Plant Pathogen Suppression the Synergistic Effect between Biofertilizer and Irradiated Oligochitosan of Tomato

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Abstract: Evaluation of the synergy effect between biofertilizer and irradiated oligochitosan (from Dr. Yokoyama) was conducted to determine the growth and yield of the test plants. Study for synergistic effect of biofertilizer and irradiated oligochitosan, some positive effect such as plant growth promoter and pathogen suppression in tomato yield in the greenhouse pot experiment. It was guessed that oligochitosan induced resistance for seedling treated. Data showed synergistic effect using disease control intensities, in this case, synergistic effect between biofertilizer and oligochitosan is clearly positive.

Keywords: Rhizobacterial biofertilizer, number of fruit, fruit weight, pathogen, resistance

1. INTRODUCTION

In order to supply food to increasing population, agricultural production should be increased and a large amount of fertilizer is required. Fertility of the soil increases due to the continuous use of the inorganic fertilizers but it also reduces the crop productivity. Soil pollution is caused due to the use of inorganic fertilizers, pesticides, and other chemicals etc (Badoni, 2006). Martinez et al. (1993) Cuba, reported that soil inoculation with Azotobacter increased tomato seed germination by 33-46 per cent, shortened the period between sowing and transplanting by 5-7 days, increased the yield by 38-60 per cent. Bio-fertilizers are the carrier-based preparations containing mainly effective strains of microorganisms in sufficient number, which are useful for nitrogen fixation. If they are used in association with macronutrients the expected yields per unit area may be much higher. Amongst these nutrients, nitrogen is the only nutrient, which play major role in synthesis of chlorophyll, amino acids and protein building blocks, which is ultimately responsible for higher source to sink ratio.

The main advantage of bio-fertilizer is that it does not pollute the soil and also does not show any negative effect to environment and human health [5].

Plant growth promoting rhizobacteria (PGPR) are free living bacteria (Azotobacter chroococcum, Pseudomonas spp., Azoarcus spp.) commonly found in soil and in association with plant roots, including important agricultural crops such as wheat. Some bacteria convert atmospheric nitrogen into soil nitrogen and some bacteria help in the solubilization of insoluble phosphates, improving P uptake from soil. In 2001 the institute successfully launched its biofertilizer product under the trade name of Rhizobacterial fertilizer, which reduces the input cost of fertilizers and increases crop yield. The Rhizobacterial fertilizer is a low cost and environment friendly product and can be used to enhance the yield of all crops as well as soil fertility.

Oligochitosan is a low molecular weight chitosan and it can be obtained by γ-ray irradiation to chitosan. It has the effect of promoting the growth of plants such as rice, barley and soybean. In order to evaluate the synergy effect between biofertilizer and irradiated oligochitosan on plant pathogen suppression, tomato seedlings, which were inoculated by Pseudomonas fluorescens strain FPH9601, were transplanted to the infected field by tomato bacterial wilt and irradiated oligochitosan 100 ppm solution was sprayed. By the application of irradiated oligochitosan, suppression effect against the pathogen by Pseudomonas fluorescens strain FPH9601 was enhanced, but it became lower in higher infected field. Oligochitosan have been reported to increase growth and several crops. The seed yield was higher in 100 ppm chitosan might be due to increase number of seeds. Fusarium diseases widely distributed in soil is known as a plant pathogen [9].

Fusarium spp crown and root rot is an important soil-borne disease, with the potential to limit productivity in glasshouse and field tomato crops. The causal agent, Fusarium oxysporum and f. spp. increased early injury to the roots and collar of tomato plants caused by Forl was also observed in Tunisia [3], where yield losses were reported to range between 20 and 60%. Fungicides are of little
use on most Fusarium diseases widely distributed in soil is known as a plant pathogen. Biological control of Fusarium wilts, in the form of natural microbial populations in soils, has been recognized for over 70 years.

2. MATERIALS AND METHODS

An experiment to evaluate the effects of multifunctional biofertilizer and irradiated oligochitosan on the growth of tomato plants was conducted in the greenhouse.

Experimental design was RCBD with 4 replications. Treatments as fallow;

- Oligochitosan 7(every week) T-1
- Oligochitosan 14 (every 2 weeks) T-2
- Rhizobacterial biofertilizer + Oligo 7 T-3
- Rhizobacterial biofertilizer + Oligo 14 T-4
- Rhizobacterial biofertilizer T-5
- Control T-6

Tomato variety Iyulskii (Russian variety) was used in this study.

Pot contained 20 kg of sterile soil. Pathogenic fungus is *Fusarium spp.* The present study was conducted to reduce soil-borne diseases caused by *Fusarium spp.*, and to increase the yield of tomato field under natural conditions through the application of biofertilizer and chitosan based-treatments at different methods.

Before one week pathogenic fungus mixed to the sterile soil and then planted seedling.

For rhizobacterial biofertilizer inoculation treatment seed were coated at sowing. In conclusion the effect of integrated use of Oligochitosan, with *Fusarium* on tomato seedling were investigated in this experiment. The performances of tomatoes seedling were spraying the oligochitosan solution (100ppm) every week and every two weeks during the 2 months.

Identified *Fusarium spp* and observed pathogenic fungal culture morphology; mycelium and spores by using Dr. Katsuhiko Ando’s “Identification of microscopic Fungi” book [6].

Biocontrolled *Fusarium spp* fungi: Mixing bacteria of PGPR and Oligochitosan from Japan, as were transferred on PDA with *Fusarium spp* fungi and observed growth of mycelium and result.

