

EFFECTS OF TEMPERATURE AND PHOTOPERIOD ON GROWTH AND
REPRODUCTION OF THE LAND SNAIL *HELIX APERTA* BORN
(GASTROPODA, PULMONATA)

Saida BENBELLIL-TAFOUGHALT^{1*}, Mohamed SAHNOUNE¹,
Annette DE VAUFLEURY² & Aissa MOALI¹

RÉSUMÉ.— *Effets de la température et de la photopériode sur la croissance et la reproduction de l'escargot Petit-gris Helix aperta Born* (Gastropoda, Pulmonata).— La croissance corporelle et l'activité reproductrice de *Helix aperta* ont été étudiées en laboratoire de l'éclosion à la maturité, à 15°C et 20°C durant 40 semaines et selon deux photopériodes: 16 h de jour / 8 h de nuit et 8 h de jour / 16 h de nuit. Seule la température, pas la photopériode, a eu des effets significatifs sur la croissance corporelle; les escargots élevés à 15°C se sont développés plus lentement et ont atteint un poids adulte plus petit que ceux élevés à 20°C. En revanche, les résultats ont montré une certaine interaction entre photopériode et température dans la reproduction (durée de l'activité reproductrice, nombre d'accouplements et de pontes) ainsi qu'un effet prédominant de la photopériode. Le nombre d'œufs par ponte est apparu lié au poids des escargots. Les meilleures conditions de croissance et de reproduction de *Helix aperta* furent la combinaison d'une température de 20°C et d'une photopériode de 16 h de jour / 8 h de nuit.

SUMMARY.— Body growth and reproductive activity of *Helix aperta* were studied in laboratory conditions over 40 weeks, from hatching to maturity at 15°C and 20°C, and under two photoperiods: 16h-Light / 8h-Dark and 8h-Light/16h-Dark. Only temperature, but not photoperiod, had significant effects on body growth; snails reared at 15°C grew slower, took more time to develop and reached a smaller adult body weight than those raised at 20°C. In contrast, the results showed some interaction between photoperiod and temperature on reproduction (length of reproductive activity, numbers of matings and layings) and revealed a predominant effect of photoperiod. Number of eggs per clutch was linked to body weight of snails. The best conditions for growth and reproduction of *Helix aperta* snails were the combination of a temperature of 20°C and a long-day photoperiod (16h L/8h D).

The influence of temperature on development and growth is particularly important in determining an organism's life strategy since growth rate and size at maturity are key traits in life-history evolution (Roff, 1992; Stearns, 1992; Charnov, 1993). In ectotherms, growth, development rates and body size at adult age are function of environmental temperature. Atkinson

¹ Laboratory of Ecology and Environment, Faculty of Nature and Life Sciences, University A. Mira of Bejaia, 06000 Bejaia, Algeria. E-mails: tafoughalt19@yahoo.fr; sahnounemohamed@yahoo.fr; rubiette_am@hotmail.com

* Corresponding author. Phone number: (213) 777.00.80.66, Fax: (213) 34.21.47.62

² Department of Environmental Biology, EA 3184 USC INRA, University of Franche-Comté, Place Leclerc. F-25030 Besançon Cedex. E-mail: annette.devaufleury@univ-fcomte.fr

(1994 & 1995) reported that, in most ectothermic species studied, size at maturity decreases with increasing growth temperature. A number of models have been proposed to explain these changes in ectotherms body size in response to thermal environment; both nonadaptive and adaptive explanations have been offered. Nonadaptive theories, which suggest that temperature-size relationship observed in ectotherms results from unequal effects of temperature on growth and differentiation, have been used to explain the larger body size at lower temperatures (von Bertalanffy, 1960; van der Have & de Jong, 1996; Van Voorhies, 1996; Arendt, 2000). Adaptive explanations for the temperature-size rule use the costs and benefits of particular life histories, given thermal constraints on production or thermal effects on survival, to describe why, in most ectotherms, natural selection favours smaller size and faster development at higher temperature (Atkinson, 1994; Berrigan & Charnov, 1994).

The optimal reaction norm for size at maturity is the product of growth rate and the duration of the growth period (Atkinson, 1994; Blanckenhorn, 1998; Stern, 2001; Davidowitz *et al.*, 2004). However, development and growth have different temperature/rate trajectories (Smith-Gill & Berven, 1979; van der Have & de Jong, 1996; Jarošík *et al.*, 2004); an individual with a particular growth rate may become either large or small, depending on the length of growth period: a slow-growing individual grows to larger body size given enough time, and a rapidly-growing individual may be small if its growth period is shortened (*e.g.* Blanckenhorn, 1999).

The effects of temperature and artificial photoperiods on body growth of helcid snails have been widely studied; it has been demonstrated that growth is strongly regulated by photoperiod in *Cornu aspersum aspersa* (Gomot *et al.*, 1982; Laurent *et al.*, 1984; Gomot & Deray, 1987). However, in *Cornu aspersum maxima*, Deray & Laurent (1987) showed that the inhibitory effect of short-day photoperiod on body growth seems to be less pronounced than in *Cornu aspersum aspersa*. But growth in this species is more sensitive to environmental temperature (Lecompte, 1995; Jess & Marks, 1998).

Many authors took an interest in the study of the effects of photoperiod and temperature on the reproductive activity of terrestrial snails, with a particular reference to *Cornu aspersum aspersa* (Stephens & Stephens, 1966; Charrier, 1980; Bailey, 1981; Enée *et al.*, 1982; Aupinel & Daguzan, 1989) and *Cornu aspersum maxima* (Bonnefoy-Claudet & Deray, 1987; Gomot *et al.*, 1990). On the whole, all these works demonstrate the importance of photoperiod on the control of reproduction.

Gomot *et al.* (1989), by using *Cornu aspersum aspersa* snails that have just entered hibernation and submitting them to four different combinations of photoperiod and temperature, demonstrated the existence of an interaction between temperature and photoperiod on the reproduction of *Cornu aspersum* and a photoperiod-dependent control of the neuroendocrine centres responsible for ovulation and egg-laying. The authors observed that combination of long-days (18L:6D) and a temperature of 20°C is the most favourable condition for egg-laying as well as effective functioning of the ovotestis and the albumen gland. In contrast, they noted that under short day photoperiod (8L:16D), egg-laying is completely inhibited at 15°C and partially at 20°C; they observed differentiation of gametes in the ovotestis of snails, but mature oocytes were never released and thus degenerated.

