ABSORCIÓN DE AGUA EN HUEVOS DE *Hylamorpha elegans* Burm. (COLEOPTERA:SCARABAEIDAE) EFECTO DE LA TEMPERATURA

WATER ABSORPTION BY THE EGG OF *Hylamorpha elegans* (Burm.) (COLEOPTERA : SCARABAEIDAE), EFFECT OF TEMPERATURE

Roberto Carrillo y Maria Eliana Fresard
Universidad Austral de Chile, Facultad de Ciencias Agrarias, rcarrill@uach.cl

ABSTRACT

Key words Water absorption, temperature, scarabaeid eggs, embryogenesis

During embryogenesis *Hylamorpha elegans* (Burm.) eggs absorb water from the soil, during embryogenesis increasing ca 2.5 in weight. Water absorption was related to the embryonic development, and therefore closely associated with temperature, it significantly was slower at 12ºC than at 20ºC. Eggs developed normally between 15 and 25ºC, but died at 30ºC. At 10ºC there was no development. Water absorption represents ca 30% of the total embryogenesis period. The distribution in southern Chile is mainly related to soil temperature conditions during the embryonic period, because soil humidity is appropriated in the area.

RESUMEN

Palabras claves Absorción de agua, temperatura, huevos de escarabaeidos, embriogénesis

La absorción de agua desde el suelo por huevos de *Hylamorpha elegans* (Burm.) en la embriogénesis incrementa su peso en alrededor de 2,5 veces. La absorción de agua esta relacionada con el desarrollo de la embriogénesis y por ello con la temperatura, a 12ºC fue mas lenta que a 20ºC. Los huevos se desarrollaron normalmente entre 15 y 25ºC, a temperaturas constantes de 30ºC no hubo sobrevivencia y a 10ºC, no se observó desarrollo. El período de absorción de agua por el huevo, representa alrededor del 30% de la duración total de la embriogénesis. El área de distribución sur de esta especie en Chile esta relacionada principalmente con las temperaturas del suelo durante la embriogénesis, debido a que la humedad del suelo es apropiada en el área.

Fecha recepción originales: 23 de febrero 2011

INTRODUCTION

Many soil-dwelling insects' eggs (e.g., acridids, scarabaeids, elaterids), absorb water from the soil during embryonic development (Laughlin,1953, Hinton, 1981). The water absorption process, is considered an adaptation by which water is conserved by the egg laying female (Whightman, 1973). The water uptake by the egg in many species of insects is dependent of embryonic development (Banks, 1950, Browning, 1965, Mori, 1986). Browning (1965), showed an inter-relationship between embryonic development and water absorption , because embryogenesis must reach a particular stage of development for the eggs to start absorbing water.

The length of the absorption period in relationship with the length of the embryogenic period is species specific, some authors suggest that species living in areas with high fluctuation in the water content of the soil, tend to show proportionally shortes periods of absorption.

When considering sacrabaeid embryogenesis, temperature and soil humidity are important. There is a temperature threshold for the embryogenic development this in some scarabaeid species living in pasture, which is around 15ºC, Roberts, 1963;Regniere et al 1981. Potter(1983), found that scarabaeid *C immaculate* eggs, present normal developing at a soil humidity 12.5% or higher, but dry up and died at lower soil moisture. Eggs are most sensitive to desiccation immediately after oviposition and during the period of water absorption (Tanaka, 2008).

The aims of this study were to determine in an important pest of berries, small grains and pastures, *H. elegans* the effect of different temperatures on egg development and to estimate the magnitude and the duration of the egg's water absorption during embryogenesis. This could help to explain in part its distribution and importance in southern Chile

MATERIAL AND METHDOS

The research was conducted at the Entomology Laboratory of the Facultad de Ciencias Agrarias of the Universidad Austral de Chile. Adut *H elegans* were collected with an entomological umbrella, by shaking in *Nothofagus obliqua* (Mirb.) Oerst branches at three different sites: Experimental Station Santa Rosa, Huachocopihue and Isla Teja Campus Adults were placed inside polyethylene bags, with *N obliqua* branches to allow adult feeding and carried to the laboratory in thermal insulated boxes. In order to obtain the eggs, special cages that measured 30x30x15 cm were used for manting and oviposition. Each cage had 10 holes 4mm diameter in its base and a fine net was used to cover each one. In the bottom of cage there was covered with 9 cm layer of soil series Valdivia. Unsexed adults were maintained at 20ºC and fed with *N obliqua* leaves. Each 24 h, soil was sieved and the eggs were collected.

To study the effect of temperature on embryogenesis, eggs (<24 h old) were placed in Petri dishes with10 cm diameter, on an aluminium plate (Alusaplast ®) and covered with 10 g of soil, 25% water by weight. Tested temperatures were 10, 15, 20, 25 and 30ºC Eggs were revised daily to determine egg hatching , for a sixty day period .

