

## **Soy Polyol Formulations As Novel Seed Treatments For The Management of Soil-Borne Diseases of Soybean**

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### **ABSTRACT**

Polyurethanes prepared from vegetable oils display a number of desirable properties useful for many commercial and industrial applications. One unique application is that of an agricultural seed treatment. Seed treatments are used to incorporate pesticides onto the seed coat and to decrease the disease susceptibility of the seed during its germination in the soil. In addition, by altering the movement of water across the seed coat and by incorporating protective pesticides in the coating, seed coating polymers can enhance the germination and survival of the seed under adverse environmental conditions. Soy polyols alone, and in combination with glycerin, polymerized with 4,4'-diphenylmethane diisocyanate (MDI) were studied for their seed treating properties and impact on soybean seed germination. The cross-linking density and properties of these polyurethane compounds were varied by changing the isocyanate/hydroxyl molar ratio. In order to optimize the coating qualities and to increase the efficiency of the coating, acetone was also studied as a diluting solvent to reduce the viscosity of the polyurethane mixture prior to polymerization on the seed coat. Optimal polymerization and resulting germination (95%) were obtained using a 1:1 isocyanate/hydroxyl molar ratio consisting of a mixture of soy polyol 180 and glycerin, and the use of an equal volume of acetone as a dilution solvent. This optimal polyurethane seed treatment had several desirable qualities including: reduced viscosity, decreased seed coating thickness, increased seed coating uniformity and permitted larger volumes of seed to be treated with the same volume of polymer. This optimal seed treatment increased the soybean seed germination by 15%, as compared with untreated seed. In addition, preliminary studies of the compatibility of these unique formulations with commercial and experimental fungicides also support the use of these polymers as seed treatments due to their enhanced stability, longevity and slow active ingredient water leaching characteristics. Compatibility of these seed coating polymers as formulations with captan, metalaxyl, thiabendazole and novel antimicrobial lipids and triterpenoid compounds display that the active ingredients can readily provide a zone of fungal inhibition around the seed as it germinates in the presence of *Macrophomina phaseolina*, causal agent of charcoal rot of soybeans. However, the release of the active ingredient from the polyol seed treatments is less affected by water leaching as compared to commercially available water-soluble seed treating polymer formulations. This is most likely due to the polyols unique polymer cross-linking characteristics. These results support the continued exploration of soy polyol derived polymers as seed coating compounds.

## INTRODUCTION

This project involved the development of novel soybean oil polyol-based polyurethanes as seed treatments. Generally, seed treatments enhance germination of seed by protecting the seed coat during handling and planting and they also act as a matrix to adhere protective chemicals to the seed coat for protection against soilborne pests. This approach to pest management reduces the amounts of pesticides used and incorporates them into the polymer to prevent leaching and non-target effects. In addition, seed treatments should increase storage and handling stability of seeds prior to and during planting.

## OBJECTIVES

- 1- Study the effect of using soy polyol 180 alone, and in combination with glycerin, polymerized with 4,4'-diphenylmethane diisocyanate (MDI) as seed treatments.**
- 2- Alter the crosslinking density and properties of these polyurethane compounds by changing the isocyanate/ hydroxyl molar ratio and by diluting the polymers with acetone prior to cross-linking.**
- 3- Study the novel polyurethane for their seed treating properties and impact on soybean seed germination.**
- 4- Evaluated the polyurethane seed treatments for compatibility with commercially available fungicides and some novel plant-derived antifungal lipids to determine their use as a formulation for fungicides.**
- 5- Evaluate the leaching of active ingredients from the polyurethane formulations on seeds.**



## MATERIALS

Soy polyol 180 having a hydroxyl number 180 mg KOH/g, glycerin and 4,4'-diphenylmethane diisocyanate (MDI) was supplied by the Kansas Polymer Laboratory, Pittsburg State University, Pittsburg, Kansas. Commercial fungicide was donated by Guftafson LLC, Plano, Texas, Novel Lipid Derivatives were purchased from Sigma Chemical Company, St. Louis, Missouri, Soybean seed was provided by Dr. James Long, Kansas State University, Southeast Agricultural Research Center, Parsons, Kansas.



## METHODS

**Polymerization protocols-** The seed coating samples were prepared using soy polyol 180 alone, and in combination with glycerin, polymerized with 4,4'-diphenylmethane diisocyanate (MDI). These mixtures were vigorously hand mixed in a 16 oz plastic cup, then the soybean seed was added. The seeds were stirred in the polymer mixture 1 min. until uniformly coated and then spread on wax paper to dry. Treated seeds were dried for 48h at 26C prior to germination testing. All seed treatments were studied for their seed treating properties and impact on soybean seed germination. Soy polyol polymerized with MDI in molar ratios of 1:1, 1: 0.7 and 1:0.4 were initially used to treat 60g soybean seed to determine optimal molar ratio. Subsequent experiments were conducted with 1:1 molar ratios that included glycerin.

**Optimal seed quantities used for treatments-** a 1:1 molar ratio consisting of 5 g of soy polyol 180, 0.5 g glycerin and 4.3 g MDI plus the addition of 0, 2.5 g or 5 g of acetone was used to treat 300g of soybean seeds.