No previous studies have been carried out on the species *Helix aperta* Born (1778) (= *Cantareus apertus* Born, 1778), an edible snail whose biological cycle is poorly documented. It is a species of Mediterranean origin, found in the south of France, Italy, Turkey, Cyprus and North Africa (Kerney & Cameron, 1979; Schütt, 2001). It is of brown greenish colour and has a mean size of 25mm. Its meat is highly prized. Sometimes known as the “burrowing snail” it is found above ground only by rainy weather; in dry conditions, it burrows 3 to 6 inches (7-15cm) into the ground and estivates in a thick whitish opercule until rain softens the soil. This snail is frequent in humid to sub-humid coastal habitats where it prefers vine and olive orchards. In Algeria, it is frequently found throughout the coastal part of the country, especially in the region of Kabylia. This species is heavily exploited for gastronomy, so its density is more and more declining in North Africa.

In this paper, we aim to examine, under laboratory conditions, the effects of temperature and artificial photoperiods on growth and subsequent reproduction of snails of this understudied species *Helix aperta* and so, determine the optimal combination of the two factors for a best productivity.

METHODS

100 adult snails identified as belonging to *Helix aperta* were collected from Bakaro (22 km east of Bejaia, Kabylia, Northern Algeria) on the second week of March 2005. The snails were reared for three generations under controlled conditions of 20°C temperature and 16-h light photoperiod.

All the rearing containers employed in this work were of transparent polystyrene, a volume of 3600 cm³ and perforated on their lids with 10 holes of 1mm diameter to provide ventilation. Hatchlings of the fourth generation were used in the experiments. These hatchlings were produced from reproductively mature snails divided into small groups of four individuals per container under optimal culture conditions (photoperiod: 16L:8D, temperature: 20°C, relative humidity: 90-95 %) following those used to raise another helioid snail, *Cornu aspersum*, an ubiquitous species living beside *Helix aperta* in Algeria (Daguzan, 1981; Enée *et al.*, 1982; Laurent *et al.*, 1984; Gomot & Deray, 1987). The experimental animals were fed with the commercial product “*Helixal*” (Etablissements Chays, France), a special meal for snails with two formulae: one for very young snails “first age” (nursery: 1 to 4 weeks) and another, “second age”, for growth and reproduction. The main components of these foods are reported in Gomot-de Vaufléury (2000).

To examine the effects of temperature and photoperiod on growth and reproduction of *Helix aperta*, four groups of 40 newly-hatched snails were weighed and randomly placed in transparent polystyrene containers described above. The density in the rearing containers was 500 snails/m² during the first 4 weeks and then 100 snails/m² for the following weeks of growth. Four replicates were used per sample and the boxes location in the rearing room was changed every day.

Equal quantities (50 g) of food were provided in each container on Petri dishes of 9 cm diameter deposited in the bottom of the containers. To maintain the humidity, wet absorbent paper was placed on the floor of the containers. Three times a week, at the same time, the containers were cleaned, the food renewed and the absorbent paper changed.

Two photoperiod lengths were selected: one of long-days (16h Light: 8h Dark, noted 16L:8D) and the other of short-days (8h Light: 16h Dark, noted 8L:16D). The lighting was provided using neon of daylight type, with an intensity of 50–100 lux. The two temperatures tested were 15°C and 20°C, in constant temperature rooms. Each of the four groups of snails was submitted to a different combination of temperature and photoperiod: Group A (20°C, 16L:8D), Group B (20°C, 8L:16D), Group C (15°C, 16L:8D) and Group D (15°C, 8L:16D). All groups were reared at a relative humidity of 90 to 95 %. Every week, all snails were weighed individually (± 0.01 g) and the mean snail weights in each group were estimated until the onset of matings, indicating the start of the reproduction phase (in *Helix aperta* species, the cessation of growth is not marked by shell-lip formation as in *Cornu aspersum*).

To study the effects of photoperiod and temperature on reproduction, the snails were randomly placed in groups of four individuals per container approximating a relative stocking density of 50 snails/m². During the reproduction period, all the snails were marked by adhesive sticks to be recognized. Pots of 10 cm of diameter and 8 cm of height filled with wet light soil were placed in each container for egg-laying. During all the time of the experiment, to follow the matings and egg-layings, two observations a day were made, the first early in the morning, the second in the afternoon. Each clutch was identified by its parentage, date of laying, number of eggs, date of hatching and number of young hatches. Twice a week, the eggs were collected using a small spoon and their numbers recorded. 30 % randomly chosen eggs were individually weighed (± 0.001 g). All the eggs were then incubated at 20°C in Petri dishes (9 cm in diameter and 1.5 cm high). To maintain the humidity during all the time of incubation, a layer of wet absorbent paper was placed in the bottom of the Petri dishes. For aeration of the eggs, the lids of the Petri dishes were perforated with 10 small holes (1mm in diameter). After hatching, the number of young hatches was counted. Each week, the number of dead animals in each group was monitored during all the time of the experiment. The length of reproduction period was estimated by recording the beginning of matings and the end of egg-laying.

The data were subjected to statistical analysis using Minitab 13.31 software (Minitab Inc., 2000) and Statistica 5.5 (StatSoft Inc., 1999). The mean weights of snails of the different groups and the mean number of eggs per clutch were expressed as means \pm standard deviation ($M \pm SD$) or standard error ($M \pm 2SE$). The differences between the mean weights of snails in the different groups, mean weights of snails within each group at the start and the end of reproduction period, mean numbers of eggs per clutch and mean egg weights per clutch were tested using Anova, Student (t) or Lsd tests. The comparison of numbers of matings and numbers of layings between the different groups and the relationships between numbers of matings and layings was done using χ^2 test. Comparison between proportions test was used to compare the different percentages estimated in the results. Pearson’s correlations were determined between wet weight of snails and the following traits: numbers of eggs per clutch and egg wet weights. Spearman’s rank coefficient was used to estimate the correlations of wet weight of snails with numbers of matings and layings.

RESULTS

The data showed that juvenile growth rates increased with increasing temperature regardless of photoperiod length (Fig. 1), but this juvenile growth, beyond the eighth week, was far faster ($P < 0.001$) at 20°C (Fig. 1, Groups A & B) than at 15°C (Fig. 1, Groups C & D). At 15°C, the long and the short photoperiods resulted in nearly confounded curves (Fig. 1, Groups C & D) whereas at 20°C, growth rate under long-day photoperiod was faster ($P < 0.001$) than under short-day photoperiod (Fig. 1, Groups A & B). At the end of growth period (23rd week after hatching), snails were found to have significantly higher wet weights at 20°C than at 15°C ($P < 0.001$); however, no significant difference ($P = 0.12$) was revealed between values obtained under long and short-day photoperiods. Long-day photoperiod seemed to have a significant positive effect on final snail weights as well as on growth rate only at 20°C but not at 15°C (Fig. 1 and Tab. I). At 15°C, snails exposed to long-days were in average only 3.87 % heavier than those reared under short-day photoperiod ($P > 0.5$), whereas at 20°C, the weights revealed to be respectively 10.26 % greater ($P < 0.05$). Under both photoperiods, the development time of snails was only of 21 weeks at 20°C but of 23 weeks at 15°C. In fact, snails raised at 20°C reached maturity and started to mate after 21 weeks of growth, while at 15°C, this was observed 2 weeks later. The end of development time coincided with the cessation of growth. Moreover, after reproduction, the mean weight of snails decreased significantly ($P < 0.001$) in Group A (20°C, long-days) and Group D (15°C, short-days). However, in Groups B (20°C, short-days) and C (15°C, short-day), the mean wet weight of snails did not change much over the reproduction period ($P > 0.05$) (Tab. II, lines 1 & 2).

