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EFFECT OF SILVER NANOPARTICLES ON SEED PROTECTION IN DIFFERENT SOILS

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ABSTRACT To investigate the possibility of using silver nanoparticles for protection of a living organism, wheat seeds were coated with these particles and planted. If germination of seeds is not affected by this new nano-treatment, the method can be used on a wide range of common treatment for seed protection. The effect of treatment on seeds with this particle protection against fungi was compared with seeds treated with a conventional preplanting fungicide (Carboxitiram). Being that soil is the most common medium for plant growth in agriculture, this can play a major role in plants life and their relationships with other factors in the field. This study looks at the effects of different soil conditions on pre-treatment. The effect on soils with different levels of nutrient, pH and humidity were investigated. Results showed that soil conditions have little affect on seed protection with silver nanoparticles against fungi, and protection is completely independent of soil conditions. Results showed that silver nanoparticles do not reduce seed germination. In other words, seed viability is not significantly affected. Moreover, seed protection cost with Silver nanoparticles and Carboxitiram was not significantly different. This effect is completely independent of soil conditions regardless of levels of nutrients, pH and humidity.

Keywords: Silver Nano Particles, Seed Treating, Preplanting Fungicide.

INTRODUCTION: Nanotechnology has the potential to revolutionize the scientific world by allowing scientists to manipulate matter at atomic or molecular scale using physics, engineering, chemistry and biology [1]. Nanotechnology is a broad and interdisciplinary area of research and development activity that has been growing at a rapid pace worldwide in the past few years [2].
In agriculture, using many monitoring and control systems already in place, nanotechnology devices for CEA providing "scouting" capabilities could tremendously improve the grower's ability to determine the best time of harvest for the crop, the vitality of crop, and food security issues, such as microbial or chemical contamination [3].
Recently nanotechnology is used in different agricultural fields like precision farming, smart delivery systems, food industry, packaging, food safety, food processing, etc [4],[5],[6].

Today we are sure that the Nanosilver can protect us against fungi and bacteria; therefore, we use it to produce our foods more securely. The new food packages containing silver nanoparticles keep our foods fresh, healthy and safe. Also, these particles are in their way toward our homes now. Recently, our refrigerators, freezers, washing machines, vacuum cleaners, etc, use these particles.
Is it possible to use these new useful particles to treat a living organism? This is the main challenge now. For example, can we protect the seeds against harmful factors like fungi in agricultural practices? Is it probable that this treatment threats our organism's life? Can environmental conditions affect the protection effect? These are the next questions if the answer to the first one had been "yes".
This study has tried answering these questions. To achieve this goal, wheat seeds were coated by silver nanoparticles. Soils with different nutrients, pH and humidity were infected by fungi and the seeds were planted in the infected soils. Then seed emergence and the protecting effect of silver nanoparticles were investigated to find the answers.

MATERIAL AND METHODS

Material: Wheat seeds (GV/D630/ALD"s"/3/Azd), soil, pure water, Nanosilver solution (100ppm), preplanting fungicide (Carboxitiram), fungi (Fusarium Graminae)

Methods: A Completely Randomized Design (CRD) was used to compare percentage of germination between different treatments according to table 1 where a, b and c designate non treated seeds, seeds treated with Carboxitiram and those Silver nanoparticles, respectively. To coat seeds with silver nanoparticles, seeds were sprayed with a 100 ppm nanosilver solution and then dried. This action was repeated 3 times. To treat seeds with Carboxitiram, a paper bag containing seeds and carboxitiram powder was well shacked.

Table 1: Experimental design for studying the effect of using Silver nanoparticles on percentage of germination

c	a	b	a	b
b	c	a	c	a
a	b	c	b	c

15 petri dishes lined with wet paper were provided and 5 seeds were placed in each one as indicated in table1. After 15 days, germination percentage was calculated using equation 1.

$$\%Germination = (\frac{Germinated\ seeds}{Planted\ seeds}).100 \qquad (1)$$

To determine the effect of Silver nanoparticles on seed protection, an experimental design as shown in table 2 was used.

