

1 VC 4. / 14

STUDIES ON THE HOST-PARASITE RELATIONSHIPS OF  
BOTRYTIS ALLII AND THE GENUS ALLIUM

By

SHAABAN ALY AHMED KHALED

B. Sc. (Cairo University) 1966

M. Sc. (Tanta University) 1978

632.4  
S. A

THESIS

21286

Submitted in Partial Fulfilment of  
the Requirement for the Degree

of

DOCTOR OF PHILOSOPHY

IN

PLANT PATHOLOGY

Department of Agric. Bot. and Plant Pathology

Faculty of Agriculture

Ain Shams University

Cairo, Egypt

1986

<

APPROVAL SHEET

Thesis for Ph. D. Degree entitled " Studies on the host-parasite relationships of Botrytis allii and the genus Allium."

Approved by:

Prof. Dr. ..W...A...Ashour.....

Prof. Dr. ....M. El-Khalil.....

Prof. Dr. ....M. M. Elzayyat.....

(Committee in charge)

Date 29 / 4 / 1986.



### ACKNOWLEDGEMENT

The writer wishes to express his sincere appreciation and gratitude to Professor Dr. M. M. El-Zayat, Prof. of Plant Pathology Division, and to Dr. M. H. Mostafa, Associate Prof. of Plant Path. Agric. Bot. and Plant Pathology Dept. Faculty of Agric., Ain-Shams Univ. for their supervision and helpfull guidance throughout the investigation and writing this thesis.

Thanks are also due to Prof. Dr. A. A. Ali, Head of Onion and Oil Crops Disease Dept., Dr. T. H. Abd-El-Moity and Nagwa, A. Osman, Plant Path. Inst., Agric. Res. Center for their generous help. Thanks are also offered to Dr. M. A. Baraka, Lecturer of Plant Path., Agric. Bot., Fac. of Agric., Suez-Canal Univ. The writer is grateful also to all staff members of Agric. Bot. and Plant Pathology Dept. Fac. of Agric., Ain-Shams Univ. and Onion and Oil Crops Diseases, Plant Path. Inst., Agric. Res. Center.

## CONTENTS

	Page
INTRODUCTION.....	1
REVIEW OF LITERATURE.....	2
MATERIALS AND METHODS.....	20
EXPERIMENTAL RESULTS.....	37
I. Isolation and identification of the causal fungus	37
II. Pathogenicity of <u>Botrytis allii</u> isolates.....	38
III. Fungi associated with onion seeds.....	41
IV. Physiological studies:.....	43
a. Variability in linear growth within <u>B. allii</u> isolates.....	43
b. Effect of carbon sources on fungal growth.....	46
c. Viability of conidia and sclerotia in the soil	48
d. <u>In vitro</u> effect of green leaves and bulbs juices on linear growth of <u>B. allii</u> .....	50
V. Reaction of different <u>Allium</u> species and cultivars to neck rot disease incidence.....	53
VI. Sensitivity of <u>Allium</u> leaves to spotting or blight- ing caused by <u>B. allii</u> .....	58
VII. Chemical changes occurred in necks and leaves of different <u>Allium</u> spp. infected with <u>B. allii</u> .....	62
1. Soluble carbohydrate and total amino acids..	62
a. Chemical changes occurred in the necks.....	62
b. Chemical changes occurred in the leaves....	64

	Page
2. Soluble phenolic compounds.....	66
a. Chemical changes in phenol contents in the necks.....	66
b. Chemical changes occurred in the leaves...	68
VIII. Pectinolytic and cellulolytic enzymes.....	70
a. In Vivo: Variability within <u>B. allii</u> isolates in enzyme activities.....	70
b. Enzyme activities of healthy and diseased onion and garlic bulbs.....	72
IX. Dissemination of <u>B. allii</u> propagules through irrigation water and air.....	74
a. Sclerotial dissemination.....	74
b. Conidial dissemination.....	76
X. Histopathological studies.....	78
DISCUSSION.....	81
SUMMARY.....	96
REFERENCES.....	102
ARABIC SUMMARY.....	-