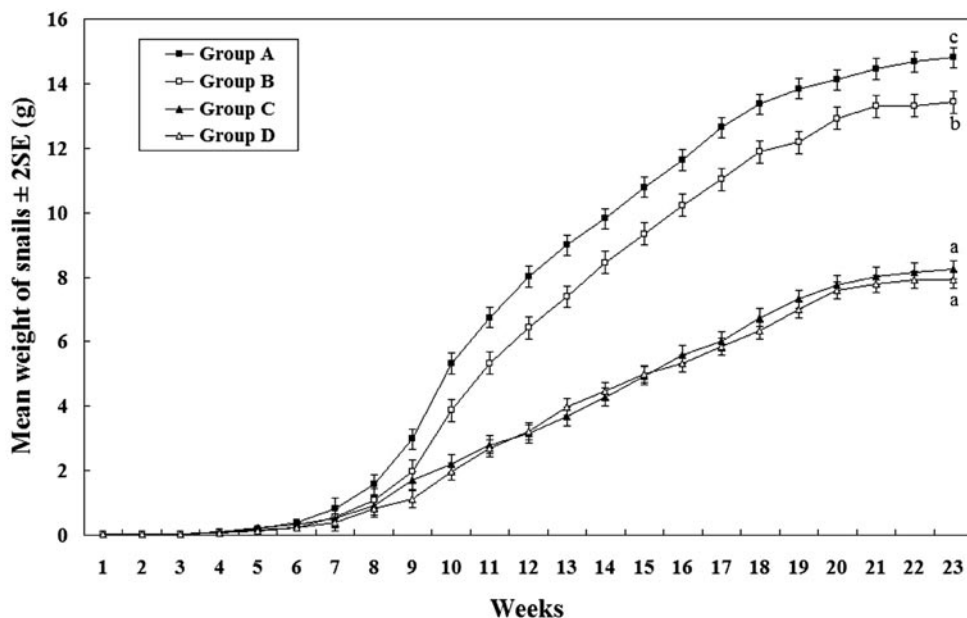


Figure 1.— Effect of four combinations of temperature (°C) and photoperiod (h) on the growth of *Helix aperta*. (Group A: 20°C, 16h-Light:8h-Dark; Group B: 20°C, 8h-Light:16h-Dark; Group C: 15°C, 16h-Light:8h-Dark; Group D: 15°C, 8h-Light:16h-Dark). The curves marked with different small letters (a-c) are significantly different ($P < 0.01$).

TABLE I

Initial and final wet weights and mortality rates of *Helix aperta* snails reared over 23 weeks under four combinations of temperature and photoperiod. The snails groups values marked with different small letters (a-c) are significantly different ($P < 0.05$)

Experimental design		Mean wet weight of snails \pm SD (g)		Mortality	
Experimental groups	Temperature ($^{\circ}$ C)/ Photoperiod ¹ (h)	Start of experiment	After 23 weeks	Final number / initial number of snails	Rates of mortality (%)
Group A	20 / (L 16:D 8)	0.024 \pm 0.001a	14.81 \pm 0.97c	38/40a	5a
Group B	20 / (L 8:D 16)	0.023 \pm 0.002a	13.43 \pm 1.07b	39/40a	2.5a
Group C	15 / (L 16:D 8)	0.023 \pm 0.001a	8.25 \pm 0.87a	38/40a	5a
Group D	15 / (L 8:D 16)	0.023 \pm 0.003a	7.93 \pm 0.81a	38/40a	5a

¹ L: Light (h); D: Dark (h).

TABLE II

Reproductive traits, rates of mortality and mean weights of *Helix aperta* reared under four combinations of temperature ($^{\circ}$ C) and photoperiod (h). The snails groups values marked with different small letters (a-c) are significantly different ($P < 0.05$); within each snail group, mean weights (descriptive traits 1 and 2) marked with different capital letters (A-B) are significantly different ($P < 0.05$)

Descriptive traits	Group A	Group B	Group C	Group D
	(20 $^{\circ}$ C/ L16:D8) ¹	(20 $^{\circ}$ C/ L8:D16) ¹	(15 $^{\circ}$ C/ L16:D8) ¹	(15 $^{\circ}$ C/ L8:D16) ¹
1. Mean weights \pm SD of snails at start of reproduction period (g)	14.81 \pm 0.97cB	13,43 \pm 1.07bA	8.25 \pm 0.87aA	7.93 \pm 0.81aB
2. Mean weights \pm SD of snails at the end of reproduction period (g)	13.46 \pm 0.98dA	12.35 \pm 1.01cA	8.12 \pm 0.52bA	7.01 \pm 1.02aA
3. Sample size at start of reproduction period	38a	39a	38a	38a
4. Sample size at the end of reproduction period	29a	35a	35a	30a
5. Percent of snails that mated	100% <i>c</i>	64% <i>b</i>	100% <i>c</i>	39,47% <i>a</i>
6. Total number of matings	132d	34b	98c	20a
7. Number of matings per snail	3.47c	0.87a	2.58b	0.53a
8. Percent of snails that laid eggs	100% <i>d</i>	53.85% <i>b</i>	84.21% <i>c</i>	13.15% <i>a</i>
9. Total number of clutches	72c	21b	56c	05a
10. Number of clutches per snail	1.89d	0.54b	1.47c	0.13a
11. Mean number of eggs \pm SD per clutch	340.4 \pm 30c	306.0 \pm 43b	188.9 \pm 31a	188.8 \pm 07a
12. Mean egg weight (mg)	26.6 \pm 1.12a	26.49 \pm 0.32a	27.01 \pm 0.21a	26.21 \pm 0.03a
13. % of hatching (= number of eggs that gave juveniles)	86% <i>a</i>	87% <i>a</i>	87% <i>a</i>	85% <i>a</i>
14. Time between mating and egg-laying (days)	20	20	20	20
15. Time of incubation (days)	13	13	13	13
16. Rate of mortality during reproduction period (%)	24% <i>a</i>	10% <i>a</i>	8% <i>a</i>	21% <i>a</i>
17. Range of reproduction activity (weeks)	20	9	11	5
18. Range of mating activity (weeks)	18	5	10	4
19. Range of laying activity (weeks)	17	6	8	2

¹ L: Light (h); D: Dark (h).

