.

Medina et al. (2004) reported that amendment of *Yarrowia lipolytica* is a useful tool to modify soil physico-chemical, biological and fertility properties that enhance the plant performance probably by making nutrients more available to plants.

Soil labile carbon makes up a fraction of the total carbon pool, but sensitive with turnover times of a few days to months. Soil labile carbon (SLC) (Parton et al., 1987) is one of the biological indicator for the soil biological quality index scaling. Li et al., (2018) reported that addition of organic manures increased the pool of stable carbon along with increased concentration and proportion of soil labile carbon in the soil

surface layer. In our study, inoculation of OT3 12 yeast isolate and OT3 8 recorded significant increase in soil labile carbon status on 15 and 30 days of incubation period. Literatures support that labile carbon pool influences the decomposition rate and acts as a major source of nutrient for soil microbes. Increase in soil labile carbon in the present investigation revealed that, the microbial activity is higher in the soils inoculated with soil yeast isolates when compared to the uninoculated control.

Other than organic carbon, some amount of protein and protein like substances present in the soil is referred as soil protein index. Organically bound nitrogen present in the soil organic carbon which is known as protein index influences the storage of nitrogen and makes it available to plants. It is also associated with soil aggregation and make the availability of water to plant growth (Moebius-Clune, 2016). From the present investigation, in the yeast isolates inoculated soils the protein content was reduced over the period of incubation and this indicates that yeast isolates have utilized the proteins present in the soil as a source of nitrogen for its growth.

The soil organic carbon, representing total soil carbon content is a measure of all carbonaceous material derived from living organisms. It is also known as the representation of soil aggregation, water holding capacity, nutrient stocking and also the energy and carbon source for the microbial diversity of the soil. The Indian agricultural soils are generally recognized as low in soil organic matter (Lal, 2002). Soil organic carbon (SOC) serves as a source and sinks for nutrients and plays a major role in maintenance of soil fertility.

Diverse microorganisms present in soil ecosystem are responsible for decomposition of the organic carbon fraction like cellulose, lignin, hemicelluloses, chitin and lipids present in soil organic matter (Khatoon et al., 2017). Microbial biomass carbon and soil organic carbon ratio was highest in soil receiving continuous poultry manure, due to increased activity of microorganisms (Kaur et al., 2005). Moreover, green manuring is considered a good agricultural practice because of its positive effect on soil fertility, quality and biodiversity (Stark et al., 2007), because green manuring increases the abundance and diversity of microbes. The soil organic carbon was found to be increased in the soil inoculated with the soil yeast isolate OT3 8. The increase in soil organic carbon due to inoculation of soil yeasts reveals the increased activity of yeast isolates in the soil. Production of polysaccharides by the inoculated yeast isolates may possibly enhance water retention in the microbial environment and regulates the diffusion of carbon sources such as glucose.

Soil pH is one of the most influential factors that affect the soil microbial community. It affects the carbon availability, nutrient availability, and the solubility of metals.

Soil pH also controls many biotic factors like fungal and bacterial biomass

(Rousk et al., 2009). In the present study, there was a slight reduction in the pH of the soil after inoculation with the yeast isolates. Medina et al. (2004) reported that application of yeast strain *Yarrowia lipolytica* changed the pH of the soil from 8.90 to 8.75. This is in correspondence with the findings of our

study. The slight reduction in the soil pH may be due to the production of organic acids by the soil yeast isolates.

Organic content of the soil is the representation of soil enzyme activity

(Beare and Bruce, 1993). Soil dehydrogenase activity is a good measure of microbial activity in the soil as it acts as the function of oxidative activity and microbial count

(Nannipieri et al., 2003). The dehydrogenase activity was directly proportional to microbial population, nutrient and organic content (Masto et al., 2007; Kumar et al., 2013) of the soil (Masto et al., 2006). The dehydrogenase activity was found to be positively influenced by the inoculation of soil yeasts. Our findings are in accordance with Medina et al. (2004),

who reported that in *Yarowia lipolytica* amended soil the dehydrogenase activity was increased. Hence, in the present study, the increase in dehydrogenase activity indicates the increment in microbial population and promotion of organic content by microbial activity. The dehydrogenase activity was also positively correlated with the other biological variables used in the study.

