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Incidence of entomopathogenic fungi of the oat bird-cherry aphid, *Rhopalosiphum padi* L. (Homoptera: Aphididae) infesting wheat plants in Assiut, Egypt

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Abstract: Four genera of entomopathogenic fungi and two hyphomycetes were identified from the cadavers of the aphid infesting wheat plants in the two growing seasons of 2013-2014. Entomophthorales was represented by four species belonging to three families; Ancylistaceae represented by *Conidiobolus*, Entomophthoraceae by *Entomophthora planchoniana* and *Pandora neoaphidis* and Neozygitaceae by *Neozygites fresenii*. The hyphomycetes fungi were represented by two species *Beauveria bassiana* and *Verticillium lecanii* which belong to the family Moniliaceae, order Moniliales. Data show that the aphid began to infest wheat plants early during the middle of January when wheat plants were in the stem-elongation stage. Thereafter, numbers of aphids increased gradually to reach a peak, when the plants were at the flowering stage during the third week of February; during the next three weeks the number of the oat aphid declined sharply. Mortality rate with the fungal pathogens was observed from the end of January up to the end of March. The number of cadavers increased gradually to reach the maximum level during the end of March.

Key words: Oat bird-cherry aphid, *Rhopalosiphum padi* L, wheat, entomopathogens, Assiut.

Introduction

The oat bird-cherry aphid, *R. padi* is considered one of the most serious cereal aphids attacking wheat plants in Assiut, Egypt. Its damage to the plants is manifested through loss of sap by sucking, reaction of plant tissues stimulated by aphid saliva; in different ways (change of color, curling of leaves and stem… etc.), excreting liquid, viscous honeydew, that is very harmful and on it sooty-moulds usually develop and, finally aphid transmission of virus diseases to plants (El-Hariry 1979, Abdel-Rahman 1997, El-Lathy 1999, and El-Fatih 2000 and 2006).

Entomopathogenic fungi are now being considered as biological alternatives to chemical control (Roberts and Yendol 1971, Samson *et al*. 1988). Entomopathogenic fungi are frequently reported as major factors suppressing populations of cereal aphids and can cause sudden decline of dense populations (Feng *et al*. 1991). Drastic reduction in the populations of various cereal aphids due to infection with Entomopathorales fungi was observed by Dedryver (1983).

The aim of the present study was to seek further information on mycopathogens of the oat bird-cherry aphid, *R. padi* L. infesting wheat plants in Assiut, Egypt.

Materials and Methods

The present investigation was carried out in Assiut Governorate (Abnoub district) about 15 Km Northeast Assiut City during 2013 and 2014 wheat-growing seasons. An area of about 2100 m^2 (about half of feddan) was cultivated with wheat (cultivar Sids 1) normally at mid-November in both cultivated seasons. The normal conventional agricultural practices were normally performed and no chemical control (insecticides or fungicides) was used during the study period. Weeds were removed by hand.

During the two seasons, the oat bird-cherry aphid numbers (all forms) were counted and recorded on 200 randomly selected seedlings or main tillers, later in the season, when the aphid numbers increase on the plants, the numbers of examined tillers were reduced to 100 tillers. Four replicates each were taken weekly from the beginning of January, when the migration of aphids onto wheat crop, at the stage of tillering or early stem-elongation stage from

overwintering sites began, and continued through the time when aphid population declined to low or undetectable levels. At the same time number of infected aphids was also counted and recorded.

Cadavers (dead aphids) were recorded, placed in 1x5 cm vials and stored at 5ºC. Aphid cadavers were examined under a dissecting microscope as soon as possible after collected to observe external symptoms and fungal reproductive structures if produced *in situ* on the plant. Desiccated and fresh cadavers were placed in a moist chamber for about 20 hrs to allow hyphae and reproductive structures to develop. Individual aphids were mounted in cotton blue or aceto-orcein and observed under a compound microscope. Identification of fungi was based on external symptoms and the morphology of spores and sporulating structures (Waterhouse and Brady, 1982), and new revision of the classification of Entomophthorales (Humber 1989 and 1991) was followed. Fungi identified as known aphid pathogens were considered to be the cause of death of their host.

