

Reproductive Biology and Postnatal Development of *Microtus rossiaemeridionalis* Ognev, 1924 (Mammalia: Rodentia) Distributed in Turkey

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Abstract: The reproductive biology and postnatal development of *Microtus rossiaemeridionalis* were investigated based on field and laboratory observations. This vole preferred humid meadows, and its burrow was complex with several entrances. The breeding season of this species was between December and October, and lasted 11 months. Pregnancy lasted 20-21 days, and average litter size was 5. The first mating was observed in a captive female at an age of 35 days, and other females subsequently mated at 40, 42, 60, and 70 days. Young were weaned at 17 days. The growth rate of weight was 0.45 g/day between birth and day 13, 1 g/day between days 18 and 24, and decreased thereafter. Length of specimens reached adult size within 45 days, whereas tail length, ear length, and weight reached adult size at 37 days, and hind foot length at 30 days.

Key Words: *Microtus rossiaemeridionalis*, reproduction biology, Turkey

Türkiye'de Yayılış Yapan *Microtus rossiaemeridionalis* Ognev, 1924 (Mammalia: Rodentia)'in Üreme Biyolojisi ve Doğum Sonrası Gelişimi

Özet: *Microtus rossiaemeridionalis*'in üreme biyolojisi ve doğum sonrası gelişimi laboratuvar ve arazide çalışıldı. Bu tür nemli çayırlıkları habitat olarak tercih etmekte ve yuvası çok girişli kompleks bir yapı göstermektedir. Bu türün üreme sezonunun Aralık – Ekim ayları arasında 11 ay kadar olduğu, hamileliğin 20-21 gün kadar sürdüğü ve doğumlardaki ortalama yavru sayısının 5 olduğu belirlendi. Laboratuvarında ilk çiftleşme 35 günlük bir dişi de gözlemlendi, bunu sırasıyla 40, 42, 60 ve 70 günlük dişiler izledi. Gençler 17 güne kadar süttten kesilmektedir. Ağırlıktaki büyüme oranı doğum ile 13. gün arasında 0.45 gr / gün, 24. güne kadar 1 gr / gün olup daha sonra azalmıştır. Örneklerin ergin bireylerin ölçü ve ağırlıklarına erişme zamanı; boy uzunluğu için 45 gün, kuyruk uzunluğu, kulak uzunluğu ve ağırlık için 37 gün ve ardayak uzunluğu için 30 gün olarak saptandı.

Anahtar Sözcükler: *Microtus rossiaemeridionalis*, üreme biyolojisi, Türkiye

Introduction

The genus *Microtus* is commonly distributed in steppe, meadow, and rocky areas in the Palaearctic region (Corbet, 1978; Harrison and Bates, 1991; Wilson and Reeder, 1993). Previously, more than 10 *Microtus* species were doubtfully recorded in Turkey (Blackler, 1916; Neuhäuser, 1936; Spitzenberger and Steiner, 1962; Osborn, 1962; Lehmann, 1969; Felten et al., 1971; Spitzenberger, 1971; Morlok, 1978; Dođramacı, 1989; Kefeliođlu, 1995; Çolak et al., 1998a; Kefeliođlu and Krystufek, 1999; Yiđit and Çolak, 2002). *Microtus epiroticus* was first described in Greece by Ondiras (1966) and was then recorded in northern Turkey by Dođramacı (1989). The taxonomic status of this species was first studied by Kefeliođlu (1995) in Turkey, after which the species was no longer considered

a valid taxa; it is now considered a synonym of *Microtus rossiaemeridionalis* Ognev, 1924 (Mitchell-Jones et al., 1999). Voles of the genus *Microtus* are usually considered agricultural pests, but there is no information given on how and when their harmful effects occur. The ecological and biological peculiarities of *M. rossiaemeridionalis* are still unknown in Turkey, and are very scant in other countries, including the ranging area of this species (Yoccoz et al., 1993). The few ecological studies on this species are mainly focused on prey-predator relationships (Korpimaki and Norrdahl, 1991; Panteleev et al., 1991; Norrdahl and Korpimaki, 1993; Korpimaki et al., 1994). The aim of this present work was to contribute to the knowledge of the reproductive biology and postnatal development of this species, and to comment on their ecological importance.

