

## SCIENTIFIC COMMUNICATION

### SEASONAL DYNAMICS OF EGG DIAPAUSE IN *MAHANARVA SPECTABILIS* (DISTANT, 1909) (HEMIPTERA: CERCOPIDAE) ON ELEPHANT GRASS

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#### ABSTRACT

This work examines the occurrence, duration and viability of diapause eggs of *Mahanarva spectabilis* (Distant, 1909) at different periods of the year. Adult insects were gathered twice a month in a greenhouse from September 2005 to May 2006 as well as in the same period in 2006-2007. The spittlebugs collected were taken to the laboratory, sexed and kept in cages containing one elephant grass plant potted in a 500 mL plastic cup with the upper part wrapped in gauze to serve as a substrate for egg laying. Around one hundred eggs obtained on each sample date were kept in climate-controlled chambers. The duration and viability of the eggs were analyzed daily. The viability was greater than 50% and 72%, respectively, in the evaluations for September 2005 to May 2006 and the same period of 2006-2007. The average embryonic period from September 2005 the first half of March 2006 varied from 15.79 to 28.24 days, except in the first half of January. In the other samples it varied from 94.80 to 139.04 days, with these being considered diapause. In the second study period (2006-2007), an embryonic period under 27 days was registered from September to the first half of November 2006 and again in January and February 2007. In the other evaluations the insects remained in the egg phase from 55.30 to 196.42 days. The largest number of diapause eggs occurred in the autumn, with 100% and 90% for the first and second evaluation periods, respectively.

KEY WORDS: *Penisetum purpureum*, forage, climatic factors, spittlebug.

#### RESUMO

DINÂMICA SAZONAL DE OVOS EM DIAPAUSA DE *MAHANARVA SPECTABILIS* (DISTANT, 1909) (HEMIPTERA: CERCOPIDAE) EM CAPIM-ELEFANTE. O objetivo da pesquisa foi avaliar a ocorrência, duração e viabilidade de ovos em diapausa de *Mahanarva spectabilis* (Distant, 1909) oriundos de diferentes épocas do ano. Adultos coletados, quinzenalmente, de setembro a maio em 2005/2006 e 2006/2007, em casa-de-vegetação, foram levados para o laboratório, separados por sexo e mantidos em gaiolas contendo uma planta de capim-elefante em copo plástico de 500 mL, com a porção superior envolvida por uma secção de gaze que serviu de substrato para oviposição. Cerca de cem ovos obtidos em cada data foram mantidos em câmaras climatizadas. Diariamente foram analisadas a duração e viabilidade de ovos. A viabilidade dos ovos de *M. spectabilis* foi superior a 50 e 72% nas avaliações de setembro de 2005 a maio de 2006 e no mesmo período de 2006 a 2007, respectivamente. O período embrionário total médio de setembro de 2005 à primeira quinzena de março de 2006 variou de 15,79 a 28,24 dias, exceto na primeira quinzena de janeiro. Nas demais amostragens variou de 94,80 a 139,04 dias, sendo esses considerados em diapausa. No segundo ciclo de avaliação (2006/2007), um período embrionário inferior a 27 dias foi registrado de setembro até a primeira quinzena de novembro/2006 e em janeiro e fevereiro/2007. Nas demais avaliações os insetos permaneceram na fase de ovo de 55,30 a 196,42 dias. O maior número de ovos em diapausa ocorreu no outono, com 100% e 90 % para o primeiro e segundo ciclo de avaliação, respectivamente.

PALAVRAS-CHAVE: *Penisetum purpureum*, forrageira, fatores climáticos, cigarrinha-das-pastagens.

Spittlebugs of the genus *Mahanarva* (Hemiptera: Cercopidae) cause serious losses in pastures, threatening the production of meat by compromising the available forage (PAULA-MORAES, 2006). This pest can

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also injury other crops such as sugarcane (GARCIA *et al.*, 2006). These insects are widely distributed in South and Central America. Their presence has been reported in various regions of the world (FEWKES, 1969). The attack of species of the genus *Mahanarva* on elephant grass, depending on the period of the year and population density, can even kill off the forage (AUAD *et al.*, 2007).

