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COMPARISON OF ALFALFA WEEVIL POPULATIONS UNDER EPIZOOTIC AND ENZOOTIC CONDITIONS OF ERYNIAPHYTONOMI IN WISCONSIN

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ABSTRACT

Comparisons of two life tables constructed to display alfalfa weevil, Hypera postica (Gryllenhal), populations in southcentral Wisconsin, U. S. A. under epizootic and enzootic conditions of fungal disease, caused by Erynia phytonomi Arthur, suggests that the "prepupal" stage provided the greates contribution to population changes under both conditions due to the high mortality rate. The principle mortality agents during this stage are E. phytonomi and the parasitoids complex of Bathyplectes curculionis and Bathyplectes anurus respectively under the two conditions.

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INTRODUCTION

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The alfalfa weevil (AW), Hypera postica, (Cyllenhal) has been an important pest of alfalfa in the United State for nearly 75 years. It was first detected near Salt Lake City Utah, in 1904 and was thought to be of European origin (Titus 1910). Since its introduction into the eastern U. S in the early 1950's (Poss & Dissell 1953), the AW has become the most serious pest of the first cutting of alfalfa, and at times may also do considerable damage to the second cutting. In Wisconsin, rank first with respect to alfalfa production in the U. S., the AW caused considerable damage during some years (Wedberg & Rohweder 1979).

However, following its spread to the eastern part of the U.S. and Canada, AW damage was greatly reduced by the dramatic appearance of a highly contagious disease organism that attacked and killed the larval and prepupal stages. This pathogen was identified as *Entomophthora phytonomi* Arthur, a fungus new to the AW in North America, but long recognized as an important natural control agent in populations of a related host, the clover leaf weevil, *H. punctata* (Harcourt *et al.* 1974, 1977). However the Ben- $\frac{3}{4}$ eev and Kenneth (1982) revision of the genus *Entomophthora* placed the fungus reported by Harcout *et al.* (1974) in the genus *Erymia*.

Other biological control agents that markedly affect AW populations are two Ichneumonid parasitoids, *Bathylectes curculionis* and *B. anurus*. both of which attack the larval stage.

MATERIALS AND METHODS

Sampling Fields

Test sites were established in alfalfa fields (var. Vernal) of relatively pure stand. No. insecticides or herbicides were used in or near the study fields.

Field A: A four-hectare field was selected in southcentral Wisconsin near the University of Wisconsin campus in Madison. The field was enclosed on all sides by wooded areas, and contained a three-year old stand of alfalfa.

Field B : A three-hectare field was chosen for study in the same area, and within 5 -m of field A. It had similar site conditions, except that it

contained a first year alfalfa stand that had been seeded the previous fall.

The sampling area (plot) in each field was restricted to one hectare $(100 \times 100 \text{ m})$ which was subdivided into nine equal subplots $(33 \times 33 \text{ m.})$, resulting in a 3×3 grid.

Estimating Populations and Mortality Factors

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Sampling techniques utilized for assessing weevil populations during the egg, larval, prepupal and pupal stages were as follows :

Numbers of eggs deposited in dead stems, green stems, and ground litter were determined weekly from 0.022 m. sq. in each subplot throughout May using a modification of the method described by Pass and Van Metere (1966). Briefly, each sample was pulverized in a Waring blender, then filtered through a series of screens (U.S. standards of 20, 30, 40) and a final sieve of 52-mesh. Material remaining on the last screen was removed with water, concentrated on a 15 cm dia, filter paper, then examined under 12X magnification for the presence of AW eggs. Eggs were picked up and placed on moist sterile filter paper in 15 cm petri dishes and held at 21'+1'C until the status of the eggs could be determined. Two different mortality factors were recognized : a) those killed by the mymarid egg parasitoid, *Anaphes luna* and b) those that did not hatch and were considered to be nonfertile.

To estimate populations and mortality factors during the larval stage, three distinct periods of development, as described by Harcourt *et al.* (1977), were recognized : a) "Period 1" (establishment phase) : represented by Ist instar larvae that had not reached the terminal buds; b) "Period 2" included both first and second instar larvae; and c) "Period 3" included third and fourth larvae. During the establishment phase, larvae often traveled considerable distances before reaching terminal buds. Thus they were subject to misadventure, such as rain, and attack by predators (Harcourt *et al.* 1977). The most common biological agents that attacked "Period 2, and 3" larvae were the parasitoids, *B. curculionis* and *B. anurus*, which emerged as adults at approximately the same time that AW eggs were deposited, and thus attacked early instar larvae. In contrast, the fungal pathogen, *E. phytonomi* was an important mortality agent during the time of "Period 3" larval activity. In southern Wisconsin, the fungal epizootic, if present, usually coincided with larval peak numbers during the last week of. May. Weather conditions

at that time usually favor both sporulation and dissemination of conidia. However, the pathogen attacked all larval instars.