Table 2: Experimental design for comparison Silver nanoparticles and carboxitiram

a	b	b	a	b
b	a	a	b	a

Where a and b indicate seeds treated with Carboxitiram and those coated with Nanosilver, respectively.

A flowerbox was provided for each plot of table 2. Disinfected soil was infected with fungi and flowerboxes were filled with this soil. In the next stage, 5 seeds according to table 2 were planted in each flowerbox.

Remaining index as shown in equation 2 was defined for comparison.

$$\%R = \left(\frac{RP}{TP}\right).100 \qquad (2)$$

Where %R is remaining percentage, RP and TP are Remained Plants and Total Planted respectively.

Table 3: Experimental design for investigation the effect of different levels of soil's nutrients on seeds protection

a_2b_1	a_2b_5	a_2b_2	a_2b_3	a_2b_3	a_1b_4	a_1b_5	a_2b_4	a_1b_1	a_2b_5
a_1b_3	a_1b_1	a_1b_3	a_1b_2	a_2b_5	a_2b_2	a_1b_2	a_1b_3	a_1b_4	a_1b_1
a_1b_2	a_1b_5	a_2b_4	a_1b_4	a_2b_1	a_1b_5	a_2b_4	a_2b_1	a_2b_3	a_2b_2

An experimental design like that shown in table 3 was used to investigate the effect of different levels of soil's nutrients on seeds protection against fungi by silver nanoparticles. Where a_1 and a_2 designate to seeds treated with Caroxitiram and silver nanoparticles, respectively. b_1 is the symbol of soil with full nutrients according to the wheat seed's demand. b_2, b_3, b_4 are the soils treated by different levels of nutrients equal to 75, 50 and 25% of b_1 level, respectively. b_5 indicates to a media for seeds growth with no nutrients. Wood trashes can make this media.

30 flowerboxes were filled with these different soils and 5 seeds were planted in each one like that shown in table 1. The effect of different levels of soil nutrients on protection of seeds were calculated by "Remaining Index" shown with equation 2.

The effect of different soil's pH and different levels of soil's humidity on the seeds protection were investigated according to table 3 and table 4.

Table 4: Experimental design for investigation the effect of different levels of soil's pH on seeds protection

a_1b_2	a_1b_3	a_2b_1	a_1b_1	a_2b_1	a_2b_1
a_2b_3	a_1b_1	a_2b_2	a_2b_3	a_1b_3	a_2b_2
a_1b_2	a_1b_2	a_2b_2	a_1b_1	a_1b_3	a_2b_3

Table 5: Experimental design for invistigation the effect of different levels of soil's humidity on seeds protection

a_2b_3	a_1b_2	a_1b_3	a_2b_2	a_1b_3	a_2b_2
a_2b_1	a_2b_2	a_1b_1	a_1b_3	a_2b_3	a_2b_3
a_2b_1	a_1b_2	a_1b_1	a_1b_1	a_2b_1	a_1b_2

Where a_1 and a_2 in these tables are the same as a and b in table 3. But b_1, b_2 and b_3 show acidic, normal and basic soils in table 4 and show different levels of humidity equal to 5, 20 and 40% of soil's dry weight in table 5, respectively. There are no more differences between these recent tests and the first experiment in process and calculation. So, equation 1 was used to determine these effects as well.

RESEARCH RESULTS

Results of germination test and discussion of statistical analysis of the data are shown in figure 1 and table 6, respectively. According to the results, there was no significant difference between treatments.

Figure 2 shows the results of protection effect test while statistical analysis of data is shown in table 7. This piece of information confirms that there is no significant difference between treatments in this test too.

Results of different levels of seeds nutrients, pH and humidity and their statistical analysis are presented in figures 3 to 5 and tables 8 to 10, respectively. According to the output of LSD test, the effects of all levels of nutrients on protection were the same.