To survive during the dry period of the year, spittlebugs have developed a form of resistance during the egg phase. During the rainy season, non-diapause and diapause eggs are laid. The incubation period of diapause eggs is variable, and can last up to 288 days (PACHECO, 1981), besides synchronizing the occurrence of the insects in the field with the growing cycle of their host plants (SUJII *et al.*, 1995).

An understanding of the mechanisms that regulate diapause and determine the pattern of its occurrence in insect pest populations is an important tool in their management (TAUBER *et al.*, 1994). Knowledge of the events that can cause changes in the metabolism, and thus the lifecycle, of these insects is essential to understand the seasonal fluctuations of their populations (SUJII *et al.*, 1995; SUJII *et al.*, 2002). Therefore, determination of the period of the year when most of the diapause eggs of spittlebugs occur can help guide decisions on when to use preventive measures to reduce egg laying (KOLLER; HONER, 1993). The population onset in the field is determined by environmental factors favorable to oviposition and hatching of nymphs.

The objective of this study was to evaluate the occurrence, duration, and viability of diapause eggs of *Mahanarva spectabilis* (Distant, 1909) in spring, summer and autumn. These data are of great importance to predict the occurrence of population peaks and the availability of insects to be kept in artificial conditions for use in experiments.

The studies were conducted at the Embrapa Gado de Leite (Juiz de Fora, Minas Gerais, Brazil). Adults of *M. spectabilis* collected in the Municipality of Valença, Rio de Janeiro, were kept on potted elephant grass in a greenhouse. Adults from this colony were then transferred every other week, from September 2005 to May 2006 (first evaluation cycle) and in the same period from 2006 to 2007 (second period), to the laboratory, sexed and maintained in clear plastic cylindrical cages (50 cm height x 10 cm diameter) at  $27 \pm 2^\circ\text{C}$ , UR of  $70 \pm 10\%$  and 12-h photophase.

A 500-mL plastic cup containing a single elephant grass plant was placed in each cage, with the upper part wrapped in gauze moistened with distilled water, to serve as a substrate for egg laying for five days. Then the gauze containing the eggs was placed over a set of sieves (400 mesh) under running water to collect the eggs.

Approximately 100 eggs obtained on each date were placed in 5-cm Petri dishes, covered with moistened filter paper, and kept in climate-controlled chambers in the same conditions under which the adults were maintained in the laboratory. The duration and viability of diapause and non-diapause eggs were analyzed daily under a stereoscopic microscope over the entire evaluation period. The discrimination of diapause and non-diapause eggs was done by the embryonic period, by which, according to CASTRO *et al.* (2005), those that remained in the egg phase for over 30 days are diapause, and under this period are non-diapause.

We correlated the total number of diapause and/or non-diapause eggs of *M. spectabilis* obtained in each sample and the average temperature, relative humidity, pluviometric precipitation and photoperiod, in the intervals before the sample date. With the average number of eggs obtained per sample, we conducted variance analysis (F-test  $< 0.05$ ) and compared the means by the Scott & Knott test ( $P < 0.05$ ). *M. spectabilis* voucher specimens were deposited in the Embrapa Gado de Leite.

The viability of the *M. spectabilis* eggs was above 60% in the evaluations between September 2005 and May 2006, except in the second half of December 2005 and in February and April of 2006, when the percentage of unviable eggs was above 47%. In the second evaluation cycle (2006-2007), the viability ranged from 72% to 100%. MARQUES (1976), working with *Mahanarva posticata* (Stal, 1855), noted a mean viability of 95.8%, and BARBOSA (1986), with the same species, found the average of 51.6%. For *Mahanarva fimbriolata* (Stal, 1854), GARCIA *et al.* (2006) reported viability of around 80% for eggs always maintained under adequate conditions of relative humidity and at a temperature of  $27^\circ\text{C}$ . These findings indicate the interspecific and intraspecific variations on the embryonic development of the genus *Mahanarva*.

