ASSESSING THE IMPACT OF NATURAL PATHOGENS ON SUGARBEET ROOT MAGGOT THROUGH SURVEYS

Ayanava Majumdar, Post-doctoral Associate
Mark A. Boetel, Associate Professor
Robert J. Dregseth, Research Specialist
{ SHAPE * MERGEFORMAT **}**
Department of Entomology, North Dakota State University, Fargo, ND

Introduction

Sugarbeet root maggot (SBRM), *Tetanops myopaeformis* (Röder) (Diptera: Ulidiidae), is a major concern for sugarbeet producers in the Red River Valley (RRV) of North Dakota and Minnesota. Almost the entire sugarbeet acreage infested with SBRM is treated with chemical insecticide. Treatments are generally directed toward the larval stage of its life cycle. Larvae and pupae are affected by a variety of biotic and abiotic environmental factors in their subterranean habitat. While there is some available information regarding the effects of abiotic elements on SBRM larvae, information on the effects of biotic factors on the SBRM life cycle is lacking due to the cryptic nature of this insect. The pupa is a vulnerable stage in the SBRM life cycle because it is unable to escape or protect itself from hazardous elements. However, the nonfeeding pupal stage of the root maggot is generally overlooked. The skin (or cuticle) of the third-instar larva becomes a natural barrier to environmental hazards. If this protective layer is breached then this stage can die or develop abnormalities.

This report summarizes findings from one preliminary and two systematic surveys conducted over a three-year period (2004-2006). The work involved individual screening of 4,117 specimens from over 18 collection sites. This report not only provides information about a newly-isolated native SBRM pathogen, but also provides information about the effect of climate on the biology of this pest.

2004 Survey. Personnel of the NDSU sugarbeet entomology regularly collect subterranean stages of the root maggot for future research. A previously unobserved phenomenon was observed on one of these trips in 2004. At a field heavily infested with SBRM, several pupae (44% of 1,200 live specimens) were found to be infected with a soil-borne pathogen – *Fusarium solani* (Martius) Saccardo (Majumdar et al. 2006). The infectious nature of the isolate was confirmed by multiple artificial inoculations of healthy pupae (Majumdar et al. 2006). The isolate was subsequently accessioned as ARSEF7382 and S2118 at two fungal collection centers (USDA-ARS Fungus Collection Center, Ithaca, NY, and Fusarium Research Center, Penn State University, PA). This common pathogen has several strains or forms that are pathogenic to insects. A *F. solani* strain is known to be pathogenic to pupae of Anthomyiid flies (Humber 1992). Based on two 15-d bioassays with eight conidial concentrations, the median lethal concentration of ARSEF7382 to SBRM pupae was found to be 1.8×10^6 conidia/ml (Majumdar et al. 2006). Transverse sections of infected pupae indicated that the fungus can penetrate the cuticle and grow within SBRM pupae. Pupae in advanced stages can also be killed by ARSEF7382.

According to Gopalakrishnan and Narayanan (1989), *F. oxysporum* pathogenic strain IMI 318632 caused 100% mortality of guava shield scale insect, *Pulvinaria* (= *Chloropulvinaria*) *psidii* Maskel, when exposed to 4.8 x 10^8 conidia/ml for 5 d. Tanada and Kaya (1993) have suggested that one particular toxin called beauvericin, produced in ample amounts by *Fusarium*, is especially lethal to Diptera (flies). Thus, our findings are supported by a number of published accounts regarding this fungus. The survey of 2004 also suggested that disease prevalence in SBRM was synchronized with rainfall amounts (Table 1) and host availability. Peak fly activity was recorded on 30 June, 2004, which provided ample time for collection of a large number of pupae from the detection site. Overall, this discovery reiterated that all stages of the root maggot life cycle should be studied in order to have a complete understanding of ecological processes that affect the survival of this key pest of sugarbeet.

Table 1. Comparison of climatic conditions during three pathogen surveys, ND, 2004-2006

	2004 / 2005 / 2006			
St. Thomas	APRIL	MAY	JUNE	JULY
Soil temperature (^{o}F)	45 / 46 / 46	54 / 54 / 59	63 / 66 / 73	70 / 75 / 79
Rainfall (inches)	1.7 / 0.7 / 1.0	3.6 / 3.6 / 0.9	0.8 / 5.1 / 0.6	3.5 / 2.5 / 1.3
Forest River	APRIL	MAY	JUNE	JULY
Soil temperature (^{o}F)	40 / 46 / 49	51 / 54 / 60	65 / 69 / 72	72 / 74 / 80
Rainfall (inches)	0.8 / 0.9 / 1.1	4.8 / 3.0 / 0.9	0.6 / 6.4 / 0.7	2.2 / 1.4 / 1.8