However basic soils and soils with less humidity (5%) were different than other treatments significantly but it was not the effect of silver nanoparticles. This subject was investigated in Conclusion section.

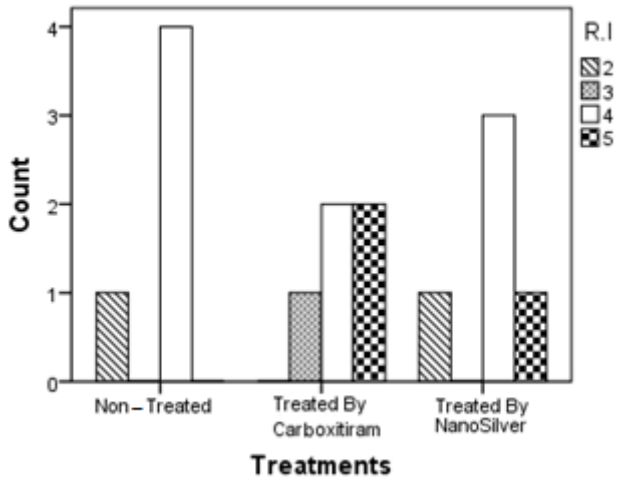

Figure 1: Result of germination test

Table 6: Statistical analysis of germination test results

Dependent Variable:R.I

	(I) Treatments	(J) Treatments	Mean Difference (I-J)	Std. Error	Sig.
LSD	Non Treated	Treated By Carboxitiram	-.600	.600	.337
		Treated By Nanosilver	-.200	.600	.745
	Treated By Carboxitiram	Non Treated	.600	.600	.337
		Treated By Nanosilver	.400	.600	.518
	Treated By Nanosilver	Non Treated	.200	.600	.745
		Treated By Carboxitiram	-.400	.600	.518

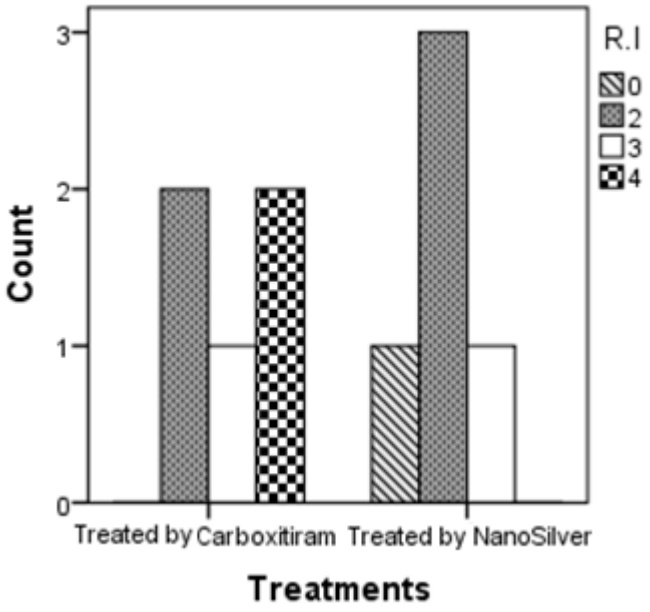

Figure 2: Result of protection effect test

Table 7: Statistical analysis of protection effect test

ANOVA

R.I

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	3.600	1	3.600	3.273	.108
Within Groups	8.800	8	1.100		
Total	12.400	9			

