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Influence of some environmental factors on the biology of two terrestrial snails, *Monacha obstructa* (Pfeiffer) and *Eobania vermiculata* (Müller), under laboratory conditions

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Abstract

The terrestrial snail became economic serious pests attacking field corps, vegetables and orchards. This work was aimed to study the effect of some environmental factors on the biology of two species of land snails, *Monacha obstructa* and *Eobania vermiculata*. Results showed that, the soil types were markedly effective on fecundity and Ovipostion period of the land snails, *M. obstructa* and *E. vermiculata*. Clay and sandy loamy soils were favorable to such snail species, Whereas the general means of total clutches and eggs laid by one pair of both snails during the entire breeding season were (6.4 and 5.4 clutches and 211.4 and 271.6 eggs) and (6.36 and 6.36 clutches and 250.02 and 273.36 eggs) for *M. obstructa* and *E. vermiculata* respectively. While the highest values of general means of clutch size were (50.29 and 42.94) for sandy loamy soil while, the highest oviposition period were recorded in clay soil with values (69.4 and 68 days) for the two land snails respectively. Also, soil moisture was effected on number of per clutch. The highest egg number were recorded at percentage 80% soil moisture (116.8 eggs for *M.obstructa*) and (89.67 eggs for *E. vermiculata*) On the other hand, data revealed that the eggs of the two species of snails did not hatch at 30°C. On other hand, the highest values of egg hatch were found at 15°C with values 81and 76% for *M. obstructa* and *E, vermiculata* respectively. As the temperature increased, the egg hatching was significantly decreased. It clear that eggs of snails are sensitive to high temperature and embryos died at 30°C.

Keywords: land snails, *Eobania vermiculata*, *Monacha obstructa*, environmental factors.

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1. Introduction

The Gastropoda is one class among the Mollusca classes, which have successfully invaded land. The members of Gastropoda are of the most diverse groups of animals, both in shape and habits. Among gastopoda, land snails (Subclass: Pulmonata) are harmful pests to many crops worldwide (Godan, 1983). Singled out-that, the glassy clover snail, *M. obstructa* (Muller) which is the major species found infested wide range of crops at most localities of Egypt. Individuals of this species cause heavy damage to economically important crops (Abdel-Aal, 2007; El-Masry,1997; Mahrous *et al*., 2006; Lokma, 2007; Nakhla *et al*., 1995; Shahawy *et al*., 2008). These snails have active dispersal capabilities, which enable it to successfully expand its range to new habitats (El-Okda, 1981). Temperature degrees, the relative humidity, photoperiod, food supply and habitat characteristics are considered the most important factors influencing the occurrence and survival of land snails (Mahrous *et al*., 2002; Martin and Sommer, 2004; Metwally *et al*., 2002; Millar and Waite, 2002; Panigrahi and Raut, 1994; Ramzy, 2009). Temperature and relative humidity are the primary factors determining snail activity (Cook, 2001). Adequate knowledge of biology and ecology of pests should be acquired before application of management purpose to control these animals. The objective of these research studies is the effect of environmental factors on biological aspect of studies on *M. obstructa* and *E. vermiculata* under laboratory conditions.

2. Materials and methods

2.1 Collection of snails

The tested animals were collected from heavily infested fields cultivated with Egyptian clover at El-waste Village, El-Fath Locality, Assuit governorate, Egypt. Also, some snails were collected from ornamental plant nurseries and seedling production located in Assuit governorate, Egypt.

2.2 Snails culture design

Juvenile of the two snail species *M. obstructa* (6-8 mm) *E. vermiculata* (12-15 mm) in shell diameter were collected from heavily infested filed and ornamental plant nurseries and seedling production at the spring season in April month 2021, this is the time when the snails have the highest density, as the juvenile, newly hatched individuals are present beside mature individuals. The collected snails were placed in glass containers (50 × 30 cm) filled with moist clay soil and lettuce and cabbage leaves as food, until it had reached maturity.

