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#### CONTENTS

Regional Disease Monitoring J. M. PI	RESCOTT		1
Long Distance Transportation an E. E. SA	d Expaosion of Wh ARI,	eat Rusts	7
Laboratory Studies of Three Spe chus urticae Koch (Acarine: I O. ECEN	cies of Fungi Patheg etranychidae)	genic to Tetrany-	13
Investigations on the Causal A Chemical Control Methods of the Grown in Greenhouses of Izmir	gents and Their P Damping - Off of Ca	athogenicities and arnation Seedlings	
E. SEZG	IN and I. Karaca		21
(Burkholder) (Dowson) of Beans Y. E. Öl	light ( <b>Pseudoman</b> : <b>KTEM</b>	as phaseolicola)	31
Elektronenmikroskopische Unters an Tomaten	uchungen über die	Stolburkrankheit	
Ü. YOR	GANCI		35
VOLUME: 5 NU	JMBER: 1	JAN. : 19	76

# Dizgi ve Baskı BİLGEHAN MATBAASI

J. Turkish Phytopath. Vol. : 5 Num. : 1 - 1976

# Regional Disease Monitoring\*

J. Michael PRESCOTT CEMMYT/Pathologist

Cereals. especially wheat and barley, are widely grown in the region bounded by Bangladesh in the east and Morrocco in the west In fact they are the main cropsof most of the countries within this region. Increased cereal production by way of growing varieties with higher yield potential, improved manage ment practices, and better protection against pests and pathogens is the goal of a modern agriculture. Pests and pathogens annually account for the destruction of at least one-fifth of the world production. This is entirely too high a price to pay. Many of the production problems, especially the pathogenes are similar throughout the region. These pathogens have called "Shifty Enemies" because of their mobility and their ability to adapt to changes. Changes in the pathogen situation are usually evident at one or a few locations prior to appearing at another. With this changing situation in mind and since many airbounde pathogens are quickly dispersed by weather sys -

tems, a disease monitoring system becomes a necessity. For this reason a regional disease surveillance program was initiated in 1971.

The actual beginnings was in the fall of 1970 when the First Turkish Trap Nursery was sent to many cooperators in Turkey and to seven locations within the region. This nurserv was comprised of commercial varieties grown in Turkey and in the adjacent countries plus a num ber of resistant and susceptible checks. The response to this first monitoring or Trap Nursery was favorable and it was decided to expand this type of disease surveillance to more countries within the region. Thus with the first Turkish Trap Nursery as a nucleus the Regional Trap Nursery (RTN) came into existence in the fall of 1971

The objectives for the RTN resulted from the experience gained in Wheat Improvement Programs in Turkey, India, Mexico, and the United States. They are:

1

\*) Paper presented at the First Turkish Plant Pathology Congress Izmir, Turkey 20-24 Oct. 1975

1) to monitor the disease situation throughout the region using com mercial and check varieties;

2) to determine the virulence potential of pathogens within the region;

3) to detect shifts or changes in virulence within the region and to provide advance warning of these shifts;

4) to assist in mapping the geographical distribution movement of the diseases within the region;

5) to determine the influence of the commercial varieties and their role in the selection and increase of virulent forms of the various diseases, particularily the rusts, within the region.

An effort was made to include in this nursery a representative number of the commercial wheat varieties. that is varieties grown on at least 100,000 ha, or more, along with appropriate checks. The number of entries selected for the nursery are not fixed and can be increased or descreased as the situation warrants each year. The RTN'71-'72 was comprised of 40 varieties, not replicated, and sent to 100 cooperators in 31 countries. The Turkish Wheat Improvement Program, CIMMYT, RF, ALAD/FF, and FAO were respon sible for this regional effort. This program was contiuned in '72 - '74 program was contiuned in '72 - '73 and '3 - '74 in this format. In the third year of this program (1973-74), there were 45 varieties included in the nursery and sent to 100 cooperators in 35 countries.

In 1974 the regional disease surveillance program was reorganized. the objectives remain the same but the manner of achieving these objectives were changed. During the first years of this program these were several specific disease monitoring nurseries being grown within the region. These nurseries included (1) The Yellow Rust Trial (Dr. R.W. Stubbs). (2) The European Leaf Rust Nursery (Dr. M. Boskovic). (3) The Egyptian Nursery (Dr. A.H. Kamel) and (4) The Regional Trap Nurserv (Drs. J.M. Prescott and E.E. Saari) To avoid duplication of effort, to save time and needed nursery space, and to provide a stronger network for the regional disease surveillance program these four nurseries were combined into one nursery The Regional Disease Trap Nursery (RDTN). The identification of races and virulence of stripe, leaf, and stem rust will be performed by Drs. Stubbs, Boskovic, and Kamel respectively. The management, dispersal of seed, field note taking, and data summerization will be done by Drs. Saari and Prescott. This pooling of scientific effort will enhance the possibility of achieveing the objectives of the regional disease surveillance program.

In 1974-75 the reorganized RDTN included 172 commercial and special rust identifying varieties or lines of

#### J. M. PRESCOTT

wheat and barley. The nursery was grown by 200 cooperators in 46 countries. This season, the RDTN is being grown by 145 cooperators in 41 countries (Table I). There are 115 varieties or lines in this nursery. These varieties have been grouped together as follows:

- I. Commercial Varieties
  - A. Durum Wheat
  - B. Bread Wheat
    - 1. Spring habit
    - 2. Winter habit
  - C. Barley
- II. Special Purpose Varieties/Lines
  - A. Stripe Rust (Puccinia Striiformis)
  - B. Leaf Rust (Puccinia recondita)
  - C. Stem Rust (Puccinia graminis f.sp. tritici)

## TABLE I. COUNTRIES PARTICIPATING IN THE 1975-76 REGIONAL DISEASE SURVEILLANCE PROGRAM

1. Afganistan	11. Greece	21. Morocco	31. Switzerland
2. Algeria	12. India	22. Nepal	32. Sudan
3. Bangladesh	13. Iran	23. Netherlands	33. Syria
4. Bulgaria	14. Iraq	24. Nigeria	34. Tanzania
5. Cyprus	15. Italy	25. Oman	35. Tunisia
6. Czechoslavakia	16. Jordan	26. Pakistan	36. Turkey
7. Egypt	17. Kenya	27. Portugal	37. Yemen. North
8. England	18. Libya	28. Romania	38. Yemen, South
9. Ethiopia	19. Lebanon	29. Saudi Arabia	39. Yugoslavia
10. France	20. Madagascar	30. Spain	40. West Germany
	41 17	Charles and the second second second	

41. Zambia

To illustrate the endemic virulence and resistance trends of the region, a comparison of the average coefficients of infection for the three rust among local, improved, and dwarf wheat varieties in four geographical areas for the three year period 1972-74 is presented in Table II. A coefficient of infection greater than 10.0 suggests that the wheat varieties in this class have less adequate resistance and should be replaced.

#### REGIONAL DISEASE MONITORING

TABLE II. COMPARISON OF THE AVERAGE COEFFICIENT OF RUST INFECTION AMONG LOCAL, IMPROVED, AND DWARF WHEAT VARIETIES IN FOUR GEOGRAPHICAL REGIONS FOR THE THREE YEAR PERIOD 1972-74.

VARIETY* GROUP		INDIAN SUBCONTENT	MIDDLE EAST	NORTH AFRICA	SOUTH EUROPEA	AVE
A.)	STEM RUST	Г:		isod V	ant B	
	LOCAL	22.35	20.16	15.94	18.67	20.41
	IMPROVED	8.94	13.19	11.32	11.53	11.57
	DWARF	1.13	4.30	2.95	2.95	3.12
B.)	STRIPE RU	ST				
	LOCAL	23.67	18.28	14.61	10.73	15.97
	IMPROVED	8.07	9.46	5.24	8.23	8.39
	DWARF	4.20	6.39	2.37	2.79	5.62
C.)	LEAF RUST	in tap, triatal)			mang	
	LOCAL	36.92	17.34	29.80	21.70	26.11
	IMPROVED	27.92	14.74	18.40	12.03	18.84
	DWARF	16.32	8.17	5.00	2.33	8.35

\* DATA BASED ON:

1972 7 LOCAL, 12 IMPROVED, 12 DWARF WHEAT VARIETIES
1973 7 LOCAL, 31 IMPROVED, 14 DWARF WHEAT VARIETIES
1974 7 LOCAL, 21 IMPROVED, 15 DWARF WHEAT VARIETIES

A value greater than 5.0 but less than 10.0 suggests reasonable degrees of resistance however, new levels or sources of resistance may be needed shortly. A value less than 5.0 suggests the presence of adequate resistance.

From the data presented in Table II it is easily seen that the local varieties do not have adequate resistance to the three cereal rusts or in any of the four zones and could sustain heavy damage whenever the conditions for an epidemic are suitable. The improved varieties res - ponse varies as to the species of rust involved. In this variety group stem rust resistance is barely adequate and in the possible damage area. Stripe rust resistance is adequate but new sources or degrees of resistance may be needed shortly. Leaf rust resistance is not sufficient and loss in production could occur in all zones within the region. In the case of the dwarf varieties, stem rust resistance is good to adequate in all zones., stripe rust resistance is reasonable but could require a change of resis-

4

tance genes, and leaf rust resistance reasobable in al zones of the region except the Indian Sub - continent where new sources of resistance are needed.

In general, the levels of resistance expressed by these commercial varieties follow a step-wise descrease with the dwarf commercial wheat varieties expressing the highest degree of resistance, the improved varieties in the middle, and the local commercially grown varieties expressing little or no resistance.

A large amount of data has now accumulated and is being assembled to that the goals of the Regional Disease Surveillence program will be attained and also be of value to the participating countries of the region.

# Long Distance Transportation and Expansion of Wheat Rusts\*

#### E.E. SAARI

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The nature of the wheat rusts dictate that a living host be present for the perpetuation of the uredospores cycle except for brief periods of time. Since the rust diseases in most areas survive via the uredospore cycle the cropping, accessory and adventive phenology is an important factor in the survival of the rust organism. In a number countries a continuing series of host plants are available throughout the year, and the uredospore cycle is restricted only by environmental factors. In such cases the situation is referred to as endemic

There, are, however, definite examples where the host plants are cyclic, the rust organism dies out in the area, and a manner of "geographical sterilization" occurs. In these situations the re - establishment of pathogen is dependent upon a new cycle of the host - usually represented by the sowing of wheat - the transport of viable initial (primary) inoculum from a distant source, and favorable weather conditions for the deposition and infection process. This type of system is referred to as exodemic.

