

## Study on the control possibilities of *Parthenolecanium rufulum* Ckll. by *Verticillium lecanii* (Zimm.) Viégas

Mehmet Işık\*

Müjgan Tunçdemir\*

Ali F. Yanılmaz\*

### Summary

The chemical tests were carried out against *Parthenolecanium rufulum* Ckll. (Hom.: Coccidae) which is a hazelnut pest, in Maçka county, Trabzon in 1981 and 1982. Spor suspension of *Verticillium lecanii* (Zimm.) which is natural enemy of the pest was used at the rates of  $1 \times 10^6$  spores/ml.,  $1 \times 10^7$  spores/ml. as a character in the tests.

The result showed that *V. lecanii* suspension at  $1 \times 10^6$  spores/ml. gave 47.28 % and 62.3 % effectiveness in 1981; and at  $1 \times 10^7$  spores/ml. gave 67.4 % effectiveness in 1982 against the first stage larvae of the pest.

It is concluded that *V. lecanii* could be easily produced under laboratory conditions at any time.

### Introduction

*V. lecanii* has been known as natural enemy of various insects. The studies carried out in the past revealed that this fungus has an effect on *P. corni* and *P. rufulum* which are hazelnut pests as well as on *Pulvinaria floccifera* Westw. which is one of the tea pest in the Eastern Black Sea Region of Turkey. The preliminary studies have been initiated on this subject based on the results obtained from the previous studies.

This fungus has been extensively studied by many researcher in various countries.

Hall (1975, 1976) recorded that *V. lecanii* suspension gave good control of aphids causing damage to the plants in glasshouse. Samsinakova and Kalalova

(\*) Bölge Zirai Mücadele Araştırma Enstitüsü, PK 3, Samsun.

(1976) stated that the production of this fungus in the laboratory was possible within 4 weeks. Barson (1977) found that *V. lecanii* suspension at  $4.4 \times 10^8$  spores/ml. gave effective control of *Scolytus scolytus* F. at 5-30°C temperatures and 100 % relative humidity. Santharam et al. (1979) noted that *V. lecanii* that has been known to be effective against Homoptera, Coleoptera and Lepidoptera killed *Honosepilachna vigintioctopunctata* F. (Col.: Coccinellidae) causing damage to tomatoes and egg-plants in India 3 days after the application. Easwaramoorthy et al. (1979) recorded that *V. lecanii* together with fenthion gave 93.7% effectiveness against *Cossus viridis* Green that causes damage to coffee plants 14 days after the application, while *V. lecanii* alone 28.4% effectiveness against the same pest. It is recorded by Miczulski (1979) that *V. lecanii* is a parasite of *Lema gallaeciana* Hayden. Murakoshi (1979) stated that *V. lecanii* isolated from *Bombyx mori* L. pupae had a fatal effect on the larvae. Ekbohm (1979) recorded that the fungi namely *V. lecanii* and *Aschersonia* spp. were used in the biological control of *Trialeurodes vaporariorum* Westw. in the glasshouse. Galani (1980) noted that the efficacy of *V. lecanii* preparations increased when Mg, Fe and K ions were added to the preparations. Hall et Latge (1980) stated that the blastospores production of *V. lecanii* exhibited no difference at temperatures ranging from 10°C to 29.5°C. Hall (1980 a) indicated that high relative humidity was needed for viability of *V. lecanii* conidia. Hall (1980 b) reported that the effect of *V. lecanii* suspension exhibited no difference in spor concentration.

In Turkey the first study on this subject has been carried out by Levendoğlu (1956). The author reported that *V. lecanii* is so effective against *P. floccifera* in Rize province that the chemical control of the pest is not necessary. Alay (1965) found that the artificial inoculations of *V. lecanii* to *P. floccifera* failed to provide effective control of the pest. Iren (1970) noted that *V. lecanii* had an effect on *P. corni* in Tirebolu and Düzce counties to great extent.

This study has been undertaken to investigate the control possibilities of *P. rufulum* by artificial spores suspension of *V. lecanii*.

## Materials and Methods

The studies were carried out in the hazelnut orchards that highly infested with *P. rufulum* in Maçka county, Trabzon.

The experiment was set-up according to randomized block design with 3 replications, 5 and 6 characters in 1981 and 1982 respectively. *V. lecanii* spores suspension was a character in these trials.

The treatments were made against first stage (mobile) larvae on 19 August in 1981 and 13 August 1982. A hazelnut bush was considered as a plot. A row of untreated bushes was left between the plots.