Regarding reproduction activity, the data showed that, at the end of the experiment, the total numbers of matings per group were very variable (Tab. II, line 6) and it seemed to be under long-day photoperiod that snails were inclined to mate more frequently ($\chi^2 = 106.54$; $df = 5$; $P < 0.001$) and had significantly more frequent egg-layings ($\chi^2 = 88.68$; $df = 2$; $P < 0.001$) (Fig. 2). An interaction between photoperiod and temperature was obvious on both matings and egg-layings (Fig. 2). The results revealed clearly that it was at 20 $^{\circ}$ C in long-days (Figs. 2-4, Group A; Tab. II) that snails had the highest reproduction activity which lasted 20 weeks, with 3.47 matings and 1.89 clutch per individual as the highest frequencies observed in this experiment (Tab. II, lines 7 & 10). Snails reared at 15 $^{\circ}$ C in short-days (Group D) showed the smallest numbers of matings (0.53) and layings (0.13) per snail and stopped copulation and egg-laying

after 4 and 2 weeks respectively (Tab. II, lines 18 & 19; Figs. 2 & 3). Snails of Group C reared at 15°C under long-day photoperiod mated and laid more frequently than those of Group B raised at 15°C under short-day photoperiod with 2.58 vs 0.87 mating per snail and 1.47 vs 0.54 laying per snail respectively (Tab. II, lines 7 & 10). Mating and egg-laying activities lasted respectively 10 and 8 weeks in Group C against 5 and 6 weeks in Group B (Fig. 3; Tab. II, lines 18 & 19).

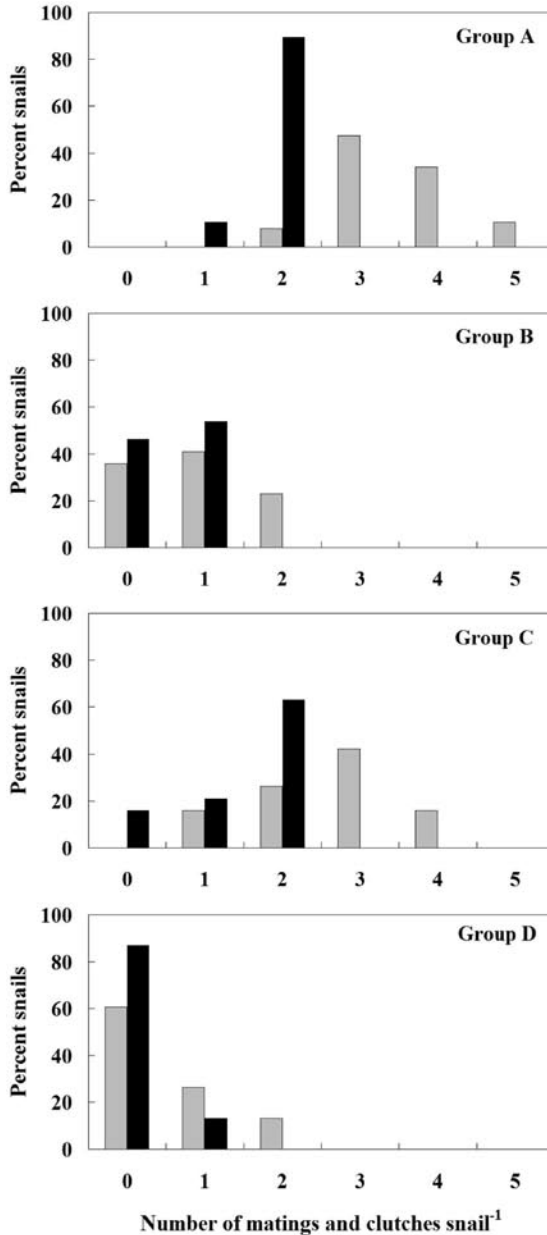


Figure 2.— Distributions of the snails of *Helix aperta* (as % of the total number of surviving snails) according to their total numbers of matings (■) and clutches (■) at four combinations of temperature (°C) and photoperiod (h). (Group A: 20°C, 16h-Light:8h-Dark; Group B: 20°C, 8h-Light:16h-Dark; Group C: 15°C, 16h-Light:8h-Dark; Group D: 15°C, 8h-Light:16h-Dark).