There are specific and well documented examples for both the endemic and exodemic models (2,3,4,5, 6,13,16,21). It should be remembered, however, that in most areas both systems probably function and tend to confuse our interpretation of results.

In any case, there are some specific examples of exodemic systems where the initial inoculum must be transported from some distant source. This aspect of long distance transport of wheat rusts has been the stimulus for international cooperation for many years. There is now additional interest in research efforts dealing with this subject, and for the purpose of this paper, I use long distance transport to mean distances in excess of 200 kilometers.

\*) Paper presented at the First Turkish Plant Pathology Congress Izmir, Turkey 20-24 Oct. 1975.

## LONG DISTANCE TRANSPORTATION AND EXPANSION OF WHEAT RUSTS

Probably the best documented long distance transport of rust spores is the example of **Puccinia graminis** f. sp. tritici in North America (13,20). Uredospore movement of 1000 km have been recorded on numerous occaions (20). Another well established system of long distance transport of stem rust uredospore occurs in Northwestern Europe. The disease cycle re-establishes itself each spring and it appears that the most likely source of inoculum is North Africa, or on occasion Southern Europe (19).

A specific exodemic situation which is being studied intensively is the Indian stem rust system. From extensive survey data re-establishment of stem rust into the plains of India occurs each year (6). The isochranes of rust appearance agree with crop phenology and there appears to be a close relationship with the isotherms of the monthly mean minimum temperature of 14°C. The environment and the absence of hosts for **P. graminis tritici** during the summer months eliminates the organism from the extensive plains area.

It has long been established that wheat and **P. graminis tritici** can be found at higher elevations in South India throghout the year (8). Stem rust also survives the summer period at the higher elevations in the northern hills of India, but the advent of cold weather shortly after sowing causes **P. graminis tritici** to "die out" The mean minimum for a three month period in the northern plains of India is below 10°C. In most cases such a low temperature would eliminate the stem rust fungus, and survey data collected indicates that the fungus indeed does not survive this period in north India (6).

The only remaining source of inoculum would be the hills (Nilig ris) in sourthern India which extents to over 2,000 meters in elevation. Wheat grown in the Niligris during the summer period is at the elevations of 1500 meters or more. From this source area, stem rust uredospores must travel a considerable distance to the wheat growing areas of the plains (in excess of 300 km).

The long distance transport of spores requires that they reach heights of 1000 meters or more (5). Once uredospore have reached a height of several kilometers the principle deposition mechanisms are down-draughts or raindrops (5).

The association of the first appearance of stem rust with a previous rainfall period in India indicated that uredospore deposition probably occurred with the rain (9, 10). A study was undertaken to determine if this form of deposition was important (11). The sampling methods used to trap uredospores of stem rust were the same as those used in North America (17).

The technique involved the use of a millipore filter attached to the end of a rain funnel (15). After each rain the filter was removed by cooperators a the different sites throughout India, and mailed to the Cereal Disease Laboratory, Indian Agricultural Research Institute, New Delhi. There the filters were processed and counts were made for the presence and number of spores trapped.

A total of 135 filters were processed over a five year period. This represents 50 test site - years (see table 1). The number of testing locations were not constant or the same throughout the study, and the number of rain samples received from each station in any year varied. A total of 83 samples or 61.5 % of the samples were found to have stem rust uredospores. Spore counts varied from a few spores to more than 5000 per sample.

At 25 test sites the rain sampling was associated with spore trapping studies using the rod impaction method (14). At the remaining 25 test sites the National Disease Trap Nursery was grown and weekly inspections of the susceptible chech variety were made for the first appearance of rust by cooperators (6).

Once stem rust spores were detected at a location subsequent rain samples with uredospores from that location were not counted in the data presented in Table 2. The percentage figures given are based upon the use of 50 test site-years. In 70 % of the cases a positive relationship exists. In 24 % of the cases spores were caught but rust wast not detected on the susceptible varieties in the nursery. The mere arrival of inoculum does not guarantee that the uredospores were viable or conditions were favorable for the infection process. In only 6 % of the cases did a negative relationship result. These negative examples could not be adequately explained but I feel confident that some unexplained experimental error is represented.

A comparison of the first date of the uredospores in rain samples and on the glass rod established an average difference of 22 days earlier detection for the rain sampling procedure. The date of rust appearance on the susceptible variety in the Trap Nursery avaraged 36 days after the first rain "ampling of uredospores.

The fact that uredospores are transported at high altitudes and become associated within cloud and rain systems suggests that Environmental Satellite Imagery or Satellite Television Cloud Photography may be a useful tool in studying long distance transport of uredospores (12). Cloud systems as viewed through such remote sensing programs provide a number of opportunities to map the movement of uredospores from a source to an area which is exodemic.

In this area of the world the most definitative exodemic wheat growing area is the Nile River Valley. Over summering of the host and the fungi (**P. graminis tritici**, **P. re**-

condita f. sp. tritici and P. striiformis f. sp. tritici) does not occur. All three rusts do, however, present a problem to wheat cultivation (1.5). Spore trapping data indicates that the rust arrives from some northernly sources (1). The rust isochranes also suggest a north to southward development in contrast to a gene-, ral northward and eastward developmental pattern in the adjacent countries (5). The environmental factors on crop and pathogen development need to be studied, and the weather systems, especially, the common low pressure cell probably has a major influence on rust development in Egypt.

If one beings to analyse certain virulence patterns and compares varietal reactions the indications are that Western Asia - parts of Iran, Syria, possibly Iraq and portions of Afghanistan - may serve as the inoculum reservoirs for Egypt (18). This area includes parts of the ancestorial home of wheat, and many related species are also present in this region. This provides for a bridging of inoculum through periods of reduced commercial acreage. In addition the cropping cycle at the higher elevations in this part of the world also provides a means by which the uredospores cycle can be perpetuate. The fact that many "islands of wheat' exists in this region, probably, provides for more than a single inoculum source. Unless all of the sources are identified and quantified, it will be difficult to use varietal response and race analysis data effectively.

Data and information is still too fragmented to draw any strong conclusions but if one reviews some of the weather systems common to this area (7) and considers both endemic and exodemic patterns a greater under-

Table 1. Summary for five	years of rain	sampling from	Nagrajan et al.	(11)	1.
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Years	Average No.	Rain	Samples with
	Stations	Samples	Uredospores
5	10	135	83

Table 2. Relationship of rain samples with spores (+) or no spores (--) and rust (+) or no rust appearance (--) from Nagarajan et al (11). See text for details.

d brieve s	Spores(+)	No spores ()	Spores(+)	No spores ()	
aanw oone	Rust (+)	No rust (—)	No rust(—)	Rust (—)	
Samples	24	11	. 12	3	
%	48	22	24	6	

10

standing of rust development seems possible. Turkey certainly must play a pivitol role in any wheat rust epidemiological patterns in the region because of all or the factors previousy mentioned. A greater understanding of what is happening here in Turkey will provide opportunities for minimizing rust attacks and losses to production. This knowledge will also provide some of vital missing bits of information that will allow other countries to develop the required background for controlling the losses caused by the wheat rusts.

I believe that First Turkish Phytopathological Society Congress represents progress toward this end. I will look forward to reading future proceedings, especially those articles contributing to our greater understanding of the wheat rust epidemiology.

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11

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# Laboratory Studies of Three Species of Fungi Pathogenic to Tetranychus urticae Koch (Acarine: Tetranychidae)

Doç. Dr. Osman ECEVIT

#### SUMMARY

Three species of fungi identified as **Cladosporium** sp., **Aspergillus ochraceus** and **A. niger** are reported as causing mortality of two-spotted spider mites, **Tetra nychus urticae** Koch. The fungi were isolated and the role of mite populations is discussed. **Cladosporium** species could be very important as a biological control agent for two-spotted spider mite. However, culture of the fungus is very difficult. **A. ochraceus** is readily disseminated in a veriety of ways, easily penetrates the mite and is easy to culture. In spite, **A. niger** is easy culture, of secondary importance as a possible biological control agent for two-spotted spider mite.

#### INTRODUCTION

During the past 30 or more years many investigations have been reported concerning pathogenic fungi attacking plant - parasitic mites but relatively few experiments to observe the activity of the fungi have been performed. In the literature many species of Phycomycetes, Ascomycetes, Basidiomycetes, and Fungi Im perfecti are recorded as pathogenic to mites, some of which seem to be important in controlling them. Petch (1940) reported that Halotydeus destructor became infected with Entomophthora acaricida. In 1944 the same author described another species, E. acaridis, from Acarina. Fisher (1950 a,b) observed that Hirsutella thompsonii caused injury to Phyllocoptruta oleivora and on non-sprayed citrus trees in June and July, these mites rapidly disappeared. In 1951 he reported that a species of Entomophthora caused

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#### TETRANYCHUS URTICAE KOCH (ACARINE : TETRANYCHIDAE)

mortalities of adult citrus red mites Panoncyhus citri, ranging from 32 -95 %. Muma (1955) observed two fungi pathogenic to citrus rust mites, P. oleivora. These were Hirsu tella thompsonii and a species of Entomophthora. Leatherdale (1965) found that Paecilomyces eriophytis (=Botrytis eriophytis) originally des cribed from an eriophyid mite, Cecidophyopsis ribis, was infective also to Eriophyes padi, Aceria hippocastani, and Panonychus ulmi.

A fungus attaking Eutetranychus banksi was first observed by Fisher (1954) and later described as Entomophthora floridana by Weiser and Muma (1966). This fungus produces single conidiophores with pyriform conidia. Selhime and Muma (1966) studied the biology of E.floridana and found that this pathogen had a 5-6 days life cycle. The death of the mites were caused by dense hyphal growth inside the body. Also, Baker and Neunzing (1968) reported that **H. thompsonii** attacked the blueberry bud mite, and Muma (1969) found that **E. floridana** caused the death of **Eutetranychus** banksi during the summer and fall, mite populations being reduced by 36.8-86.3 per cent.

Carner and Canerday (1968) observed an entomogenous fungus in fecting Tetranychus telarius and T. urticae. They identified it as Entomophthora fresenii, however this identification has been questioned by Wilding and Weiser (Carner, pres. comm. 1969, cf., Lipa, 1971). Recently Sannasi and Oliver (1971) studied the integument of the velvet mite, Dinathrombium giganteum and the histological changes caused by the fungus, Aspergillus flavus. Also, Kenneth, et al. (1972) found that Triplosporium floridanum caused mortality of spider mites in Israel.