Source: NDAWN, The North Dakota Agricultural Weather Network, NDSU, Fargo

2005 Survey. This survey was funded by the State Board of Agricultural Research and Education. It was the first intensive insect survey of its kind in the RRV. The objective was to locate natural epizootics of *F. solani* in SBRM populations and detect additional pathogens. A detailed account of the survey methodology and insect screening procedure were provided by Majumdar et al. (2006), which closely follows a published procedure of handling and diagnosis of insect pathogens (Lacey and Brooks 1997). A total of 2,400 pupae were collected in early July from many fields (Table 2) that were planted to sugarbeet the previous year. Field histories for the survey fields were collected from sugarbeet producers. It appears that potato crops grown prior to sugarbeet can increase the likelihood of finding *F. solani*-infected SBRM pupae. The low temperatures and high soil moisture that prevailed in June of 2005 appeared to be favorable for *Fusarium* epizootics in SBRM populations. This finding agrees with published reports of *Fusarium* epizootics in other insects (Venugopal et al. 1989, Villacaros and Robin 1989, Pandit and Tarannum 2002). The survey data also suggested that high early-season (i.e., May) rainfall amounts favors infection, probably by exposing SBRM pupae to high levels of viable spores; however, the unusually high (5.1 inches) rainfall in June probably reduced the availability of susceptible hosts (Table 1). This resulted in a low (3.8%) overall infection level of 92 infected pupae out of 2,400. Peak fly activity occurred late (22 June) in 2005. This provided ample time to plan and conduct the survey as the insects emerged from the soil. Adverse climatic conditions

(and other abiotic factors) caused mortality of 268 pupae, which was equivalent to 11% of sampled pupae (Table 2).

Table 2. *Fusarium solani* epizootics in *T. myopaeformis* pupae collected in eastern North Dakota, 2005

Township	Collection site coordinates	Elevation (m)	Soil temperature (^{o}C)	Previous crop	Total pupae collected	Infected pupae	Infection rate (%)	Emergence failure (%)
St. Thomas	N 48.33, W 97.28	257	20	Potato	740	33	4	7
	N 48.36, W 97.31	262	15	Bean	13	1	7	15
	N 48.37, W 97.29	262	16	N/A[a]	70	2	3	36
	N 48.37, W 97.32	262	15	Potato	173	1	<1	8
	N 48.41, W 97.27	261	17	Bean	153	4	3	3
	N 48.71, W 97.27	261	19	N/A[a]	195	6	3	19
	N 48.92, W 97.32	252	16	Potato	346	18	5	12
Lodema	N 48.38, W 97.32	264	15	Potato	390	23	6	10
Cavalier	N 48.49, W 97.38	265	16	Barley	300	2	<1	14
Forest River	N 48.15, W 97.24	N/A[a]	N/A[a]	Bean	20	2	10	35
Total					2400	92		
Average							4%	11%

[a] Not Available

2006 Survey. This survey was also funded by the State Board of Agricultural Research and Education. The 2006 sugarbeet growing season was marked by unusually low rainfall during the critical survey month of June. The rainfall received in May (0.9 inch), combined with higher soil temperatures, was adequate to cause rapid development and emergence of SBRM adult flies. Thus, the peak fly activity occurred early (2 June). About 163 flies were trapped per day on sticky stakes at peak fly activity (Boetel et al. in this report). This rapid emergence early in the season affected our ability to survey effectively. As a result, we were able to collect and screen only 517 pupae from over 10 fields spread across five townships. Pertinent data from four townships has been provided in Table 3. No pupae were found at fields near Cavalier and Auburn, ND (not presented). Pupae were hand-collected in the field and maintained under controlled environmental conditions until diagnosis. Screening was conducted within a week of collection to ensure pupal viability. Pupae were surface-sterilized using a 1% sodium hypochlorite solution followed by drying in a gentle stream of compressed air. Pupae were transferred to individual clean plastic soufflé cups containing four to five pieces of sterile moist filter paper to provide high humidity to pupae. After a 15-d incubation period, diseased pupae were microscopically examined to confirm *Fusarium* infection. Thereafter, diseased pupae were kept on culture media where *F. solani* forms distinctive colonies. The dry summer months and rapid fly emergence made it challenging to determine survey sites and collect root maggot pupae in dry soil. A total of 27 pupae from 517 collected specimens (= 5.2%) had *Fusarium* infections.