The total mean embryonic phase for all eggs (diapause or not) from September 2005 through the first half of March 2006, of the first evaluation cycle, varied from 15.79 to 28.24 days (Table 1), except in the first half of January, when 48% of the eggs were diapause (Fig. 1A), with the average being 65.92 days for this date. In the other samples, the embryonic period was significantly longer ( $P < 0.05$ ), varying from 94.80 to 139.04 days. All these eggs were considered diapause. Lower values were observed by CASTRO *et al.* (2005), who recorded durations of 50.7 and 48.2 days for diapause eggs of the Cercopidae *Zulia carbonaria* (Lallemand) in 1999 and 2000, respectively. The mean duration of the embryonic period for the non-diapause eggs was 18.61 days. This is close to the results reported by MARQUES (1976) for *M. posticata* (17.4 days) and by FREIRE *et al.* (1968) and GARCIA

*et al.* (2006) for *M. fimbriolata* (20 days). In contrast, the diapause eggs remained in the egg phase for an average of 154.50 days (Table 1). A higher average (196.2 days) was reported by MORALES (1993) for *Aeonolamia varia* (Fabricius, 1787).

In the second evaluation cycle (2006-2007) the mean duration of the non-diapause and diapause eggs was 18.76 and 138.41 days, respectively. The embryonic period for all the eggs taken together was under 27 days in the samples from September to the first half of November and also in the second half of January and February. This interval was

significantly shorter ( $P < 0.05$ ) than in the other samples. The duration and percentage of diapause eggs varied from 55.30 to 196.42 days and from 40% to 100%, respectively (Table 1), (Fig. 1B). For the non-diapause eggs the duration varied from 13.71 to 26.15 days.

From September to December 2005, the percentage of diapause eggs ranged from 0% to 12%. There was a significant increase (50%) in January 2006, but this increment was not maintained. Starting from the second half of March until May 2006, nearly all the eggs were diapause (Fig. 1 A).

Table 1 - Duration (days) of diapause eggs (DE) and non-diapause eggs (NDE) of *Mahanarva spectabilis* and viability (%), in the 2005-2006 and 2006-2007 evaluation periods. Juiz de Fora, Minas Gerais, Brazil.

2005-2006					
Collection date d/m/y)	Duration			Viability (%)	
	NDE	DE	Total		
20/9/2005	19.40	129.00	21.26	d	77
1/10/2005	17.98	-	17.98	d	67
18/10/2005	18.05	-	18.05	d	78
2/11/2005	15.79	-	15.79	d	64
15/11/2005	19.93	91.00	24.24	d	78
01/12/2005	16.07	-	16.07	d	73
14/12/2005	17.12	224.30	23.96	d	53
31/12/2005	17.06	279.50	24.35	d	87
14/1/2006	17.18	123.35	65.92	c	62
30/1/2006	18.89	201.25	28.24	d	75
15/2/2006	17.28	151.67	27.89	d	51
1/3/2006	17.17	-	17.17	d	63
17/3/2006	30.00	143.32	129.88	a	69
30/3/2006	-	139.04	139.04	a	66
17/4/2006	-	94.80	94.80	a	50
4/5/2006	-	122.30	122.30	a	91
Mean	18.61	154.50			
2006/2007					
Collection date d/m/y)	Duration			Viability (%)	
	NDE	DE	Total		
29/9/2006	16.41	-	16.41	f	98
13/10/2006	18.43	-	18.43	f	93
27/10/2006	15.66	-	15.66	f	100
9/11/2006	26.15	-	26.15	f	75
21/12/2006	19.03	163.04	75.76	e	89
8/1/2007	21.82	169.97	136.71	c	72
17/1/2007	13.71	121.00	14.98	f	82
7/2/2007	21.92	-	21.92	f	78
5/3/2007	19.00	158.93	140.17	c	96
22/3/2007	-	196.42	196.42	a	84
11/4/2007	-	161.63	161.63	b	84
30/4/2007	15.49	106.81	70.46	e	96
16/5/2007	-	112.56	112.56	d	69
27/5/2007	-	55.30	55.30	e	100
Mean	18.76	138.41			

Means within a column followed by the same letter are not significantly different ( $P < 0.05$ ).