#### MATERIAL and METHODS

In our laboratory, cultures of phytoseiid mites were being isolated in petri plates and two-spotted spider mites used as prey. Fungi were observed attacking the spider mites in these cultures and also it was noted that the stock culture of the spider mites was at a very low density and declining. Examination of infested leaf samples under the stereo microscope showed that the spider mites were infected with fungi. Slide mounts of the fungi were made using Hoyer's medium as the mountant. Examination of the slides indicated that at least two different species were present.

Isolation of each fungus was made on potato-dextrose medium in the Biological Control of Insects Research Laboratory at Columbia, Mo., and identification of the made by a specialist at he Peoria, Illinois USDA laboratory.

#### O. ECEVIT

Uninfected spider mite cultures were established in our laboratory and infected with each of the fungus isolated taken from the P-D media. Also, fungi isolated from the infected stock culture were found capable of infecting live mites. Unfortunately, the original source of the infected stock culture of two-spotted spider mites is unknown.

#### **RESULTS** and **DISCUSSION**

Three species of fungi were found to be infecting the two-spotted spider mites. They have been idetified as Cladosporium sp.; Aspergillus ochraceus; and Aspergillus niger. Frequently it was found that more than one species of fungi were growing on the same mite specimen. For this reason, isolation and establishing pure cultures had been difficult.

**Cladosporium** sp.- In our opinion this pathogen must be the most important of the three. Examination of numerous slides reveals that spores



Fig.1. Two-Spotted Spider Mite killed by Cladosporium sp.

#### TETRANYCHUS URTICAE KOCH (ACARINE : TETRANYCHIDAE)

of Cladosporium grow on the surface of the mite and the hyphae enter through the anus or mouthparts. It seems probable also this fungus is able to dissclve the body wall of the mite. Death of the mite follows penetration, by whatever method, and the mite, inside and out, is completely covered (Fig. 1). The fungus is brown in color.

This fungus seems also to cause mortality of mite eggs. The fungus first forms a covering over the egg, then penetrates inside (Fig.2). There are two ways in which the fungus might penetrate the egg, first, it may dissolve the shell and penetrate in that manner or, second, it may penetrate via the micropylar area. The probability seems to favor the latter because the fungus is almost always found at only one point inside the egg.

This **Cladosporium** species could be very important as a biological control agent for the two-spotted spider mite. However, culture of the fungus is very difficult. In our experience only a pair of culture plates were achieved and none of them grew very well. Another drawback is that dissemenation of the fungus is very slow and depends on mite movement primarily and very little by spreading over the leaf surface.

Aspergillus ochraceus.- One of the most important genera of fungi attaking insects and mites in Aspergillus. Hundreds of spores of A. ochraceus have been observed on the



Fig. 2. The egg of Tw-Spotted Spider Mite killed by Cladosporium sp.

body surfaces of the mites. This fungus seems able to penetrate the body wall of the spider mite and invade the body. From the spore a structure grows for a short distance and penetrates the cuticle. From ths a stalk (Fig. 3) emerges on the end of which a spore-bearing body forms. A single dead mite may have from 1 - 5 such structures.

This fungus is readily disseminated in a variety of ways, easliy penetrates the mite, and is easy to culture.

Aspergillus niger.- This species is very similar in penetrating ability, dissemination characteristics, and easy of culture to A. ochraceus (Fig. 4). There is also very little difference in conidiophore structure between the two. This fungus usually appears in cultures later than the two preceding and is therefore believed to be a more secondary cause of mite mortality. However, it has the same ability to penetrate into mites and to spread rapidly once it becomes established. In spite of its easy culture it is, in our opinion, of secondary importance as a possible miological control agent for spider mites.

In summary, many attempts have been made to use fungi for spider mite control and unquestionably fungi play a very important role in regulating mite populations. However, fungi are dependent to a high degree on local weather conditions and on the microenvironment in the immediate vicinity of the target spe-



Fig.3. Two-Spotted Spider Mite killed by Aspergillus ochraceus.

#### TETRANYCHUS URTICAE KOCH (ACARINE : TETRANYCHIDAE)



Fig. 4. Two-Spotted Spider Mite killed by Aspergillus niger.

cimens. As Carner and Canerday (1968) reported, attempts to grow these fungi on common mycological media are usually unsuccessful. My isolation attempts were unsuccessful more often than not, supporting the conclusion of these authors.

In a greenhouse experiment, I infested potted bean plants with **T**. **urticae**. Solution containing the several fungi were then sprayed on the plants, and each pot isolated with polyethylene plastic covers. Every week leaf samples were taken and examined for mite infection by the fungi but the experiment failed completely. Obviously a spray technique of application is not effective as compared to infestation by infected live mites or contaminated leaves.

It is very difficult to find only one species of fungus on a given mite as, for example, Fig. 1 shows only **Cladosporium** sp., Fig. 3 shows only **A. ochraceus**, and Fig. 5 shows only **A. niger.** Usually **A. ochraceus**, **A. niger**, and **Cladosporium** sp. are found in combination. The associations in

An Empusa in a mits. Proc.

all probability have considerable, but at present unknown, significance.

#### ACKNOWLEDGEMENTS

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#### ÖZET

# Tetranychus urticae (ACARINA: TETRANCYHIDAE)'DA PATHOGENİK ÜÇ FUNGUS TÜRÜ ÜZERİNDE LABORATUVAR ÇALIŞMALARI

Tetranychus urticae (Koch)'da ölüm meydana getiren üç tür pathogenik fungus bulunarak Cladospori um sp., Aspergillus ochraceus ve Aspergillus niger olarak tanılanmıştır. Funguslar izole edilerek akar populasyonları üzerindeki tesirleri tesbit edilmiştir. Cladosporium sp., T. urticae ile yapılacak mücadelede çok önemli olacağa benzemektedir. Bununla beraber bu fungusun kültüre alınmasının çok zor olduğu görül müştür. **A. ochraceus** muhtelif yollardan kolaylıkla yayılabilen, kolay kültüre alınabilen ve akar'ın vucuduna nûfuz edebilen bir fungustur. Buna karşılık, **A. niger** kolay kültüre alınabilmekle beraber, kanaatıma göre **T. urticae'nın** kontrolunda ikinci derecede rol oynayacaktır.

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in greenhouses which covered with

Emel SEZGIN\*

# Investigations on the Causal Agents and Their Pathogenicities and Chemical Control Methods of the Damping-Off of Carnation Seedling3 Grown in Greenhouses of Izmir

Commercial flower prowing has be-

İbrahim KARACA\*\*

MORTOLADORIZI

This study was conducted on the causel agents and their pathogenicities and chemical control methods of the damping-off of carnation seedlings grown in greenhouses of İzmir in 1972-1974. In this study 41 genera of fungi were isolated from diseased samples collected at two different times.

ABSTRACT

and

The genera of Sclerotium, Macrophomina, Fusarium, Acremoniella, Cladorrhinum, Stilbella, Rhizoctonia, Nigrospora, Melanospora, Verticillium, Alternaria, Sordaria, Pythium and Actinomucor were selected for pathogenicity tests among all of the isoated fungi according to the results of the preliminary laboratory tests. In the pathogenicity tests, these fungi were used alone and combined with saprophytic fungi such as Aspergillus, Myrothecium, Penicillium, Trichoderma, Chaetomium and Gliocladium. When all the tested pathogens were combined with saprophytic fungi, it was generally determined that the incidence of the disease increased.

In the chemical control tests the genera of Sclerotium, Rhizoctonia Fusarium Macrophomina, Cladorrhinum, Melanospora, Pythium, Acremoniella, Stibella and Actinomucor and Methyl bromide, Orthocide soil treater Dexanal and Benlate chemicals were used. There have not been obtained any satisfactory effectivenesses from the chemicals against the causal agents of the damping-off carnation seedlings.

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#### INTRODUCTION

Commercial flower growing has become a great importance in Turkish agriculture. Flower growing were do ne in three sections such as cuttings flower growing, greehnouse flower growing and flower bulbs and corms growings. The value of these sections were 12.000.000, 35.000.000 TL. res pectivelly in 1968-1972 and 1974 (OS-MANLIOĞLU, 1974).

In Ege Region green house flower growing began in 1936 and it was done in an area of 2000 m<sup>2</sup> and soon become enlarged According to 1971-1972 statistics, total flower growing area is 220.766m<sup>2</sup> in Balçova and Narlidere. Generally flowers are grown in greenhouses which covered with polyethylene as a family enterprise. The most growing flower is carnation (Dianthus, caryophyllus L.). There are many diseases and pests probems of carnation in our region. One of them is damping-off disease which causes damage in early stage of carnations. In some years this damage is very high because of growers don't use any chemicals against soil born fungi.

This study was done to determine root mycoflora, their pathogenicities and chemical control methods of the damping-off disease of carnation in 1972-1974.

#### MATERIAL AND METHODS

Survery were done at two times in order to collect diseased cuttings and seedlings. During the surveys 2925 samples were collected from 117 greenhouses. The roots of diseased samples were washed with tap water and cut into small pieces with a sterilized scalpel. Their surface sterilized with 0.5 % sodium hypochloride for one minute and washed in sterilized water and then dried between sterile blotting papers. The root pieces placed in petri dishes. The isolation mediums were %2 PDA, czapeckdox agar and 8 % water agar. The petri dishes were incubated at 20°C

Pathogenicity tests were performed in pots. Test plants were grown from cuttings taken from healthy plants and rooted in sterilized soil. Genera of Fusarium, Rhizoctonia, Pythium, Sclerotium, Macrophomina, Alternaria, Cladorrhinum, Nigrospora, Acremoniella. Stilbella, Actinomuccor. Sordaria. Verticillium and Melano spora were selected for pathogenicity tests among all of the isolated fungi according to the preliminary laborato ry tests. These genera were both applied singly and in mixture with saprophytic fungi such as Gliocladium. Aspergillus, Chaetomium, Penicillium,

### Myrothecium and Trichoderma.

Inoculations were done according to the WILES (1952) method. Tested fungi were cultured on PDA for 10 days then 5 petri dishes of each fungi were blended in 250 cc sterilized water for 5 min. at high speed in waring's blender. Roots of the experimental seedlings were dipped in these inoculum for 5 min. and the seedlings were transplanted to pots. Roots of check plants were dipped only in sterilized water. Results of pathogenicty tests were obtained 50 days after inoculation.