**Germination and Growth Studies-** Soy polyol based urethane treated, Magnacoat<sup>TM</sup> treated and untreated soybean seed were used to study the impact of treatments on

germination. Each in vitro germination test consisted of 20 seeds of each treatment group, wrapped in 4 layers of damp paper toweling and sealed in resealable bags. Seeds were grown for 8 days at 26C and data was recorded as a percentage of seed tested. All tests were repeated for statistical analysis.

**Fungicide Compatibility Studies-** Allegiance<sup>TM</sup>, Rival<sup>TM</sup> and two experimental fungicides, Sesamol (Lipid) and a Terpenoid were incorporated into the polyol based urethane mixture prior to polymerization. Concentrations ranging from 0µg/ml-1000µg/ml of each fungicide was incorporated into the seed coating polymers.

#### **Leaching Assays-**

**Zone Of Inhibition Study:** Treated seeds were used in a modified Kirby-Bauer technique which utilized the plant pathogenic fungus Macrophomina phaseolina (causal agent of soybean charcoal rot) to determine the leaching of fungicide from the seed coats and subsequent fungal growth inhibition. Ten seeds per treatment were assayed and the experiment was duplicated for statistical analysis.

**UV Absorption Quantified Leaching:** Five treated seeds were rinsed with 5 mls of distilled water solution and the supernatant run-off was analyzed using UV absorption assays as compared to control standards for each fungicide. Concentrations of leached active ingredients were extrapolated from the standards.

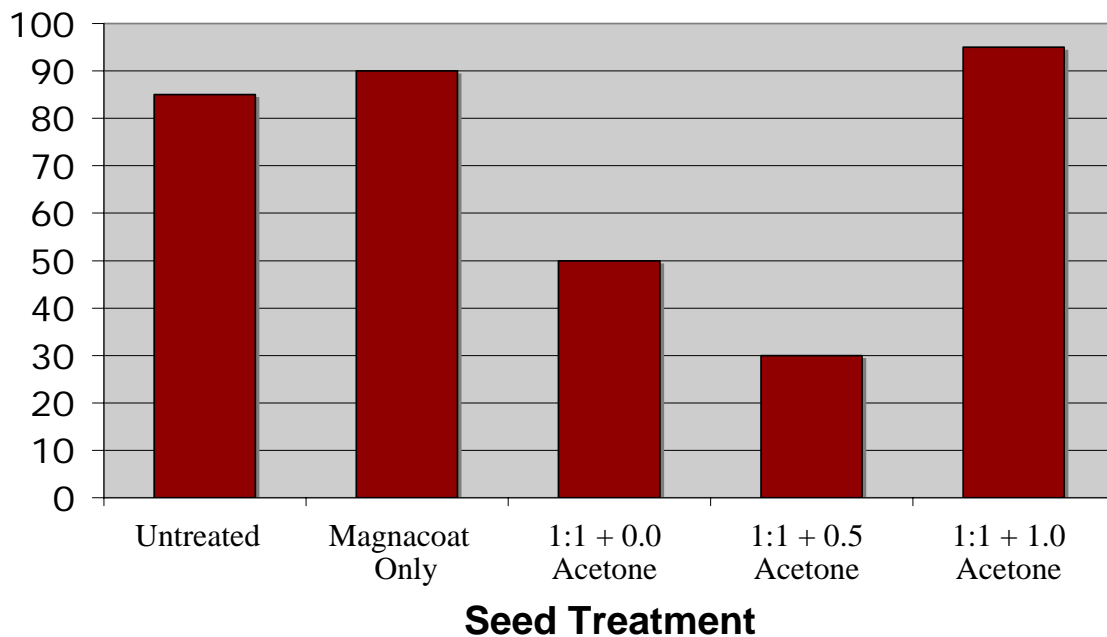
Figure 1. Plate Assay Showing Zone Of Fungal- Inhibition By Allegiance Leaching From The Polyurethane Coating Around A Soybean Seed

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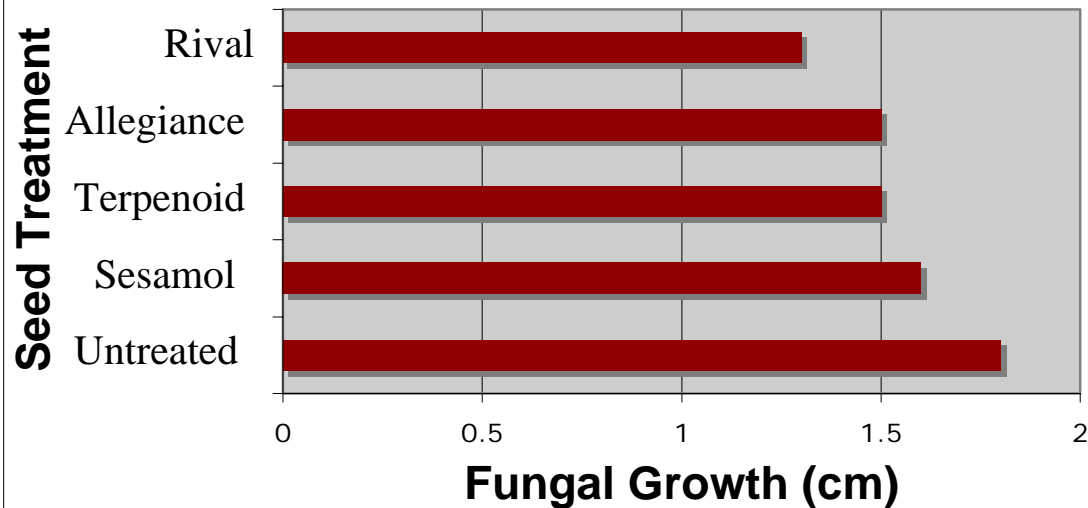
## **RESULTS**

