

## **CORN PLANT AND SOİL RESPONSE TO MYCOAPPLY® SUPERCONCENTRATE MYCORRHIZAL INOCULATION**

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### **SUMMARY**

This project includes the results of a study conducted with Ankara University and Araştırma Agricultural Institute greenhouse and laboratory in Turkey. MycoApply® Superconcentrate Mycorrhizal inoculum was tested for plant and soil response.

In this project the soil taken from Ankara's Lodumlu region was used in greenhouse trials. The test plant in this project was corn (*Zea mays*). Corn plants were planted into four treatments in the study; (1) Sterilized soil, (2) sterilized soil with sterilized Mycorrhizal inoculum (3) non sterilized soil and (4) non sterilized soil with non sterilized Mycorrhizal inoculum. MycoApply® Superconcentrate Mycorrhizal inoculum diluted with water and perlite was used as the Mycorrhizal treatment following operating instructions in the label information.

In the results of this project the Mycorrhizal colonization percentage of the non sterilized Mycorrhizal inoculated roots was 51% compared to 18% for the controls. The analysis and statistical evaluation of the plants shows that the corn plant biomass (vegetative component) increased significantly (Duncan 0.01) with the non sterilized soil and non sterilized Mycorrhizal inoculum treatment compared to all other treatments. Biomass increases for non sterilized MycoApply® inoculated corn test material was 63% greater compared to non

sterilized soil without inoculation. Sterilizing the Mycorrhizal inoculum eliminated the growth response. This further supports that the corn growth response was due to the living Mycorrhizal inoculum. Visual observations were made about corn plants in greenhouse and it was observed that the plant leaves without unsterilized Mycorrhizal inoculation were yellow and lacked vigor. The corn plants which were grown with inoculation of unsterilized MycoApply® Superconcentrate mycorrhizal had bigger plants and taller leaves. Soil parameters were carefully measured before and after the study and no significant differences in soil conditions were observed.

## **1. Project Name: CORN AND SOİL RESPONSE TO MYCOAPPLY® SUPERCONCENTRATE MYCORRHIZAL INOCULATION**

**1.1. Project Leaders:** Songül Dalcı- Biologist Aynur Dilsiz-Agricultural Engineer

**1.2. Projects Goal and Reason:** The aim of the project is establishing a greenhouse test for the effectiveness of an agricultural mycorrhizal inoculant

## **2. Project Description**

**2.1 The Research Location:** The research was conducted in Araştırma Toprak Gübre ve Su Kaynakları Merkez Araştırma Enstitü's greenhouse and laboratory in Turkey.

**2.2 Materials and Method:** In the research trial the material used was 15g MycoApply® Superconcentrate Mycorrhizal inoculum from Mycorrhizal Applications, Inc in Grants Pass, Oregon USA. In the greenhouse six replications of each treatment was established. After the plants were harvested roots and plants were sampled separately. Mycorrhizal colonization was determined by taking three plant root systems from every treatment clearing and staining the roots with Trypan Blue stain for Mycorrhizal fungi. Spore density was determined by wet elimination method in test material: spores were extracted and average spore count ascertained.

## **2.3 Trial Treatments**

a0: SS

a1: MS+Field soil

a2: Field Soil

a3: sterilized MS+SS

(SS: Sterilized soil, MS: MycoApply® Superconcentrate)

## **2.4 Greenhouse Trial:**

Three kg pots were used in the trial. First the pots were sterilized then the soil which was used in sterilized treatments was placed in an autoclave for 121 degrees Celcius for 2 hours. Trial soil was taken from Ankara's Lodumlu region. In the trials, corn seeds were sterilized with sodiumhipoclorith(10%) and rinsed with distilled water. Seedlings were germinated in sterile perlite. Sterile perlite was used for mixing with the MycoApply® Superconcentrate to have sufficient inoculum to treat the number of seedlings. The final spore concentration of the mycorrhizal inoculum was determined in the methods below.

After root growth the seedling roots were inoculated by approximately 10-15 grams of MycoApply® Superconcentrate by dipping method and planted to trial pots. After an eight week growing period the plants were harvested. In greenhouse, temperature was 24-28 degrees C, lightened for a 12 hours photo period(800-100  $\mu$ mol natural light) and relative humidity was 60% for growing the corn plants. For plants to grow normally the Hoagland solution which includes necessary elements was given to plants once every 15 days.