Table 3. *Fusarium solani* epizootics in *T. myopaeformis* pupae collected in eastern North Dakota, 2006

Township	Collection site coordinates	Elevation (feet)	Soil Temperature (^{o}F)	Previous crop	Total pupae collected	Infected pupae	Infection rate (%)	Emergence failure (%)
St. Thomas	N 48.36, W 97.31	863	60	Potato	29	0	0	27
	N 48.36, W 97.30	850	60	Potato	42	4	9	12
	N 48.33, W 97.28	826	70	Potato	3	0	0	0
Nash	N 48.29, W 97.33	858	72	Potato	377	19	5	34
	N 48.28, W 97.22	806	72	Potato	12	2	16	58
Forest River	N 48.17, W 97.21	802	67	Wheat	51	2	4	14
	N 48.15, W 97.24	834	74	N/A[a]	3	0	0	0
Total					517	27		
Average							5%	30%

[a] Not Available

Surprisingly, 31 larvae were collected after the peak fly activity. Many larvae were collected from a field site near Nash, ND. A high number of pupae (154 of 517 samples or about 30%) died from unknown reasons. High soil temperatures and low moisture levels likely affected

pupal development. It is also probable that prevalence of *F. solani* was low throughout the RRV in 2006 due to low rainfall amounts (i.e., dry soil conditions) during May and June.

Conclusions

The survey points out many important aspects about fungus activity and SBRM biology. The abundance of root maggots makes them a likely target for natural enemies, such as soil microorganisms. Coevolution of native pathogens exposed to SBRM (a native pest) could be a probable reason for the development of specialized pathogen strains, such as ARSEF7382, that are aggressive decimators of root maggot populations under suitable conditions.

Epizootics of insect-pathogenic fungi are favored by rainfall. There is a lag period between rainfall and disease development. High soil moisture could increase inoculum levels of the pathogen and aid in persistence of the fungal spores. Laboratory studies indicate *F. solani* spores to be highly germinative on artificial media.

Availability of a healthy host population is important for successful natural epizootics and their detection during surveys. Asynchrony between SBRM and *F. solani*, triggered by climatic variations as seen in the surveys, may result in low or undetectable levels of infection.

Effects of cultural practices on SBRM pathogens have not been documented. This study provides evidence regarding the buildup of fungus inoculum in relation to crop rotation. Sublethal effects of *Fusarium* infection on the survival and reproductive biology of SBRM adults is a topic for further research. Pathogenicity of the isolate toward larvae and adult flies also needs to be assessed. Molecular techniques could be used in future for rapid identification of *Fusarium* strains (Hennequin et al. 1999). This could pave the way for genetic characterization of insecticidal components produced by this fungus. Area-wide surveys should continue in order to discover more potential antagonists of the SBRM.

Acknowledgements

Thanks are extended to V. R. Panavala and R. Gooduru for assistance with collecting and screening SBRM pupae. Grants from the North Dakota State Board of Agricultural Research and Education (SBARE) supported much of this work.

References cited

Gopalakrishnan, C., and K. Narayanan. 1989. Occurrence of *Fusarium oxysporum* Schlecht and its pathogenicity on guava scale *Chloropulvinaria psidii* Maskell (Hemiptera: Coccidae). Curr. Sci. 58: 92–94.

Hennequin, C., E. Abachin, F. Symoens, L. Lavarde, C. Reboux, N. Nolard, and P. Berche. 1999. Identification of *Fusarium* species involved in human infections by 28SrRNA gene sequencing. J. Clin. Microbiol. 37: 3586–3589.

Humber, R. A. 1992. Collection of Entomopathogenic Fungal Cultures: Catalog of Strains, 1992. U.S. Department of Agriculture, Agricultural Research Service, Bulletin ARS-110.

Lacey, L. A., and W. M. Brooks. 1997. Initial handling and diagnosis of diseased insects, pp. 1–15. *In* L. A. Lacey (ed.), Manual of techniques in insect pathology. Academic Press, San Diego.

Majumdar, A., M. A. Boetel, and R. J. Dregseth. 2006. Survey, isolation, and screening for native pathogens of sugarbeet root maggot (Diptera: Ulidiidae). 2005 Sugarbeet Research and Extension Reports. Coop. Ext. Serv. North Dakota State University. 36: 228–231.

NDAWN Center. 2006. North Dakota Agricultural Weather Network. Department of Soil Science, North Dakota State University, ND. Website: http://www.ndawn.ndsu.nodak.edu/ (5 January, 2007).

Pandit, N. C., and Z. Tarannum. 2002. Record of fungi associated with diseased insects of jute and mesta crop. J. Entomol. Res. 26: 175–178.

Tanada, Y., and H. K. Kaya. 1993. Insect Pathology. Academic Press, San Diego.

Venugopal Rao, N., A. S. Reddy, and K. Tirumala Rao. 1989. Natural enemies of cotton whitefly, *Bemisia tabaci* Gennadius, in relation to host populations and weather factors. J. Biol. Control. 3: 10–12.

Villacarlos, L. T. and R. P. Robin. 1989. Entomogenous fungi infecting *Heteropsylla cubana* Crawford (Homoptera: Psyllidae) in Leyte, Philippines. Trop. Pest Management. 35: 120–122.