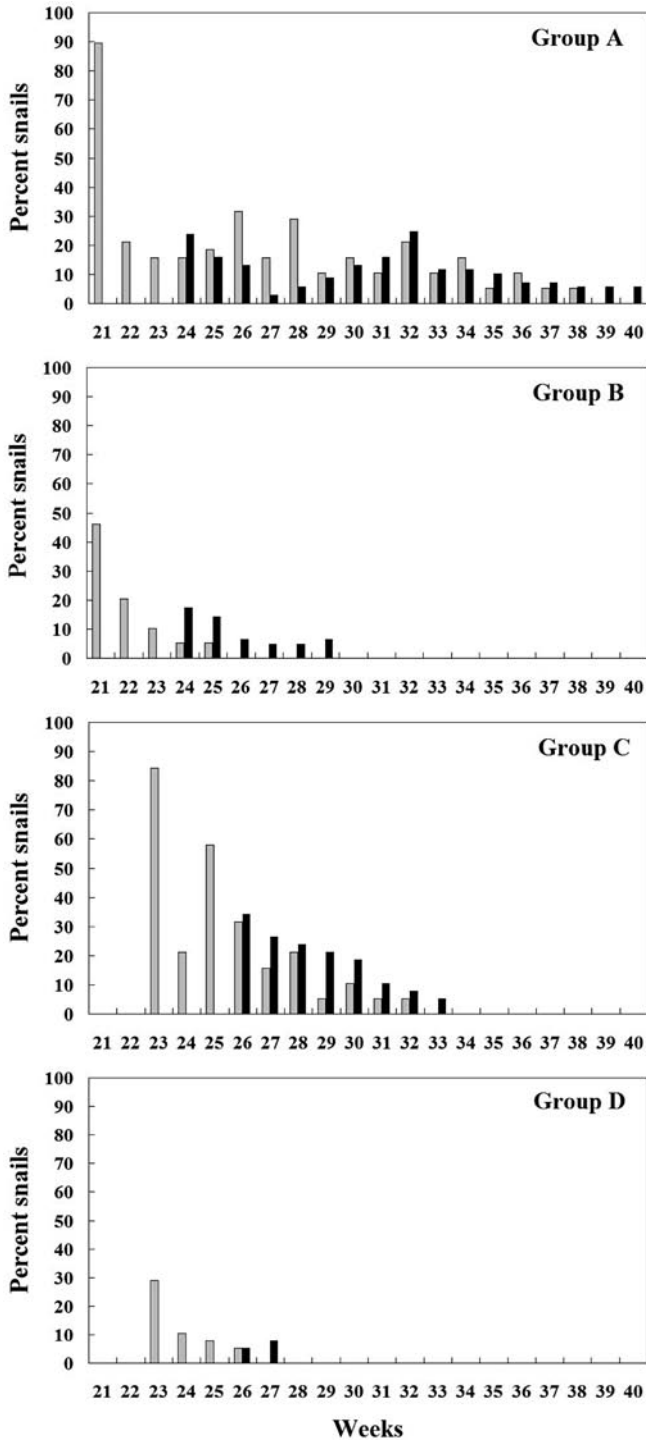


Figure 3.— Weekly variations of mating (■) and laying frequencies (■) in *Helix aperta*, (as % of the total number of surviving snails) at four combinations of temperature (°C) and photoperiod (h). (Group A: 20°C, 16h-Light:8h-Dark; Group B: 20°C, 8h-Light:16h-Dark; Group C: 15°C, 16h-Light:8h-Dark; Group D: 15°C, 8h-Light:16h-Dark).

The first mating took place on the 21st week in Groups A and B and on the 23rd in Groups C and D at frequencies of 89.47 %, 46.15 %, 84.21 % and 28.95 % respectively (Fig. 3). As to the first clutch, it was laid on the 24th week for Groups A and B and on the 26th for Groups C and D at the respective frequencies of 23.68 %, 17.42 %, 34.21 % and 5.26 % (Fig. 3). Numbers of matings per snail ranged from 2-5, 0-2, 1-4 and 0-2 in Groups A, B, C and D respectively; likewise, the numbers of egg-layings per individual were 1-2 for Groups A and C, and 0-1 for Groups B and D (Fig. 2). The modal values of mating numbers per snail and their frequencies were respectively, for the four groups listed in order, 3 (47.37 %), 1 (41.03 %), 3 (42.11 %) and 0 (60.53 %) (Fig. 2); similarly, the modal values of laying numbers per snail were 2 (89.47 %), 1 (53.85 %), 2 (63.16 %) and 0 (86.84 %) (Fig. 2). Even if the ranges of mating and laying numbers per snail were the same for Groups B and D, the corresponding frequencies were significantly different ($P < 0.05$). The four groups showed also important differences regarding the numbers of eggs laid per snail over the reproduction period (Fig. 4). On the basis of the total quantity of eggs produced, the four groups classified, in decreasing order, as follows: Group A, Group C, Group B and then Group D (Fig. 4; Tab. II, line 11). Groups A and B showed two peaks of egg numbers per snail, the first one at the 24th week for both groups and the second one at the 29th week for Group B and the 32nd week for Group A, with the highest values in Group A (Fig. 4). As to Groups C and D, they presented only one peak of egg numbers per snail at the 26th and 27th week respectively.

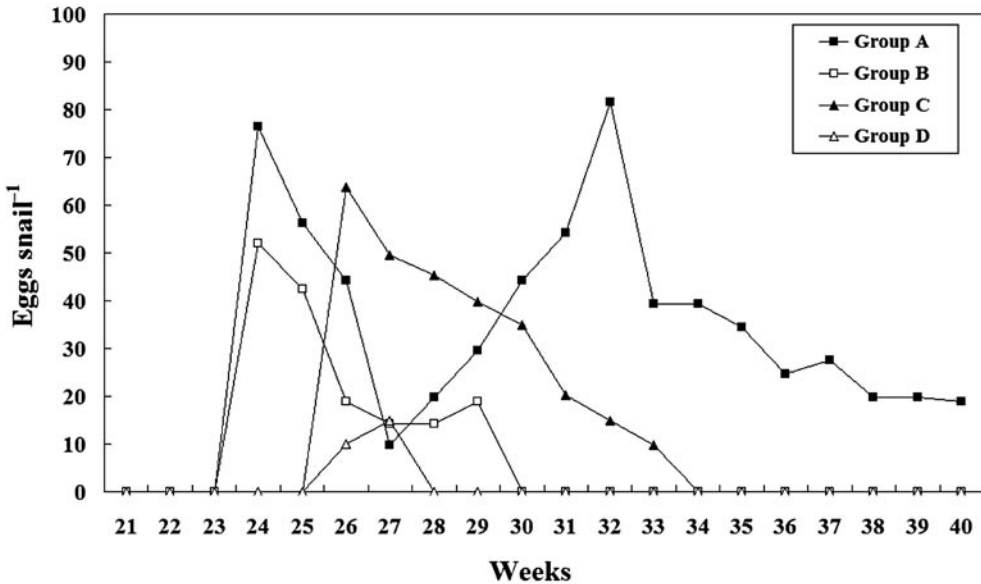


Figure 4.— Weekly oviposition (eggs per snail) of *Helix aperta* at four combinations of temperature and photoperiod: (Group A: 20°C, 16h-Light:8h-Dark; Group B: 20°C, 8h-Light:16h-Dark; Group C: 15°C, 16h-Light:8h-Dark; Group D: 15°C, 8h-Light:16h-Dark).

However, there was a highly significant association between numbers of matings and layings per snail ($\chi^2 = 71.81$; $df = 5$; $P < 0.001$; $n = 153$). The snails that laid eggs were the ones that mated the most. Furthermore, there was no relation between the weights of snails and the numbers of matings in all groups ($R = 0.033 - 0.098$, $P = 0.555 - 0.845$). Similarly, the results showed no influence of body weight on the number of layings in all groups ($R = 0.068 - 0.108$, $P = 0.519 - 0.679$).

The results show clearly that snails reared at 20°C laid more eggs per clutch. In fact, the mean number of eggs per clutch was 340.4 ± 30 and 306.0 ± 43 in Groups A and B respectively, 188.9 ± 31 in Group C and 188.8 ± 07 in Group D (Tab. II, line 11). The effect of tempera-

ture on egg number by clutch was significant at both photoperiods ($P < 0.001$). In contrast, the length of photoperiod had no effects on the mean number of eggs per clutch ($P = 0.87$) at 15°C , but a significant difference ($P = 0.001$) was detected in the mean number of eggs per clutch between the two photoperiods at 20°C (Tab. II, line 11). There was a strong correlation between the weight of the snails and the number of eggs by clutch ($R > 0.966$, $P < 0.001$) in all the four groups but no significant one was detected between the weight of the snails and that of the eggs ($R = 0.120$, $P = 0.473$). No significant difference in the weights of the eggs was detected between the four studied groups ($P = 0.591$); the mean egg weight was practically the same (Tab. II, line 12). The period between mating and egg-laying (20 days) and the time of incubation (13 days) were exactly the same in all the four groups. Hatching success of eggs was not variable and the percentage of hatching was nearly identical in the four groups (Tab. II, line 13).