In the chemical control test, the genera of Sclerotium, Rhizoctonia, Melanospora, Fusarium, Macropho mina, Cladorrhinum, Pythium, Acremoniella, Stilbella and Actinomucor which they shown above 30 % patho-

genicity in the pathognicity tests were used. These and 6 saprophytic Myrothecium, (Aspergillus, fungi Penicillium, Trichoderma and Gliocladium were combined and then added to pots. After 7 days when the fungi colonized in the pot soil methyl  $(100 \text{ cc} / 1\text{m}^3),$ bromide Benlate (methyl 1- (butyl-carbomyl)- 2-benzimidazole carbomate) 0,5 /lt. Orthocide soil treater (10 % N-trichvoromethyl - mercapto - 4- cyclohexene-1,2 dicarboximide (captan) and 60 % PCNB) 10 Kg/hec. and Dexonal (P-Dimethyl ominolbenzol - diazonat rium sulfanat 2,5 % + 10 % PCNB) 200 mg /kg soil. Were applied. 5 seedlings per pot were sown. After two months from aplications effects of the chemicals were determined by counting the healthy and diseased seedlings.

#### **RESULTS AND DISCUSSION**

A — Isolated genera and their rates of occurence :

The genera of fungi isolated from diseased samples were collected at two different times and their persentage, occurence in the total isolates are shown in table.

41 genera of fungi were isolated from diseased samples. Among these genera the most commonly fungus was **Fusarium** which were isolated from all of the greehouses which were surveyed. The genera of **Fusari**um is common for soils and rhizosphere in Turkey and other countries (MENON, WILLIAMS, 1957; PAR -KINSON et al., 1963; KUBIKOVA, 1968; BAGGA, 1970; BORA, 1972; TURHAN, 1973; SARIBAY, 1974).

The other genera of fungi followed **Fusarium** have different rates of occurences at both surveys. This can be attributed to the ecological conditions **especially** to soil temperature. For example **Sclerotium** species give damage to seedlings especially during the summer months (AN -DREUCCI 1955) and **Aspergillus** spe-

