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STUDIES ON LEAF BEETLES (*OULEMA* SPP.) DEVELOPMENT FOR SHORT-TERM FORECASTING – EVALUATION OF EFFECT OF TEMPERATURE AND HUMIDITY ON DURATION OF EGG INCUBATION

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Abstract: Two species of leaf beetles from *Chrysomelidae* family i.e. the cereal leaf beetle *Oulema melanopus* (Linnaeus 1758) and the blue leaf beetle *Oulema gallaeciana* (Heyden 1970) can cause considerable damage to cereal crops. Improvement of short-term forecasting was focused on determination of optimal time of control of these pests that takes place at mass larval hatching. Thus long term studies were carried out to evaluate a correlation between duration of egg incubation, temperature and air humidity. The length of egg incubation was examined at temperatures of 16°C, 20°C and 25°C and air humidity 60% in a growth chamber and under field conditions. Based on linear and curvilinear multinomial regression analyses the influence and significance of such independent variable (x) as: sum of temperatures (sums of mean daily temperatures), mean air humidity, sum of effective temperatures and mean effective temperature versus duration of egg incubation of *O. melanopus* and *O. gallaeciana* (dependent variable – y) were evaluated. Analyses of 32 interactions (x, y) showed that only four were insignificant i.e. mean air humidity in the growth chamber at 20°C had no effect on the length of egg incubation of *O. melanopus* and at 25°C on the length of egg incubation of *O. gallaeciana*. Also under field conditions sums of effective temperatures showed no effect on duration of egg incubation of both species.

Key words: *Oulema* spp., cereal crops, egg incubation, effective temperatures

INTRODUCTION

More frequent appearance of *Oulema* spp. on cereal crops in Poland causes losses of economical importance that has been recorded since 1986 (Walczak at el. 1987; Walczak 1990; Kaniuczak 1987, 1993). Mass occurrence of larval stage of these pests can result in significant decrease of area of leaf tissue of second leaf (46% of winter wheat and 26% of spring wheat) (Kaniuczak 1997).

Efficient pest control requires a selection of the most suitable, product and correctly determined time of chemical treatment. According to Jørgensen and Nielsen (1998) the later factor affects efficacy of disease and pest control more than an applied dose. Determination of the optimal time of *Oulema* spp. control is not easy due to extended time of laying eggs and brood of larvae. Thus cereal crop producers frequently decide to control *Oulema* spp. when feeding of larvae has already damaged substantial portion of leaf tissue.

Based on previous experiments and observations regarding biophenology of *Oulema* spp. two conclusions were withdrawn:

a) beetles of *O. gallaeciana* occur on cereal crops in the Wielkopolska region later than of *O. melanopus*, however, both species are controlled at the same time i.e. at mass larval hatching,
b) mass brood of larvae from mass laid eggs occurs at the same time what an appearance of larvae of ca. 4 mm length of both species of *Oulema* spp. that develop from freshly laid eggs. Both developmental stages indicate an optimal term of leaf beetle control (Bubniewicz et al. 1993; Zalecenia ochrony roślin 2001; Walczak 2002).

The objective of this research was to evaluate impact of temperature and air humidity on the length of egg incubation of *Oulema* spp.

MATERIAL AND METHODS

The insects were reared in an environmental growth chamber in the Institute of Plant Protection in the years 1997–2001. Their development was observed from egg lying until hatching of larvae at a specific set up temperature (16°C, 20°C, and 25°C) and ca. 60% air humidity. The programmed temperatures due to technical features of the growth chamber oscillated ±2°C and air humidity ±5%.

In the years 1999–2001 *Oulema* spp. were reared under the field conditions in net cages placed on winter wheat plants grown in the Winna Góra Experimental Station. The MetPole system recorded meteorological data (mean, daily air temperature and mean, daily relative air humidity).

Each spring the collected beetles were identified to *O. gallaeciana* and *O. melanopus* species and then only mating pairs were selected to ensure that females would be laying eggs. The insects were placed either on winter wheat plants, cv. Korweta growing in pots that were laid in the growth chamber (controlled environment) or on plants growing in the cages (field conditions).

Every day, at the same time, observations were performed in the growth chamber and field conditions to register development of both species; from egg lying until hatching of larvae. For further studies only information regarding those eggs from which larvae hatched was included.