Statistical analysis

Data were statistically analyzed using analysis of variance (F test) and means were compared according to Duncan's multiple range test.

Percentage of infection (mortality %) caused by entomopathogenic fungi was calculated in each sampling date according to Feng *et al*. (1992) as follows:

$$\text{Mortality \%} = \frac{\text{Number of infected aphids}}{(\text{Total number of alive aphids}) + \text{No. of cadavers}} \times 100$$

Results and Discussion

1-Entomopahogens identified from the aphid

From the survey studies through 2013 and 2014 wheat growing seasons, five species and one genus of entomopathogens, including four entomophthorales and two hyphomycetes were identified from the oat bird-cherry aphid, *R. padi*, infesting wheat plants. Entomophthorales was represented by four species belonging to three families: Ancylistaceae was represented by one genus, *Conidiobolus*, Order: Entomophthoraceae by *Entomophthora planchoniana* and *Pandora neoaphidis*, and Neozygitaceae by *Neozygites fresenii*. The hyphomycetes fungi were represented by *Beauveria bassiana* and *Verticillium lecanii* from order Moniliales.

These fungi mentioned above are surveyed worldwide as they are well-known species as biological control agents of cereal aphids (Feng *et al*. 1990, 1991 and 1992, Abdel-Rahman 2001 and Hammam 2003, 2009, Moubasher *et al*. 2010). Mycopathogens are considered to be the best means for biological control of aphids (Latge and Papierok, 1988), and numerous accounts of cereal aphids killed by entomophthoralean fungi were documented in Europe (Dean and Wilding 1971, 1973, Dedryver 1983, Papierok and Havukkala 1986) and South America (Lazzari 1985). Regional lists of aphid pathogenic fungi have been published in Australia (Milner *et al*. 1980) and Finland (Papierok 1989). Five entomopathogenic fungi were reported from 34 aphid hosts in eastern Canada and the United States (Remaudiere *et al*. 1978, Humber and Soper 1986).

2-Incidence of entomopathogens recorded

Data in Table 1 show the relative incidence of six entomopathogens which infect the oat aphid, namely *Conidiobolus* sp., *E. planchoniana*, *P. neoaphids*, *N. fresenii*, *B. bassiana* and *V. lecanii* during 2013 and 2014 wheat growing seasons.

In 2013 season, 147 cadavers were collected from *R. padi* naturally-infected with entomopathogens. The six species were identified.

Statistical analysis showed that *E. planchoniana* followed by *P. neoaphidis* were the most dominant species encountered in 38.26%, and 33.20% of samples respectively. Three species: *N. fresenii*, *B. bassiana* and *V. lecanii* showed a moderate level of infection, while *Conidiobolus* sp. was scarce.

In 2014 season, 659 cadavers of the oat aphids were collected during the whole season. The six species were also collected with the same sequence of dominance in 2013 season.

3- Entomopathogens and their host, *R. padi*

Data in table 2 show that in 2013 season, the aphid began to appear on wheat plants (0.02 individual / tiller) during the first week of January. Its population reached a peak of 4.10 aphids / tiller during the first week of March, then sharply declined to reach 0.80 aphids / tiller during the end of March. The cadavers appeared on wheat plants during the period extended from the beginning of February up to the end of March. The percentage of infection was relatively low, generally <6% during the first of March. Then the level of infection dramatically increased as the aphid population increased. Maximum infection (26.52%) was recorded during the second week of March correlated with 0.31 aphid / tiller.

In 2014 season, the aphid first appeared on wheat plants during the first week of January up

to the end of March with a peak of abundance (6.70 individuals / tiller) during the first half of March. The cadavers were detected after three weeks of aphids observed at the end of January (0.01 cadaver / tiller). Percentage of mycosis increased from 0.79% correlated with 1.25 aphids / tiller during the first week of February to 3.43% correlated with 1.04 aphids / tiller during the end of March. Maximum infection (41.51%) was markedly higher in the season of 2013 and was recorded during the middle of March correlated with 3.10 aphids / tiller.