## **2.5 Laboratory Analysis**

**2.5.1 Mycorrhizal spore counting:** After the test material was diluted with perlite it was suspended with distilled water, The test material was centrifuged and passed through an array of three sieves and spores washed and counted with stereomicroscope in petri dishes. Every test subject was counted 3 times, and the average calculated and the spore count indicated.(Philips and Hayman,1970)

**2.5.2 Mycorrhizal colonization rate (%):**After greenhouse trial, the plants were harvested after an 8 weeks period and fresh roots were cleared and painted with Trypan blue for determining the colonization rate for each plant root and activity rate calculated as a percentage.(Nicholson,1963)

**2.5.3 Analysis of dry substance inside plants:** After the plants were harvested the plants top part washed for weight determination and the plants dried inside drying ovens with 65 C until the plants became their constant weight then the plants weights measured with accurate scales for determining the differences between implanted subjects and the others.

**2.5.4 Soil analysis:** For the soil sample in this operation the following analysis was made before and after the greenhouse trial in the institute's laboratory.

**-Saturation with water (%):** As Richards (1954) declared pure water added to soil until it saturated.

**-Total Salt (%):** By using conductive tool the water saturated soil's electrical conductivity was determined. (Richards 1954)

**-Soil reaction (pH):** Calculated with the water and saturated soil. (Richards 1954)

**-Soil texture:**(Uzunoğlu 1992) The study was done with respect to wet elimination Bouyoucos method and for determining the constitution class soil classification triangle used with respect to analysis results.

**-Lime (%):** As Çağlar (1949) declared, the lime analyzed by using Scheibler Calsimeter with HCl(1+3).

**-Organic element (%):** As Richards (1954) declared, the analysis made with respect to modified Walkey-Black method by using FeSO<sub>4</sub> titration.

**-The Phosphorus used by the plant (kg/da):** As Olsen and friends (1954) declared, 0.5M Sodiumcarbonate extracted for calculations.

**-The Potassium used by the plant (kg/da):** As Richards (1954) declared 1N Ammonium acetate extracted for calculations.

## **2.6 Analysis and evaluation methods**

In Yurtsever (1984) determined in greenhouse trials variance analysis was used and the differences between subjects evaluated with a Duncan test.

## **3.Research Findings:**

### **3.1 Soil Analysis**

In greenhouse trial the necessary soil for test material was taken from Ankara's Lodumlu region (0-20 cm). For determining the test materials effects on soil, soil samples taken from pots before and after the harvest and were analyzed. Table 3.1a

Tablo 3.1 Deneme toprağının bazı fiziksel ve kimyasal analiz sonuçları

DURUSU Ltd.Şti /MycoAply Superconcentrate	Su ile Doymuşluk %	Bünye Sınıfı	EC dS/m	Toplam Tuz %	Su ile Doymuş Toprakta pH	Kireç (CaCO <sub>3</sub> ) %	Bitki ere Yararışlı Besin Maddeleri (kg/da)		Organik Madde %	Alg K+ organik madde %
							Fosfor (P <sub>2</sub> O <sub>5</sub> )	Potasyum (K <sub>2</sub> O)		
Deneme öncesi toprak	58	CL	1,005	0,037	7,75	23,41	6,02	109,02	1,00	58,17
Doğal toprak+M	62	CL	1,097	0,044	7,71	23,23	7,30	123,02	1,27	
Doğal toprak-M	63	CL	1,035	0,042	7,74	23,20	6,74	109,02	1,22	
Steril toprak+M	61	CL	1,308	0,051	7,70	24,07	7,46	149,86	0,81	
Steril toprak-M	62	CL	1,173	0,046	7,76	24,28	8,67	102,24	0,95	

The soil which was used for greenhouse trial included moderate amounts of clay, low organic matter, light alkali and high lime. Phosphorus and potassium levels were also low. In the soil analysis after trial, there was no change detected in these basic parameters. Mycorrhizal inoculated soils had no visual change in soil structure. Analysis of the soil after the trial showed that the mycorrhizal inoculated planting has no negative effect on the soil.

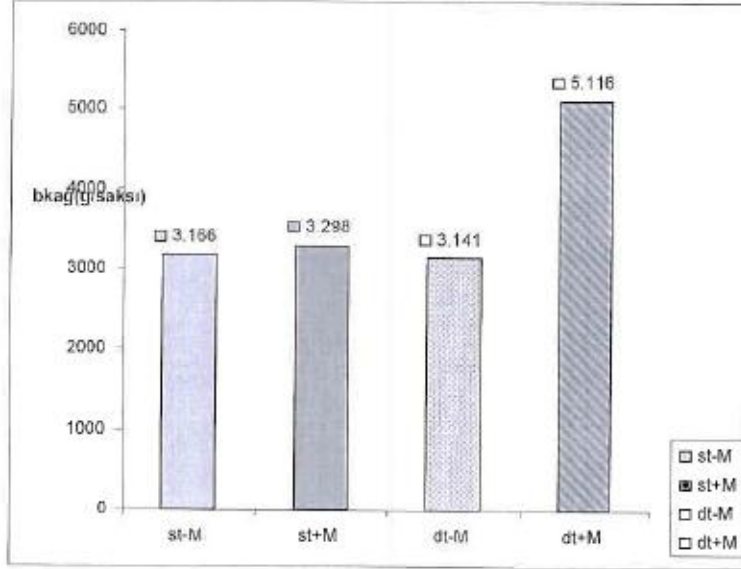
### 3.2 Plants dry weight analysis:

In greenhouse test vegetative parts were evaluated by considering plant dry weights. Plant dry weights showed in Table 3.1 and statistical evaluation is given in Table 3 .2. The differences between treatments are given in Sekil 1. **The non sterile soil with Mycorrhizal inoculation significantly (Dogal Toprak +M treatment) (Duncan 0.01) increased corn plant dry weight. This significant increase in biomass production was nullified by sterilizing the Mycorrhizal inoculum (Steril Toprak-M).**

**Tablo 3. 1** Sera denemesi MS uygulaması mısır bitkisi kuru ağırlıkları (g/saksı)\*

		I	II	III	IV	V	VI	Ort.(g/saksı)
Steril Toprak	- M	2.519	3.548	3.087	3.526	2.985	3.328	3.166
	+M	3.508	3.495	2.256	4.436	3.796	2.296	3.298
Doğal Toprak	- M	3.271	3.670	3.414	2.841	2.452	3.198	3.141
	+M	5.257	4.376	5.533	5.240	4.686	5.605	5.116

\*: +M: Mikorizalı, -M : Mikorizasız, MS: Mycoapply superconcentrate

**Şekil 1.** Sera denemesi mısır bitkisi kuru ağırlıkları (g/saksı)\*

\*: st: Steril toprak, dt: Doğal toprak, +M: Mikorizalı, -M : Mikorizasız,

**Tablo 3.2** Mısır bitkisi kuru ağırlıkları Duncan grupları

Konular	Ortalama	Duncan Grupları (0.01)
Doğal toprak (+M)	5.12	A
Steril toprak (+M)	3.30	B
Steril toprak (-M)	3.17	B
Doğal toprak (-M)	3.14	B

\* p<0.01

Corn plant dry weight values variance analysis was determined and after the statistical evaluation that MycoApply® inoculated pots biomass values (**Doğal Toprak +M treatment**) are statistically significant (0.01) (Table 3.2) compared to corn plants in pots without Mycoapply® inoculation or sterilized MycoApply® inoculation. None of the other treatments were statistically different.

### 3.3 Visual evidence

Visual observations were made about the corn plants in the greenhouse and it was observed that the plant leaves without unsterilized Mycorrhizal inoculation were yellow and lacked vigor. The corn plants which were grown with inoculation of unsterilized MycoApply® Superconcentrate had bigger plants and taller leaves. In sterilized control subject; leaf length was shorter and down leaves has more dead leaves than Mycorrhizal sterilized soil subject.

### **3.4 Mycorrhiza colonizaton rate (%):**

For determining the Mycorrhizal fungus' activity in inoculated plant; in other words, fungus' symbiotic relation with plant can be understood by microscopic inspection on stained plant roots. In this scientific research trial occurred by getting random root samples from pots and clearing and staining them with Trypan blue (%0.05). Slide method was used for checking plant roots colonization under stereo microscope. 10 pieces of 1cm root parts on each glass of microscopes, colonization rate was ascertained and the infection rate was found in terms of % for 50 root pieces. By getting a root from each pot the infection rate was found and the average of parallels was calculated. Unsterilized soil with unsterilized Mycorrhiza colonizaton rate was 51%. The unsterilized soil without Mycorrhizal inoculation colonization rate was determined as 18%. The plants with high Mycorrhizal colonization rate had a high biomass rating.

### **3.5 Mycorrhizal Spore Count:**

After the test material became a suspension with distilled water by using 50ml centrifuge tube, this material mixed with %60(w/v) sucrose solution and centrifuged with 3000 rpm for 2 minutes. By washing with pure water and by passing through a 45 µm sieve transferred to a petri dish and the mycorrhiza spore count was made by stereo microscope. After the spore count, the spore count was found as 13.157 spore/gram for diluted MycoApply® Superconcentrate test material.

### **4. Conclusion:**

In greenhouse by using 3kg plastic pots for trial, corn plant roots were inoculated with MycoApply® Superconcentrate and were planted. After an 8 weeks process plants was harvested and necessary analyses were made. With respect to these analyses results with MycoApply® Superconcentrate inoculated corn had 63% more plant

biomass than the treatments without MycoApply® Superconcentrate or sterilized MycoApply® Superconcentrate. By checking the soil analysis results before and after trial; there was no specific change determined. The Mycorrhizal colonization rate was found as 51% compared to the 18% in non-inoculated or sterilized controls. Using both laboratory and visual inspection it was determined that the MycoApply® inoculation greatly stimulated the growth and vigor of corn.