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To estimate larval mortality factors, samples consisting of 10 stems per subplot (90 stems per field) were taken twice weekly through the period of AW activity. Larvae were removed, recorded, and then separated into two groups. one consisting of Ist. and 2nd. instar forms, and the other, 3rd. and 4th. instar forms. Larvae from both groups were then placed individually in ventilated, 1 oz., paper cups with fresh alfalfa foliage, along with date and sampling site information. Larvae were provided with fresh alfalfa and examined for pupal parasitoids or disease symptoms daily. Parasitoids were based on those larvae which were successfully parasitized and displayed no other abnormalities, in particular any interactions with the pathogen.

Larvae exhibiting disease symptoms were separated into two groups : a) those that were brown to tan, and b) those that were blacc in color (Ben Zeev and Kenneth, 1982). The brown or tan diseased larvae were placed on water agar (15 g/1 H20) in sterile disposable petri dishes and held at 20 '+1' C and 14:10 (L:D) photoperiod. The plates were examined daily for the presence of conidial spores. All black larvae were placed singly in petri plates and examined under a binocular microscope (60X) to determine if they contained internal resting spores.

Numbers of prepupae (cocoons) were determined twice weekly by Morris (1963) and Harcourt *et al.* (1977). The Ix values were based on direct sampling. Sufficient population samples were taken to estimate population sizes and mortalities at density levels before, during, and after various stage peaks. To determine populations of "eggs", "Period 2 & 3 larvae", and "pupae", at least 10 foliage and litter samples were secured throughout the AW activity period.

Using the graphical method modification of Southwood and Jepson (1962), and mean stage durations from data cited by Guppy and Muker (1974), the different developmental stage estimates were integrated to obtain a Ix value for each stage interval per 0.09 m. sq. However, the Ix value for "Period 1 larvae" was calculated indirectly, and is the product of egg density times the survival rate of eggs, the same as was done for the "prepupal" stage. The dx values were counted from the percent mortality caused by either

disease or parasitoids, and then adjusted to represent the number of individuals killed per 0.09 m. sq. (Appendix no. 14, 15) However the dx value for "period 1 larvae" was obtained indirectly by subtracting "Period 2" from "Period 1 larvae" in the Ix column. Since total mortality observed in the laboratory by examining larvae and pupae was less than the actual field mortality obtained from observing Ix values, a mortality factor was counted for the unknown.

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RESULTS AND DISCUSSION

Figure 1 shows the annual epizootic, host densities, and percent of parasitism of the two populations in fields A and B. Highest percent of infection by E. *phytonomi*, and larval mortality appeared to coincide with the larval population density peak. The pathogen effectively reduced the AW population during the course of the epizootic (May 29-June 3) when over 50° of the feeding larvae population at that time were infected and died. However, observing the data in Table 1 does not indicate that E. *phytonomi* effectively reduced the overal larval population, probably due to the late time of the disease outbreak. In Field B, where no epizootic was recorded, the parasitoids B. curculionis and B. anurus increased the rates of parasitism and caused approximately 35% mortality of the total AW population. Analysis of life tables

Life table analysis usually beging with an attempt to determine the relative importance of the different age intervals under study by establishing a simple linear correlation between them and generation survival. Following Watt (1963 and Harcourt *et al.* (1977), we attempted to identify the stage in the cycle that made the greatest contribution to a population trend. Population survival was expressed as the product of a servival of successive stages.

 $Swag = Se Sp1 Sp2 Sp3 Spp Sp \dots (1)$

Where Swg = within - generation survival, Se = eggs survival, Sp1 = larvae Period 1 survival. Sp2 = larvae Period 2 survival. Sp3 = larvae Period 3 survival. Spp = prepupae survival. Sp = pupae survival. Therefore .

 $.081 = (.746) (.704) (.828) (.698) (.297) (.909) \dots$ from Table 1.

.157 = (.764) (.794) (.814) (.863) (.421) (.875) from table 2

As illustrated in Tables 1 & 2, the "prepupal" stage interval provided the largest contribution to population changes both under disease epizootic and enzootic conditions.

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In comparison with other results, Harcourt *et al.* (1977) found that variation within-generation mortality was mostly contributed by the larval stage, which is described as "Period 3 larvae" in this study. The principle mortality agent during this stage interval was E. *phytonomi*, which caused 83% mortality. One may expect different results because of differences in epizootic magnitude and host density. However, we believe that Harcourt and co-workers were overestimated the disease mortality since they estimated larval mortality at the time of the disease epizootic, and also because their estimations were based on diseased dead larvae that had exhibited conidial showers, or were about to shower.

Under enzootic disease conditions, *Bathypleites* spp. iaused 47% mortality (Table 2) in the prepupal stage in comparison to 19% mortality for the same stage indicated by Harcourt *et al.* (1977). On this evidence, it would seem that *Bathyplectes* spp. may become promising AW control agents in Wisconsin.