2.3 Effect of soil mechanical properties on the fecundity of M. obstructa and E. vermiculata

Four types of soil (different mechanically and physically) were tested egg

production of *M. obstructa* and *E. vermiculata* different soil types of clay and sandy soils uses as follows:

- Clay soil (sand 52%, silt 36%, clay 12%).
- Sandy loamy soil (sand 82.5%, silt 7.9%, clay 9.6%).
- Sandy soil (sand 92.8%, silt 2.4%, clay 4.8%).
- Soil mix from clay soil and sandy soil (clay soil 50% + sandy soil 50%)

The clay soil was obtained from heavily infested fields El-wasta and Bossra district, El-Fath locality, Assiut governorate, Egypt. While sandy soil obtained from newly reclaimed from New Assiut Valley, Egypt. In September 2021, the snails were paired with individual's convergent size and placed in small plastic pots filled with 500-1000 g of soil according to the type of soil test to depth of about 5cm the soil moisture was kept at 75% of field capacity. Five replicates were used for each snail specie, disc of fresh lettuce leaves was added to each pots daily. The pots were kept clean by removing left –over food was introduced. The soil within each pot was examined for the snail clutches and eggs. Clutches were removed and the number of eggs in each clutch was counted.

2.4 Effect of soil moisture on numbers of clutches and eggs

Adult snails were collected in the same manner for describable each snail specie. Each snail was paired with another of similar shell diameter. The pairs were placed on small plastic pots containing 700 g of sleeved sterile soil. Four levels of soil moisture *i.e*., 20, 40, 60 and 80% of filed capacity were tested on sandy loamy soil, The soil was remoisten as required and discs of lettuce leaves were placed. The pots were kept after each week (for 12 weeks during November 2021 till last January 2022), number clutches and number egg of clutches were determined. Data were subjected to statistical analysis using the F test.

2.5 Effect of temperature on egg hatching and incubation period

Adult snails of *M. obstructa* and *E. vermiculata* were collected by hand from their host plant. The two species were kept in glass and plastic boxes (50 × 30 × 30 cm) contained moist soil to depth of (8:10 cm). Both species fed on fresh lettuce and cabbage leaves and remained was removed daily. The soil in each culture boxes was remoisted as required. Boxes were covered with muslin cloth and fixed with rubber bands to prevent snail from escaping. The soil within each culture box was searched daily for clutches of both species. The newly deposited clutches were singly removed, and ten eggs were placed in class petri dishes 10cm in diameter and incubated at 15, 20, 25 and 30^{o}C the bottom of petri dishes was equipped by disc of cotton and covered with moist paper toweling to maintain egg viability. Each treatment

was replicated ten times. Eggs were examined daily to calculate the incubation period and the newly hatched juveniles were counted and removed to avoid eggs cannibalism (Baur and Baur, 1993). Data were statistically analyzed using F test and standard error.

2.6 Effect of soil moisture on egg hatching and incubation period

Newly deposited eggs were collected as mentioned previously. Plastic pots 10-cm diameter were filled to depth of 2-cm wish sieved sterile clay soils. Four soil moisture levels *i.e*., 20, 40, 60 and 80 of field capacity were maintained during the experiment. Each treatment was replicated three times. Ten eggs were normally buried in the tins to the same depth According to (Awesu, 1988). The tins dishes were placed on bench in the laboratory and examined daily. Present of hatching and incubation period were calculated. The new hatching was removed to avoid eggs cannibalism. Air temperature and relative humidity were recorded during the experiment. Stander error and F test were used.

3. Results and Discussion

3.1 Effect of soil type on fecundity of land snails

Data in Table (1) cleared that, the soil type was in fluent on fecundity of the land snail *M. obstructa* under laboratory conditions during the period from November 2021 to March 2022. Results indicated that the total number of clutches and eggs laid by one pair of *M. obstructa* during the entire breeding season were affected by soil type. The highest numbers of clutches were deposited in soil clay followed by sandy loamy and mix clay + sand (1:1). The parallel values were 6.4 and 5.4 and 4.5 respectively, while the highest numbers of eggs were deposited in clay soil sandy loamy soil followed by Clay soil and mix soil clay + sand (1:1) with values 271.6, 211.4 and 124.6 respectively. On the other hand, in the case sand soil, the total number of clutches and eggs laid by *M. obstructa* were significantly decreased. Whereas, the total numbers of clutches and eggs laid by one pair were 2.4 and 109.6 respectively. The average of clutch size (number of eggs / clutch) was calculated for each treatment, the highest value (50.29 eggs/ clutch) was detected with sandy loamy soil, while the lowest one (27.6 eggs/ clutch) was found with soil mix clay + sand (1:1). Regarding the total numbers of clutches and eggs deposited by one pairs of *M. obstructa* in each month during the oviposition period, it is obvious that the highest values were laid during January (9.2 and 347.2) and December (6.2 and 227.4) followed by February (3.4 and 131.6) and March (0.2 and 11.0). However, *M. obstructa* wasn't deposited clutches during November. Concerning the effect of soil type on oviposition period, it was noticed that the longest period was detected in clay soil with means of 69.4

days followed by soil mix soil clay + sand (1:1) and sandy loamy, the means of oviposition periods were 55.2 and 51.8 days, respectively. However, the shortest oviposition period (17.9 days) was calculated in sandy soil.