The analysis of variance and interpretation of data were done as per procedures given by Fisher and Yates (1963), Panse and Sukhatme (1967) and Gomez and Gomez (1984). Level of significance used in ‘F’ test was P=0.05 critical difference (CD) values were calculated only wherever the ‘F’ test was found significant.

3. RESULTS AND DISCUSSION

Concerning synergistic effects among biofertilizers and oligochitosan, we investigated two targets; 1) plant growth promotion activities and 2) disease control intensities.

3.1. Plant Growth Promotion Activities

Survey tomato biofertilizer effect on the growth of the highest measured at 40 days after planting. Effect of treatment and biofertilizer+Oligo 14 on number of fruits per plant was found significant (Fig.1).

In biofertilizers application the higher number of fruits per plant was noticed (23) over without biofertilizers application (17). Among the treatments significantly higher number of fruits per plant was noticed in biofertilizer with oligochitosan every two week (26).

The significant difference in seed yield per plant was noticed in treatment and biofertilizers, where the interaction effect was also found to be significant (Fig1). Significantly higher fruit yield per plant was noticed in biofertilizer with oligochitosan every two week application (1147.3 g) as compared to without biofertilizers application (761.2 g).
It is well known that oligochitosan related materials induce plant defense mechanisms to plant pathogens. Data showed synergistic effect using disease control intensities, in this case, synergistic effect between biofertilizer and oligochitosan is clearly positive /Table 1/.

### Table 1. Synergistic effect between biofertilizer and irradiated oligochitosan of yield under contaminated soil by *Fusarium sp*

<table>
<thead>
<tr>
<th></th>
<th>Rhizobacterial biofertilizer</th>
<th>Rhizobacterial biofertilizer + Oligochitosan 14</th>
<th>(Rhizobacterial biofertilizer + Oligochitosan 14)/(Rhizobacterial biofertilizer)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Height of plant, cm</td>
<td>67.4</td>
<td>73.3</td>
<td>108.8</td>
</tr>
<tr>
<td>Number of fruit per plant</td>
<td>22</td>
<td>24</td>
<td>109.1</td>
</tr>
<tr>
<td>Weight of fruit per plant</td>
<td>1114.0</td>
<td>1147.3</td>
<td>103</td>
</tr>
</tbody>
</table>

However, concerning synergistic effect using plant growth parameters, there are very difficult to distinguish efficacy of oligochitosan to plant growth from those of biofertilizer tested.

### 3.2. Results of Disease Control Intensities

Plant growth promoting Rhizobacteria (PGPR) belonging to *Pseudomonas* spp. are being exploited commercially for plant protection to induce systemic resistance against various pests and diseases. Mixtures of different PGPR strains have resulted in increased efficacy by inducing systemic resistance against several pathogens attacking the same crop. The performance of PGPR has been successful against certain pathogens, insect and nematode pests under field conditions [9].
In addition to the cytological modifications, induction of resistance can result from biochemical reaction. In this case, addition of the non-pathogenic strain of *F. oxysporum* to tomato plants before inoculation with *Fusarium oxysporum* f. sp. *lycopersici* increased the chitinase, glucanase, and glucosidase activities in treated plants (7).

The result of in vitro experiment, where Oligochitosan with concentration of 100 ppm and Rhizobacteria BF added to potato dextrose medium, infected fungus showed, that both two agents are little effective on inhibition growth of *Fusarium sp* and fungus spores was obviously lower than that of the control without the treatment (fig3).

![Control, Rhizobacterial BF, Oligochitosan](image1.jpg)

**Fig3. Antifungal effects of Oligochitosan and Rhizobacterial BF**

Also soil testing from the pot experiment was conducted for detecting *Fusarium on* potato dextrose medium without fungus infects at 28°C 7 days later when grown in an incubator.

The combination of Rhizobacteria seed treatment and spaying Oligochitosan in every 7 days and along the seed treatment Rhizobacteria BF had disease control effect, if compared to control and to other variant of the experiment (fig4). Thus, spores of *Fusarium* fungus wasn’t observed on culture medium.

![T1, T2, T3, T4, T5, T6](image2.jpg)

**Fig4. Soil testing contaminated by Fusarium spp**

In some literature has been reported the similarly results as of our experiment. Plant growth promoting Rhizobacteria (PGPR) belonging to *Pseudomonas spp*. are being exploited commercially for plant protection to induce systemic resistance against various pests and diseases. Mixtures of different PGPR strains have resulted in increased efficacy by inducing systemic resistance against several pathogens attacking the same crop. Seed-treatment with PGPR causes cell wall structural modifications and biochemical/physiological changes leading to the synthesis of proteins and chemicals involved in plant defense mechanisms [7].
According to microscopic fungus, spores of *Fuzarium spp* are detected in control and variant of spraying Oligochitosan in every 7 days on tomato plant. The other spores were low activity and opaque (fig 4, 5).

**Fig5. Observation of spores by electron microscop**

4. CONCLUSION

The oligochitosan and biofertilizer have synergy effect on growth of tomato plant in the greenhouse and it can be used for getting maximum fruit yield.

The higher fruit yield per plant was noticed in biofertilizer with oligochitosan every two week application. Fruit yield was increased 1.5 times as compared to without biofertilizers application.

These results suggest that combination of both agents can be used for promoting growth and increasing yield and promising natural fungicides for disease control caused by *Fuzarium spp* on tomato plant.

REFERENCES

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