**Technical Summary**

**Use of Silver nanoparticles as an alternative to conventional fungicides to manage white mold in soybeans**

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The goals of this project were

1(a): to evaluate the inhibition effects of laboratory prepared silver nanoparticles against *Sclerotinia sclerotiorum* isolates in-*vitro*

1(b): to evaluate the efficacy of silver nanoparticles in field condition using multiple dosages, alone and in combination of recommended dosage of BallardPLUS®, (has active ingredient of a biological control organism, *Bacillus pumulis* QST 2808) to control *Sclerotinia sclerotiorum*, and 2: to determine the possible entry of silver nanoparticles into food chain through residue analysis and electron microscopic visualization.

**Objective 1(a):** to evaluate the inhibition effects of laboratory prepared silver nanoparticles (SNP’S) against *Sclerotinia sclerotiorum* isolates *in-vitro*

**Silver Nanoparticles (SNP’s):** The SNP’s were prepared in the laboratory of the Co-PI Dr. Bezbaruah using methodologies of sodium borohydride reduction of silver nitrate and using green tea plant leaf extract with size control and modification of the nanoparticle surface. Green tea nanoparticle were successfully prepared in Dr. Brezbaruah’s lab and the efficacy of laboratory prepared SNP’s was tested and compared with commercial SNP’s under field condition. *S. sclerotiorum* isolates have been recovered from sclerotia collected in the previous year from infected soybean stems. Sclerotia were plated on Potato Dextrose Agar culture media to recover the isolates by hyphal tipping after three days of growth. Out of twenty three isolates of *S. sclerotiorum* recovered; five isolates have been randomly picked and were tested for SNP’s sensitivity by determining the EC50 values (the effective concentration at which 50% of fungal growth reduction occurs) twice at Langdon Research and Extension Center /Plant Pathology Lab. The concentrations of SNP’s used in the sensitivity test were 0, 1, 10, 100, 500 and 1000 µg/ml and the most effective concentrations were selected for the field research study.

**Results:**

Based on the EC50 values the SNP’S showed growth reduction at very high concentration (1000 µg/ml) on *S. sclerotiorum* isolates. The five white mold isolates tested have the EC50 values ranging from 211 µg/ml to 703 µg/ml (Fig. 1) indicating SNP’s as of alone do not have fungicidal properties below 100 µg/ml. Based on these results we selected concentrations of 500 and 1000 µg/ml for field studies.

**Figure 1:** EC50 values of white mold isolates tested on Potato Dextrose Agar culture media amended with silver nanoparticles.

**Objective 1 (b):** to evaluate the efficacy of SNP’s in field condition using multiple dosages alone and in combination of recommended dosage of BallardPLUS® (has active ingredient of a biological control organism, *Bacillus pumulis* QST 2808) to control *Sclerotinia sclerotiorum*

A field study was laid out at Langdon Research Extension Center in a Randomized Complete Block Design (RCBD) arranged in split-plot with application timings (R1 and R4 stage of soybeans) as whole plots and fungicides (alone and combination of biologicals plus SNP’s) as sub-plots and was replicated four times. All the treatments were compared with non-treated control along with standard checks of conventional fungicide Endura, and biological fungicide BallardPlus which were applied at R1 stage of soybean as per commercial recommendation practices to manage white mold using standard doses. Supplemental irrigation was provided to enhance white mold disease incidence during flowering season.

**Results:**

The mean white mold incidence on soybean was 9.5% in non-treated control with mean severity of 2.5% and has higher index (percent white mold incidence x severity/100) when compared with the other treatments of the current research trial. Whereas, the lowest incidence of white mold was in the combination treatment of commercial grade SNP’s at 1000 µg/ml with biological fungicide the Ballard Plus when applied at R1 stage of the soybean. The white mold disease incidences and severities obtained in the treatments were converted to white mold index as shown in Figure 2. None of the treatments were significantly different statistically from each other at p≥0.05 at any applied growth stage and there were no differences observed among SNP’s prepared in the laboratory and commercial obtained SNP’s.

**Note:**

1. SNP: Commercial grade Silver nanoparticle

2. LabSNP: Silver nanoparticles prepared in the lab

3. BPLUS: is BallardPlus (trade name of biological fungicide (*Bacillus pumulis* QST 2808))

4. Concentration of SNP’s used in the field study: 500, 1000 µg/ml

**Figure 2**: Efficacy of silver nanoparticles tested (SNP’S) alone and in combination of biological fungicide BallardPlus (BPLUS) on white mold (mean Index) at R1 and R4 growth stages of soybeans under field condition

**Objective 2**: To determine the possible entry of silver nanoparticles into food chain through residue analysis and electron microscopic visualization.

**Work in progress:** To determine the possible entry of SNP’s into food chain through residue analysis and electron microscopic visualization; seed samples from the treatments of the field research are being processed in Dr. Bezbarauh’s laboratory after the crop harvest.

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