Table (1): Effect of soil types on fecundity of the land snail *M. obstructa* under laboratory conditions. during the period from November 2021 to March 2022.

Soil type	Numbers of clutches and egg laid by one pair															Oviposition period	Total No. of clutch	Total No. of egg	Mean of clutch size
	November 21			December 21			January 22			February 22			March 22						
	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size				
Clay	0	0	0	2.8	84.6	27	2.6	91	35	1	35.8	35.8	0	0	0	69.4	6.4	211.4	32.96
Sandy loamy	0	0	0	1.8	104	57.8	2.2	107.6	48.9	1.2	49	40.83	0.2	11	55	51.8	5.4	271.6	50.29
Sandy	0	0	0	0	0	0	2.4	109.6	45.6	0	0	0	0	0	0	17.9	2.4	109.6	45.6
Mix clay + sand (1:1	0	0	0	1.6	38.8	24.25	2	39	19.5	1.2	46.8	39	0	0	0	55.2	4.5	124.6	27.6
Total	0	0	0	6.2	227.4	109.0	9.2	347.2	149	3.4	131.6	115.6	0.2	11	55	194.3	18.7	717.2	156.5

On the other hand, the effect of soil type on fecundity of the land snail *E. vermiculata* under laboratory conditions during the period from November 2021 to March 2022 illustrated in Table (2). From the result found that, the highest numbers of clutches were deposited in Clay soil and sandy loamy followed by Mix soil clay + sand (1:1). with values 6.36, 6.36 and 6.02 respectively, while the highest numbers of eggs were deposited in Clay soil followed by loamy soil and mix soil with values 273.36 and 250.02 and 195.35 respectively. While in the total number of clutches and eggs laid by one pair of *E. vermiculata* for the sand soil were 4.8 and 124.6 respectively. On the other hand, the highest average of clutch size was recorded in clay soil with value (42.94 eggs/clutch), while the lowest one was found with sandy soil with value (25.9 eggs/clutch). Also, results revealed that, the total numbers of clutches and eggs deposited by one pairs of *E. vermiculata* in each month during the oviposition period, it is obvious that the highest values were laid during December followed by January and February with numbers 9.62 (361.14), 9.36 (300.35), 3.88 (161.16) respectively. While wasn't deposited clutches during March. Also, it was found that, the longest oviposition period was detected with clay soil sandy loamy with means of 57.3 days, while the shortest oviposition period (48.7 days) was recorded with mix soil. Many authors in good line with report the result obtained herein. Kassab and Daoud (1964) showed that *H. vestalies* laid the eggs in clutch, each contained25 to 30egg or more in the soft soil. Godan (1983) indicate that mixture of damp loam and peat (1:1) was a suitable substrate for snails' egg laying. filed prefer the heavy soil is a habitat, while fine loose earth was chosen for egg laying Moreover Ismail (1997) investigated that the breeding season of *M. cartusiana* under laboratory condition started from November to the beginning march and the fecundity of such snail was obviously increase in clay soils as compared to sandy soil. Also, Mohamed (1999) found that the snails and slug

reared by using six types of soil: clay + sand + peat, clay + sand, clay soil, loose soil, sandy soil and tilled soil. mixed soil was (clay + sand + peat) more preferable soil for snail and slug fecundity, total clutch and egg were 9.3 clutches and 740.4 eggs in snail, while it was 3.5 clutches and 236.5 eggs in slug.

Table (2): Effect of soil types on fecundity of the land snail *E. vermiculata* under laboratory conditions during the period from November 2021 to March 2022.

Soil type	Numbers of clutches and egg laid by one pair															Oviposition period	Total No. of clutch	Total No. of egg	Mean of clutch size
	November 21			December 21			January 22			February 22			March 22						
	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size	No. of clutch	No. of egg	clutch size				
Clay	0.68	20.68	30.41	2.68	108.67	40.54	2	84.33	42.16	1	36.34	36.34	0	0	0	68	6.36	250.02	39.31
Sandy loamy	0	0	0	3	141	47.15	2.68	100.68	37.57	0.68	31.68	48	0	0	0	57.3	6.36	273.36	42.94
Sandy	0	0	0	1.6	38.8	24.25	2	39	19.5	1.2	46.8	39	0	0	0	52	4.8	124.6	25.9
Mix clay + sand (1:1	0	0	0	2.34	72.67	31.06	2.68	76.34	28.48	1	46.34	46.34	0	0	0	48.7	6.02	195.35	32.45
Total	0.68	20.68	30.41	9.62	361.14	143	9.36	300.35	127.71	3.88	161.16	169.68	0	0	0	226	23.54	843.33	140.6