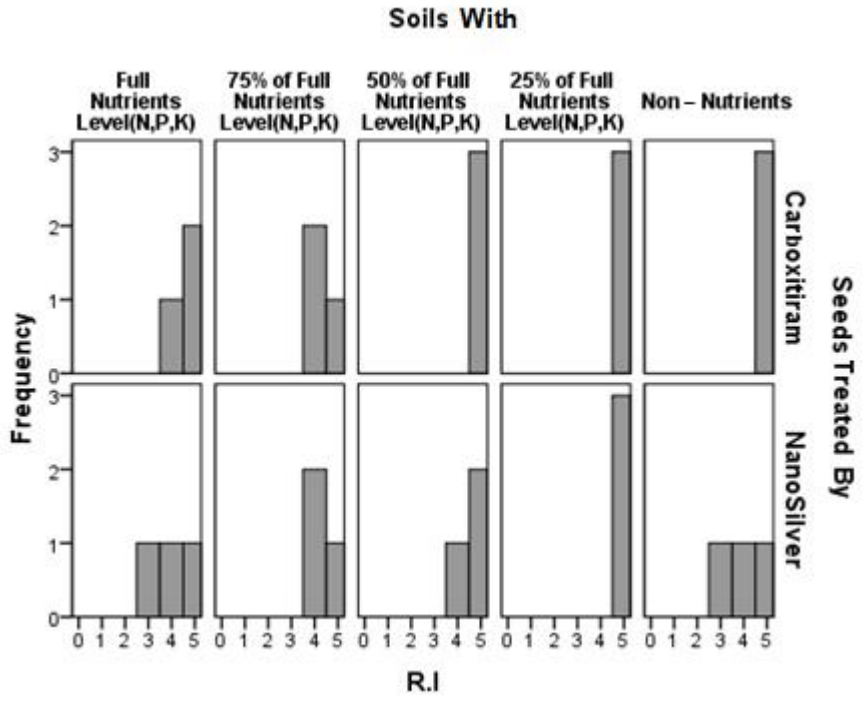

Figure 3: Result of the effect of different levels of soil's nutrients on seeds protection

Table 8: Statistical analysis of the effect of different levels of soil's nutrients on seeds protection

Post Hoc Tests\Soils\Multiple Comparisons

Dependent Variable:R.I

	(I) Soils	(J) Soils	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
						Lower Bound	Upper Bound
LSD	Soil Treated by Full Nutrients Level(N,P,K)	Soil Treated by 75% of Full Nutrients Level(N,P,K)	.00	.333	1.000	-.70	.70
		Soil Treated by 50% of Full Nutrients Level(N,P,K)	-.50	.333	.149	-1.20	.20
		Soil Treated by 25% of Full Nutrients Level(N,P,K)	-.67	.333	.059	-1.36	.03
		Non-Treated Soil	-.17	.333	.623	-.86	.53

Soil Treated by 75% of Full Nutrients Level(N,P,K)	Soil Treated by Full Nutrients Level(N,P,K)	.00	.333	1.000	-.70	.70
	Soil Treated by 50% of Full Nutrients Level(N,P,K)	-.50	.333	.149	-1.20	.20
	Soil Treated by 25% of Full Nutrients Level(N,P,K)	-.67	.333	.059	-1.36	.03
	Non-Treated Soil	-.17	.333	.623	-.86	.53
Soil Treated by 50% of Full Nutrients Level(N,P,K)	Soil Treated by Full Nutrients Level(N,P,K)	.50	.333	.149	-.20	1.20
	Soil Treated by 75% of Full Nutrients Level(N,P,K)	.50	.333	.149	-.20	1.20
	Soil Treated by 25% of Full Nutrients Level(N,P,K)	-.17	.333	.623	-.86	.53
	Non-Treated Soil	.33	.333	.329	-.36	1.03
Soil Treated by 25% of Full Nutrients Level(N,P,K)	Soil Treated by Full Nutrients Level(N,P,K)	.67	.333	.059	-.03	1.36
	Soil Treated by 75% of Full Nutrients Level(N,P,K)	.67	.333	.059	-.03	1.36
	Soil Treated by 50% of Full Nutrients Level(N,P,K)	.17	.333	.623	-.53	.86
	Non-Treated Soil	.50	.333	.149	-.20	1.20
Non-Treated Soil	Soil Treated by Full Nutrients Level(N,P,K)	.17	.333	.623	-.53	.86

	Soil Treated by 75% of Full Nutrients Level(N,P,K)	.17	.333	.623	-.53	.86
	Soil Treated by 50% of Full Nutrients Level(N,P,K)	-.33	.333	.329	-1.03	.36
	Soil Treated by 25% of Full Nutrients Level(N,P,K)	-.50	.333	.149	-1.20	.20

Based on observed means. The error term is Mean Square (Error) = .333.