To study the morphological changes on eggs as well as variations on fresh and dry matter two experiments were carried out. In the first one the temperature was maintained constant at 20ºC, in the second one temperature was maintained at 12ºC for 12 days , and then changed to 20ºC.for the rest of the experiment. To evaluate morphological changes, every other day 10 eggs were removed at random with a fine brush and their lengths and widths were measured by using a stereoscopic microscope with a graduated micrometer objective To calculate volume changes the formula V = ab/6 a=length, b=width was used (Niikawa & Takeda, 1966.) .To determine fresh weight and dry matter 5 eggs were cleaned of soil particles and weighed individually, in an electronic balance. Afterwards they were exposed to 105ºC temperature in individual containers for 24h, and they were weighed at the end of this period.

The t student test(P 0.05)was used to determine statistical differences of fresh weigth of the eggs between days in both temperature regimens.

RESULTS

Physical changes of eggs during the embryogenesis.

Similar to other studied species of scarabaeids *H elegans* eggs absorb water from the soil. At 20ºC, the eggs did not absorb water from the soil during the first three days of their development. Water absorption occurs from the third day up to the seventh day, from here onwards the eggs no longer absorbed water. (Table 1) Water absorption produced drastic changes in the fresh weight, that were 2.5 times higher after water absorption. Volume, length and width also increased. However the length's increase rate was different and lower than that of the width's increase; as a result , the eggs' shape changed from ovoid to almost spherical. Considering a total embryogenic period of 15 days, water absorption occurred in a maximum of 4 days, equivalent to 36% of the total time of this process. Eggs showed a small loss of dry weight during the development probably due the expenditure of energy during normal activities of the developing embryo, this has been observed also in other insects those eggs absorb water (Eluwa,1970).

Eggs kept at 12ºC, showed no changes in the percentage of dry matter, length / width relationship and volume, which could indicate that water absorption from the soil was absent or very reduced at this soil temperature. (Table 2) When temperature was changed to 20ºC, there was rapid decrease in dry matter percentage and an increase in the eggs' fresh weight. The smaller period of time in the experiment 2 to produce physical changes on the eggs suggest that at 12ºC the process of embryogenesis had already started and that development threshold for this species was between 10 and 12ºC, because at 10ºC, there was no eggs' morphological changes..

Effect of temperature on egg development

Egg embryogenesis occurred between 15 and 25ºC, while at 30ºC all the eggs died At 10ºC , no morphological changes or eclosion occured in a 60 days observation period. The length of time between egg oviposition and egg eclosion, was done by the lineal equation y= 54.33-1.6x, in which temperature explained 99,4% of the developmental speed change.

DISCUSSION

Results confirm the water absorption by scarabaeid eggs is part of its embryogenic process. The water absorption is dependent of the embryogenic development of the egg if soil mois-

Table 1 Morphological and physical changes of *H. elegans eggs* during embryogenesis at 20ºC.
Tabla 1 Cambios físicos y morfológicos de huevos a *H. elegans* **durante la embriogénesis.**

Days from oviposition	Fresh weight mg)	Dry weight (mg)	Dry matter (%)	Length (mm)	Width (mm)	Length/ width ratio	Volume (mm_)
1	1.95±0,4a	0.985	50.52	1.78±0.14	1.41±0.12	1.262	1,31
3	2.08±0,5a	1.029	49.49	1.74±0.13	1.44±0.16	1.208	1.31
5	3.20±0,5a	1.035	32.33	1.83±0.15	1.63±0.13	1.122	1.56
7	4.87±0,7b	0.947	19.44	2.14±0.15	1.91±0.19	1.120	2.14
9	5.00±0,7b	1.039	20.78	2.15±0.14	1.95±0.12	1.102	2.19
11	4.75±0,6b	0.912	19.20	2.19±0.11	1.95±0.12	1.123b	2.23
13	4.94±0,3b	0.938	18.99	2.28±0.12	2.02±0.06	1.128b	2.41
15	4.84±0.9b	0.910	18.81	2.30±0.12	2.04±0.07	1.127b	2.46

Values of fresh weight followed with different letters are statistically different t-student (P 0.05)

Table 2. Effect of temperature change from 12ºC to 20ºC in *H. elegans* **eggs morphological and physical changes.**
Tabla 2. Efecto del cambio de la temperatura de incubación de 12 a 20ºC en huevos de *H. elegans.*

Days from oviposition	Fresh weight mg)	Dry weight (mg)	Dry matter (%)	Length (mm)	Width (mm)	Length/ width ratio	Volume (mm_)
1	0.7346	1.62±0.2a	45.35	1.61±0.08	1.31±0.07	1.239	1.103
3	0.8750	1.75±0.2a	50.00	1.62±0.14	1.29±0.07	1.255	1.094
5	0.5703	1.40±0.3a	40.74	1.59±0.26	1.24±0.20	1.282	1.031
7	0.9687	2.02±0.4b	47.95	1.73±0.07	1.41±0.11	1.226	1.277
9	0.9643	2.02±0.3b	47.74	1.76±0.18	1.36±0.08	1.294	1.253
11	0.9704	2.10±0.3b	46.21	1.75±0.11	1.41±0.09	1.241	1.291
13*	1,0018	2.26±0.3c	44.33	1.85±0.16	1.58±0.06	1.170	1.539
15	0.9397	3.28±0.3df	28.65	1.95±0.11	1.74±0.07	1.120	1.776
17	0.9252	3.99±0.4e	23.19	2.21±0.07	1.98±0.08	1.116	2.280
19	0.8799	3.96±0.7e	22.22	2.04±0.14	1.80±0.18	1.133	1.921
21	0.9310	4.19±0.7e	22.22	2.10±0.17	1.87±0.12	1.122	2.055
23	0.8000	4.21±0.5e	19.00	2.16±0.14	1.86±0.13	1.161	2.103
25	0.7840	3.94±0.4e	19.90	2.11±0.07	1.83±0.11	1.153	2.020
27	0.7373	3.92±0.3e	18.81	2.06±0.84	1.83±0.09	1.125	1.973
29	0.7181	3.83±0.4e	18.75	2.13±0.09	1.89±0.10	1.126	2.107