Mortality rate was very low during the growth phase of snails and did not exceed 5 % for all the four groups (Tab. I); it seemed to be unaffected by temperature or photoperiod ($P = 0.46$) but it tended to increase rapidly towards the end of the experiment in Groups A and D (Tab. II, line 16). Mortality was high for Group A (24 %) after intense reproductive activity and also in Group D (21 %), in which very few eggs were produced (Tab. II, line 16). However, in Groups B and C, there were relatively lower rates of mortality (10 % and 8 % respectively).

DISCUSSION

The present study contributes to a better understanding of the effects of temperature and photoperiod on growth, sexual maturity and fecundity of *Helix aperta* snails reared under laboratory conditions. It provides the first data on the life history of this ecologically and economically important land snail.

Our results revealed that length of photoperiod had no effect on juvenile growth in the species *Helix aperta*; and no significant interactive influence of temperature and photoperiod on growth was detected (Fig. 1, Tab. I). Furthermore, the prolonged development time at 15°C compared to 20°C , regardless of photoperiod (Tab. I), suggests that length of photoperiod has also no effect on development rate. These findings are similar to those reported by Hunter & Stone (1986) in their study of *Cepaea nemoralis* where they concluded that gametogenesis and functional development of the reproductive system are neither prevented nor retarded by exposing the animals to short-days (8h-daylength) for the entire pre-reproductive life. Similarly, Jess & Marks (1998) reported that, in *Cornu aspersum maxima*, the mean adult weight and sexual maturity of snails (incidence of shell-lip and onset of oviposition), were unaffected by photoperiod and no interaction between photoperiod and temperature was detected. In contrast, Henderson & Pelluet (1960) observed that, in *Deroceras reticulatum*, spermatogenesis was strongly stimulated under 24h-daylength. In *Cornu aspersum aspersa*, Gomot *et al.* (1982) and Gomot & Deray (1987), demonstrated that the young snails of this species, when exposed to long-days (18h or 24h of light per day) from birth, matured earlier and achieved heavier body weights than those reared under short-days.

As expected from our experiments, both growth and development time of juvenile snails of *Helix aperta* species were faster at 20°C than 15°C (Fig. 1, Tab. I). In several groups of ectotherms, temperature is indicated as the major proximal factor explaining the variation in growth rates and development (Smith-Gill & Berven, 1979; van der Have & de Jong, 1996; Jarošík *et al.*, 2004) and body size is the product of growth rate and duration of growth period (Atkinson, 1994; Blanckenhorn, 1998; Stern, 2001; Davidowitz *et al.*, 2004). In our results, at low temperature, snails achieved smaller weights with little prolonged development time. At high temperature, however, snails were forced into a trade-off in which they favoured early maturation over large size, with selection favouring high growth rates at high temperatures and a short growing season. Consequently, the basic trade-off between development time and body size at maturity frequently assumed in life-history theory (Stearns, 1992; Roff, 2002) did not occur. The heavy weights in the snails reared at 20°C were due to plastic growth (higher

growth rates) and the reduction in adult size at lower temperature (15°C) may be a consequence of short growing season, development time being prolonged only by 2 weeks. As shown by Davidowitz & Nijhout (2004) at lower temperatures, the duration of the growth period is more important in determining body size, while growth rate is more important in determining size at higher temperatures.

Our results are closely comparable with those reported earlier for *Cornu aspersum maxima* in which body size at maturity increases with increasing growth temperature (Lecompte, 1995) and hatched snails reared at 20°C matured earlier and achieved heavier body weights than those at 15°C (Jess & Marks, 1998).

Natural selection would favour earliest age at maturity in all thermal environments and the faster growth at a higher temperature would result in a larger size at maturity (Angalita, 2004). In our results, higher growth rates and higher weights at 20°C favoured fecundity. Similarly, Stearns (1992) and Roff (2002) reported that fecundity increases with increasing body size. Our results showed strong positive correlation between the weight of *Helix aperta* mature snails and their fecundity (number of eggs per clutch). Snails reared at 20°C laid more eggs per clutch than those raised at 15°C. Furthermore, rapid growth and larger body size are expected to be advantageous, not only in fecundity but also in other aspects of life history for many organisms (“the bigger is better” hypothesis, reviewed by Arendt, 1997; Blanckenhorn, 2000). However, reduction in adult weights at lower temperature resulted in smaller numbers of eggs per clutch because slower growth and the resulting smaller body size reduce fecundity of each individual (Arendt, 1997; Blanckenhorn, 2000).

The mean weight of snails after reproduction (Tab. II, lines 1 & 2) decreased significantly ($P < 0.001$) in Group A (20°C, long-days) presumably because the animals required higher energy amounts to reproduction and also in Group D (15°C, short-days) which produced the smallest number of eggs but not in Groups B (20°C, short-days) and C (15°C, long-days) ($P > 0.05$).

Although the interactive effect of photoperiod and temperature has an influence on the number of matings and layings, with predominance of the effects of photoperiod, our results showed clearly that snails reared in long-day photoperiod had higher numbers of matings and layings and longer periods of reproduction (Figs. 2 & 3; Tab. II, lines 6, 9 & 17). In contrast, for snails reared in short-days, egg-laying stopped very early (after two weeks time) at 15°C (Fig. 3, Group D) and lasted only 6 weeks at 20°C (Fig. 3, Group B). Numbers of matings and egg-layings were higher in Group C (15°C, Short-day photoperiod) than in Group B (20°C, Long-day photoperiod) (Tab. II, lines 6 & 9; Fig. 2, Groups B & C).