## INTRODUCTION

Onion (Allium cepa L.,) is one of the main crops in A. R. E. Egyptian onion is greatly desired in the foreign markets for its good quality and early appearance. Hence it is exported to different foreign countries (Anonymous 1983). Onion is liable to be attacked by different species of Botrytis namely, B. allii Munn., B. squamosa Walker, B. byssoides J. C. Walker, and B. cinerea Pers. ex. Fr. causing seedling death, leaf spotting, leaf blight (die back) and rotting of neck and bulbs during storage (Walker 1925, Hancock and Lorbeer 1963 and Croxall 1946). Neck rot disease, incited by B. allii spreads widely on onions grown for seed production and also in onion bulbs during storage, causing great losses (Croxall 1946 and Netzer and Dishon 1966).

The present work was carried out to study the host parasite relationships between the fungus B. allii and the genus Allium. including Allium cepa (onion), Allium sativum (garlic) and Allium-korrat (korrat) Also this investigation aimed to study the persistence of sclerotia and conidia in the soil, the behaviour of the fungus in the host, the enzymatic activities and other physiological studies. Chemical analysis was carried out on leaves and necks of the investigated Allium varieties that were showing neck rot disease and leaf spotting or blighting . The disease dissemination was also studied.

# REVIEW OF LITERATURE



## REVIEW OF LITERATURE

### I. Causal organism of neck rot:

Dana (1923) stated that neck rot of onion was incited by Botrytis allii Munn. in Washington, Heald and Dana (1924) reported that neck rot of onion caused by B. allii occurred in both growing onions and during storage. Walker (1925), distinguished three species of Botrytis, each of which may incite a neck rot of onion bulbs, the respective diseases differ somewhat in symptoms. They were given distinctive names as follows: gray mould neck rot (B. allii Munn); mycelial neck rot (B. byssoides J. C. Walker); and small sclerotial neck rot (B. squamosa J. C. Walker).

Siemaszko (1929), recorded that B. allii caused the gray mould disease of onion in Poland.

Bremer (1935), noted that B. allii and Sclerotium cepivorum, caused storage injuries to onions in Germany.

Croxall (1946), stated that most of the losses of stored onion bulbs were due to B. allii, and the major part of the remainder was due to B. cinerea.

Kamel (1952), reported that B. allii is the causal organism of neck rot disease of onion bulbs in Egypt.

Hancock and Lorbeer (1963), showed that B. cinerea conidia may occur frequently on necrotic onion leaves in high or heavy blight infestation, while B. squamosa occurs only in fields with severe blight and B. allii was encountered infrequently. Isolates of B. squamosa and B. cinerea caused more extensive leaf spotting in the green-house than B. allii.

Kennelly (1963), isolated B. allii, S. cepivorum and Aspergillus niger from rotted garlic cloves in Dunedin (Newzealand). B. allii neck rot was found on garlic bulbs from Taiwan among the quarantine interception in Australia (Anonymous, 1972).

Leguizamon and Barriga (1976), mentioned that in Colombia, bulb rots of garlic in the field were generally caused by Penicillium corymbiferum, B. allii and Fusarium oxysporum.

Georgy (1977), demonstrated that B. allii infected onion and garlic varieties as well as korrat in Egypt. Radwan (1980), stated that B. allii was one of the most predominant fungi isolated from garlic cloves, also it constituted the highest percentage among the isolated fungi.

## II. Fungi associated with onion seeds:

Neergaard (1945), reported that spores of Alternaria porri f. sp. porri were detected from the seeds of onion and leek in Denmark.

Anderson (1974), mentioned that spores of Botrytis allii were commonly associated with onion and chives seeds.

Maude and Presly (1977), indicated that infected onion seeds were a source of the pathogen causing neck rot disease.

Hassan and Osman (1985), isolated Alternaria sp., Aspergillus sp., Botrytis sp., Chaetomium sp., F. oxysporum, Penicillium sp., and Verticillium sp. from onion seeds in Egypt.

## III. Physiological studies:

### A). Fungal growth on PDA medium:

Baraka (1978), recorded that B. allii grew well on both onion bulb extracts, potato dextrose and nutrient broth dextrose, while the least growth was on Richard and Czapek's liquid media. Isolates of the fungus differed in mycelial growth and dry weight.

