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THE EFFECT OF SAFE MATERIALS IN CONTROL OF CUCUMBER DOWNY MILDEW UNDER FIELD CONDITION

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ABSTRACT

Downy mildew on cucurbits (*Pseudoperonosporacubensis*) is an important disease in cucumbers cultivated in wet areas and greenhouses which causes considerable damages to this product annually. In one hand, no influential and stable controlling approach has been found for a number of pathogens and in another hand the emergence of resistance phenomena toward various kinds of synthetic pesticides specially antibiotics, poisoning resulted from chemical pesticides in animals, beneficial insects and aquatics and also harmful effects of pesticides residues cause numerous problems regarding human health and environment. Therefore, introducing minerals and low-risk compounds to control the disease in this fresh product seems critical and essential. The current study was performed in a site in the research station of Ghrakhil in 2014. The experiment consisted of completely random blocks accompanying 7 treatments repeated for 3 times. Compounds used included: potassium silicate, salicylic acid, kaliban, chitosan, volck oil, equation pro fungicide and control elements. Solution spraying was done immediately after observing the initial signs of downy mildews in terraces. Considering the distribution of this disease on the control treatment bushes, we analyzed the experimental treatments by counting the number and size of blotches, number of healthy and infected leaves, determining the kinds of infection in each terrace, and finally the crop yield. The results demonstrated a significant difference of 1% between fungicide treatments regarding the disease intensity control. Also they showed that control treatments had a significant difference of 1% regarding disease control percent and finally the product yield. The maximum and minimum amount of infection were observed in the control treatment with an infection average of 76/86 in group A and an amount of 400gr/hain the equation pro fungicide treatment and an infection average of 14/58 in group D, respectively. The highest infection occurrence percent in the control treatment was observed in group A with an infection percent of 73/83 and the lowest infection occurrence percent was seen in the equation pro fungicide treatment with an amount of 400 gr/ha and the average infection occurrence percent of 10 in group D. The highest amount of efficiency was reported in the equation pro fungicide treatment with an amount of 400 gr/ha and an average function amount of 2/233 kilogram in group A and the lowest amount was observed in the control treatment with an average efficiency amount of 0/7 kilogram in group D.

Keywords: Downy Mildew, Infection Type, Super Dominus, Treatment, Disease

INTRODUCTION

Cucumber crop (*Cucumis sativus*) is considered one of the most economical plants of cucurbits family. This vegetable has a documented history of more than 5000 years. However, it is highly influenced by fungal diseases in farms or greenhouses. Under appropriate conditions, downy mildew spreads rapidly and cause significant damages. Downy mildew (*Pseudoperonosporacubensis*) is considered as a common fungal disease in cucumbers cultivated in geographically potential places (Ranjbar, 2008). Although applying appropriate farming methods and cultivating relatively resistant plants may reduce the average amount of damage resulted from downy mildew, its pervasive outbreak necessitates chemical compounds. Some of the non-chemical controlling methods consisted of using biologic fungicides containing bacteria (*B.subtillis*) or fungi (*Trichoderma harzianum*). Using the following materials may control downy mildew: Materials which stimulate plant's resistance such as chitosan, salicylic acid, vegetable oils such as canola, soya, corn, sunflower, olive, safflower and neem oil, using the blue wavelength under the thin blue layer

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(decreasing the fungus sporulating), extracts of plants including garlic, ginger, proxides, bicarbonates and compost tea.

Repeated application of fungicide compounds causes undesired side effects for human and environment. The pressure of foundations and supervising agencies to maintain the quality of food, agricultural products and the environment in the field of agriculture and food industry lead to the reconsideration of substituting chemical pesticides with a new and modern approach in the scientific societies throughout the world. This subject was seriously discussed in annual meeting of researchers and scientists from various scientific fields. Moreover, the amount of performed studies and researches in progress illuminate a new revolution in providing the health and safety of agricultural products. Therefore further researches are required to identify new compounds with better effects and less harmful consequences.