3.3 Effect of soil moisture on snails' eggs production

Data illustrated in Table (3) showed that, of *M. obstructa* snail failed to lay eggs during November month at 20, 40 and 60% soil moisture while, numbers of clutches and eggs laid by snails-pair in clay soil were 0.2 clutches and 9 eggs at 80% soil moisture level. When soil moistures were 40, 60 and 80% the numbers of clutches and eggs were 1.8 (27.8), 2.2 (60) and 2 and116.8 during December. While January recorded (1 and 18.4), (1and 19) and (3.2 and114) respectively. While in *E. vermiculata*, the snail failed to lay eggs at soil moisture levels 20% but when soil moisture increased to 40 and 60%, numbers of clutches and eggs laid by snails-pair in clay soil were (0.67 and 12.67), (2.333 and 28), (1.33 and 19.67), at 40% soil moisture (0.33 and 29.67), (2.33 and 47) and (1.67 and 42) at 60% of soil moisture during November, December and January, respectively. As the soil moisture increased to 80% of field capacity numbers of clutches and eggs laid by one pair of snails were increased, the numbers of clutches and eggs were (1.33 and 41.33), (1.67 and 74) and (2.33 and 89.67) during November, December and January months, respectively. Generally, it is obvious that as the soil moisture increased, numbers of clutches and eggs laid by *M. obstructa* and *E. vermiculata* were markedly increased in soils. The obtained results agreement with that obtain by Carrick (1942) showed that *Deroceras agrestis* preferred soil with high water content (50: 70% saturation) for egg laying. Godan (1983) indicated that health young Gastropoda developed only when the water of the soil was 60 to 85%. In dry or extremely wet soil, they died during embryonic development or else the young animals were too weak to hatch out. Also, Ali and Suleman (1992) reported that *M. obstructa* laid its eggs in humid shady places either in shallow or under the fallen leaves, logs stones etc. Moreover, Ismail (1997) showed that *M. cartusiana* failed to lay eggs at soil moisture level 25 and 50% of field capacity in both clay and sandy

soils. As the soil moisture increase to 75 and 100% of field capacity, numbers of clutches and eggs were markedly increased in both clay and sandy soils.

Table (3): Effect of soil moisture on egg production for *M. obstructa and E. vermiculata* in clay soil during the period from November 2021 to January 2022 under laboratory conditions in clay soil.

Moisture levels	*M. obstructa*						*E. vermiculata*					
	November 21		December 21		January 22		November 21		December 21		January 22	
	No. of clutch	No. of egg	No. of clutch	No. of egg	No. of clutch	No. of egg	No. of clutch	No. of egg	No. of clutch	No. of egg	No. of clutch	No. of egg
20	0	0	1.2	8.8	0	0	0	0	0	0	0	0
40	0	0	1.8	27.8	1	18.4	0. 67	12. 67	2.333	28	1.33	19.67
60	0	0	2.2	60	1	19	0.33	29.67	2.33	47	1.67	42
80	0.2	9	2	116.8	3.2	114	1.33	41.33	1.67	74	2.33	89.67
Mean	0.05	2.25	1.8	53.35	1.3	37.85	0.56	23.67	1.60	37.30	1.33	37.83

Generally, it is necessary to mention here that eggs of terrestrial pulmonates are in great danger of desiccation, since they contain more than 85% water (Bayne, 1968) and it must be borne in mind that measures undertaken to increase soil water content create more favorable conditions for the reproduction of land snails.