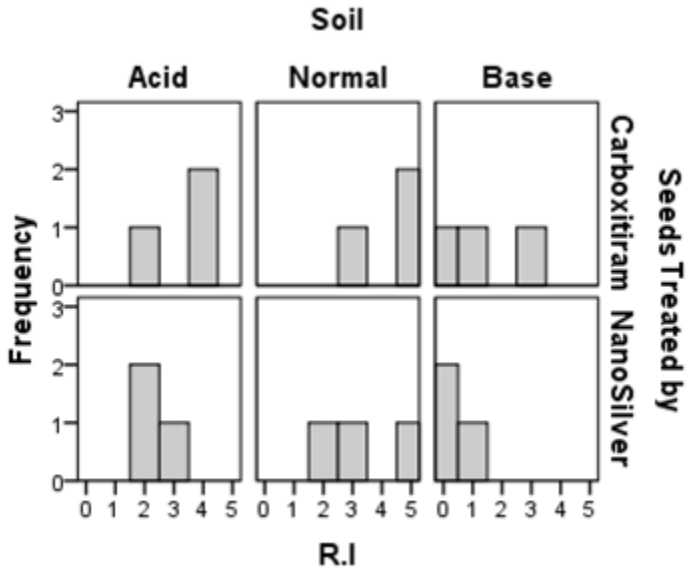

Figure 4: Result of the effect of different levels of soil's pH on seeds protection

Table 9: Statistical analysis of the effect of different levels of soil's pH on seeds protection

Post Hoc Tests\Soil\Multiple Comparisons

R.I(LSD)

(I) Soil	(J) Soil	Mean Difference (I-J)	Std. Error	Sig.	95% Confidence Interval	
					Lower Bound	Upper Bound
Acid	Normal	-1.00	.667	.159	-2.45	.45
	Base	2.00*	.667	.011	.55	3.45
Normal	Acid	1.00	.667	.159	-.45	2.45
	Base	3.00*	.667	.001	1.55	4.45
Base	Acid	-2.00*	.667	.011	-3.45	-.55
	Normal	-3.00*	.667	.001	-4.45	-1.55

Based on observed means. The error term is Mean Square(Error) = 1.333.

*. The mean difference is significant at the 0.05 level.

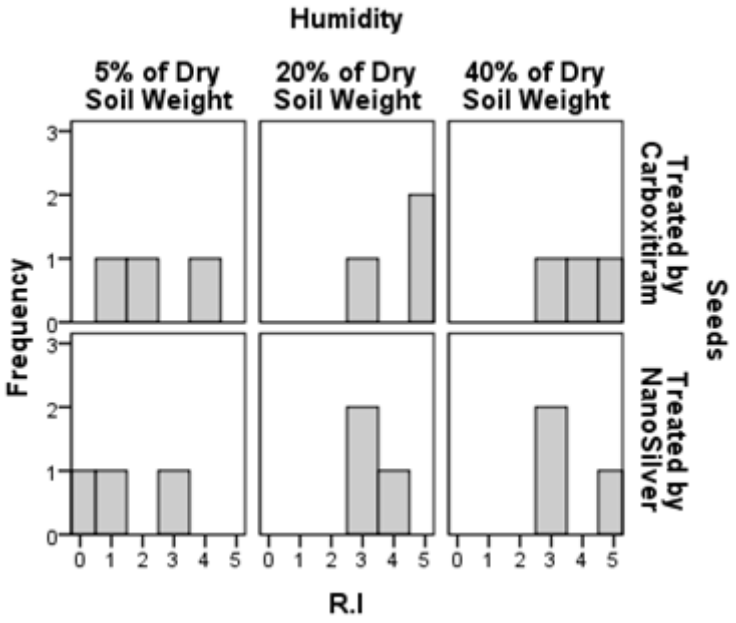

Figure 5: Result of the effect of different levels of soil's humidity on seeds protection