Downy mildew spreads in all farms in which cucurbits family is cultivated (Palti and Hernneth, 1980). This fungus is an obligate parasite which cannot grow in a synthetic medium and only survives on a living plant.

This infection initially emerges on the higher area of leaves (mostly in the nervures margin) as some oily-angled light green blotches with a diameter of 1 up to 2 centimeters. After a couple of days, blotches turn to yellow and brown and withered then the bush loses its natural growth and production. Generating high amounts of sporangiophore and sporangium in the underside epidermis of leaves is one of the sensible signs of this disease. Studies demonstrate that sporangiums of the pathogen fungus only produce zoospore after germination contrary to the sporangiums of the peronospora kind. The spread of pathogen fungus by the oval like sporangium is done by a porous pupil at the end which 3-8 zoospore with two lateral flagella are created as a result of its germination.

The zoospores' diamond reaches 10-12 micron after being encysted which generate a germ tube after germination (Arshi, 2000). The amount of damage differs from 0 to 100% depending on the time of infection, weather conditions and the sensitivity of the host plant (Mahrishi and Siradhana, 1988). The reduction amount in cucumber production as a result of downy mildew has been calculated as 61% and the early infection of farm causes a severe decrease in the yield amount (Tsai, 1992). The used fungicides not only did not have a definite influence on controlling the disease, but imposed negative effects on the health and safety of nutrition and environment. In this study, we aimed to use safe compounds to control downy mildew on cucumbers in farms.

METHODS AND MATERIALS

Experiment

The Gharakhil station of the agricultural research center of Mazandaran was selected for field experiments because of its appropriate weather conditions. This station is located at 10 kilometers of south-east of GhaemShahr which is 14/7 meter upper the sea level and has longitude of 52 degrees and 53 minutes east, latitude of 36 degrees and 27 minutes north and the average rainfall of 745 mm. EC and PH amount of soil are 0/6 Ds/m and 7/6, respectively. First seeds are kept in water for 24 hours and then transferred to the target land after germination. After preparing soil, planting seeds was done through the piling method. one day after cultivating, the experimental treatments were irrigated through sprinkler irrigation. Also the experimental terraces were weeded 3 times and irrigated 4 times.

Treatments applied in this experiment:

1. The fungicide cymoxanil+ famoxadone (equation pro) 400gr/ha
2. Chitosan 0/07%
3. Salicylic acid 150 mgr/l
4. potassium silicate 30 mM
5. Kaliban 85% (potassium bicarbonate) 5gr/l
6. Volck oil 4 in 1000
7. Control (just spraying with water)

To secure consumers health regarding kinds of pesticides we just used the equation pro (formulation: cymoxanil 30%, famoxadone 22/5% and WDG 52/5%) which has a curing and hindering effect on the

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disease and also has low and acceptable amount of residue in plants sprayed. Also, other treatments include safe chemical compounds.

Sampling and Microscopic Analysis of the Field Samples in the Laboratory

After the last spray, leaves of all terraces were gathered separately and transferred to the laboratory. Microscopic observations of the samples gathered from the farm demonstrated that all the leaves gathered from the cucumber farm contained sporangiophore and sporangium of *Pseudoperonosporacubensis*. Fungus hyphae without transverse walls and sporangiophores with a length of 200-360 and width of 5-6/5 micrometer were gathered in multiple groups including the pores in the underside area of leaves having bifurcated divisions. Oval sporangiums had a dimension of 38-19 x 20-13.

Experiment Analysis

Analyzing the experiment was performed two weeks after spraying and the disease progress on the control through determining disease occurrence percent (D1) and disease intensity (Ds). To calculate the disease occurrence percent we used the formula $DI = \frac{\sum x}{N} \times 100$.

In this formula, x is the number of infected bushes and N is the total number of analyzed bushes. Disease intensity was calculated by the following formula

$$DS = \frac{\sum (xi \times ni)}{N \times v} \times 100$$

In this relation xi and ni are the kind of infection and the number of infected leaves in the ith degree of the disease. N is the total number of analyzed leaves and v is the highest level of disease (Cardoso *et al.*, 2004).