Using knap-sack sprayer in 1981 and atomizer in 1982 the hazelnut bushes were sprayed so that complete coverage was achieved. *V. lecanii* suspension at  $1 \times 10^8$  spores/ml. in 1981 and at  $1 \times 10^7$  spores/ml. in 1982 was prepared and sprayed to the marked branch of each bush in the evening at the same day. Pinolen (Nu film P(R)) was used at 40 cc./decare in 1981.

Another experiment was set-up according to the randomized block design with 3 replications and 4 characters in 1981.

The countings were made on 10 September and 11 September 1982.

In 1981 the alive and infected larvae on the upper side of the 25 leaves taken from each plots at random were counted and evaluations were made according to Abbott formula.

In 1982 the alive and infected larvae along the midrib (1.5 cm. in width) of 25 leaves taken from each plots were counted. Evaluations were made by Abbott formula based on the rates of artificial and natural infections.

*V. lecanii* suspension used in the experiment was prepared according to the following method: The spores of the fungus was grown in a medium composed of 2.5 % glucose, 2.5 % starch, 2.0 % corn-steep liquor, 0.5 % NaCl, 2.0 %  $\text{CaCO}_3$  for 4-5 days (Samsinakova and Kalalova, 1976). At the end of this period the spores obtained from the culture were used for the production. Sabouraud dextrose agar (SDA) medium was put into many more petri dishes for production. These petri dishes were inoculated with spores and incubated at 20-22°C for 4 days (Hall and Burges, 1979).

SDA medium was composed of 40 gr. dextrose, 10 gr. pepton, 18 gr. agar, 1 lt. distilled water (Stockdale, 1971).

After 4 days, the fungal material developed in the petri dishes was removed. This material was centrifuged for 15-20 minutes and water added. This suspension was filtered through a cheese cloth. The concentration of the inoculum was measured with a hemocytometer. The concentration of the suspension was adjusted by adding water and spores.

## Results and Discussion

The results obtained from the experiment are shown in Table 1. It could be seen from the data that in 1981, *V. lecanii* suspension as a character gave

on average 47.28 % effectiveness against *P. rufulum* while it gave 67.4 % effectiveness in 1982.

The results of trial in which lateral branches were sprayed in 1981, showed that *V. lecanii* suspension gave on average 62.3 % efficacy against the pest.

According to the evaluations based on both natural and artificial infections by *V. lecanii*, the effectiveness was 81,66 % in lateral branches in 1981, 74,6 % and 75,9 % in the plots in 1981 and 1982 respectively.

The meteorological data recorded during the studies are shown in Table 2 and 3. It is evident from the data that the temperatures were favorable for the development of the fungus (Hall, 1980; Hall and Ladge, 1980). But there were not enough relative humidity for development of the fungus in the orchard, compared to the glasshouse.

Hall and Burges (1979) recorded that in the glasshouse at high relative humidity *V. lecanii* gave good control to the aphids. But in our trials *V. lecanii* suspension gave approximately 70 % effectiveness against *P. rufulum*. It seems possibly due to the low relative humidity level.

Levendoglu (1956) and İren (1970) stated that the natural infection rate of the fungus was so high that no chemical control against *P. corni* and *P. floccifera* was necessary. But in our opinion, this high natural infection rate occurs only under certain conditions.

On the other hand, we are not agree with the Alay (1965) states that *V. lecanii* were ineffective against *P. floccifera*. Because the rate of alive spores in the suspension and the time elapsed after the suspension has been prepared was unknown.

It is concluded from the results that *V. lecanii* is so effective against *P. rufulum* that the chemical control of this pest is not necessary when *V. lecanii* suspension is applied to the pest.

It has also been found that under laboratory conditions this fungus could be easily produced.

It was no important difference between the plots sprayed and unsprayed with Pinolen.

Table 1. The results of the trials with *Verticillium lecanii* against *Parthenolecanium rufulum* in 1981 and 1982 in Mačka, Trabzon.