Table 10: Statistical analysis of different levels of soil's humidity on seeds protection

Post Hoc Tests\Humidity\Multiple Comparisons

R.I (LSD)

(I) Humidity	(J) Humidity	Mean Difference (I-J)	Std. Error	Sig.
5% of Dry Soil Weight	20% of Dry Soil Weight	-2.00*	.694	.014
	40% of Dry Soil Weight	-2.00*	.694	.014
20% of Dry Soil Weight	5% of Dry Soil Weight	2.00*	.694	.014
	40% of Dry Soil Weight	.00	.694	1.000
40% of Dry Soil Weight	5% of Dry Soil Weight	2.00*	.694	.014
	20% of Dry Soil Weight	.00	.694	1.000

Based on observed means. The error term is Mean Square (Error) = 1.444.

*. The mean difference is significant at the 0.05 level.

Multiple Comparisons

R.I (LSD)

(I) Humidity	(J) Humidity	95% Confidence Interval	
		Lower Bound	Upper Bound
5% of Dry Soil Weight	20% of Dry Soil Weight	-3.51	-.49
	40% of Dry Soil Weight	-3.51	-.49
20% of Dry Soil Weight	5% of Dry Soil Weight	.49	3.51
	40% of Dry Soil Weight	-1.51	1.51
40% of Dry Soil Weight	5% of Dry Soil Weight	.49	3.51
	20% of Dry Soil Weight	-1.51	1.51

Based on observed means.

The error term is Mean Square (Error) = 1.444.

CONCLUTIONS

Since results showed that treating seeds with silver nanoparticles does not reduce germination, it is possible to use this treatment in agricultural practices. In other words, silver nanoparticles did not destroy the seed's living process. Now the answer to the first question asked in introduction is found. Treating a living organism like a seed with Nanosilver while keeping its life is possible.

By results of protection test, also it is found that using Silver nanoparticles instead of conventional poison is possible to protect seeds against fungi. This recent point can help us to use these new treated seeds in precision planters to keep environmental health and reduce the cost of agricultural productions.

According to results, it is clear that different levels of soil's nutrients cannot concern the protection effect significantly. However, at the first glance, there seems to be some significant differences between basic soils with acidic or normal soils, as well the significant difference existed between soils with less humidity (5%) and the others, but with a deeper look, since the effect of silver nanoparticles and Carboxitiram were the same, it will be understood that these differences just depended on soil conditions .Therefore, it can be suggested that the silver nanoparticles protection effect is independent from soil conditions.

REFRENCES

[1] Roco,M. C.,Williams, R. S., Alivasatos, P., Eds.Nanotechnology Research Direction: IWGN Workshop Report, Kluwer Academic Publishers: Norwell, MA, 1999.

[2] F.Kulzer and M. Orrit, "single-Molecule Optics", (2004), Annual Review of Physical chemistry, Vol. 55, 585-611

[3] The US Department of Agriculture, Nanoscale science and engineering for agriculture and food system.

[4] Down on the farm, ETC group, 2004: http://www.etcgroup.org/documents/ETC DOTFarm2004.pdf

[5] http://www.syngentaproffessionalproducts.com/to/prod/primo/

[6] J.Dunn, "A MINI REVOLUTION", Food Manufacture, 2004: http://www.foodmanufacture.co.uk/news/fullstory.php/aid/472/A mini revolution.html