In this relation ten leaves were selected from each bush and scoring from 1 to 9 was performed using the approach designed by Thomas *et al.*, (1987):

Table 1: Analysis degrees regarding downy mildew on cucumbers (Thomas *et al.*, 1987)

Degree	Disease sign	Signs descriptions
1	0	Blotches are seen but does not generate sporangium so belong to the incompatible class
3	-1	
5	++	Sporangiums were generated in alimited number. Only a Few sporangiophores were recognizable by a microscope and were categorized as the compatible class
7	+	Sporangiums were scattered (about 5×10^3 spore in each cm2 of the blotch area). blotches were scattered and were in higher rank comparing the previous class
9	+++	Numerous sporangiums were generated (about 5×10^3 spore in each cm2 blotch area). Blotches were seen all over the leaf surface and categorized as the highly compatible class.

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Statistical Analysis

Analyzing the research data was performed through the MSTATC statistical software. Data was pluralized and means were compared by the multi-range Danchon method after the statistical analysis.

RESULTS AND DISCUSSION

Results

Results indicated a significant difference of 1% between various treatments of fungicide regarding the infection occurrence percent (table 2). Comparing the average of infection occurrence percent in various treatments, we discovered that the highest percent was in the control treatment with a mean infection occurrence percent of 64.33 put in group A. the lowest observed infection occurrence level belonged to the equation pro fungus treatment with an amount of 400gr/ha and an infection occurrence percent of 10 put in group D. the mean of other experimental treatments on the occurrence percent were between these two groups (table 7).

Table 2: Variance analysis of treatments influence on the infection occurrence percent in Mazandaran (at the level of control infection occurrence of 50%)

F	Mean squares MS	Sum squares S.S	Degree freedom D.F	Sources of change S.O.V
1.0695	33.333	66.667	2	repeat
33.7998	1053.429	6320.571	6	treatment
	31.167	374.000	12	error
		6761.238	20	total

Study indicated a significant difference of 1% between various fungus treatments regarding the infection intensity in the infection level of 50% (table 3). Comparing the infection intensity means in various experimental treatments showed that the highest amount of infection was in the control treatment with an average of 61.40 which belonged to group A. the lowest amount of observed infection belonged to the equation pro fungicide with an amount of 400 gr/ha with an average of 12.53 categorized in group D. the average of other experimental treatments on the disease intensity were located between these two groups (table 7).

Table 3: Variance analysis of treatments effects on the disease intensity in Mazandaran (at the control infection level of 50%)

F	Degree freedom D.F	Sum squares S.S	Mean squares MS	Sources of change S.O.V
1.9907	2	65.284	32.642	repeat
45.5819	6	4484.412	747.402	treatment
	12	196.763	16.397	error
	20	4746.458		total

Results indicated a significant difference of 1% between various treatments of fungicide regarding the infection occurrence level of 90% control infection (table 4). Comparing the infection occurrence percent in various experimental treatments, we realized that the highest infection occurrence percent was 83.33 in the control treatment belonged to the group A and the lowest infection occurrence percent was 400 gr/ha in the equation pro fungicide with a mean infection occurrence of 10 belonged to group D. the average of other experiment treatments on the disease occurrence percent were located between these two groups (table 7).

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Table 4: Variance analysis of treatments effects on the disease occurrence in Mazandaran(at the control infection level of 90%)

F	Mean squares MS	Sum squares S.S	Degree freedom D.F	Sources of change S.O.V
0.0157	1.000	2.000	2	repeat
25.2303	1611.937	9671.619	6	treatment
	63.889	766.667	12	error
		10440.286	20	Total

Table 5: Variance analysis of treatment influence on disease intensity in Mazandaran (in control infection intensity of 90%)

F	Mean squares MS	Sum squares S.S	Degree freedom D.F	Sources of change S.O.V
4.3762	160.000	320.001	2	repeat
50.1371	1833.092	10998.549	6	treatment
	36.562	138.739	12	error
		11757.289	20	total

Considering the results, a significant difference of 1% was found between various fungicide treatments regarding the crop yield (table 6). Comparing the mean of yield in various experiment treatments demonstrated that the highest yield belonged to the equation pro treatment with an amount of 400 gr/ha and an average yield of 2.233 categorized in group A. the lowest product's yield was observed in the control treatment with an average yield of 16.63 belonged to group D. the average of other experiment treatments on the crop yield were located between these two groups (table 7).