Microbial biomass carbon can act as a potential indicator to soil quality by responding to management practices. Application of farmyard manure increases the microbial biomass carbon of the soil as reported by (Marschner et al., 2003). Organic management increases the microbial biomass carbon and microbial activity in soil which tends to make nutrients available for crop growth (Zaman et al., 1999; Marinari et al., 2006; Tu et al., 2006; Wang et al., 2007). The application of organic manures in soil was found to increase the MBC, because the manures supply the essential nutrients for microbial growth. In our present study, a slight reduction in MBC was observed and this may be due to the exploitation of organic carbon source present in the soils for the growth of the yeast isolates.

Soil microorganisms produce various intra and extracellular polysaccharides,

which play an important role in the life of microorganisms and have great practical application. The properties of the exopolysaccharides contained in the extracellular polymeric substances (EPS) have been well documented, but the properties of colloidal polysaccharides produced by certain microorganism still remain unexplored (Markosyan et al., 2017). Wherever the multiplication of microorganisms is high, those soils contain high amount of soil polysaccharides and these have a major contribution to stabilize the soil aggregate formation. Microbiota of soil is highly responsible for soil polysaccharide synthesis. Hence it is very tedious process to separate the pure polysaccharide from the soil. The extraction of these polymers was related to soil stability and aggregation. In the present study it was evident that due to inoculation of yeast isolate OT3 8, the soil colloidal polysaccharide was increased. From the results of the present study it is evident that the soil yeast isolates possess the ability to produce soil colloidal polysaccharides. Markosyan et al. (2017) investigated the properties of a colloidal polysaccharide of the iron oxidizing chemolithotrophic bacteria *Leptospirillum ferriphilum* newly isolated in Armenia and reported that it consists of three monomers *viz*., glucose, fructose, mannose.

Conclusion

Identification of potential soil yeasts will serve as a good alternative to improve the soil quality. Application of microbial inoculants for the improvement of soil quality is one of the efficient and eco-friendly methods for improving soil health. The research on soil yeasts is still in its infant stage and the soil yeasts remains as an unidentified component as microbial inoculants. From our study it is evident that the soil yeasts possess potential plant growth promoting traits and mechanisms for soil quality improvement. Further studies on effect of soil yeasts on plant growth promotion through pot culture and field trials will pave way for identifying a potential yeast candidate and can be exploited for sustaining the production of crop plants and soil quality improvement.

Declarations

Competing interests: The authors declare no competing interests

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Tables

Table 1. Molecular identification of soil yeast isolates

Isolate	Source	Accession number of Homologous microorganisms	Homologous microorganisms	Identity
OT3 5	Orchard Soil (papaya)	FN652303	*Candida tropicalis*	98%
OT3 2	Orchard Soil (grapes)	EF198000	*Pichia kudriavzevii*	99%
OT3 8	Orchard Soil (grapes)	LC389030	*Pichia kudriavzevii*	99%
OT3 12	Orchard Soil (Sapota)	CP028531	*Candida tropicalis*	98.5%
RT2 4	Gardenland Soil (Redfort)	KT715474	*Pichia kudriavzevii*	98.5%

Table 2. Effect of soil yeast isolates on the stability of macro aggregates and micro aggregates of soil

Treatment	Macro-aggregate stability (4-0.25 mm) (%)		Micro aggregate stability (0.25-0.05 mm) (%)	
	15 DAI	30 DAI	15 DAI	30 DAI
OT3 5	3.34±(0.78)^{c}	5.60±(0.76)^{e}	90.37±(4.01)^{a}	111.03±(11.86)^{a}
OT3 2	4.26±(0.58)^{c}	5.92±(0.41)^{e}	90.45± (6.99)^{a}	127.03±(14.00)^{a}
OT3 8	9.84±(0.89)^{b}	31.31±(2.91)^{b}	45.50±(1.80)^{bc}	61.07±(14.47)^{bc}
OT3 12	6.78±(0.32)^{bc}	9.42±(0.24)^{e}	100.83±(1.68)^{a}	103.28±(5.54)^{a}
RT2 4	9.06±(0.39)^{b}	25.85±(1.43)^{c}	39.28±(3.10)^{c}	110.21±(14.98)^{a}
Positive control	13.34±(2.69)^{a}	40.22±(2.08)^{a}	21.71±(2.00)^{d}	35.79±(5.64)^{c}
Negative control	3.80±(0.57)^{c}	4.86±(0.60)^{e}	52.11±(1.42)^{b}	68.89±(6.62)^{b}
Uninoculated control	4.10±(0.87)^{c}	20.56±(1.16)^{d}	53.71±(2.16)^{b}	54.94±(1.52)^{bc}

DAI – Days after inoculation
Data represent by mean±(SE). Values are means of three replicates, and the values with the same lower case letter within a column indicate, there is no significant difference according to Duncan's test ($P < 0.05$).