The second se		Survey I (1-15.5.1972)			Survey II (15-7-15.8.1972)			
Fusarium       177       32.89       100       849       38.22       100         Rhizoctonia       47       8.73       60.4       350       15.75       91.8         Aspergillus       6       0.92       2.3       234       10.53       33.7         Pythium       45       8.36       48.8       125       5.62       48.7         Gliocladium       66       12.26       74.4       102       4.59       64.8         Melanospora       1       0.18       2.3       101       4.54       35.1         Macrophomina       17       3.15       23.2       -97       4.36       63.5         Sclerotium       2       0.37       4.6       65       2.92       2.9.7         Alternaria       64       11.89       88.3       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.62         Cladornhinum       16       2.97       2.09       23       1.03       17.5         Trichoderma       3       0.55       4.6       19	Genera of Fungi	No of recovered isolates	Relative intensity (%)	Rates of presence in the nurseries (%)	No of recovered isolates	Relative intensity (%)	Rates of presence in the nurseries (%)	
Rhizoctonia         47         8.73         60.4         350         15.75         91.8           Aspergillus         6         0.92         2.3         234         10.53         33.7           Pythium         45         8.36         48.8         125         5.62         48.7           Gliocladium         66         12.26         74.4         102         4.59         64.8           Melanospora         1         0.18         2.3         101         4.54         55.1           Macrophomina         17         3.15         23.2         -97         4.36         63.5           Sclerotium         2         0.37         4.6         65         2.92         29.7           Alternaria         64         11.89         88.3         42         1.89         44.5           Mucor         7         1.30         11.6         30         1.35         24.3           White steril mycel         3         0.55         6.9         28         1.62         27.02           Cladorhinum         16         2.97         20.9         23         10.3         17.5           Trichoderma         3         0.55         4.6         <	Fusarium	177	32.89	100	849	38.22	100	
Aspergillus       6       0.92       2.3       234       10.53       33.7         Pythium       45       8.36       48.8       125       5.62       48.7         Gliocladium       66       12.26       74.4       102       4.59       64.8         Melanospora       1       0.18       2.3       101       4.54       35.1         Macrophomina       17       3.15       23.2       -97       4.36       63.5         Sclerotium       2       0.37       4.6       65       2.92       29.7         Alternaria       64       11.89       88.3       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.26       27.02         Cladorrhinum       16       2.97       20.9       23       10.3       17.5         Trichoderma       3       0.55       4.6       19       0.85       25.6         Papulospora       2       0.37       2.3       18       0.81       16.2         Rhizopus       5       0.74       6.9       <	Rhizoctonia	47	8.73	60.4	350	15.75	91.8	
Pythium         45         8.36         48.8         125         5.62         48.7           Gliocladium         66         12.26         74.4         102         4.59         64.8           Malanospora         1         0.18         2.3         101         4.54         35.1           Macrophomina         17         3.15         23.2         -97         4.36         63.5           Sclerotium         2         0.37         4.6         65         2.92         29.7           Alternaria         64         11.89         88.3         42         1.89         44.5           Mucor         7         1.30         11.6         30         1.35         24.3           White steril mycel         3         0.55         6.9         28         1.26         27.02           Chaetomium         -         -         -         27         1.21         16.2           Cladorrhinum         16         2.97         20.9         23         1.03         17.5           Trichoderma         3         0.55         4.6         19         0.85         25.6           Papulospora         2         0.37         2.3         18	Aspergillus	6	0.92	2.3	234	10.53	33.7	
Gliocladium         66         12.26         74.4         102         4.59         64.8           Melanospora         1         0.18         2.3         101         4.54         35.1           Macrophomina         17         3.15         23.2         -97         4.36         63.5           Sclerotium         2         0.37         4.6         65         2.92         29.7           Alternaria         64         11.89         88.3         42         1.89         44.5           Mucor         7         1.30         11.6         30         1.35         24.3           White steril mycel         3         0.55         6.9         28         1.26         27.02           Cladorrhinum         -         -         -         27         1.21         16.2           Cladorrhinum         16         2.97         20.9         23         1.03         17.5           Trichoderma         3         0.55         4.6         19         0.85         25.6           Papulospora         2         0.37         2.3         18         0.81         16.2           Penicillium         6         1.11         4.6         14 <td>Pythium</td> <td>45</td> <td>8.36</td> <td>48.8</td> <td>125</td> <td>5.62</td> <td>48 7</td>	Pythium	45	8.36	48.8	125	5.62	48 7	
Melanospora       1       0.18       2.3       101       4.54       35.1         Macrophomina       17       3.15       23.2       -97       4.36       63.5         Sclerotium       2       0.37       4.6       65       2.92       29.7         Alternaria       64       11.89       88.3       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.26       27.02         Chaetomium       —       —       —       7       1.31       16.2       27.02         Chaetomium       —       —       —       7       1.31       16.2       27.02         Chaetomium       —       —       —       7       1.31       16.2       27.02         Chaetomium       6       2.97       20.9       23       1.03       17.5         Trichoderma       3       0.55       4.6       19       0.85       25.6         Papulospora       2       0.37       2.3       18       0.81       21.1         Myrothecium       — <td< td=""><td>Gliocladium</td><td>66</td><td>12.26</td><td>74.4</td><td>102</td><td>4 59</td><td>64.8</td></td<>	Gliocladium	66	12.26	74.4	102	4 59	64.8	
Macrophomina       17       3.15       23.2       -97       4.36       63.5         Sclerotium       2       0.37       4.6       65       2.92       29.7         Alternaria       64       11.89       88.3       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.26       27.02         Chaetomium       —       —       —       7       1.21       16.2       27.02       23       1.03       17.5         Trichoderma       3       0.55       4.6       19       0.85       25.6       25.6         Papulospora       2       0.37       2.3       18       0.81       16.2         Rhizopus       5       0.74       6.9       17       0.76       13.5         Actinomucor       32       5.94       23.2       16       0.72       12.1         Myrothecium       —       —       —       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         He	Melanospora	1978911	0.18	2.3	101	4.54	25.1	
Sclerotium       2       0.37       4.6       65       2.92       29.7         Alternaria       64       11.89       88.3       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.26       27.02         Chaetomium       —       —       —       27       1.21       16.2         Cladorrhinum       16       2.97       20.9       23       1.03       17.5         Trichoderma       3       0.55       4.6       19       0.85       25.6         Papulospora       2       0.37       2.3       18       0.81       16.2         Rhizopus       5       0.74       6.9       17       0.76       13.5         Actinomucor       32       5.94       23.2       16       0.72       12.1         Myrothecium       —       —       —       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       3       0.55       6.9       4	Macrophomina	17	3.15	23.2	-97	4 36	62 5	
Alternaria       64       11.89       83.       42       1.89       44.5         Mucor       7       1.30       11.6       30       1.35       24.3         White steril mycel       3       0.55       6.9       28       1.26       27.02         Chaetomium       —       —       —       27       1.21       16.2       27.02         Cladorrhinum       16       2.97       20.9       23       1.03       17.5         Trichoderma       3       0.55       4.6       19       0.85       25.6         Papulospora       2       0.37       2.3       18       0.81       16.2         Rhizopus       5       0.74       6.9       17       0.76       13.5         Actinomucor       32       5.94       23.2       16       0.72       12.1         Myrothecium       —       —       —       14       0.63       12.1         Penicillium       6       1.11       4.6       14       0.63       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9 <td< td=""><td>Sclerotium</td><td>2</td><td>0.37</td><td>4.6</td><td>65</td><td>9.09</td><td>00.0</td></td<>	Sclerotium	2	0.37	4.6	65	9.09	00.0	
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White steril mycel3 $0.55$ $6.9$ $28$ $1.26$ $27.02$ Chaetomium——— $27$ $1.21$ $16.2$ Cladorrhinum $16$ $2.97$ $20.9$ $23$ $1.03$ $17.5$ Trichoderma3 $0.55$ $4.6$ $19$ $0.85$ $25.6$ Papulospora2 $0.37$ $2.3$ $18$ $0.81$ $16.2$ Rhizopus5 $0.74$ $6.9$ $17$ $0.76$ $13.5$ Actinomucor $32$ $5.94$ $23.2$ $16$ $0.72$ $12.1$ Myrothecium——— $-14$ $0.63$ $12.1$ Penicillium6 $1.11$ $4.6$ $14$ $0.63$ $13.5$ Stilbella7 $1.30$ $11.6$ $12$ $0.54$ $9.4$ Helminthosporium4 $0.74$ $9.3$ $9$ $0.40$ $12.1$ Sordaria2 $0.37$ $2.3$ $8$ $0.36$ $8.1$ Cladosporium3 $0.55$ $6.9$ $4$ $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ $4$ $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ $4$ $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ 2 $0.09$ $2.7$ Stachybotris——— $1$ $0.04$ $1.3$ Monocrosporium—— $1$ $0.04$ $1.3$ Monocrosporium—— $1$ $0.04$ </td <td>Mucor</td> <td>7</td> <td>1.30</td> <td>11.6</td> <td>30</td> <td>1.09</td> <td>44.0</td>	Mucor	7	1.30	11.6	30	1.09	44.0	
Chaetomium271.2627.02Cladornhinum162.9720.9231.0317.5Trichoderma30.554.6190.8525.6Papulospora20.372.3180.8116.2Rhizopus50.746.9170.7613.5Actinomucor325.9423.2160.7212.1Myrothecium140.6312.1Penicillium61.114.6140.6313.5Stilbella71.3011.6120.549.4Helminthosporium40.749.390.4012.1Sordaria20.372.380.368.1Cladosporium30.556.940.189.4Dark steril mycel20.372.340.182.7Doratomyces10.182.320.092.7Stachybotris10.041.3Beauveria10.041.3Monocrosporium10.041.3Monocrosporium10.041.3Monocrosporium10.041.3Monocrosporium10.041.3Monocrosporium<	White steril mycel	S. Long	0.55	6.0	- 00 - 00	1.30	24.3	
Cladorrhinum162.9720.9231.2116.2Trichoderma30.554.6190.8525.6Papulospora20.372.3180.8116.2Rhizopus50.746.9170.7613.5Actinomucor325.9423.2160.7212.1Myrothecium140.6312.1Penicillium61.114.6140.6313.5Stilbella71.3011.6120.549.4Helminthosporium40.749.390.4012.1Sordaria20.372.380.368.1Cladosporium30.556.940.189.4Dark steril mycel20.372.340.182.7Stachybotris10.041.3Beauveria10.041.3Basidiomycet10.041.3Epicoccum10.041.3Ulocladium30.556.9Trichothecium20.372.3Doratomyces10.041.3-Beauveria10.041.3Phoma10.182.3<	Chaetomium	lioz gal	0.00	0.9	20	1.20	27.02	
Distribution10 $2.87$ $20.9$ $23$ $1.03$ $17.5$ Trichoderma3 $0.55$ $4.6$ 19 $0.85$ $25.6$ Papulospora2 $0.37$ $2.3$ 18 $0.81$ $16.2$ Rhizopus5 $0.74$ $6.9$ $17$ $0.76$ $13.5$ Actinomucor $32$ $5.94$ $23.2$ $16$ $0.72$ $12.1$ Myrothecium $   14$ $0.63$ $12.1$ Penicillium6 $1.11$ $4.6$ $14$ $0.63$ $13.5$ Stilbella7 $1.30$ $11.6$ $12$ $0.54$ $9.4$ Helminthosporium4 $0.74$ $9.3$ $9$ $0.40$ $12.1$ Sordaria2 $0.37$ $2.3$ $8$ $0.36$ $8.1$ Cladosporium3 $0.55$ $6.9$ $4$ $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ $4$ $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ $2$ $0.09$ $2.7$ Stachybotris $   1$ $0.04$ $1.3$ Beauveria $   1$ $0.04$ $1.3$ Acremoniella $   1$ $0.04$ $1.3$ Monocrosporium $  1$ $0.04$ $1.3$ Dotatina $3$ $0.55$ $6.9$ $ -$ Trichothecium2 $0.37$ $2.3$ $ -$ <td>Cladorrhinum</td> <td>16</td> <td>9.07</td> <td>20.0</td> <td>41</td> <td>1.21</td> <td>16.2</td>	Cladorrhinum	16	9.07	20.0	41	1.21	16.2	
Papulospora       2       0.37       2.3       19       0.83       25.6         Papulospora       2       0.37       2.3       18       0.81       16.2         Rhizopus       5       0.74       6.9       17       0.76       13.5         Actinomucor       32       5.94       23.2       16       0.72       12.1         Myrothecium       —       —       14       0.63       12.1         Penicillium       6       1.11       4.6       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       4       0.74       9.3       9       0.40       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       2.7         Stachybotris       —       —       1       0.04       1.3	Trichoderma	3	0.55	20.9 A C	23	1.03	17.5	
Participital2 $0.31$ 2.318 $0.81$ $162$ Rhizopus5 $0.74$ $6.9$ $17$ $0.76$ $13.5$ Actinomucor $32$ $5.94$ $23.2$ $16$ $0.72$ $12.1$ Myrothecium $14$ $0.63$ $12.1$ Penicillium6 $1.11$ $4.6$ $14$ $0.63$ $13.5$ Stilbella7 $1.30$ $11.6$ $12$ $0.54$ $9.4$ Helminthosporium4 $0.74$ $9.3$ $9$ $0.40$ $12.1$ Sordaria2 $0.37$ $2.3$ $8$ $0.36$ $8.1$ Cladosporium3 $0.55$ $6.9$ $4$ $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ $4$ $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ $2$ $0.09$ $2.7$ Stachybotris $1$ $0.04$ $1.3$ Beauveria $1$ $0.04$ $1.3$ Volutina $1$ $0.04$ $1.3$ Epicoccum1 $0.04$ $1.3$ Ulocladium3 $0.55$ $6.9$ Trichothecium2 $0.37$ $2.3$ Migrospora1 $0.18$ $2.3$ Phoma1 $0.18$ $2.3$ Ulocladium3 $0.55$ $6.9$ Phoma </td <td>Papulospora</td> <td>2</td> <td>0.33</td> <td>4.0</td> <td>19</td> <td>0.85</td> <td>25.6</td>	Papulospora	2	0.33	4.0	19	0.85	25.6	