Table 6: Variance analysis of treatment influences on the yield of cucumber in Mazandaran

F	Mean squares MS	Sum squares S.S	Degree freedom D.F	Sources of change S.O.V
0.7618	0.046	0.092	2	repeat
15.3717	0.932	5.592	6	treatment
	0.061	0.728	12	error
		6.412	20	total

Table 7: A mean comparison of calculated characteristics in the cucumber plant

treatment	Mean of infection occurrence percent in the control infection level of 50%	Mean of infection intensity in the control infection level of 50%	Mean of infection occurrence percent in 90% control infection	Mean of infection intensity in 90% control infection	Mean of product (cucumber) yield and grouping
Equation fungicide pro	10.00 ^D	12.53 ^D	10.00 ^D	16.63 ^D	2.233 ^A
chitosan	17.33 ^D 22.33 ^{CD}	19.20 ^{CD} 21.43 ^{CD}	22.67 ^{CD} 36.00 ^{BC}	22.53 ^{CD} 34.10 ^{BC}	1.500 ^{BC} 1.633 ^{AB}
Salicylic acid					
Potassium silicate	35.00 ^{BC}	28.10 ^{BC}	44.67 ^B	34.07 ^{BC}	1.500 ^{BC}
bicarbonatepotassium (kaliban)	42.67 ^B	32.17 ^B	50.00 ^B	40.73 ^B	0.9000 ^{CD}
Volck oil	45.00 ^B	32.53 ^B	45.33 ^B	42.67 ^B	0.7667 ^D
Control without the use of fungicide	64.33 ^A	61.40 ^A	83.33 ^A	92.33 ^A	0.7000 ^D

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Considering the results of the current experiment all the performed treatments influenced on decreasing the disease occurrence regarding the infection intensity in control infection levels of 50% and 90%. Also the best treatment was demonstrated to be the equation pro fungicide which showed the lowest level of disease occurrence (diagram1).