Table 3. Effect of soil yeast isolates on total water stable aggregates and dry aggregate ratio

Treatment	Total water stable aggregates (%)		Dry aggregate ratio	
	15 DAI	30 DAI	15 DAI	30 DAI
OT3 5	25.19±(3.40)^{bc}	32.36±(1.03)^{de}	3.06±(0.32)^{a}	3.08±(0.25)^{ab}
OT3 2	26.96±(2.93)^{b}	33.25±(1.12)^{cde}	3.11±(0.39)^{a}	3.46±(0.39)^{a}
OT3 8	33.05±(1.21)^{a}	39.70±(2.70)^{bc}	2.91±(0.09)^{a}	2.96±(0.19)^{ab}
OT3 12	36.32±(0.69)^{a}	58.92±(4.13)^{a}	2.30±(0.10)^{ab}	2.46±(0.28)^{bc}
RT2 4	19.70±(0.79)^{cd}	45.71±(1.40)^{b}	2.94±(0.47)^{a}	2.88±(0.33)^{ab}
Positive control	16.67±(1.55)^{d}	39.99±(0.93)^{bc}	1.90±(0.29)^{b}	1.95±(0.25)^{c}
Negative control	27.13±(1.98)^{b}	29.57±(2.07)^{e}	1.85±(0.16)^{b}	1.82±(0.14)^{c}
Uninoculated control	16.46±(2.00)^{d}	28.44±(0.41)^{e}	0.78±(0.13)^{c}	0.85± (0.11)^{d}

DAI – Days after inoculation
Data represent by mean±(SE). Values are means of three replicates, and the values with the same lower case letter within a column indicate, there is no significant difference according to Duncan's test ($P < 0.05$).

Table 4. Effect of soil yeast isolates on soil labile carbon and soil protein index

Treatment	Soil labile carbon (%POXC in SOC)		Soil protein index (SPI)(µg g^{-1} of soil)	
	15 DAI	30 DAI	15 DAI	30 DAI
OT3 5	22.15±(3.87)^{a}	15.69±(1.23)^{bc}	7.59±(0.55)^{abc}	4.66±(0.08)^{ab}
OT3 2	23.18±(1.96)^{a}	14.85±(3.66)^{bc}	6.89±(0.16)^{c}	4.98±(0.46)^{ab}
OT3 8	14.08±(3.36)^{bc}	29.07±(5.31)^{ab}	7.20±(0.40)^{bc}	5.89±(0.20)^{ab}
OT3 12	20.51±(1.10)^{ab}	39.85±(8.84)^{a}	7.67±(0.09)^{abc}	4.40±(0.94)^{ab}
RT2 4	10.88±(1.67)^{c}	21.05±(7.71)^{bc}	7.96±(0.24)^{ab}	6.22±(0.86)^{ab}
Positive control	8.90±(4.70)^{c}	17.71±(2.06)^{bc}	8.29±(0.46)^{a}	6.01±(0.43)^{ab}
Negative control	11.77±(2.97)^{c}	15.82±(2.73)^{bc}	7.99±(0.16)^{ab}	6.39±(0.51)^{a}
Uninoculated control	7.72±(1.26)^{c}	11.48±(0.15)^{c}	3.78±(0.09)^{d}	4.06±(0.97)^{b}

DAI – Days after inoculation
Data represent by mean±(SE). Values are means of three replicates, and the values with the same lower case letter within a column indicate, there is no significant difference according to Duncan's test ($P < 0.05$).