Using a 1:1 molar ratio consisting of 5 g of soy polyol 180, 0.5 g glycerin and 4.3 g MDI, a series of acetone dilutions were made based on the 5 g weight of polyol in each reaction. Acetone was added at weight ratios of 0, 0.5 and 1.0 to the polymer reactions and all treatments were mixed until initial cross-linking had begun before spreading and drying seed. A seed weight of 300g was used to determine if the acetone diluted polymer would impact germination frequency. Figure 3 results indicated that 5g of acetone (1 ratio) used to dilute the polymer mixture, could effectively coat 300g of seed with a resulting seed germination frequency of 95%. This is 5% higher than the Magnacoat<sup>TM</sup> treated seed. This less viscous polyol-based polymer seed treatment coated seeds uniformly with little excess polymer and the resulting coated seeds had germination frequencies that were uniform and comparable to Magnacoat<sup>TM</sup> treated seed. No phytotoxicity was noted in any of the germinated seedlings

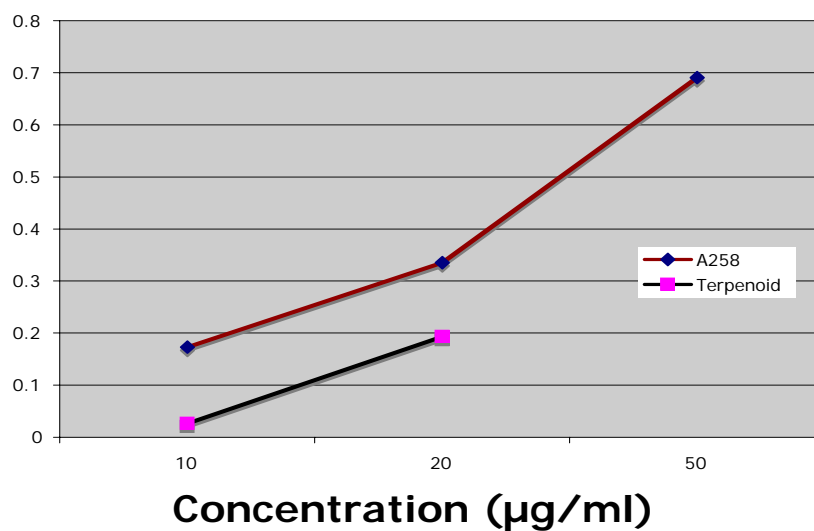
**Figure 2. Acetone Dilutions Impact on Seed Coating And Germination**



**Figure 3. Fungal Growth Inhibition By Seed Treatments**



**Figure 4. Leaching Of Terpenoid From Seed Polymer Coating**



**Table 1. Leaching Concentrations of Active Ingredients From Polyurethane Polyol Seed Treatments As Determined By Ultraviolet Absorption**

<b>S e e d T r e a t m e n t s</b>	<b>L e a c h e d A c t i v e I n g r e d i e n t ( <math>\mu\text{g} / \text{m l}</math> )</b>
<b>Al leg i a n c e H i g h R a t e (0.49 m l/Kg S e e d )</b>	<b>0.02</b>
<b>S e s a m o l L i p i d 1000<math>\mu\text{g}</math> /ml</b>	<b>9.61</b>
<b>T e r p e n o i d 1000<math>\mu\text{g}</math> /ml</b>	<b>11.19</b>

## **DISCUSSION AND CONCLUSIONS**

The results indicate that the use of soy polyols as components of polyurethane seed treatments produces a treatment comparable in efficacy to commercially available seed treatments. Incorporation of glycerin and acetone to reduce the viscosity of the polymer seed treatments resulted in polyurethanes that could efficiently and uniformly coat soybean seed. Soy polyol based polyurethanes yielded treated seeds of comparable quality to those found commercially available, with easy handling and low dust. In addition, these seed treatments did not produce any phytotoxic symptoms in the treated seedlings. These polyurethanes uniformly coated the seed and enhanced the natural germination process of the seed by 12-15% as compared to untreated seed. This represents a significant increase in germination. In addition, these polyurethane seed treatments allow fungicides to leach from their matrix, thus providing a protective zone around the seed. Therefore these results provide a strong economic reason for further development of novel seed treatments for agronomically important crops such as soybeans.

**Special Thanks to Dr. Irene Zegar and Dr. Robert Pavlis, Pittsburg State University Department of Chemistry for their analytical expertise and support.**