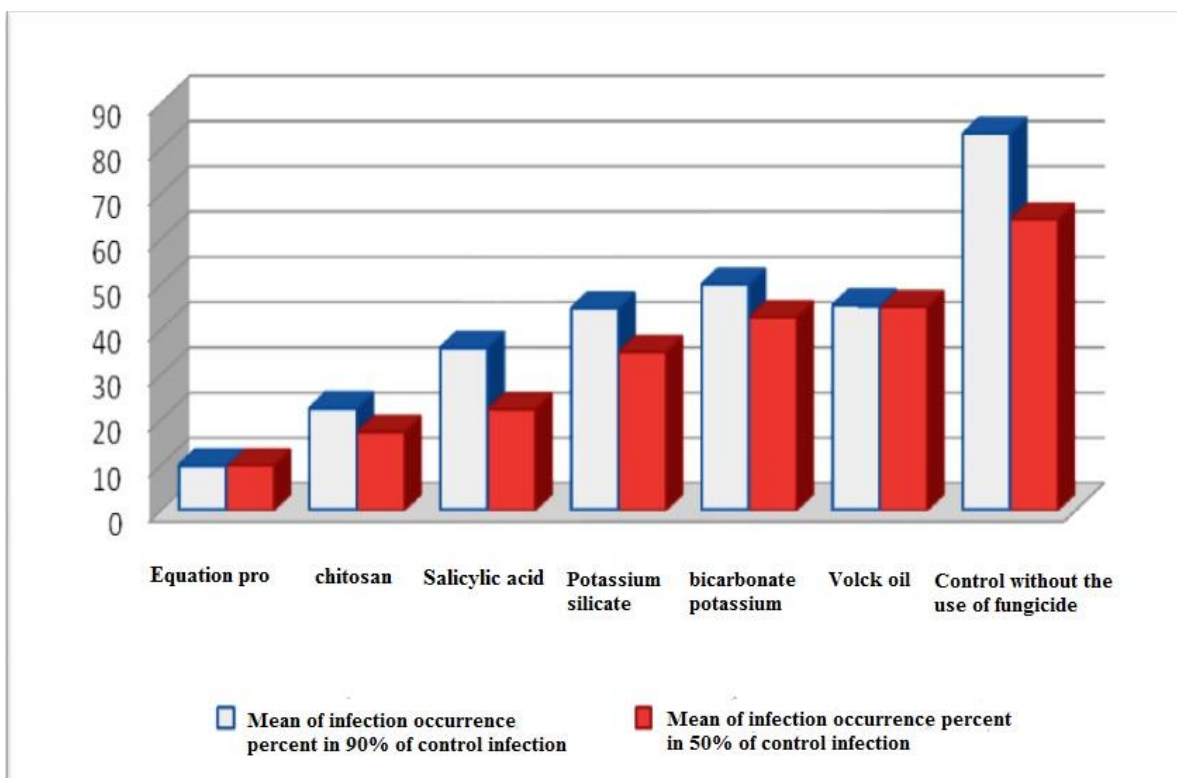


Diagram 1: The influence of treatment time on the disease occurrence percent in the control infection levels of 50% and 90%

Discussion

The extensive use of chemical pesticides caused to forget the supreme achievement of human experiences and studies in fields of medicine, veterinary and herbal medicine for a period of 50 years. Increasing the production and consumption of organic products and observing the interesting and fascinating results of using herbals in medicine and non-chemical pesticides (vegetable oil, bio-control elements, minerals, natural essences or herbal extracts, secondary metabolites and etc) in agriculture and food industry and also the pressure of organs and supervising agencies regarding the quality of food, agricultural products and environment drew the attention of the scientific society again to the subject of substituting chemical pesticides from a new perspective (IFOAM, 2002). In the current study the effect of using volck oil, bicarbonate potassium, potassium silicate, salicylic acid, chitosan and equation pro fungicide on downy mildew on cucumber was studied. To fight with downy mildew a common approach is using fungicides (Gisi, 2002). Fungicides play an important and critical role on controlling the disease in epidemic conditions (Bhedad, 1990; Mozaffari *et al.*, 2000; Elahinia, 2007). In this experiment we used common fungicides and minerals to control the disease. Results indicated that the equation pro fungicide (Famoxadone+cymoxanil) was the most efficient comparing other treatments. Results of the current study are in correspondence with results obtained by other researchers. Samavatian experimented 11 numbers of chemical pesticides to eliminate downy mildew in Esfahan. He observed that the equation pro fungicide had a better reducing and curing effect (Samavatian, 2009). Colucci used 26 fungicides in his experiments and asserted that among them famoxadone+cymoxanil (equation pro), zuxamide, ciazofamid and

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propamocarb were more beneficiary comparing the control (Colucci, 2008). Colluci and Holmes compared the performance of fungicides including mancozeb, cymoxanil, mancozeb+cymoxanil, dimethomorph+mancozeb, mancozeb+oxadixyl, cymoxanil+mancozeb and famoxadone+ cymoxanil (equation pro) regarding controlling downy mildew on cucumber in field conditions (Colucci and Holmes, 2010).

It is suggested to use the fungicide famoxidine+cymoxanil (equation pro) to reduce the amount of chemical pesticides' residues and securing the health of consumers as it was proved to be the most efficient fungicide in controlling the disease. Also after the equation pro acid salicylic is the most influential fungicide which may be applied in a synthetic management with this disease.

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