Based on means of daily temperatures another three meteorological features were calculated; sum of temperatures, sum of effective temperatures and mean effective temperature. Physiological thresholds of egg incubation (*O. gallaeciana* – 10.2°C; *O. melanopus* – 10.6°C) were used to calculate the sums of effective temperatures (Ali et al. 1977). Data of mean, daily air humidity allowed calculating mean air humidity for egg incubation of *O. gallaeciana* and *O. melanopus*.

The results collected from the growth chamber surveys were analyzed statistically and used for determination of short-term prognosis. Individuals of *Oulema* spp. were regarded as an experimental unit. Curvilinear multinomial regression of the third degree (Elandt 1964, chapter 9 and 11) was applied to evaluate correlation between a length of egg incubation of *Oulema* spp., (two species; y) and each of the following factors (four independent variables); sum of temperatures (x_1), mean air humidity (x_2), sum of effective temperatures (x_3) and mean effective temperature (x_4), at each programmed temperature in the growth chamber and under field conditions. For each pair of examined variables (x, y) critical, significant level for regression (degree of freedom), coefficient of determination and standard deviation were calculated.

The same analyses were performed for the results collected from the field trials.

RESULTS AND DISCCUSION

In the years 1999–2000, 66 individuals of *O. gallaeciana* and 80 of *O. melanopus* were reared in the growth chamber at temperature of 16°C. In the years 1997–1998 and in 2001, 85 individuals of *O. gallaeciana* and 116 of *O. melanopus* were reared under controlled conditions at 20°C. In the years 1999–2001, 110 individuals of *O. gallaeciana* and 131 of *O. melanopus* were reared at temperature of 25°C. Totally, over the course of the studies 261 individuals of *O. gallaeciana* and 327 of *O. melanopus* were examined.

In the years 1999–2001 under field conditions, incubation of 26 eggs of *O. gallaeciana* and 23 of *O. melanopus* was observed on winter wheat crop.

Table 1 presents mean values of length of egg incubation, sums of temperatures, mean air humidity, sums of effective temperatures and means of effective temperatures for egg incubation of *O. gallaeciana* and *O. melanopus* in the growth chamber and field conditions. Duration of egg incubation of both species reared in the growth chamber at each examined temperature was shorter than in field conditions as length of egg development extended with decrease of mean air temperature (the lowest mean temperature was in field conditions).

Table 1. Mean values of length of *Oulema* spp. egg incubation and mean values of selected climatic factors as independent variable

Conditions	Mean time of egg incubation (days; y)	Mean sum of temp. [°C (x_1)]	Mean of means of RH [% (x_2)]	Mean sum of effective temp. [°C (x_3)]	Mean of means of sum effective temp. [°C (x_4))	Mean air temp.
			Oulema gallaeciana			
Growth chamber 16°C	12.6	212.7	60.5	81.3	6.4	16.5
Growth chamber 20°C	7.2	156.8	61.6	84.2	11.7	21.8
Growth chamber 25°C	5.5	133.8	57.9	78.1	14.3	24.5
Field trials	15.2	219.3	68.3	65.8	4.4	14.6
			Oulema melanopus			
Growth chamber 16°C	12.2	205.6	60.4	74.6	6.1	16.6
Growth chamber 20°C	7.3	167.7	62.0	88.5	11.8	22.4
Growth chamber 25°C	5.6	136.2	58.2	77.4	13.9	24.5
Field trials	17.0	248.7	67.0	71.1	4.3	14.8

Also mean sums of temperatures for egg incubation of *Oulema* spp. increased with decrease of temperature in the growth chamber. The highest means were registered under field conditions along with the longest time of egg development. Mean sums of temperatures for egg incubation of both *Oulema* spp. in field conditions were similar to those values that were collected from studies carried out at 16°C in the growth chamber. Besides, in field trials, mean sums of temperatures at time of egg incubation were higher for *O. melanopus* than for *O. gallaeciana* (248.7°C and 219°C, receptively). The same correlation was found for the temperature of 20°C (*O. melanopus* 167.7°C, *O. gallaeciana* 156.8°C) and for 25°C (*O. melanopus* 136.2°C, *O. gallaeciana* 133.8°C). This relationship resulted form the length of development period that was longer for *O. melanopus* than for *O. gallaeciana*.