Table 5. Effect of soil yeast isolates on soil microbial biomass carbon (MBC) content and dehydrogenase activity

Treatment	Soil Microbial Biomass Carbon (MBC) ($\mu g\ g^{-1}$)		Dehydrogenase activity (μg TPF $g^{-1}day^{-1}$)	
	15 DAI	30 DAI	15 DAI	30 DAI
OT3 5	602.96±(58.76)^{bc}	569.96±(54.36)^{e}	0.45±(0.04)^{bc}	0.50±(0.05)^{ab}
OT3 2	684.44±(48.89)^{bc}	634.44±(83.96)^{d}	0.54±(0.01)^{ab}	0.52±(0.06)^{ab}
OT3 8	570.37±(16.30)^{c}	667.37±(73.89)^{cd}	0.64±(0.11)^{a}	0.62±(0.07)^{ab}
OT3 12	977.78±(74.68)^{a}	906.78±(25.10)^{a}	0.53±(0.02)^{ab}	0.57±(0.04)^{ab}
RT2 4	619.26±(71.03)^{bc}	635.26±(59.26)^{d}	0.46±(0.03)^{bc}	0.51±(0.08)^{ab}
Positive control	700.74±(32.59)^{bc}	689.74±(63.64)^{c}	0.62±(0.01)^{a}	0.77±(0.19)^{a}
Negative control	847.41±(81.48)^{ab}	813.41±(38.47)^{b}	0.35±(0.02)^{bc}	0.42±(0.06)^{b}
Uninoculated control	472.59±(181.47)^{c}	462.59±(60.48)^{f}	0.31±(0.02)^{c}	0.32±(0.04)^{b}

DAI – Days after inoculation
Data represent by mean±(SE). Values are means of three replicates, and the values with the same lower case letter within a column indicate, there is no significant difference according to Duncan's test ($P < 0.05$).

Figures

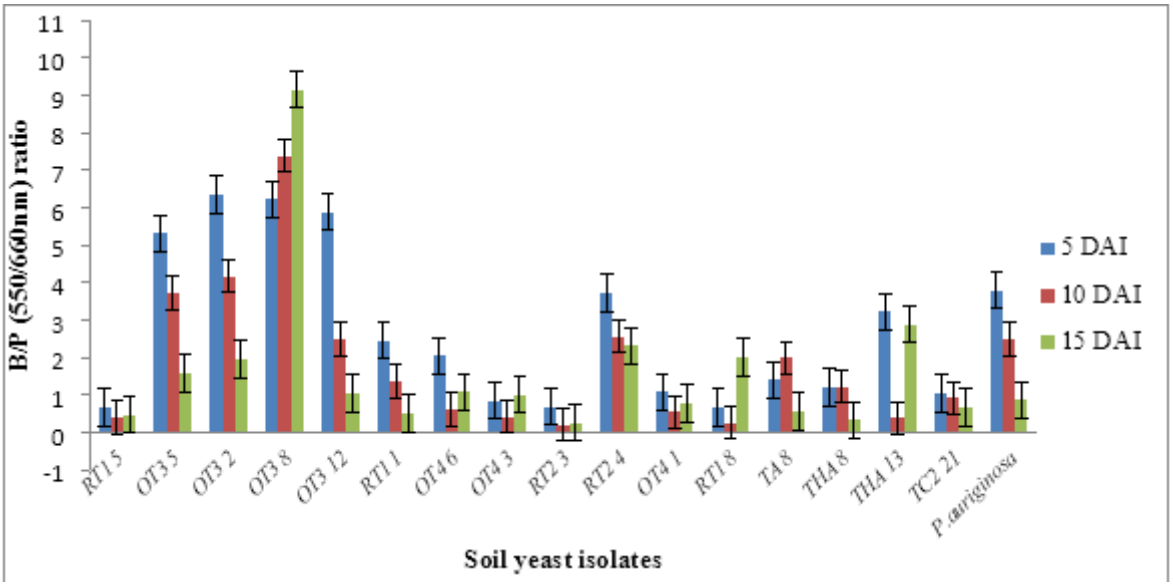

Figure 1

Comparison between the biofilm forming ability of soil yeast isolates; B/P ratio – Ratio of biofilm producing cells and planktonic cells, DAI-Days After Inoculation, Soil yeast isolates – RT1 5, OT3 5, OT3 2, OT3 8, OT3 12, RT1 1, OT4 6, OT4 3, RT2 3, RT2 4, OT4 1, RT1 8, TA 8, THA 8, THA 13, TC2 21, Pseudomonas aeruginosa (Positive biofilm former)