These results suggest that photoperiod has the greatest effect on all the process of mating and egg-laying of the species *Helix aperta*; it means that reproduction is strongly regulated by the length of photoperiod. This observation confirms those made by other authors on *Cornu aspersum*. Stephens & Stephens (1966) observed that the species *Cornu aspersum aspersa* receiving 9h of light per day never laid eggs. In the same species, Bailey (1981) detected also a close relationship between photoperiod and reproductive activity. Enée *et al.* (1982) reported that reproductively active *Cornu aspersum aspersa* snails collected from natural habitats in France stopped laying after only 4 weeks when exposed to short-days (6-12h of light), whereas those exposed to long-days (18h light) did continue laying for as long as 13 weeks. Our results demonstrate the existence of an interaction between temperature and photoperiod on matings and egg-laying in *Helix aperta* with a predominant effect of photoperiod. Previously, Gomot *et al.* (1989) observed that oviposition was completely or partially inhibited under short-day photoperiods at 15°C and 20°C, respectively. The authors have shown that interactions between photoperiod and temperature exist, and increased daylight compensates the negative effect of low temperature on reproduction of *Cornu aspersum aspersa*. Similar observations have been recorded for *Helix pomatia* (Gomot, 1990). Furthermore, our results indicated highly significant association ($\chi^2 = 71.81$; $df = 5$; $P < 0.001$) between the number of matings and that of layings. In fact, snails which mated a lot were those which laid the most. These results confirm those reported for different gastropod snails where it has been clearly shown that the number of clutches is strongly regulated by the number of matings. Baur (1988) and Baur & Baur

(1992) showed that repetitive mating in the species *Arianta arbustorum* had a positive effect on the number of egg-layings. According to Stearns (1989), such a relation could be due to the "intermediate structure" of the variation, including physiological mechanisms under endocrine control. Numbers of matings and clutches are not correlated to the weight of snails (Madec *et al.*, 1998, 2000).

Our experiments demonstrate clearly that the number of eggs per clutch was very significantly related ($R > 0.966$; $P < 0.001$) to the weight of the snails in all the four groups. This could explain the differences in the mean number of eggs per clutch between snails reared at different temperatures and between those reared at 20°C under different photoperiods (Groups A & B) (Tab. II, line 11); snails reared at 20°C were heavier and more reproductive, those reared at 15°C were lighter and less reproductive. Similarly, snails in Group A were heavier and have greater mean number of eggs per clutch than those in Group B (Tab. II, line 11). Our results revealed no significant correlation between the mean weight of eggs and the weight of snails ($R = 0.120$, $P = 0.473$). Comparable correlations between weight and fecundity of animals were noticed in *Cornu aspersum maxima* on farm snails after an artificial hibernation of 3 months (Madec *et al.*, 1998) and, in *Cornu aspersum aspersa*, on wild snails collected from nature in France (Madec *et al.*, 2000). The authors detected a strong correlation between the number of eggs by clutch and the body size of snails, but no relation between the body size of snails and the weight of their eggs. In *Cornu aspersum*, Dupont-Nivet *et al.* (1998) found that snails with comparable past and present conditions of life showed positive phenotypic and genetic correlations between egg number and (1) time from emergence from hibernation to laying and (2) time from mating to laying, but no relation of (1) and (2) with egg weight.

Our results indicate also that the physiological factors that control all of (1) the mating and egg-laying and (2) the number of eggs by clutch were not identical, with no correlation between these two aspects of reproduction activity. Matings and egg-layings are only sensitive to interferences between the environmental factors and the neuroendocrine stimuli of the snails of the species *Helix aperta*; but the number of eggs per clutch is strongly related to the weight of snails, it depends probably on the quantity of energy stocked regardless of temperature and photoperiod.

In conclusion, the best conditions for growth and reproduction of *Helix aperta* snails are the combination of temperature of 20°C and a long-day photoperiod (16h Light: 8h Dark).

ACKNOWLEDGEMENTS

We would like to thank very much Pr Hayet Louaileche for her linguistic help during the writing of this paper and Mr Mohand Akli Tafoughalt for his valuable technical support regarding the utilization of Minitab Software.

REFERENCES

- ANGILLETTA, M.J.JR., STEURY, T.D. & SEARS, W.M. (2004).— Temperature, growth rate and body size in ectotherms: Fitting pieces of a life-history puzzle. *Integr. Comp. Biol.*, 44: 498-509.
- ARENDT, J.D. (1997).— Adaptive intrinsic growth rates: an integration across taxa. *Q. Rev. Biol.*, 72: 149-177.
- ARENDT, J.D. (2000).— Allocation of cells to proliferation vs. differentiation and its consequences for growth and development. *J. Exp. Zool.*, 288: 219-234.
- ATKINSON, D. (1994).— Temperature and organism size — a biological law for ectotherms? *Adv. Ecol. Res.*, 25: 1-58.
- ATKINSON, D. (1995).— Effects of temperature on the size of aquatic ectotherms: Exceptions to the general rule. *J. Therm. Biol.*, 20: 61-74.
- AUPINEL, P. & DAGUZAN, J. (1989).— Étude du rôle de la photopériode sur l'activité métabolique des jeunes escargots Petit-gris (*Helix aspersa* Müller) et mise en évidence d'une phase photosensible. *Haliotis*, 19: 47-55.
- BAILEY, S.E.R. (1981).— Circannual and circadian rhythms in the snail *Helix aspersa* and the photoperiodic control of annual activity and reproduction. *J. Comp. Physiol.*, 142: 89-94.
- BAUR, B. (1988).— Repeated mating and female fecundity in the simultaneously hermaphroditic land snail *Arianta arbustorum*. *Invertebr. Reprod. Develop.*, 14: 197-204.
- BAUR, B. & BAUR, A. (1992).— Effects of courtship and repeated copulation on egg production in the simultaneously hermaphroditic land snail *Arianta arbustorum*. *Invertebr. Reprod. Develop.*, 21: 201-206.