Mean value of means of air humidity in the growth chamber was ca. 60% for each range temperature of *Oulema* spp. egg incubation. It varied slightly and ranged from 57.9% to 62.0%. Under field conditions mean air humidity for egg incubation was 67.7% for *O. melanopus* and 68.3% for *O. gallaeciana*.

Mean values of sums of effective temperatures for three temperatures programmed in the growth chamber varied from 78.1°C to 84.2°C for *O. gallaeciana* egg incubation and 77.4°C to 88.5°C for *O. melanopus* (mean 81.2°C and 80.2°C, respectively). Under field conditions sums of effective temperatures were lower for egg incubation. The mean for *O. gallaeciana* was 65.8°C and for *O. melanopus* 71.1°C. Differences between means of sums of effective temperatures in field and controlled conditions did not exceeded 19°C. However, due to low number of individuals reared under field conditions it is hard to define sums of effective temperatures for *Oulema* spp. egg incubation.

According to data describing Halle region (Ali et al. 1977), sums of effective temperatures for *Oulema* spp. egg incubation in this area are close to the results presented in this paper. These authors defined the sum of effective temperatures in a growth chamber at established temperatures of 15°C, 20°C and 25°C and relative humidity 50%–60%. It was equal to 87.1°C for *O. gallaeciana* and 79.9°C for *O. melanopus*. In presented here studies the difference between the highest and lowest mean sums of effective temperatures in the growth chamber was only equal to 6.1°C for *O. gallaeciana* and 13.9°C for *O. melanopus*. Despite high differences in mean daily temperatures in growth chamber, differences in the sums of effective temperatures were insignificant indicating their stability. Means of sums of effective temperatures obtained for large number of reared individuals (588) are more representative that those referring to only 49 observed under field conditions. In the growth chamber at temperature 16°C, 20°C and 25°C and air humidity 57.9%–62% these values were 81.2°C for *O. gallaeciana* and 80.2°C for *O. melanopus*.

Freier et al. (1985) studied also effect of sums of effective temperatures while developing simulation model for forecasting of population dynamic of *Oulema* spp. and harmfulness of pests to winter wheat. They calculated sums of effective temperatures including temperature thresholds (6°C for female migration period and 9°C for egg lying) excluding physiological thresholds, as was done in presented here studies. Thus these results could not be compare with presented above on own data.

Mean values of mean effective temperature for egg incubation of *Oulema* spp. decreased along with decline of programmed temperature in the growth chamber. The lowest mean effective temperatures for egg incubation for both species were registered in field conditions where mean air temperature was the lowest compared to the growth chamber (14.6°C and 14.8°C, respectively).

In the next step of the investigation, the effect of separate four examined independent variables on duration of egg incubation was studied at three different temperatures and separately for each species in field conditions and growth chamber. Linear and curvilinear multinomial regressions were applied for this analysis.

For each pair of examined variables the following parameters were calculated: degree of multinomial (using degree of freedom), coefficient of determination and standard deviation (Table 2).

Available national and foreign literature lacks information concerning studies that applied curvilinear multinomial regressions to analyze the correlation between duration of presented in this paper developmental stage of *Oulema* spp. and effect of abiotic factors.

Analysis of results from the growth chamber test carried out at temperature 16°C (Table 2)

Each of independent variables analyzed separately affected significantly duration of egg incubation of *O. gallaeciana* and *O. melanopus*. Comparing influence of independent variables on dependent variable of both *Oulema* species it was concluded that as far as duration of egg incubation is considered, both species respond similarly to changes of sums of temperatures (97.1% and 97.8%) and sum of effective temperatures (72.5% and 79.5%). Mean air humidity showed higher influence on *O. melanopus* development (51.2%) compared to *O. gallaeciana* (42.5%). Instead, mean effective temperature affected more length of egg incubation of *O. gallaeciana* (42.4%) than *O. melanopus* (30.7%).

Analysis of results from the growth chamber test carried out at temperature 20°C (Table 2)

Influence of each independent variable on duration of egg incubation, besides the correlation between mean air humidity and egg incubation of *O. melanopus*, was very significant. The examined *Oulema* species reacted alike to changes of sum of temperatures (95.8% and 91.1%) and sum of effective temperatures (89.6% and 79.9%). Mean effective temperature affected more development of *O. gallaeciana* eggs (50.3%) compared to *O. melanopus* (14.8%). Mean air humidity modified length of egg incubation of *O. gallaeciana* (19.3%) and had no significant influence on development of *O. melanopus*.