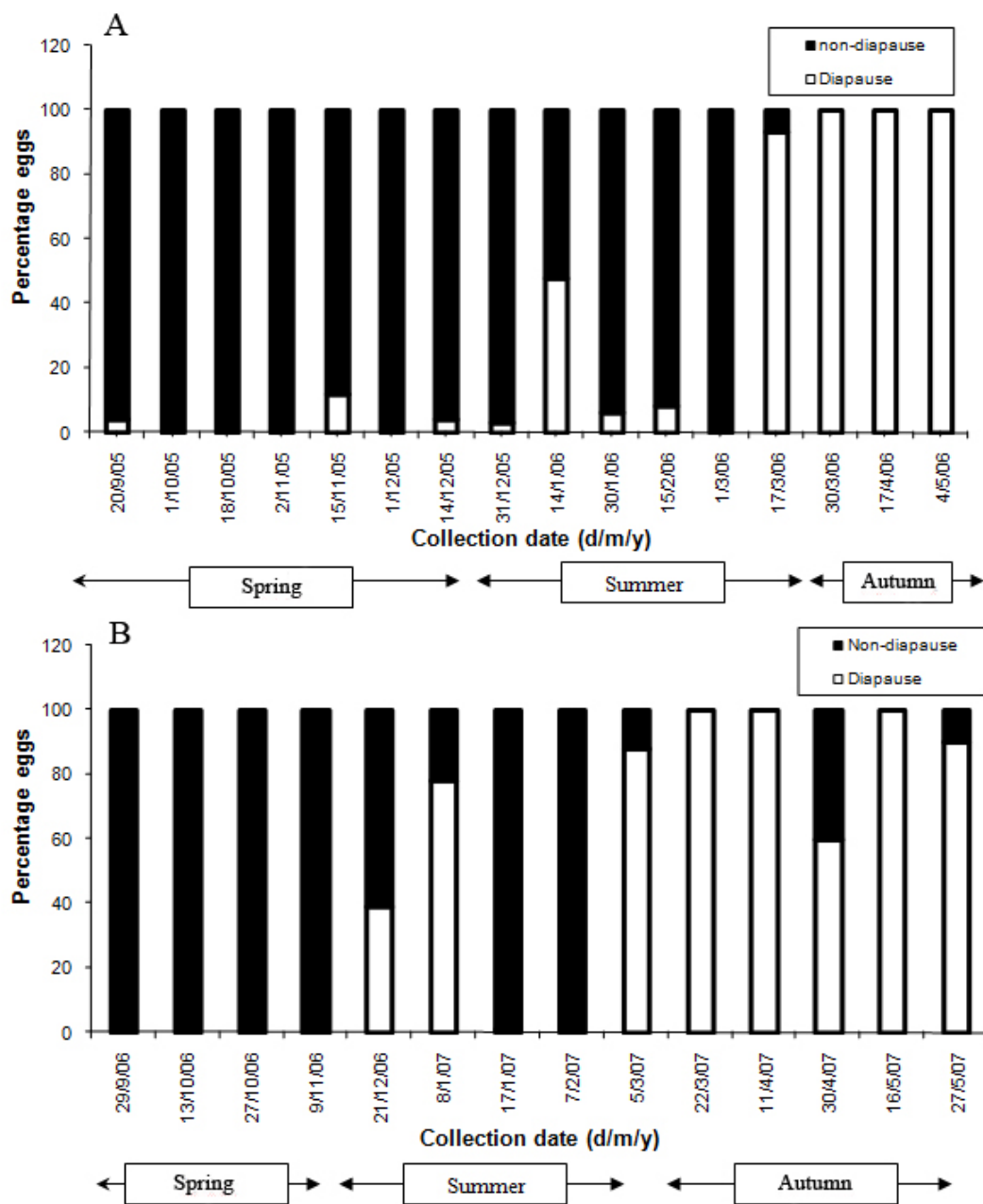


Fig.1 - Percentage of diapause and non-diapause eggs in the 2005-2006 (A) and 2006-2007 (B) evaluation periods. Juiz de Fora, Minas Gerais, Brazil.