Years	Characters	Alive	Parasitized	Rate of alive	Effectiveness	Average effectiveness	Rate of parasitism	The rate of increase in natural infection	Average effectiveness
1981	V. lecanii, $1 \times 10^8$ spores/ml. + Pinolen	867	1225	42.3	54.1	47.28	57.7	94.2	74.6
		141	419	25.1	71.04		74.9	82.2	
		257	91	73.9	16.7		26.1	57.4	
	V. lecanii, $1 \times 10^8$ spores/ml. (Lateral branches)	24	478	4.7	96.1	62.3	95.3	96.5	81.66
		74	176	29.7	65.7		70.3	81.08	
		840	439	65.6	26.1		34.4	67.4	
	Control	630	22	96.7	—	—	3.3	—	—
		2223	343	86.7	—		13.3		
		118	15	88.8	—		11.2		
1982	V. lecanii, $1 \times 10^7$ spores/ml.	937	1098	46.04	41.4	67.4	53.96	60.3	75.9
		186	798	18.9	79.7		81.1	91.8	
		127	722	14.9	81.2		85.1	75.8	
	Control	932	262	78.6	—	—	21.4	—	—
		1260	89	93.4	—		6.6		
		1090	257	79.5	—		20.5		

Table 2. — Meteorological data between from 19 August to 10 September in 1981 in Maçka, Trabzon.

Days	Mean daily* temp. (C°)	Mean daily** R.H. (%)	Rainfall (mm.) (7 am to 7 pm 24 hour)
19	23.8	70	—
20	23.2	72	—
21	24.1	73	—
22	24.5	73	—
23	23.8	70	0.6
24	22.6	71	—
25	23.4	70	—
26	20.9	77	—
27	19.8	76	1.7
28	17.4	81	0.3
29	20.3	74	—
30	16.8	82	10.8
31	16.9	84	2.2
1	17.3	82	11.4
2	21.5	60	1.2
3	21.3	70	—
4	20.4	74	—
5	19.0	87	—
6	18.4	90	—
7	20.6	77	—
8	21.4	72	—
9	23.8	70	—
10	21.9	72	—

\* : max: 32.0°C, min: 14.0°C

\*\* : max: 90 , min: 58

Table 3. — Meteorological data between from 13 July to 11 August in 1982 in Maçka, Trabzon.

Days	Mean daily* temp. (C°)	Mean daily** R.H. (%)	Rainfall (mm.) (7 am to 7 pm 24 hour)
13	20.9	72	—
14	19.9	86	—
15	20.7	81	—
16	21.4	81	2.7
17	20.5	87	1.5
18	19.4	89	1.6
19	19.2	87	0.4
20	20.1	79	—
21	19.3	73	—
22	20.6	83	—
23	20.1	82	—
24	20.5	74	—
25	21.1	77	0.8
26	21.3	79	—
27	21.2	79	—
28	19.8	85	—
29	19.5	84	9.1
30	17.7	83	2.4
31	15.8	87	2.1

\* : max: 29.0°C, min: 12.5°C

\*\* : max: 95, min: 44

## Ö z e t

### **Verticillium lecanii (Zimm.) Viegas ile Parthenolecanium rufulum Ckll.'un mücadele imkânları üzerinde araştırmalar**

Bir fındık zararlısı olan *Parthenolecanium rufulum* Ckll. (Hom.: Coccidae)'a karşı Maçka (Trabzon) ilçesinde 1981 ve 1982 yıllarında yapılan ilaç denemelerinde bu zararlının doğal paraziti olan *Verticillium lecanii* Zimm. mantarının  $1 \times 10^6$  ve  $1 \times 10^7$  spor/ml.'lik süspansiyonu karakter olarak kullanıldı.

1981 Yılında  $1 \times 10^6$  spor/ml.'lik mantar süspansiyonu Pinolen yapıştırıcısıyla birlikte bir ilaç gibi değerlendirildiğinde, ortalama % 47.28 etkili olmuştur. Yan dallarda yapıştırıcı kullanılmadan yapılan deneme ise % 62.3 oranında bir tesir göstermiştir. Aynı denemelerde enfeksiyon oranı üzerinden yapılan değerlendirmeye göre etkiler, sırasıyla % 74.6 ve % 81.66 olmuştur.

1982 Yılında ise  $1 \times 10^7$  spor/ml.'lik *V. lecanii* süspansiyonu ile yapılan denemeden ortalama % 75.9 olmuştur. Değerlendirmelerde ölü bireyler dikkate alınmamıştır. Bu etkiler, zararlının zarar seviyesinin altına düşmesi için yeterlidir.

*V. lecanii* mantarının laboratuvarında istenilen zamanda ve miktarda kolaylıkla üretimi sağlanmıştır.

Pinolen yapıştırıcısıyla ilaçlanan ve ilaçlanmayan parseller arasında etki yönünden bir farklılık tesbit edilememiştir.