Analysis of results from the growth chamber test carried out at temperature 25°C (Table 2)

Three independent variables, either sum of temperatures or sum of effective temperatures or mean effective temperature, each separately, had very significant effect on the length of egg incubation of *O. gallaeciana* and *O. melanopus*. On the other hand, mean air humidity significantly influenced on egg development of

Table 2. Characteristics of correlations between duration of egg incubation of *Oulema* spp. and meteorological parameters based on linear and curvilinear multinomial regressions analyses

Conditions	Dependent variables	*Oulema gallaeciana*					*Oulema melanopus*				
		Deg. of multinomial	Deg. of freedom for deviation	Critical significant level	Coef. of determination	Standard deviation	Deg. of multinomial	Deg. of freedom for deviation	Critical significant level	Coef. of determination	Standard deviation
Growth chamber 16°C	sum of temp.	1	64	<0.001	97.1	0.28	1	78	<0.001	97.8	0.24
	mean air humidity	1	64	<0.001	42.5	1.24	1	78	<0.001	51.2	1.10
	sum of effective temp.	1	64	<0.001	72.5	0.86	2	77	<0.001	79.5	0.72
	mean of effective temp.	1	64	<0.001	42.4	1.24	1	78	<0.001	30.7	1.31
Growth chamber 20°C	sum of temp.	2	82	<0.001	95.8	0.27	3	112	<0.001	91.1	0.37
	mean air humidity	1	83	<0.001	19.3	1.18	1	114	0.613	0.2	1.24
	sum of effective temp.	2	82	<0.001	89.6	0.43	3	112	<0.001	79.9	0.56
	mean of effective temp.	2	82	<0.001	50.3	0.93	1	114	<0.001	14.8	1.14
Growth chamber 25°C	sum of temp.	2	107	<0.001	99.5	0.06	2	128	<0.001	99.5	0.05
	mean air humidity	1	108	0.063	3.2	0.89	1	129	0.035	3.4	0.69
	sum of effective temp.	2	107	<0.001	98.5	0.11	2	128	<0.001	98.4	0.09
	mean of effective temp.	1	108	<0.001	12.9	0.84	2	128	0.001	10.0	0.67
Field conditions – Winna Góra	sum of temp.	1	24	<0.001	82.8	0.88	1	21	<0.001	77.6	1.35
	mean air humidity	2	23	<0.001	88.1	0.75	2	20	0.001	75.3	1.45
	sum of effective temp.	1	24	0.598	1.2	2.12	1	21	0.516	2.0	2.82
	mean of effective temp.	2	23	<0.001	65.7	1.27	1	21	<0.001	49.4	2.02

The Least Significant Difference at p equal $0.01<p<0.05$ – differences are significant, and at $p < 0.01$ differences are very significant

O. melanopus and showed no significant effect on egg incubation of *O. gallaeciana*. Comparing impact of independent variables on dependent of temperatures – 99.5% and 99.5% or sum of effective temperatures – 98.5% and 98.4%) had variable of both examined *Oulema* species it was stated that two variables, each separately (sum
similar effect on duration of egg incubation. Mean effective temperature showed less influence (12.9% and 10.0%). The response of both species to changes of mean air humidity varied. It affected egg incubation of *O. melanopus* less than three other independent variables (3.4%) and had no significant effect on *O. gallaeciana* development.

Analysis of results from field trials (Table 2)

Significant effect of sum of temperatures, mean air humidity and mean effective temperature on egg incubation of *O. gallaeciana* and *O. melanopus* was found under field conditions. Sum of effective temperatures had no influence on egg incubation of both species. *O. gallaeciana* reacted more to changes of independent variables, excluding sum of effective temperatures, (82.8%, 88.1%, 65.7%) compared to *O. melanopus* (77.6%, 75.3%, 49.4%).