* Temperature change
* Values of fresh weight followed with different letters are statistically different t-student (P 0.05)

ture is over 3% water by weight in other scarabaeids (Potter, 1983), so eggs kept under the developmental threshold do not absorb water, in spite the soil moisture was adequate(12.5%) In the experiments when the temperature was 12ºC, there was a small change in the egg volume and width, due probably to the change in the egg water permeability, when temperature was modified to 20ºC, the water movement into the egg was accelerated. Comparing both tables it is possible to observe that the first experiment maintaining the temperature constant at 20ºC, the water movement into the egg was observed at the fifth day, when the egg was maintained previously at 12ºC and then modified at 20ºC , the effect was observed at the second day indicating that some embryonic development has occurred at 12ºC with the consequent effect on the chorion permeability . The chorion of eggs just oviposited presents an impermeable layer of fatty acids (Browning, 1965 Hinton, 1981), during the embryogenic development the serosa secretes the serosal cuticle under the viteline membrane, once the secretion of the serosal layer the fatty acid layer is lost or modified in different points, allowing the movement of water into the egg, until the hydrostatic pressure inside the egg prevents the water entrance (Potter, 1983).In acridids the serosal layer presents polarized permeability, avoiding through this way the lost of water from the egg. Potter (1983) , has showed in the scarabaeid *C. immaculate* eggs the development of the serosal layer during the embryogenesis and its enzymatic destruction and reabsorption before eclosion.

Has been indicated that sacarabaeids living in places with drought conditions, their eggs are able to absorb water from the soil rapidly and scarabaeids living under conditions rather humid are able to absorb water more slowly, results indicate that for this species the water absorption is rather slowly (30% of the embryonic development), in *C. immaculate* that lives in areas hot and dry water absorption lasted only

20% of the egg period. Results suggest that this species is well adapted to mild weather conditions, confirming its distribution in Chile from V to X Region , in areas with abundant soil moisture (natural or irrigated), plant cover and mild weather. Therefore summer temperature must be the main factor affecting its distribution in Chile, high soil temperatures limits its distribution to the north and lower soil temperatures during summer to the south of X Region of Chile.

LITERATURE CITED

BANKS, C 1950 The absorption of water by the eggs of *Corixa punctata* Illig.(Hemiptera:Corixidae) under experimental conditions. J. exp. Biol 26:131-136

BROWNING, T.O. 1965 Observations on the absorption of water, diapause and embryogenesis in the eggs of the cricket *Teleogryllus commodus* (Walker). J. exp Biol. 43:433-439

ELUWA, M.C. 1970. The biology of the West African bush-cricket, *Euclypoda acutipennis* Karsch (Orthoptera:Tettigonidae). Biol. J. Linn. Soc 2:1-39.

HINTON, H.E. 1981. Biology of insect eggs. Vol 1 Oxford, Pergamon. 473p.

LAUGHLIN, R. 1953. Absorption of water by the egg of the garden chafer, *Phyllopertha horticola* L. Nature 171:577

MORI, H. 1986. Water absorption by eggs and serosal specialization as clues to evolutionary trends in Heteroptera. Annals of the Entomological Society of America 79:456-459.

NIIKAWA, K.; TAKEDA, M. 1966 Water absorption by diapause and non diapause eggs in two Velarifictorus species (Orthoptera:Gryllidae). Applied Entomological Zoology 31:105-110

POTTER, D.A. 1983. Effect of soil moisture on oviposition water absorption, and survival of southern masked chafer (Coleoptera:Scarabaeidae) eggs. Environmental Entomology 12.1223-1227.

POTTER, D.A.; GORDON, F.C. 1984. Susceptibility of *Cyclocephala immaculate* (Coleoptera:Scarabaeidae) eggs and immatures to heat and drought in turf grass. Environmental Entomology 13:794-799

REGNIERE, J.; RAAB, R.L; STINNER, R.E.1981 *Popilia japonica*: the effect of soil moisture and texture on survival and development of eggs and first instar grubs. Environmental Entomology 10:654-660.

ROBERTS, R.J. 1963 Improved methods for obtaining and rearing first-instar *Cyclocephala immaculate* larvae for experimentation. Journal of Economic Entomology 56:538-541

TANAKA, S 2008. Uptake and loss of water in diapause and no diaspause eggs of crickets Physiological Entomology 11:343-351.