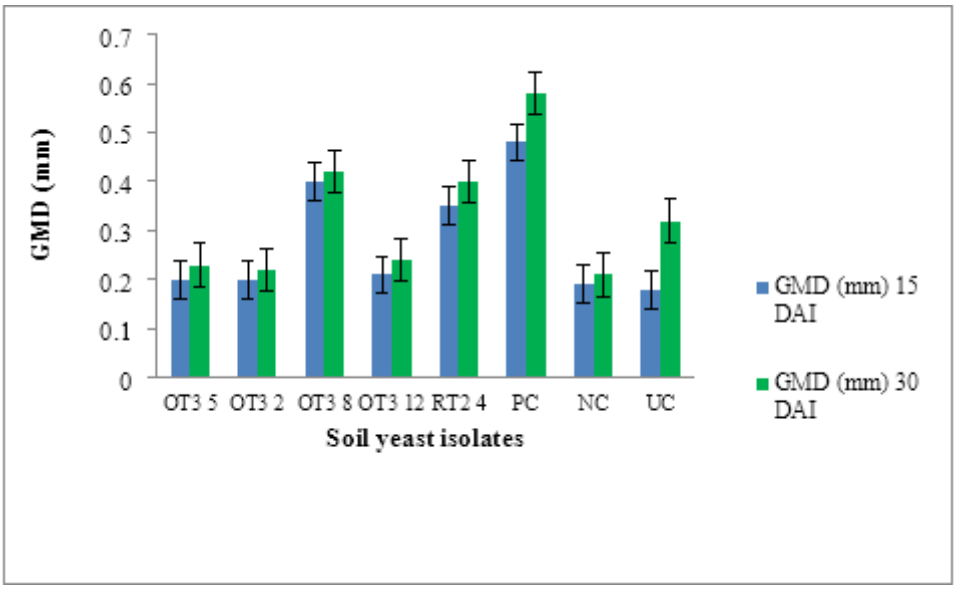

Figure 2

Changes in Geometric Mean Diameter (GMD) due to inoculation of soil yeast isolates; GMD – Geometric Mean Diameter, DAI-Days After Inoculation, Soil yeast isolates – OT3 5, OT3 2, OT3 8, OT3 12, RT2 4, PC-Positive Control (Aeromonas hydrophila), NC-Negative Control (Baker's yeast- Saccharomyces cerevisiae), UC-Uninoculated control (without inoculums)

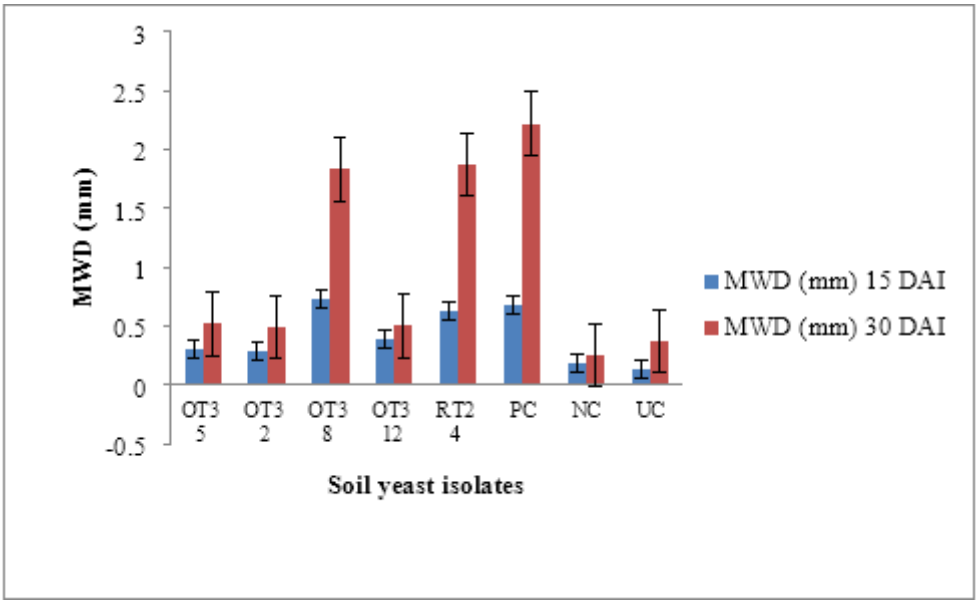

Figure 3

Changes in Mean weight diameter (MWD) due to inoculation of soil yeast isolates; MWD – Mean Weight Diameter, DAI-Days After Inoculation, Soil yeast isolates – OT3 5, OT3 2, OT3 8, OT3 12, RT2 4, PC-Positive Control (Aeromonas hydrophila), NC-Negative Control (Baker's yeast- Saccharomyces cerevisiae), UC-Uninoculated control (without inoculums)