Khaled (1978), stated that the best rate of growth and sporulation of the tested isolates of B. allii were obtained on natural media specially that contained onion bulb extract. He added that the tested synthetic media gave the lowest rate of growth and sporulation.

B). Effect of carbon sources on mycelial dry weight:

El-Refaeli (1973), stated that the highest amount of growth for Pyricularia oryzae was on medium with glucose, starch or dextrin, where the lowest was on lactose.

Baraka (1978), recorded that B. allii grew well on both onion bulbs extracts, P. D. and N. P. D., while the least growth was on Richard's and Czapek's liquid media. Isolates of the fungus differed in mycelial dry weight.

C). Survival of conidia and sclerotia of B. allii and related species:

Viennot-Bourgin (1952), demonstrated that after exposure of B. allii cultures for 48 hours to temperatures ranging from 0 C to 10 C, the fungus remained viable. He added that sporulation was ceased at temperatures ranging from 4 C to 10 C, while at 8 C to 10 C the organism completely lost its parasitic ability.

Maude and Presly (1977), found that naturally formed sclerotia of B. allii decayed within 21 weeks in field soil in the absence of host debris. It was suggested that a large proportion of sclerotia of Botrytis spp. would survive one winter in the field, and a small proportion might survive a second winter.

Ellerbrock and Lorbeer (1977 a), indicated that conidia of B. squamosa produced during disease epidemics were found to disappear from soil 1-2 weeks after crop harvest.

D). Effect of onion and garlic extracts on fungal growth:

Walker et al., (1925), detected two types of toxins present in onion scales and affecting various bulb parasites including B. allii, Fusarium cepae and Aspergillus niger. one of the two toxins was neither removed nor readily inactivated by heat, while the other one relatively disappeared from the extracted juice at room temperature within few hours.

Nikolaeva (1959), studied the effect of phytoncides from clematis, onion and garlic on the microflora of Esparcette beans. An aqueous infusion of dry onion scales was effective in controlling white rot (Sclerotinia

sclerotiorum ) and to some extent grey rot (B. cinerea) of vegetables in storage.

Mozes and Pall (1961), reported that extracts from a number of higher plants, including onion, garlic, horse chestnut and potato were phytoncidal to specific fungi.

Russel and Mussa (1977), found that garlic extract, prepared as a juice crushed from cloves inhibited growth of F. oxysporum.

Radwan (1980), recorded that Balady, Chinese and American garlic juice inhibited the growth of F. moniliforme isolated from maize, Helminthosporium oryzae from rice, Alternaria citri from cotton, S. bataticola from peanuts and A. niger from onion. In addition he found also that the juice of the garlic cultivars tested, suppressed the formation of sclerotia of Macrophomina phaseoli around paper discs.

El-Shamy, (Mona) et al., (1984), indicated that extract of Chinese garlic diluted with sterile water to 100, 75, 50 and 25% inhibited germination of F. oxysporum f. sp. nivium spores upon PDA medium from 44% to zero for discs dipped in the full strength extract and sterile water, respectively. The inhibition zones decreased by decreasing

the concentration of extract, when these extracts were added to PDA medium at the rate of 1 ml. per 10 ml of medium.

#### IV. Varietal resistance:

Walker and Lindegren (1924), Walker (1926) and Walker et al., (1944) reported that the coloured cultivars of onion were less susceptible to B. allii, B. byssoidea and B. squamosa, than the white ones. It was suggested that the resistance of the cultivars was due to the presence of water soluble toxins in the cells of the outer scales.

Van Beekon (1940), found that the Dutch coloured onion cultivars were resistant to neck rot disease caused by Botrytis spp.

Georgy (1977), stated that high percentage of infection with white rot disease was observed on different onion varieties when compared with both korrat and garlic specially Chinese garlic.

Baraka (1978), found that twelve onion cultivars were susceptible to infection with B. allii. The degree of disease reaction ranged from 75 to 100 %. Hybrids golden and tropicana showed the least percentage of infection.