- BERRIGAN, D. & CHARNOV, E.L. (1994).— Reaction norms for age and size at maturity in response to temperature—a puzzle for life historians. *Oikos*, 70: 474-478.
- BLANCKENHORN, W.U. (1998).— Adaptive phenotypic plasticity in growth, development and body size in the yellow dung fly. *Evolution*, 52: 1394-1407.
- BLANCKENHORN, W.U. (1999).— Different growth responses to temperature and resource limitation in three fly species with similar life histories. *Evol. Ecol.*, 13: 395-409.
- BLANCKENHORN, W.U. (2000).— The evolution of body size: what keeps organisms small? *Q. Rev. Biol.*, 75: 385-407.
- BONNEFOY-CLAUDET, R. & DERAY, A. (1984).— Influence de la durée d'hibernation sur l'activité reproductrice de l'escargot *Helix aspersa* Müller. *CR. Soc. Biol.*, 178: 42-49.
- CHARRIER, M. (1980).— *Contribution à la biologie et à l'écophysiologie de l'escargot Petit-gris Helix aspersa Müller (Gasteropode Pulmoné Stylommatophore)*. Thèse 3^e Cycle, Rennes.
- CHARNOV, E. (1993).— *Life history invariants: Some explorations of symmetry in evolutionary ecology*. Oxford University Press, Oxford.
- DAGUZAN, J. (1981).— Contribution à l'élevage de l'escargot Petit-gris *Helix aspersa* Müller. I. **Reproduction et éclosion** des jeunes en bâtiment et en conditions thermohygrométriques contrôlées. *Annales de Zootechnie*, 30: 249-272.
- DAVIDOWITZ, G., D'AMICO, L.J. & NIJHOUT, H.F. (2004).— The effects of environmental variation on a mechanism that controls insect body size. *Ecol. Evol. Res.*, 6: 49-62.
- DAVIDOWITZ, G. & NIJHOUT, H.F. (2004).— The physiological basis of reaction norms: The interaction among growth rate, the duration of growth and body size. *Integr. Comp. Biol.*, 44: 443-449.
- DERAY, A. & LAURENT, J. (1987).— Lumière et dynamique de croissance chez l'escargot *Helix aspersa maxima* (Gros-gris) en conditions hors sol contrôlées. *C. R. Soc. Biol. Paris*, 181: 79-186.
- DUPONT-NIVET, M., MALLARD, J., BONNET, J.C. & BLANC, J.M. (1998).— Quantitative genetics of reproductive traits in the edible snail *Helix aspersa* Müller. *J. Exp. Zool.*, 281: 220-227.
- ENÉE, J., BONNEFOY-CLAUDET, R. & GOMOT, L. (1982).— Effet de la photopériode artificielle sur la reproduction de l'escargot *Helix aspersa* Müller. *C. R. Acad. Sci. Paris*, 294: 357-360.
- GOMOT, A. (1990).— Photoperiod and temperature interaction in the determination of reproduction of the edible snail *Helix pomatia*. *J. Reprod. Fertil.*, 90: 581-585.
- GOMOT, L. & DERAY, A. (1987).— Les escargots. *La Recherche*, 186: 302-311.
- GOMOT, L., ENÉE, J. & LAURENT, J. (1982).— Influence de la photopériode sur la croissance pondérale de l'escargot *Helix aspersa* Müller en milieu contrôlé. *C. R. Acad. Sci. Paris*, 294: 749-752.
- GOMOT, P., GOMOT, L. & GRIFFOND, B. (1989).— Evidence for a light compensation of the inhibition of reproduction by low temperatures in the snail *Helix aspersa*. Ovotestis and albumen gland responsiveness to different conditions of photoperiods and temperatures. *Biol. Reprod.*, 40: 1237-1245.
- GOMOT, P., GRIFFOND, B. & GOMOT, L. (1990).— Action de la température sur la synthèse d'ADN des cellules mâles et la spermatogenèse d' *Helix aspersa* en hibernation. *J. Therm. Biol.*, 15: 267-280.
- GOMOT-DE VAUFLEURY, A. (2000).— Standardized growth, toxicity testing (Cu, Zn, Pb and Pentachlorophenol) with *Helix aspersa*. *Ecotoxicol. Environ. Saf.*, 46: 41-50.
- HENDERSON, N.E. & PELLUET, J. (1960).— The effect of visible light on the ovotestis of the slug *Deroceras reticulatum* Müller. *Can. J. Zool.*, 38: 173-178.
- HUNTER, R.D. & STONE, L.M. (1986).— The effect of artificial photoperiod on growth and reproduction in the land snail *Cepaea nemoralis*. *Int. J. Invertebr. Reprod. Dev.*, 9: 339-344.
- JAROŠÍK, V., KRATOCHVÍL, K., HONĚK, A. & DIXON, A.F.G. (2004).— A general rule for the dependence of developmental rate on temperature in ectothermic animals. *Proc. R. Soc. London B (Suppl.)*, 271: 219-221.
- JESS, S. & MARKS, R.J. (1998).— Effect of temperature and photoperiod on growth and reproduction of *Helix aspersa* var. *maxima*. *J. Agr. Sci.*, 130: 367-372.
- KERNEY, M.P. & CAMERON, R.A.D. (1979).— *A field guide to the land snails of Britain and NW Europe*. William Collins Sons and Co. Ltd., London.
- LAURENT, J., DERAY, A. & GRIMARD, A.M. (1984).— Influence de la photopériode et du degré d'hétérogénéité de la population sur la dynamique de croissance et la maturité sexuelle de l'escargot *Helix aspersa*. *C. R. Soc. Biol.*, 178: 421-441.
- LECOMPTE, O. (1995).— *Etude préliminaire de la plasticité de quelques traits de vie chez deux races du Pulmoné terrestre Helix aspersa*. Unpublished DEA, University of Rennes.
- MADEC, L., DESBUQUOIS, C. & COUTELLE-CVRETO, M.A. (2000).— Phenotypic plasticity in reproductive traits: Importance in the life history of *Helix aspersa* (Mollusca: Helicidae) in a recently colonized habitat. *Biol. J. Linn Soc.*, 69: 25-39.
- MADEC, L., GUILLER, A., COUTELLE-CVRETO, M.A. & DESBUQUOIS, C. (1998).— Size-fecundity relationships in the land snail *Helix aspersa*: preliminary results on a form outside the norm. *Invertebr. Reprod. Develop.*, 34: 83-90.
- MINITAB, INC. (2000).— *Minitab for Windows, Release 13.31*. Minitab Inc., State College, PA, USA.
- ROFF, D.A. (1992).— *The evolution of life histories*. Chapman and Hall, New York.
- ROFF, D.A. (2002).— *Life history evolution*. Sinauer Associates, Inc., Sunderland, Massachusetts.

- SCHÜTT, H. (2001).— Die Türkische Landschnechen 1758-2000. *Acta Biologica Benrodis*, Suppl., 4: 550.
- SMITH-GILL, S.J. & BERVEN, K.A. (1979).— Predicting amphibian metamorphosis. *Am. Nat.*, 113: 563-585.
- STATSOFT, INC. (1999).— *Statistica pour Windows, kernel version 5.5*. StatSoft France, Maisons-Alfort, France.
- STEARNS, S.C. (1989).— Trade-offs in life-history evolution. *Funct. Ecol.*, 3: 259-268.
- STEARNS, S.C. (1992).— *The evolution of life-histories*. Oxford University Press, Oxford.
- STEPHENS, C.I. & STEPHENS, G.C. (1966).— Photoperiodic stimulation of egg-laying in the land snail *Helix aspersa*. *Nature*, 212: 1582.
- STERN, D. (2001).— Body-size evolution: How to evolve a mammoth moth. *Curr. Biol.*, 11: 917-919.
- VAN DER HAVE, T. M. & DE JONG, G. (1996).— Adult size in ectotherms: Temperature effects on growth and differentiation. *J. Theor. Biol.*, 183: 329-340.
- VON BERTALANFFY, L. (1960).— Principles and theory of growth. Pp. 137-259 In: W.W. Nowinskii (ed.), *Fundamental aspects of normal and malignant growth*. Elsevier, Amsterdam.
- VAN VOORHIES, W.A. (1996).— Bergmann size clines: A simple explanation for their occurrence in ectotherms. *Evolution*, 50: 1259-1264.