In the second evaluation cycle (2006-2007), from September to November 2006 all the *M. spectabilis* eggs were non-diapause. In the second half of December and the first half of January the proportions of diapause eggs were 39% and 78%, respectively, decreasing to 1% and 0%, in the next two samples. However, starting in March, the majority of the eggs were diapause. According to RODRÍGUEZ *et al.* (2003), except for March and May, the proportion of diapause eggs of *Prosopis simulans* (Walker, 1858) was above 70%, in evaluations running from January to May 2001.

The highest number of diapause eggs occurred in the autumn, with 100% and 90% in the first and second evaluation cycles, respectively. In the other seasons, the percentage of diapause eggs was 3% and 0% in the spring and 26% and 40% in the summer of 2005-2006 and 2006-2007, respectively (Fig. 1 A and B). For *Z. carbonaria* (Lallemand), CASTRO *et al.* (2005) reported that from January 1999 to August 2000, only 0.4% of eggs were diapause, suggesting that this species has another defense mechanism to withstand the dry season.

We observed similar embryonic periods in the different evaluation cycles (Table 1), but this did not occur for the proportion and percentage of diapause eggs (Fig. 1 A and B). We found the occurrence of diapause eggs from December 2006 to the first half of January 2007, while in this same period of the first evaluation cycle the majority of eggs were not diapause. Also, the predominance of diapause eggs in the dry season occurred later in the 2005-2006 cycle. Starting from March the majority of the eggs were diapause in both evaluation cycles, and there was a significant correlation between the periods of the occurrence of diapause eggs between 2005-2006 and 2006-2007 ( $r = 0.83$ ,  $z = 2.34$ ,  $F = 0.0096$ ).

Despite the evidence of a defined period for the occurrence of diapause eggs in *M. spectabilis*, these were present in different proportions and were observed on 70% and 62% of the dates in the sample, in the first and second evaluation cycles, respectively. KOLLER; HONER (1993) found non-diapause and diapause eggs of *Deois flavopicta* (Stall, 1954), in all the collections in the period from 1984 to 1988. EVANS (1972) mentioned that the proportion of females that lay diapause eggs increases as the generations progress, with a limit of 94% occurrence.

Considering that the recently emerged females in this study were gathered and immediately placed under constant conditions of temperature, relative humidity and photoperiod suggests that the climate conditions to which these females were submitted during the egg laying period is not a stimulating factor for diapause eggs production. This hypothesis has also been proposed by LUKEFAHR et al. (1964), SAUNDERS (1987) and KOLLER; HONER (1993) who mentioned that diapause is induced during the nymph stage and controlled by the physiology of the female.

The correlation of the climatic factors to which the nymphs were submitted and the occurrence of diapause eggs was only significant in the 2005-2006 cycle. In this period there was a significant correlation with the minimum humidity inside the greenhouse ( $r = 0.66$ ,  $z = -2.62$  and  $F = 0.0044$ ), and for the photoperiod ( $r = 0.4792$ ,  $F = 0.048$ ) and average humidity ( $r = 0.6017$ ,  $F = 0.0186$ ) outside the greenhouse.

We observed that diapause was interrupted without environmental stimulus, since the eggs were kept under controlled conditions of humidity, temperature and photoperiod. This corroborates the findings of EVANS (1972) and BECK (1980).

EVANS (1972) and TAUBER et al. (1986) reported that the metabolic activity of diapause eggs decreases progressively until the embryos reach a dormant state. We observed that in diapause eggs the embryo developed until phase 2, according to the classification of PECK (2002), and then there was an interruption in development until after the occurrence of events capable of causing changes in metabolism, and

consequently emergence from the diapause state. This was also mentioned by RODRÍGUEZ et al. (2003).

Since the production of diapause eggs is an important strategy for the establishment of spittlebugs as forage pests, the ability to predict the occurrence of population peaks, the availability of specimens maintained in breeding facilities for experimental testing and the emergence of nymphs is a valuable tool in efforts for integrated management of these cercopids, providing input for decisions on the best control method.

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