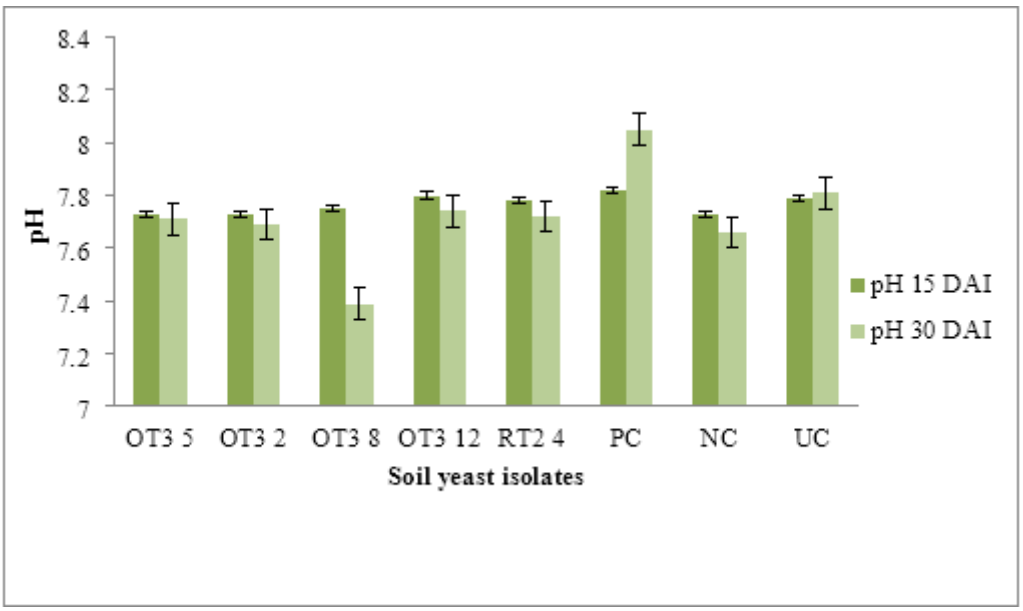

Figure 4

Changes in soil pH due to inoculation of soil yeast isolates; pH – hydrogen ion concentration, DAI-Days After Inoculation, Soil yeast isolates – OT3 5, OT3 2, OT3 8, OT3 12, RT2 4, PC-Positive Control (Aeromonas hydrophila), NC-Negative Control (Baker's yeast- Saccharomyces cerevisiae), UC-Uninoculated control (without inoculums)