#### Literature cited :

- Alay, K., 1965. *Pulvinaria floccifera*'ya karşı *Verticillium lecanii* ile biyolojik savaş imkânları üzerinde araştırmalar. *Bitki Koruma Bülteni*, 5(3) : 113-120.
- Barson, G., 1976. Laboratory studies on the fungus *Verticillium lecanii*, a larval pathogen of the Large elm bark beetle, *Scolytus scolytus*. *Annals of Applied Biology*, 83(2) : 207-214 (RAE, 65(1) : 442).
- Easwaramoorthy, S., A. Regupathy, G. Santharam and S. Jayaraj, 1978. The effect of subnormal concentrations of insecticides in combination with the fungal pathogen, *Cephalosporium lecanii* Zimm. in the control of Coffee green Scale, *Coccus viridis* Green. *Zeitschrift für Angewandte Entomologie*, 86(2) : 161-166 (RAE, 67 : 3387).
- Ekbon, B.S., 1979. Biological control of the Glasshouse white fly *Trialeurodes vaporariorum*. Brief history and future prospects. *Entomologisk Tidskrift*, 100(1) : 1-10 (RAE, 67(12) : 4822).
- Galani, G., 1979. Investigations on the variation in pathogenicity of the fungus *Verticillium lecanii* (Zimm.) Viégas to the larvae of the insect *Trialeurodes vaporariorum* Westw. *Analele Institutului de Cercetari pentru productia plantelor*, 15 : 243-248 (RAE, 68 (11) : 5662).



- Hall, R.A., 1975. Aphid control by a fungus, *Verticillium lecanii*, within an integrated programme for *Chrysanthemum* pests and diseases. Glasshouse Crops Research Institute Worthing Road, Littlehampton, Sussex BN 16 3 PU. 94-98 S.
- , 1976. A Bioassay of the pathogenicity of *Verticillium lecanii* conidiospores on the Aphid, *Macrosiphoniella sanborni*, *Journal of Invertebrate Pathology*, 27 : 41-48.
- , and H.D. Burges, 1979. Control of aphids in glasshouses with the fungus, *Verticillium lecanii*. *Annals of Applied Biology*, 93(3) : 235-246.
- , et J.P. Latge, 1980. Étude de quelque facteurs stimulant la formation invitro des blastospores. C.R.A. cad. SC. Paris. Serie D. 291 : 75-78.
- , 1980 a. Effect of relative humidity on survival of washed and unwashed conidiospores of *Verticillium lecanii*. *Acta Oecologica. OEcol. Applic.*, 1(3) : 265-274.
- , 1980 b. Control of aphids by the fungus *Verticillium lecanii*. Effect of spore concentration. Glasshouse Crops Research Institute, Worthing Road, Littlehampton, BN. 16 3 PU, England.
- İren, S., 1970. Düzce ve Tirebolu'da fındıklara arız olan *Parthenolecanium corni* (Bouché)'yi parazitleyen *Cordiceps clavulatus* (Schw.) Ellis et Ev. ve *Verticillium lecanii* (Zimm.) Viégas üzerinde bir araştırma. Tarım Bakanlığı, Ziraî Mücadele ve Karantina Genel Müdürlüğü, 32 s.
- Levendoglu, R., 1956. Doğu Karadeniz Bölgesinde entomofit mantarlar. *Tomurcuk*, 5 (50) : 15.
- Miczulski, B., 1978. Further studies regarding natural control factors effecting *Oulema* spp. (Coleoptera; Chrysomelidae) in Poland. *Roczniki Nauk Rolniczych*, E, 7 (1) : 133-141 (RAE, 67 (2) : 591).
- Murakoshi, S., M. Ichinoe, A. Suzuki, Kanaoka, A. Isogai and S. Tamura, 1978. Presence of toxic substance in fungus bodies of the entomopathogenic fungi *Beauveria bassiana* and *Verticillium lecanii*. *Applied Entomology and Zoology*, 13 (2) : 97-102 (RAE, 67 (3) : 1238).
- Samsinakova, A. and S. Kalalova, 1976. Mass cultivation of entomophagus fungus *Verticillium lecanii*. *Ceska Mykologie, casopis ceskoslovenske vedeckie Spolecnosti promykologii Rocnik 30 Sesit 2. Seperatum*: 118-120.
- Santharam, G., E. Easwaramoorthy and S. Jayaraj, 1978. Preliminary laboratory evaluation of *Cephalosporium lecanii* Zimm. as a pathogen of brinjal leaf beetle, *Henosepilachna vigintioctopunctata* Fabr. *Current science Entomology*, 47 (13) : 477 (RAE, 67 (8) : 3362).
- Stocdale, M.P., 1971. «Diseases of keratinized tissues. 429-460» *Methods of in Microbiology* edited by C. Booth. Volume 4 Academic Press London and Newyork, 795 s.