Antimopus       3       0.74       0.9       17       0.76       13.5         Actinomucor       32       5.94       23.2       16       0.72       12.1         Myrothecium       -       -       -       14       0.63       12.1         Penicillium       6       1.11       4.6       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       4       0.74       9.3       9       0.40       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       2.7         Stachybotris       -       -       1       0.04       1.3         Beauveria       -       -       1       0.04       1.3         Volutina       -       -       1       0.04       1.3         Basidiomycet	Rhizonus	5	0.57	2.3	18	0.81	16.2	
Action       32       5.94       23.2       16       0.72       12.1         Myrothecium       -       -       -       14       0.63       12.1         Penicillium       6       1.11       4.6       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       4       0.74       9.3       9       0.40       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       2.7         Stachybotris       -       -       1       0.04       1.3         Beauveria       -       -       1       0.04       1.3         Acremoniella       -       -       1       0.04       1.3         Basidiomycet       -       -       1       0.04       1.3         Monoccrosporium       -	Actinomucor	29	0.74	0.9	17 10	0.76	13.5	
Mytohlerium $    14$ $0.63$ $12.1$ Penicillium6 $1.11$ $4.6$ $14$ $0.63$ $13.5$ Stilbella7 $1.30$ $11.6$ $12$ $0.54$ $9.4$ Helminthosporium4 $0.74$ $9.3$ 9 $0.40$ $12.1$ Sordaria2 $0.37$ $2.3$ 8 $0.36$ $8.1$ Cladosporium3 $0.55$ $6.9$ 4 $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ 4 $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ 2 $0.09$ $5.4$ Stemphylum4 $0.74$ $6.9$ 2 $0.09$ $2.7$ Stachybotris1 $0.04$ $1.3$ Beauveria1 $0.04$ $1.3$ Volutina1 $0.04$ $1.3$ Basidiomycet1 $0.04$ $1.3$ Difference1 $0.04$ $1.3$ Ulocladium3 $0.55$ $6.9$ Trichothecium2 $0.37$ $2.3$ Nigrospera1 $0.18$ $2.3$ Phoma1 $0.18$ $2.3$ Cephalosporium1 $0.18$ $2.3$	Murothogium	34	5.94	23.2	16	0.72	12.1	
remember       6       1.11       4.6       14       0.63       13.5         Stilbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       4       0.74       9.3       9       0.40       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       2.7         Stachybotris       —       —       1       0.04       1.3         Beauveria       —       —       1       0.04       1.3         Volutina       —       —       1       0.04       1.3         Basidiomycet       —       —       1       0.04       1.3         Monocrosporium       —       —       1       0.04       1.3         Ulocladium       3       0.55       6.9       —       —         Trichothecium       2       0.37       2.3	Dopioillium	<u> </u>		_	14	0.63	12.1	
Stinbella       7       1.30       11.6       12       0.54       9.4         Helminthosporium       4       0.74       9.3       9       0.40       12.1         Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       2.7         Stachybotris       —       —       1       0.04       1.3         Beauveria       —       —       1       0.04       1.3         Volutina       —       —       1       0.04       1.3         Basidiomycet       —       —       1       0.04       1.3         Monocrosporium       —       —       1       0.04       1.3         Ulocladium       3       0.55 </td <td>Stilbollo</td> <td>0</td> <td>1.11</td> <td>4.6</td> <td>. 14</td> <td>0.63</td> <td>13.5</td>	Stilbollo	0	1.11	4.6	. 14	0.63	13.5	
Itermintnosportum4 $0.74$ $9.3$ 9 $0.40$ $12.1$ Sordaria2 $0.37$ $2.3$ 8 $0.36$ $8.1$ Cladosporium3 $0.55$ $6.9$ 4 $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ 4 $0.18$ $9.4$ Dark steril mycel2 $0.37$ $2.3$ 4 $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ 2 $0.09$ $5.4$ Stemphylum4 $0.74$ $6.9$ 2 $0.09$ $2.7$ Stachybotris———1 $0.04$ $1.3$ Beauveria———1 $0.04$ $1.3$ Acremoniella———1 $0.04$ $1.3$ Volutina———1 $0.04$ $1.3$ Epicoccum———1 $0.04$ $1.3$ Monocrosporium———1 $0.04$ $1.3$ Ulocladium3 $0.55$ $6.9$ ——Trichothecium2 $0.37$ $2.3$ ——Nigrospora1 $0.18$ $2.3$ ——Phoma1 $0.18$ $2.3$ ——Heterosporium1 $0.18$ $2.3$ ——Cephalosporium1 $0.18$ $2.3$ ——	Uolminth and ani	1	1.30	11.6	12	0.54	9.4	
Sordaria       2       0.37       2.3       8       0.36       8.1         Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       9.4         Doratomyces       1       0.18       2.3       2       0.09       5.4         Stemphylum       4       0.74       6.9       2       0.09       2.7         Stachybotris       —       —       —       1       0.04       1.3         Beauveria       —       —       —       1       0.04       1.3         Acremoniella       —       —       —       1       0.04       1.3         Volutina       —       —       —       1       0.04       1.3         Epicoccum       —       —       —       1       0.04       1.3         Monocrosporium       —       —       —       1       0.04       1.3         Ulocladium       3       0.55       6.9       —       —       —	Sondaria	4	0.74	9.3	dr. 69	0.40	12.1	
Cladosporium       3       0.55       6.9       4       0.18       9.4         Dark steril mycel       2       0.37       2.3       4       0.18       2.7         Doratomyces       1       0.18       2.3       2       0.09       5.4         Stemphylum       4       0.74       6.9       2       0.09       2.7         Stachybotris       —       —       1       0.04       1.3         Beauveria       —       —       1       0.04       1.3         Acremoniella       —       —       1       0.04       1.3         Volutina       —       —       1       0.04       1.3         Basidiomycet       —       —       1       0.04       1.3         Monocrosporium       —       —       1       0.04       1.3         Monocrosporium       —       —       1       0.04       1.3         Ulocladium       3       0.55       6.9       —       —       —         Trichothecium       2       0.37       2.3       —       —       —         Nigrospora       1       0.18       2.3       —       — <t< td=""><td>Sordaria</td><td>2</td><td>0.37</td><td>2.3</td><td>8</td><td>0.36</td><td>8.1</td></t<>	Sordaria	2	0.37	2.3	8	0.36	8.1	
Dark steril mycel2 $0.37$ $2.3$ 4 $0.18$ $2.7$ Doratomyces1 $0.18$ $2.3$ 2 $0.09$ $5.4$ Stemphylum4 $0.74$ $6.9$ 2 $0.09$ $2.7$ Stachybotris———1 $0.04$ $1.3$ Beauveria———1 $0.04$ $1.3$ Acremoniella———1 $0.04$ $1.3$ Volutina———1 $0.04$ $1.3$ Basidiomycet———1 $0.04$ $1.3$ Epicoccum———1 $0.04$ $1.3$ Monocrosporium———1 $0.04$ $1.3$ Ulocladium3 $0.55$ $6.9$ ——Trichothecium2 $0.37$ $2.3$ ——Nigrospora1 $0.18$ $2.3$ ——Phoma1 $0.18$ $2.3$ ——Heterosporium1 $0.18$ $2.3$ ——Cephalosporium1 $0.18$ $2.3$ ——	Cladosporium	1.319	0.55	6.9	4	0.18	9.4	
Doratomyces       1       0.18       2.3       2       0.09       5.4         Stemphylum       4       0.74       6.9       2       0.09       2.7         Stachybotris       -       -       1       0.04       1.3         Beauveria       -       -       1       0.04       1.3         Acremoniella       -       -       1       0.04       1.3         Volutina       -       -       1       0.04       1.3         Basidiomycet       -       -       1       0.04       1.3         Epicoccum       -       -       1       0.04       1.3         Monocrosporium       -       -       1       0.04       1.3         Monocrosporium       -       -       1       0.04       1.3         Ulocladium       3       0.55       6.9       -       -         Trichothecium       2       0.37       2.3       -       -         Nigrospera       1       0.18       2.3       -       -         Phoma       1       0.18       2.3       -       -       -         Verticillium       1       0.18	Dark steril mycel	2	0.37	2.3	4	0.18	2.7	
Stemphylum4 $0.74$ $6.9$ 2 $0.09$ $2.7$ Stachybotris1 $0.04$ $1.3$ Beauveria1 $0.04$ $1.3$ Acremoniella1 $0.04$ $1.3$ Volutina1 $0.04$ $1.3$ Basidiomycet1 $0.04$ $1.3$ Epicoccum1 $0.04$ $1.3$ Monocrosporium1 $0.04$ $1.3$ Chlamydomyces1 $0.04$ $1.3$ Ulocladium3 $0.55$ $6.9$ Trichothecium2 $0.37$ $2.3$ Nigrospora1 $0.18$ $2.3$ Phoma1 $0.18$ $2.3$ Heterosporium1 $0.18$ $2.3$ Cephalosporium1 $0.18$ $2.3$	Doratomyces	N, 1 <b>5</b> 73;	0.18	2.3	2	0.09	5.4	
Stachybotris $    1$ $0.04$ $1.3$ Beauveria $   1$ $0.04$ $1.3$ Acremoniella $   1$ $0.04$ $1.3$ Volutina $   1$ $0.04$ $1.3$ Basidiomycet $   1$ $0.04$ $1.3$ Epicoccum $   1$ $0.04$ $1.3$ Monocrosporium $   1$ $0.04$ $1.3$ Chlamydomyces $   1$ $0.04$ $1.3$ Ulocladium $3$ $0.55$ $6.9$ $  -$ Trichothecium $2$ $0.37$ $2.3$ $ -$ Nigrospera $1$ $0.18$ $2.3$ $ -$ Phoma $1$ $0.18$ $2.3$ $ -$ Heterosporium $1$ $0.18$ $2.3$ $ -$ Cephalosporium $1$ $0.18$ $2.3$ $ -$	Stemphylum	4	0.74	6.9	2	0.09	2.7	
Beauveria       —       —       —       1       0.04       1.3         Acremoniella       —       —       —       1       0.04       1.3         Volutina       —       —       —       1       0.04       1.3         Basidiomycet       —       —       —       1       0.04       1.3         Basidiomycet       —       —       —       1       0.04       1.3         Epicoccum       —       —       —       1       0.04       1.3         Monocrosporium       —       —       —       1       0.04       1.3         Monocrosporium       —       —       —       1       0.04       1.3         Chlamydomyces       —       —       —       1       0.04       1.3         Ulocladium       3       0.55       6.9       —       —       —         Trichothecium       2       0.37       2.3       —       —       —         Nigrospora       1       0.18       2.3       —       —       —         Phoma       1       0.18       2.3       —       —       —         Verticillium	Stachybotris	other g	adi	0137_180*	1 1	0.04	1.3	
Acremoniella       -       -       1 $0.04$ $1.3$ Volutina       -       -       1 $0.04$ $1.3$ Basidiomycet       -       -       1 $0.04$ $1.3$ Epicoccum       -       -       1 $0.04$ $1.3$ Monocrosporium       -       -       1 $0.04$ $1.3$ Monocrosporium       -       -       1 $0.04$ $1.3$ Chlamydomyces       -       -       1 $0.04$ $1.3$ Ulocladium       3 $0.55$ $6.9$ -       -         Trichothecium       2 $0.37$ $2.3$ -       -         Nigrospora       1 $0.18$ $2.3$ -       -         Phoma       1 $0.18$ $2.3$ -       -         Verticillium       1 $0.18$ $2.3$ -       -         Cephalosporium       1 $0.18$ $2.3$ -       -	Beauveria	arian	wed Paw	_	1	0.04	1.3	
Volutina       -       -       1 $0.04$ $1.3$ Basidiomycet       -       -       1 $0.04$ $1.3$ Epicoccum       -       -       1 $0.04$ $1.3$ Monocrosporium       -       -       1 $0.04$ $1.3$ Monocrosporium       -       -       1 $0.04$ $1.3$ Chlamydomyces       -       -       1 $0.04$ $1.3$ Ulocladium       3 $0.55$ $6.9$ -       -         Trichothecium       2 $0.37$ $2.3$ -       -         Nigrospera       1 $0.18$ $2.3$ -       -         Phoma       1 $0.18$ $2.3$ -       -         Verticillium       1 $0.18$ $2.3$ -       -         Heterosporium       1 $0.18$ $2.3$ -       -         Cephalosporium       1 $0.18$ $2.3$ -       -	Acremoniella	ences a	of <del>oc</del> ent	isonted	mei Lvere	0.04	1.3	
Basidiomycet       -       -       1       0.04       1.3         Epicoccum       -       -       1       0.04       1.3         Monocrosporium       -       -       1       0.04       1.3         Monocrosporium       -       -       1       0.04       1.3         Chlamydomyces       -       -       1       0.04       1.3         Ulocladium       3       0.55       6.9       -       -         Trichothecium       2       0.37       2.3       -       -         Nigrospera       1       0.18       2.3       -       -         Phoma       1       0.18       2.3       -       -         Verticillium       1       0.18       2.3       -       -         Heterosporium       1       0.18       2.3       -       -       -         Cephalosporium       1       0.18       2.3       -       -       -	Volutina	attribut	can be	ng these	om Al zelo	0.04	1.3	
Epicoccum       -       -       1       0.04       1.3         Monocrosporium       -       -       1       0.04       1.3         Chlamydomyces       -       -       1       0.04       1.3         Ulocladium       3       0.55       6.9       -       -       -         Trichothecium       2       0.37       2.3       -       -       -         Nigrospera       1       0.18       2.3       -       -       -         Phoma       1       0.18       2.3       -       -       -         Verticillium       1       0.18       2.3       -       -       -         Heterosporium       1       0.18       2.3       -       -       -         Cephalosporium       1       0.18       2.3       -       -       -	Basidiomycet	ns estiec	condition	amant v	vinor <b>1</b> mor	0.04	1.3	
Monocrosporium       —       —       —       1       0.04       1.3         Chlamydomyces       —       —       1       0.04       1.3         Ulocladium       3       0.55       6.9       —       —       —         Trichothecium       2       0.37       2.3       —       —       —         Nigrospora       1       0.18       2.3       —       —       —         Phoma       1       0.18       2.3       —       —       —         Verticillium       1       0.18       2.3       —       —       —         Heterosporium       1       0.18       2.3       —       —       —         Cephalosporium       1       0.18       2.3       —       —       —	Epicoccum	n <del>da</del> xe r	ture. For	batafoai	ich fyere	0.04	1.3	
Chlamydomyces	Monocrosporium	nage lo	mh.svin	dointer a	azoolaare	0.04	1.3	
Ulocladium       3       0.55       6.9	Chlamydomyces	nutia ad	t same	. Francis	0.01100	0.04	1.3	
Trichothecium       2       0.37       2.3            Nigrospora       1       0.18       2.3             Phoma       1       0.18       2.3             Verticillium       1       0.18       2.3            Heterosporium       1       0.18       2.3            Cephalosporium       1       0.18       2.3	Ulocladium	3	0.55	6.9	ne <del>pli</del> ne	nat <del>ma</del> nsion		
Nigrospera       1       0.18       2.3          Phoma       1       0.18       2.3           Verticillium       1       0.18       2.3	Trichothecium	2	0.37	2.3				
Phoma         1         0.18         2.3             Verticillium         1         0.18         2.3             Heterosporium         1         0.18         2.3	Nigrospora	1	0.18	2.3				
Verticillium         1         0.18         2.3            Heterosporium         1         0.18         2.3             Cephalosporium         1         0.18         2.3	Phoma	1	0.18	2.3		1		
Heterosporium         1         0.18         2.3	Verticillium	1	0.18	2.3	_	10 <u>1</u>		
Cephalosporium 1 0.18 2.3	Heterosporium	1	0.18	2.3	_			
	Cephalosporium	1	0.18	2.3				