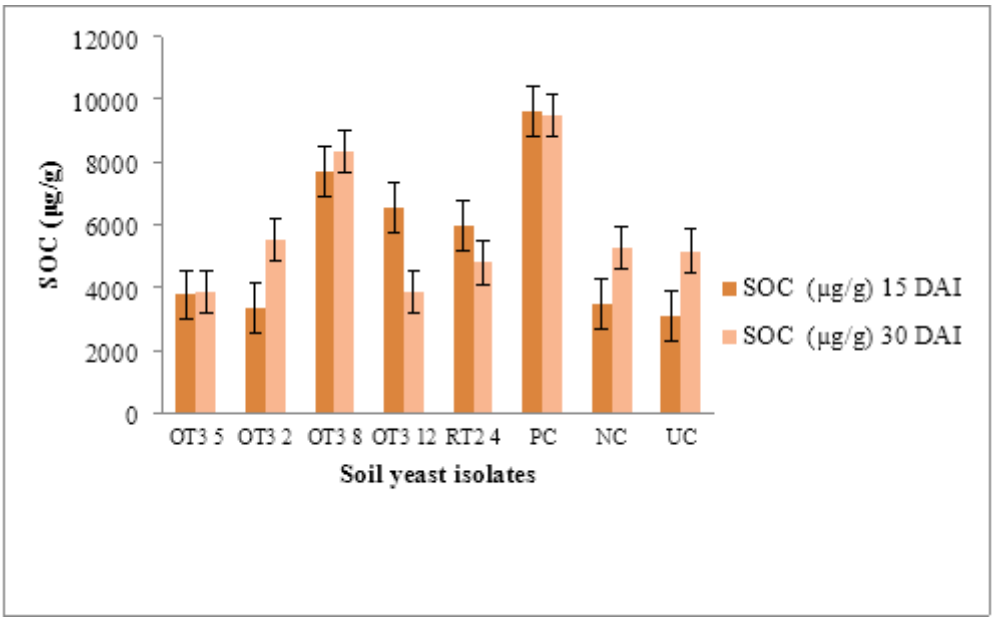

Figure 5

Changes in Soil organic carbon due to inoculation of soil yeast isolates; SOC – Soil Organic Carbon, DAI-Days After Inoculation, Soil yeast isolates – OT3 5, OT3 2, OT3 8, OT3 12, RT2 4, PC-Positive Control (Aeromonas hydrophila), NC-Negative Control (Baker's yeast- Saccharomyces cerevisiae), UC-Uninoculated control (without inoculums)

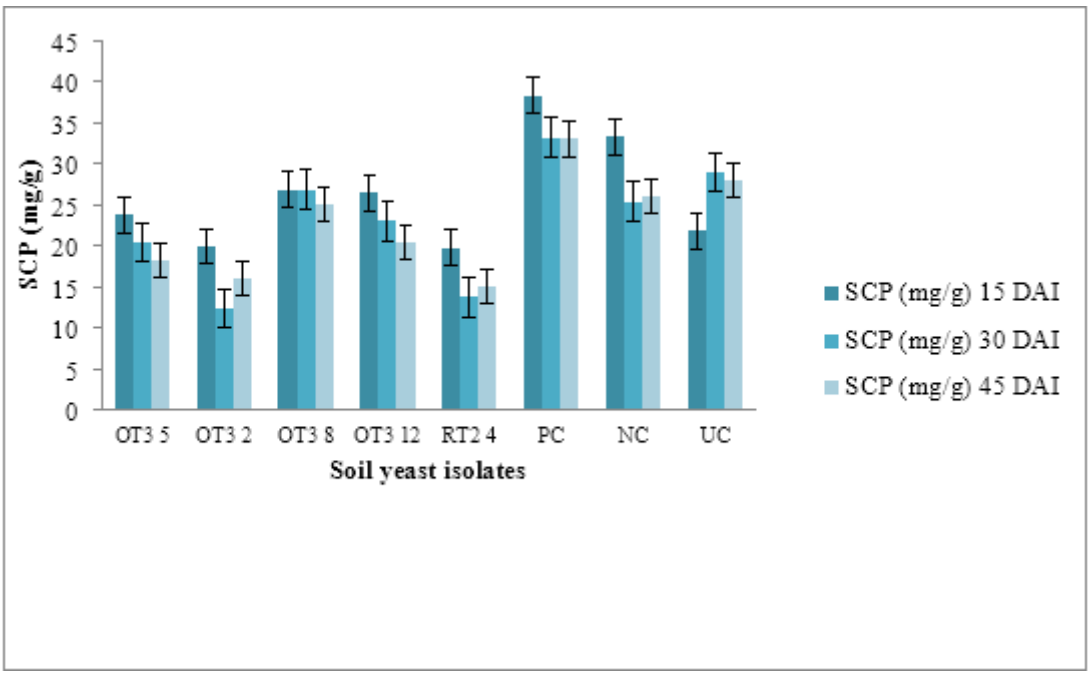

Figure 6

Changes in soil colloidal polysaccharides content due to inoculation of soil yeast isolates; SCP – soil colloidal polysaccharides content, DAI-Days After Inoculation, Soil yeast isolates – OT3 5, OT3 2, OT3 8, OT3 12, RT2 4, PC-Positive Control (Aeromonas hydrophila), NC-Negative Control (Baker's yeast-Saccharomyces cerevisiae), UC-Uninoculated control (without inoculums)