Although the two study sites (Field A and B) were similar in many respects, the epizootic which occured in Field A probably was due to differences in host densities and the age of the alfalfa stand. Host densities in Field B probably did not reach the level necessary to initiate a disease epizootic. Nordin *et. al.* (1983) proposed that a density of 1.7 larvae/sfem is required to initiate an epizootic of *Erynia* sp. Grop stand age may also play a role in the availability of inoculum source, which is believe to be the resting spires. Field A probably had more inoculum than Field B, since the later was seeded the previous fall. This conclusion may support the idea that this pathogen forms resting spores in the overwintering host stage in the field

Table (1)

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Life table for alfalfa weevil under enzootic disease

condition of Erynia phytonomi in southcentral Wisconsin in 1986.

x		dxF	dx	100qx	Sx		
Stage	No. falive	Factor re	No. •dying	a dx as	Survival		
interval	begining	-ponsible for dx	during x	Percent-	Percent- rate		
	anot x			age of lx	within x		
Eggs	122	Infertility	22	18.0	5 10 1000 56		
		A. luna		7.3			
		Total	31	25.4	.746		
Period 1	91	Establishment	27	29.6	.705		
Period 2	× 64	loss E. phytonomi		14.0			
		Unknown	2	3.1			
		Total	11	17.1	.828		
Period 3	53	E. phytonomi	14	26.0			
		Unknown	ź	3.7			
		Total	16	30.1	.698		
Period	· · · · 37	E. phytonomi		24.3			
		B. anurus	•	16.2			
		B. curculionis	84 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1 - 1	24.3			
	Martine 1	Unknown	2	5.4			
		Total	.26	70.2	.297		
Pupae	11	Unknown	1	9.000.0	.909		
Summer adult	10						
Gen. total	ls		112	91.8	.082		

• Numbers per 0.09 m. sq.

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Table (2)

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Life table for alfalfa weevil under epizootic disease

conditions of Erynia phytonomi in southcentral Wisconsin in 1986.

x	lx	dxF	dx	100qx	Sx
Stage 1	No. *alive	Factor re	No. •dying	dx as	Survival
interval begining of x		-ponsible	during x	Percent-	rate
		for dx		age of lx	within x
Eggs	89	Infertility	15	16.8	:23
		A. luna	and the second second	6.7	
	潮游到41.1113	Total	21	23.5	.764
Period 1	68	Establishment	14	20.5	.794
		loss			
Period 2	54	Unknown	10	18.6	.814
Period 3	44	Unknown	6	13.6	.863
Prepupa	38	B. anurus	14	36.8	
		B. curculionis	Service and the service of	10.5	Second 1
		Unknown	- wondat	10.5	
		Total	22	57.9	.421
Pupae	16	Unknown	2	12.5	.875
Summer	14	111 P			
adult					
Gen. totals	- 4		75	84.2	.157

* Numbers per 0.09 m. sq.

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Erynia phytonomi,

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مقارنة المجاميع السكانية لحشرة منوسة الجت تحت الظروف الوبائية وغير الوبائية في وسكونسون

حسن ف حسن وجي ١ أم ٢ بوش قسم وقاية النبات ، كلية الزراعة ، جامعة بغداد و مختبر روسل ، جامعة وسكونسن ماديسون الولايات المتحسدة

الخيلاصية

عند مقارنة جداول الحياة المنجزة التي توضع ديناميكبة المجاميع السكانية لحشرة موسة الجت (Gryllenhal) ، في ولاية وسكانسن في الولايات المتحدة الاميركية ، تحت الظروف الوبائية وغير الوبائية وسكانسن في الولايات المتحدة الاميركية ، تحت الظروف الوبائية وغير الوبائية للمرض الفطري Erynia phytonomi Arthur ، اتضع ان الدور قبل العذري 'Prepupae' يعتبر المرحلة المهمة والمؤثرة في تغيير الكثافات العددية للحشرة وذلك نسبة للوفيات العالية خلال هذا الدور · العامل الاساسي لهذه الوفيات يرجع الى اصابة البشرة بالفطر قالى حالة الاصابية ، وبشكل خاص في الحالات الوبائية للمصرض اضافة الى حالة الاصابية بالطغيلي Bathyplectes anurus, Bathyplectes curculionis

عند غياب الحالة الوبائية للمرض الغطري .

Rassan F. Rasan and G. M. Bou

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معند مقررة مدارل المراة المحرية التي توقيع وبالمركزة البانيي المكاني المسرة مريدة الرمان المعاد الأمراكة والمعادية الفرول الولاية وم الرواية وسكاسي از الولايات المعند الأمراكة والعدة الفرول الولاية وم الرواية المرعى العاري المالية المعادية والعارية والعرب العارية والد المدر المع المسلوي العالية المرابية المعالية ملان مثل المور المالي المعادية المعادي الرالية المرابية المعادية مالية ملان مثل المور المالي المعادية المسلوي الرالية المرابية المعادية المحادية المعادية المعادية المسلوي الرالية المدرية المولية ملان مثل المور المالي المعادية المعادية المسلوي الرالية المدرية المعادية المحادية المحادية المعادية المعادية المعادية المعادية المسلوي الرالية المدرية المعادية المحادية المعادية المعادية

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Figure 1. Relationship between <u>Hypera postica</u> density and both the percent of infection by <u>Erynia phytonomi</u> and parasitism by <u>Bathyplectes</u> spp. in Field A & B.

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