Table. Isolated genera and their rates of occurence in total number of isolates and sampled in the greenhouses.

cies were more common in hot soils than cold soils. These two fungus were obtained commonly in the second survey which the soil temperature was higher  $(31,2 \degree C)$  than during the first survey.

Among these fungi Stilbella, Monocrosporium and Volutina are new genera and Acremoniella atra Sacc. and Beaveria alba are new species for Turkish mycoflora (Fig. 1,2,3).

## B — Pathogenicity tests :

In pathogenicity tests, genera of Fusarium, Rhizoctonia, Pythium, Sclerotium, Alternaria, Verticillium, Macrophomina, Cladorrhinum, Ac remoniella, Nigrospora, Stilbella, Melanospora, Sordaria and Actino muccor caused various degree of discoloration and decay of the roots and drying and death on above ground soil parts of the plants.

According to the results of the pathologenicity tests the genera of fungi showed wide ange of differences in their pathogenicities. The isolates of Sclerotium, Fusarium, Macrophomina, Stilbella, Cladorrhinum, Rhizoctonia, Nigrospora, Melano spora, Verticillium, Alternaria, Sor-Pythium, and Actinomucor daria, were shown % 52, 48, 48, 44, 40, 40, 36, 36, 32, 20, 16, 16 12 and 12 pathogenicity respectively. The isolates of Pythium, Alternaria, and Verticillium has shown slight pathogenicity. According to HENDRIX and CAMP-BELL (1973) not all of the isolates of Pythium species are pathogenc on plants. Verticillium and Alternaria species are more important on latter stage than seedling stage on carnations (GUBA, 1945; SCHMIDT, 1952; HELMERS, 1960; ROBINSON, 1961). There were no report about the pathogenicity of Nigrospora, Melanospora, Cladorrhinum, Acremoniella and Stilbella on carnation seedlings in literature. But they were found pathogenic in our tests. However Melanosporia and Nigrospora may caused lesions and decline on some plant roots (VIENNOT - BOURGIN, 1949; DICKSON, 1956; TARR, 1963). Also Cladorrhinum foecondissimum caus ed necrosis on pea roots in vitro conditions (DOMSCH und GAMS, 1970).

Although we used some antagonistic fungi according to the various report (WILSON, 1955; CATANI and PETERSON, 1963) generally the incidence of the disease increased when the tested pathogens were combined with saprophytic fungi (Fig.4). These results were shown that the saprophy tic fungi have not antagonistic effect on pathogenic genera in this study. It depends on the various causes. However, antagonism were developed easily on artificial medium between two organisms (GAUMANN, 1950) it can only take place under some favourable conditions in soil. Such as pH, soil temperature humidity, organic matter content, inoculum potential of the antagonistic organisms and persistence of their toxic subs tances (WAKSMAN 1952, DEVAY 1956). Also some saprophytic fungi may actually be pathogenic under certain conditions or combinations. On the other hand saprophytic fungi may help penetration of pathogen by increasing the sensitivity of the host plant through certain biochemical activities

C - Chemical control tests :

The results of the chemical tests were not satisfactory. The effectiveness of Methyl bromide, Orthocide soil treater, Banlate and Dexonal were 57,7 %, 43,3 %, 25,1 % and 21,2 % respectively.

Different effectiveness were recorded from the same chemicals used in our studies according to the genus of the fungi and the inoculum density of the same species (DROSIHN et al., 1968; TORGESON, 1969).

Thse findings were also confirmed by our results.

#### ÖZET

# İZMİR İLİ ÇİÇEK SERALARINDA KARANFİL FİDELERİNDE ÇÖKER-TEN HASTALIĞINA SEBEB OLAN FUNGAL ETMENLERİN SAPTAN-MASI, PATOJENİSİTELERİ VE İLAÇLI SAVAŞ YOLLARI ÜZERİNDE ARAŞTIRMALAR

Araştırma, 1972 ve 1974 yıllarıarasında İzmir ili çiçek seralarında karanfil fidelerinde çökerten hastalığına sebep olan fungal etmenleri, patojenisitelerini ve kimyasal savaş yollarını araştırmak amacıyla yapılmıştır. Çalışma sırasında 117 sera gezilmiş ve bunlardan toplam 2925 hastalıklı bitki örneği alınmıştır.

İzolasyon çalışmaları sonunda 41 fungus genusu izole edilmiştir. Bu genuslardan Stilbella, Volutina, Monocrosporium yurdumuz için yeni genuslar, Acremoniella atra ile Beauveria alba ise yeni türlerdir.

Patojenisite testlerine alınan Sclerotium, Macrophomina, Fusarium Acremoniella, Cladorrhinum, Still bella, Rhizoctonia, Nigrospora, Me-

lanospora, Verticillium, Alternaria, Pythium ve Actinomucor genusları survey sırasında izole edilen tüm genuslar arasından bir ön test sonucu seçilmişlerdir. Bu genuslar tek başlarına ve ayrı bir karekter olarakda Aspergillus, Myrothecium, Penicillium, Trichoderma ve Gliocladium gibi survey sırasında en çok izole edilen saprofit genuslarla birlikte patojenisite testlerine alınmışlardır. Patojenisite testlerine alınan tüm genuslar saprofit genuslarla birlikte verildiklerinde karanfil fidelerinde oluşan kuruma oranı artmıştır.

İlâçlı savaş denemelerine patojenisite testlerinde % 30 un üzerinde patojenisite gösteren Sclerotium, Rhizoctonia, Fusarium, Macrophomina, Cladorrhinum, Melanospora, Pythium Acremoniella, Stilbella ve Actinomucor genusları ile metil bromit, orthocide soil treater dexonal ve benlate preparatları alınmıştır. Deneme sonuçlarına göre ilaçlar karanfil fidelerinde çökerten hastalığına karşı yüksek bir etki göstermemislerdir.

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Fig. 3. The colony of Volutina sp. On PDA



(appr. X 115)



(appr. X 465)

Fig. 6. The severity of disease of the pathogen and their combinations with the saprophytes.

# Studies on Serology of Halo Blight (Pseudomonas phaseolicola) (Burkholder) (Dowson) of Beans\*

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#### INTRODUCTION

Bean production in Turkey shows a considerable increasing trend. In 1974 bean production was 230.000 tons pink calavance production was 60.000 tons and dried beans was 145.000 tons the yield being 145 kgs. per decar. (ANONYMUS 1974).

KARAHAN (1971) reports that **Pseudomonas phaseolicola** (Burkholder) (Dowson) exists in Turkey and the primary infections of the disease caused by infested seeds.

GUTRIE et al. (1965) declare that the causal organism of Halo Blight can easily be spread to long distances with seed, dust, rain, animals and equipment. They also add that one infected plant in 16.000 is

good enough to result in a complete crop loss under epiphytotic conditions. They also indicate that while the pathogen bacteria isolated from leaves, pods and seeds of the bean can be detected by classical bacteriological methods shorten this period. According to the same authors, rabbit or sheep can be used in producing antiserum and it can be obtained by injecting rabbit twice a week, totally 8 times, and four days after the last injection antiserum is obtained. Thus obtained antiserum can be used to detect the P. phaseolicola in infested seeds.

This study has been realized to enable the identification of Halo Blight by serological techniques.

<sup>•)</sup> This paper was summarized from first phytopathological congress report which organized by the Turkish Phytopathological Society on 20-24 October, 1975 in İzmir.

#### STUDIES ON SEROLOGY OF HALO BLIGHT

#### MATERIAL AND METHODS

One year old female new Zeland rabbits are used in the course of the study. **P. phaseolicola** culture with number 52 which has been obtained from NCPPB is used as antigen. Preparation of the antigen and injections were made according to the method of GUTRIE et al. (1965) **P. phaseolicola** culture was grown on NA (Nutrient Agar) for 36 hours. Then colonies of bacteria were taken, suspended in sterile saline and added formalin with concentration of 0.5 %. The concentration of antigen which is preparat was adjusted to the Mc. Farland tube no.3. Injections were interveinously made. In Table I the amount of the antigen and the type of the injections made are given.

Table I. Numbers of Injection and the Amount of Antigen Inject

Numbers of Injection	method	Amount	
lidw tests 1 solibai sele vad	Interveinous	0.25 ml.	
and hatelo 2 arretted materia	and and a set of the s	0.25 ml.	
and and 2 many here along	» ••••	0.50 ml.	
3	ed ner	1.0 ml.	
nainen sidt 4 stands short im	»	1.0 ml.	
5	>	1.0 ml.	
6	a to ild »	1.0 ml.	
7	»	1.0 ml.	
8	»	1.0 ml.	

Four days after the last injection rabbit was bled and the antibody

concentration was checked with tube agglutination method.

#### RESULTS

Until the seventh injection, the method was strictly followed both for the amount of the antigen injected and for the recommended days. However, during the seventh injection the rabbit was subject to a shock, therefore injection were postponed for some time. Later when rabbit recovered, injection started again. Four days after the eighth injection, antibody concentration of the blood taken from the ear of the rabbit, was determined (Table 2).

#### Y.E. ÖKTEM

Series	1/50	1/100	1/400	1/800	1/1600	1/3200	1/6400	Kontrol
Reaction	+	+	+	+	+	+	1.50.610	ing <del>na sana</del> Magyaraka

Table 2. Control of antibody contcentration with tube agglutination method

In the control of the antibody concentration by this method, results was found to be 1/3200. A very vague agglutination was found in 1/6400 which is evaluated as negative. There was nc change in control tubes. Finding the result obtained satisfactory the rabbit was sacrificed. Blood taken from arters were separated. The antiserum thus preparad was controled with slide agglutination method against different groups of bacteria such as Agrobacterium tumefaciens, A. radiobacter, E. coli and Pseudomonas sp. All of the above mentioned cultures gave negative reaction with antiserum. Stock antiserum is kept in deep-freeze.