Comparison of the results from the growth chamber studies and field trials

Analyzing individual independent variable at each temperature examined in the controlled and field conditions with their impact on duration of egg incubation the following conclusion were withdrawn:

1. Sum of temperatures significantly affected *Oulema* spp. development in the field and controlled conditions. Mean sum of effective temperatures increased with a decrease of mean temperature. Mean sum temperatures in field conditions were the closest to these from the test performed at 16°C in the growth chamber. In field conditions higher mean sum of temperature was suitable for egg incubation of *O. melanopus* compared to *O. gallaeciana*. The results from tests carried out in the growth chamber at 20°C showed the same correlation.
2. Mean air humidity affected significantly and very significantly egg incubation, except *O. melanopus* reared at 20°C and *O. gallaeciana* at 25°C in the growth chamber. Low correlation or insignificance of effect of mean air humidity on egg incubation in growth chamber compared to field conditions resulted from relatively high humidity at programmed temperatures. Mean air humidity ranged from 57.9% to 62.0% at temperature 16°C, 20°C and 25°C. Only these small fluctuations of air humidity allowed investigating influence of these temperatures on length of egg incubation. Under field conditions at mean temperature 14.6°C–14.8°C, mean air humidity was 67%–68.3%. A decrease of the first value caused an increase of the second one.
3. Sum of effective temperatures very significantly affected duration of egg incubation of *Oulema* spp. at all temperatures investigated in the growth chamber however, had no effect in field conditions. Despite high differences in mean daily temperatures in growth chamber, mean sums of effective temperatures were stable. Differences between mean sums of effective temperatures in growth chamber and field conditions did not exceed 19°C.

4. Mean effective temperature significantly affected egg incubation of both *Oulema* species in field and controlled conditions. Mean values of mean effective temperatures were lower at lower programmed temperature in the growth chamber. The lowest values were registered in field conditions where mean temperature was lower than in growth chamber.

CONCLUSIONS

1. Different ranges of temperatures in growth chamber and field conditions affected means of sums of temperatures and means of effective temperatures. However, they did not show significant effect on mean of sums of effective temperatures for egg incubation.
2. Small fluctuations of air humidity in the growth chamber allowed to investigate effect of programmed temperatures on duration of egg incubation.
3. The conducted studies with statistical analyses confirmed that variability of temperature and air humidity affects length of developmental stages of *Oulema* spp. Investigated correlations that included four meteorological parameters and three temperatures established in the growth chamber and also field conditions showed that only four correlations out of 32 examined had insignificant effect of independent variables on dependent variables ($p>0.05$).
4. These results will be used to perform the short-term forecasting of *Oulema* spp. with application of curvilinear multinomial regressions.

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POLISH SUMMARY

BADANIA NAD ROZWOJEM SKRZYPIONEK (*OULEMA* SPP.) DLA POTRZEB PROGNOZOWANIA KRÓTKOTERMINOWEGO – OCENA WPŁYWU TEMPERATURY I WILGOTNOŚCI POWIETRZA NA DŁUGOŚĆ INKUBACJI JAJ

W Polsce, obok innych szkodników zbóż, szkody gospodarcze mogą powodować dwa gatunki *Oulema* spp., tj. skrzypionka zbożowa – *Oulema melanopus* (Linnaeus 1758) i skrzypionka błękitek – *Oulema gallaeciana* (Heyden 1870) z rodziny Ch*rysomelidae*. Doskonalenie prognozowania krótkoterminowego *Oulema* spp. skoncentrowano na wyznaczeniu optymalnego terminu zwalczania, który przypada w okresie masowego wylęgu larw. W tym celu przeprowadzono wieloletnie badania w poszukiwaniu zależności między długością inkubacji jaj a temperaturą i wilgotnością powietrza. Badano długość inkubacji jaj w fitotronie w 16°C, 20°C i 25°C i około 60% wilgotności powietrza oraz w warunkach polowych. Wykorzystując metodę regresji wielomianowej prostoliniowej i krzywoliniowej oceniono wielkość i istotność wpływu (zmiennych niezależnych – x) sumy ciepła (sumy średnich dobowych temperatur powietrza), średniej wilgotności powietrza, sumy temperatur efektywnych i średniej temperatury efektywnej na długość inkubacji jaj *O. gallaeciana* i *O. melanopus* (zmiennych zależnych – y). Spośród przebadanych 32 związków (x, y) nie stwierdzono istotnego wpływu tylko w przypadku 4 związków tj.: średniej wilgotności powietrza w fitotronie w temperaturze 20°C na długość inkubacji jaj *O. melanopus* i w 25°C na długość inkubacji jaj *O. gallaeciana*, a także sumy temperatur efektywnych w warunkach polowych na długość inkubacji jaj obu gatunków.