3.3 Effect of soil moisture and temperature degrees on incubation period and egg hatching of snails

The effect of four levels of soil moisture (20, 40, 60 and 80%) on percentage of egg hatching, and incubation period of *M. obstructa* was studied under laboratory conditions tabulated in Table (4). Data revealed that eggs failed to hatch when soil moisture was 20% of field capacity, while increase soil moisture to 40, 60 and 80% was significantly increased eggs hatching to 63.4, 73.3 and 93.4 respectively. While the mean of incubation period was 18.4, 16.8 and 14.5 days at 40, 60 and 80% soil moisture, respectively. The hatching period was 2.4 days at soil moisture 40 and 60% wile when soil moisture was 80% the hatching period was 2 days. On the other hand, the effect of temperature on egg hatching and incubation period of *M. obstructa* was studied in Petri dishes each contained 10 eggs placed on moist paper toweling and incubated at 15, 20, 25 and 30°C. Results showed that, percentages of egg hatching were 81, 73 and 57% at 15, 20 and 25°C, respectively, while eggs did not hatch at 30°C. The highest values of egg hatch were found at 15 °C. As the temperature increased, the egg hatching was significantly decreased. None hatched eggs at 30°C. It seems that eggs of *M. obstructa* are sensitive to high temperature and embryos died at 30°C **.** While the effect of temperature on the incubation period of *M. obstructa*, it was found that the incubation period was correlated with temperature. The mean time needed for the eggs to hatch (incubation period) was 16.6, 15.5 and 9.54 days at 15, 20 and 25°C, respectively.

Table (4): Effect of clay soil moisture and temperature degrees on egg hatching and incubation period of *M. obstructa* under laboratory conditions.

Treatment	Egg hatching (hatchability %)	Incubation period (days)	Hatching period (day)
Soil moisture level (%)			
20	0	0	0
40	63.4±0.58	18.4±4.9	2.4±0.57
60	73.3±0.75	16.8±4.42	2.4±0.57
80	93.4±0.845	14.5±0.75	2
L.S.D.	16.30	6.15	0.76
Temperature (°C)			
15	81±2.76	16.6±2.84	1.9±0.31
20	73±2.04	15.5±0.72	2
25	57±1.33	9.54±3.71	2.4±0.51
30	0	0	0
L.S.D.	9.92	1.61	0.27

On the other hand, the hatching period (time elapsed between the date on which the first egg hatched and the date on which the last egg hatched) was affected by temperatures where, the hatching period was 1.9, 2 and 2.4 days at 15, 20 and 25°C, respectively. Egg hatching and incubation period were significantly decreased. At 30°C, none of the tested eggs were hatched. Concerning to the effect of soil moisture and temperature on egg hatching and incubation period of *E. vermiculata* under laboratory conditions illustrated in Table (5). Data showed that the highest egg hatching percentages were 66.7% at 80% soil moisture followed by 60 and 40% soil moisture with values 56.6 and 43.4% egg hatching percentages, while eggs failed to hatch when soil moisture was 20% of field capacity. However, the mean of incubation period (The mean time needed for the eggs to hatch) of *E. vermiculata* at 40, 60 and 80% soil moisture were 23, 22.5 and 18.5 days respectively. Also, the time elapsed between the date on which the first egg hatched and the date on which the last egg hatched (the hatching period) was 2 days at two level of soil moisture 60 and 80%.

Table (5): Effect of clay soil moisture and temperature degrees on egg hatching and incubation period of *E. vermiculata* under laboratory conditions.

Treatment	Egg hatching (hatchability %)	Incubation period (days)	Hatching period (day)
Soil moisture level (%)			
20	0	0	0
40	43.4±0.72	23±5.19	2.7±0.57
60	56.6±1.08	22.5±0.75	2
80	66.7±2.08	18.5±0.75	2
L.S.D.	13.31	4.89	0.45
Temperature (°C)			
15	76±1.70	19.5±0.72	2
20	58±1.93	15.8±3.81	1.8±0.45
25	44±1.36	12.2±2.90	1.8±0.44
30	0	0	0
L.S.D.	21.51	3.20	0.42

On the other hand, the effect of temperature on the percent of egg hatching of *E. vermiculata*, it was obvious that the egg hatching

percentages were 76, 58 and 44% at 15, 20 and 25°C, respectively, while eggs did not hatch at 30°C. The highest incubation period of *E. vermiculata* was 19.5 days at 15°C followed by 20 and 25°C temperatures with values 15.8 and 12.2 days respectively, the hatching periods were 1.8 days at 20 and 25°C, while when temperatures was 15°C the hatching period was 2 days. The obtained results are in harmony with those reported by many authors measured egg hatching and incubation period. Mohamed Ghada (2004) who found that 20°C gave the best hatching rate for *Eobania vermiculata*, *M. cartusiana* and *M. obstructa* (Mohamed, 1999, Al-Akra, 2001). Also, Baur and Baur (1993) reported that the hatching optima for *Cepaea nemoralis* were at 22 and 25°C. Our finding indicated that 20°C temperature and soil moisture 50% of this experiment accelerate development rate and reduced the duration of the developmental stages is similar to findings of Le Clave (1989) who reported that the length of incubation period for *Helix aspersa* was negatively correlated with temperature.

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