#### DISKUSSION

The antiserum gave a strong reaction in the slide agglutination against **P. phaseolicola** with ratio 1/10 and 1/20. In the control process with different groups of bacteria the nega tive result achieved from the cultures apart from **P. phaseolicola** proves that the antiserum is specific thus the identification of the pathogen can be made in a short period. However, GUTRIE et al. (1965) recommend to use the gel diffusion test and other tests for the most satisfactory result with large number of seed samples. The antiserum has been preparad for this purpose and we do believe that it will give satisfactory result when it is started to be used.

#### ÖZET

## FASULYA YAPRAK YANIKLIĞI ETMENİ (Pseudomonas phaseolicola) (Burkholder) (Dowson)'NİN SEROLOJİSİ ÜZERİNDE ÇALIŞMALAR

Yurdumuzda taze ve kuru fasulye üretimi sürekli olarak artmaktadır. İklim koşullarının, bakteriyel hastalıkların gelişmesine uygun olduğu yıllarda üretim kayıpları meydana gelmektedir. Bu çalışma fasulyenin önemli hastalıklarından olan Fasulye Yaprak Yanıklığı etmeni (**Pseudomonas phaseolicola**)'nin se rolojik yöntemlerle saptanması amacıyla yapılmıştır.

Çalışmada bir yaşlı Yeni Zelanda

#### STUDIES ON SEROLOGY OF HALO BLIGHT

tavşanı, antijen olarakta NCPPB'dan temin edilen **Ps. phaseolicola** kültürü kullanılmıştır. Enjeksiyonlar damardan (Interveinous) haftada iki defa uygulanmıştır.

Birinci enjeksiyonda 0.25 ml., ikincide 0.5 ml. ve diğer altı enjeksiyonda 1'er ml. antijen enjekte edilmiştir. Sekizinci enjeksiyondan dört gün sonra kulaktan kan alınıp antibady konsantrasyonu tüp aglütinasyon metoduyla 1/3200 olarak saptanmıştır. Konsantrasyon yeterli görülmüş ve tavşan kesilmiştir. Arterlerden alınan gerekli seperasyonu yapılmıştır. Elde edilen antiserum deep-pfreezde saklanmıştır. Antiserumun bir kısmı 1/10 ve 1/20 oranında serum fizyolojikle seyreltildikten sonra lam aglütinasyon metoduyla **P. phaselicola** kültürü kontrol edilmiş ve kuvvetli bir reaksiyon vermiştir. Buna karşılık **A. tumefaciens, A. radiobacter, E. coli ve pseudomonas** sp. gibi değişik gruplara ait bakterilerde aynı me todla uygulama yapılmış ve kültürlerin hiç birinde aglütinasyon görülmemiştir.

Alınan sonuçlar hazırlanmış olan antiserumun **P. phaseolicola** için spesifik olduğunu göstermektedir. Patojenin teşhisi kolaylıkla kısa süre içerisinde yapılabilmektedir.

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# Elektronenmikroskopische Untersuchungen über die Stolburkrankheit an Tomaten

#### Von Ülkü YORGANCI\*

#### ZUSAMMENFASSUNG

In dieser Arbeit wurden die stolburkranken Tomatenpflanzen elektronen mikroskopisch untersucht. Diese Kranheit wurde in der Türkei vorher symptomatologisch beobachtet und berichtet.

Die Blütenknospen von Tomaten mit typischen Stolbursymptomen wurden mit Glutaraldehyd und OSO<sub>4</sub> fixiert, in Epon eingebettet. Die hergestellten ultradünnen Schnitte wurden mit Uranylacetat und Bleicitrat nachkontrastiert, mit dem Elektronenmikroskop aufgenommen. Im Phloem besonders in den Siebröhren von kranken Tomatenpflanzen wurden pleomorphe Gebilde beobachtet. Die Dimensionen dieser Gebilde varierten zwischen etwa 70-630 nm. Die kleineren Partikel enthalten dichtes, granuliertes Plasma. Dagegen haben die Grösseren ribosomenähnliche Strukturen im Plasma. Lerschiedene Formen kommen nebeneinander in einer Siebröhre vor.

Das Auftreten von Ackerwinde (Convolvulus arvensis) in grossen turen im Plasma. Verschiedene Formen kommen nebeneinander in einer fallend.

Die Mycoplasmen sind als Erreger von manchen Pflanzenkrankheiten erst im Jahre 1967 entdeckt worden, und diese Entdeckung ist wohl eines der wichtigsten Ereignisse der letzten Jahre. Als Krankheitserreger bei Tieren und Menschen sind die Mycoplasmen schon sehr lange bekannt. Obwohl sie aber überall zu finden sind, (d.h. dass sie ubiquiter sind) wurden die Veröffentlichungen von Doi et al. (1967), die Mycoplas-

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men können auch Krankheiten an Pflanzen verursachen, mit grosser Überraschung aufgenommen. Diese Autoren stellten die Mycoplasmen im Phloem von kranken Pflanzen elektronenmikroskopisch fest. Später folg ten weitere Arbeiten über andere Pflanzenkrankheiten, bei denen die Mycoplasmen als Erreger eine Rolle spielen.

An Tomaten gibt es eine Krankheit die als Stolbur oder big bud bekannt ist und im späteren Stadium mit grossen, unfruchtbaren Blütenknospen ins Auge springt. Als Ursache dieser Krankheit wurde bis vor einigen Jahren ein Stamm der Falschblütigkeit von Vaccinium (Vaccinium false blossom virus) angenommen.

Aber in den letzten Jahren berichteten verschiedene Autoren da rüber, die Erreger der Stolburkrankheit seien mycoplasmaähnliche Gebilde, (Gianotti et al., 1968; Bor - ges V. de Lourdes, 1969; Bowyer et al., 1969; Marchoux et al., 1969; Panjan et al., 1969; Granett et Provvidenti, 1974), was wurde das von diesen Autoren elektronenmikroskopisch festgestellt.

In der Türkei wurde über das Vorhandensein dieser Krankheit an Tomaten von Tanrıkut (1953) und Tekinel (1973) und an Kartoffeln von Sahtiyancı (1966) symptomatologisch berichtet.

Für die Feststellung der Stol burkrankheit wurden die Tomatenpflanzen im August kontrolliert. Die Symptome stimmten mit denen, die von Klinkowski et Uschdraweit (1968) gegeben wurden, völlig überein. Die Blätter von infizierten Pflanzen waren löffelförmig verbeult. Ihre Nerven und der Rand der Blattunterseiten waren rötlich. Die Blütenknospen standen aufrecht. Die Kelchsegmente



Abb 1. Die Symptome an den Blütenknospen der mit Stolbur infizierten Tomatenpflanze

waren ganz oder teilweise verwachsen der Kelch war blasenförmig erweitert und viel grösser als normal. Dagegen waren Petalen und die Stamina sehr klein und teilweise grün. Das Gynaceum war häufig vergrössert, vor allem war das Gynophor ausserordentlich verlängert.

In dieser Arbeit wurden im August kleine Stücke von den Basalregionen der Blütenknospen und Kelch blätter ausgeschnitten. Die Fixie rung erfolgte in Glutaraldehyd und die Nachfixierung in 1 % igem OSO<sub>4</sub>. Die Gewebestücke wurden nach Luft in Epon eingebettet (Köktürk, 1967) Die ultradünnen Schnitte wurden mit dem Reichertultramikrotom OmU, hergestellt, mit Uranylacetat und Bleicitrat (Reynolds, 1963) nachkontrastiert und mit dem Elektronen mikroskop der Fa. Zeiss 9a aufgenommen.

Besonders in Schnitten von den Basalregionen der vergrösserten Blüten enthielten einige Zellen im Phloem vorwiegend in den Siebröhren, pleomorphe Gebilde. Die Dimensionen dieser Gebilde varierten zwischen etwa 70-630 nm. Die kleineren



Abb 2. Pleomorph mycoplasmaähnliche Gebilde in den Siebröhren von Tomatepflanzen nach Infektion mit Stolburkrankheit

#### STOLBURKRANKHEIT AN TOMATEN

Partikel enthalten dichtes, granuliertes Plasma. Dagegen haben die grösseren ribosomenähnliche Strukturern im Plasma. Verschiedene Formen kommen nebeneinander in einer Siebröhre vor (Abb. 2).

Sinha et Paliwal beschrieben die kleineren, dichten Gebilde als Elemantarkörpechen, die Grösseren, mit fädigen Inneren als reife Zellen.

Das Auftreten von Ackerwinde (Convolvulus arvensis) in grossen Massen in Tomatenfeldern mit typi schen Stolbursymptomen ist sehr auffallend. Ackerwinde ist in der Literatur als Wirtzpflanze und Krankheitsreservoir von Stolbur angegeben Ausserdem war Cuscuta campestris, die auch als Vektor bekannt ist, auf einem Tomatenfeld in Muradiye sehr verbreitet.

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#### ÖZET

## DOMATESLERDE STOLBUR HASTALIĞI ÜZERİNDE ELEKTRONMİKROSKOBİK İNCELEMELER

Bu çalışmada daha önce Türkiye'de simptomatolojik olarak gözlenmiş olan Stolbur hastalığı elektronmikroskobik olarak incelenmiştir.

Tipik Stolbur belirtisi gösteren domates çiçek tomurcukları gluta aldehit ve Osmium tetroksit'le fikse edildikten sonra Epon'a gömülmüştür. Hazırlanan ince kesitler uranylasetat ve kurşun sitratla boyandıktan sonra elektronmikroskobunda incelenmişlerdir. Stolburla infekteli domates bitkilerinin kalbur borularında çok sayıda, büyüklükleri 70 - 630 nm arasında değişen mikroplazma benzeri pleomorf cisimcikler gözlenmiştir. Küçük partiküller koyu, granüle plazma içermektedir. Daha büyük olanlarda, plazmada ribozom benzeri yapılar bulunur. Çeşitli şekillere bir kalbur borusunda yanyana rastlanabilir.

Stolburla bulaşık tarlalarda çok miktarda literatürde konukçu ve hastalık kaynağı olarak bildirilen **Convolvulus arvensis** (tarla sarmaşığı) bulunması ilginctir.

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# All Correspondance Should Be Made To FİTOPATOLOJİ DERNEĞİ Ege Üniversitesi Ziraat Fakültesi Eiteneteleji ya Zirai Botanik Kürsüsü

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