In general, the mean data of the two seasons showed that the first case of infection by the entomopathogens was observed at the beginning of February with a percent mortality of 0.43% up to the end of March with a percent mortality of 8.45%. Maximum mortality (38.52%) was detected on March 17th.

Several species of entomopathogenic are known to cause fatal diseases in aphids, including *Conidiobolus* sp. *V. lecanii*, various species of *Beauveria*, *P. farinosus* (Holm ex S.F. Gery) Brown & Smith (Roberts & Yendol 1971, Samson *et al*. 1988). Entomopathogenic fungi are frequently reported as major factors suppressing populations of cereal aphids and can cause sudden decline of dense populations (Feng *et al*. 1991). Three entomopathoralean fungi species killed 65-80% of common cereal aphids in in eastern England (Dean and Wilding 1973). Drastic reduction in the populations of various cereal aphids due to infection with Entomopathorales fungi was observed by Dedryver (1983).

Members of order Entomophthorales are excellent candidates for biological control of aphids (Latge and Papierok 1988). Worldwide, *P. neoaphidis* is the most common and frequently the dominant-pathogen of aphids (Waterhouse and Brady 1982). This fungus can cause collapse of unmanaged aphid population within few weeks of the onset of disease (Feng *et al*. 1990). Without management however, the fungus is not effective as control agent alone because it does not attack until the aphid population has peaked and has already caused considerable damage (Feng *et al*. 1991).

Table 1: Numbers and percentages of entomopathogens naturally infecting the oat bird-cherry aphid in the field during 2013 and 2014 wheat-growing seasons.

Fungi species	2013		2014		Total	
	No.	(%)	No.	(%)	No	(%)
Conidiobolus sp.	2c	1.54	3c	0.45	5c	0.62
Entomophthora planchoniana Cornu	56a	38.26	291a	44.16	347a	43.05
Pandora neoaphidis (Remaudierel & Hennebert) Humber	49a	33.20	230a	34.90	279a	34.62
Neozygites fresenii (Nowakowski) Batko	14b	9.53	50b	7.59	64b	7.94
Beauveria bassiana (Balsamo) Vuill.	11b	7.35	34b	5.16	45b	5.58
Verticilium lecanii (Zimmermann) A.W. Viegas	15b	10.12	51b	7.74	66b	8.19
Total	147	100	659	100	806	100

Means vertically followed by the same letter are not significantly different < 0.05 level of probability.

Table 2: Average number of the oat bird-cherry aphid on wheat plants and natural infection rate with entomopathogens in the field during 2013 and 2014 wheat growing seasons.

Inspection date	Growth stage (ZGS)*	2013 season			2014 season		
		No / tiller		Infection (%)	No / tiller		Infection (%)
		Aphids	Cadavers		Aphids	Cadavers	
Jan. 6	28	0.02	0	0	0.02	0	0
13	30	0.07	0	0	0.18	0	0
20	32	0.21	0	0	0.34	0	0
27	33	1.05	0	0	1.25	0.01	0.79
Feb. 3	37	1.62	0.01	0.61	3.19	0.04	1.24
10	39	2.31	0.04	1.60	4.31	0.07	1.59
17	43	3.40	0.09	2.57	4.97	0.10	1.97
24	57	3.87	0.13	3.05	4.99	0.24	4.59
March 3	65	4.10	0.22	5.09	4.13	1.30	23.94
10	71	1.64	0.46	21.90	6.70	2.00	22.99
17	73	0.97	0.35	26.52	3.10	2.20	41.51
24	75	0.31	0.11	26.19	1.51	0.52	25.62
31	77	0.80	0.06	6.98	1.04	0.11	9.57
Total	----	20.37	1.47	6.73	35.73	6.59	15.57

*(ZGS) = A decimal code for growth stage of cereal (Zadoks *et al*. 1974)

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