Materials and Methods

The field observations and laboratory study were performed between 1998 and 2001 in western Turkey. Live specimens were captured around Yeşilköy (lat 38°18'39.09"N, long 31°36'01.62"E) and Beyşehir (lat 37° 33'53.39"N, long 31°35'12.08"E) (Konya) by Sherman live traps and excavating burrows. Burrows (n = 10) were also excavated to determine burrow structure. Live specimens were transferred to the laboratory to observe their reproductive biology and postnatal development, and then were housed as pairs and mixed groups in cages under natural light. The animals were provided with nesting materials, food (wheat seeds and fresh grass), and water. To determine the birth of offspring, mating, and behavior, adult voles housed as pairs were inspected daily and nightly. The gestation period was estimated both by separating pairs after mating occurred and as the minimal time interval between successive births. After birth occurred, 4 external measurements (total body length, tail length, hind foot length, and ear length) and weights of newborns were recorded throughout the postnatal period (hind foot measurement included claw length). Growth rates were calculated in accordance with the formula $N_1 - N_{1+t} / t$, where N_1 is the first value of any external characteristic or weight, N_{1+t} is the second value of any external characteristic or weight in a time interval, and t is the time (days) between the first and the second time.

Biometric comparisons were performed by t-test (Microsoft Excel t-test Two-Sample Assuming Unequal Variance). All voles were kept in ambient conditions similar to the climate of their natural environment. In addition to ecological and biological observations, karyological studies from bone marrow were performed in accordance with the conventional colchicine hypotonic citrate technique to confirm species identification ($2n = 54$, $FN = 56$, $FN_a = 52$). Skins and skulls of the voles have been deposited in the collection at Ankara University, Department of Biology, Faculty of Science, Ankara University.

Results

Habitat, burrowing, and behavior: Live specimens of *M. rossiaemeridionalis* were obtained from Yeşilköy and Beyşehir, and altitudes of 1050 and 1130 m, respectively. According to the field observations this vole preferred to live in humid fields and usually occupied meadows, watery plains, rivers, and lake sides with soft soil and covered by grassy vegetation. We recorded *M. rossiaemeridionalis* at 16 different localities in western Anatolia, which were not grain fields; however, these voles very frequently built burrows with several entrances in clover fields, and were active throughout the year. This diurnal vole was commonly seen out of the burrow during the day. Unsuccessful trapping during the night lent support to the diurnal lifestyle of this vole. The vole spent most of its time in the burrow systems in the colder season. Intensive daily activity above ground resulted in narrow pathways between the entrances of burrows. This vole lives in a complex burrow, with several entrances about 4.5-6 cm in diameter and blind alleys. The burrow was a maximum of 10-12 m long and 20-25 cm deep. The passages descended from the entrance holes at an angle of 45°-70°. There was usually one nest chamber 14 x 10 cm, and this chamber contained dry grasses as nesting material. There were no food store chambers in the burrows (Figure 1). The vole usually occupied a burrow alone, but sometimes there was a small colony with a female and offspring. Nonetheless, burrow systems were found very close to each other. According to the laboratory observations, voles housed as a large colony in a single cage had good tolerance for each other, and fighting did not occur. These findings demonstrate that this vole lives colonially in its habitat.

Reproduction biology: In the laboratory, 13 pairs housed were separately and monitored throughout 1 year. Births were first observed at the end of December

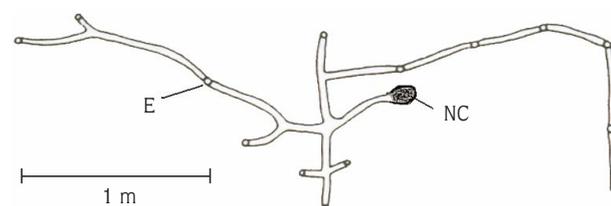


Figure 1. Burrow structure of *M. rossiaemeridionalis*, planar view (E = entrance, NC = nest chamber).

and lasted until the middle of September. The 13 pairs had 32 births during the breeding season, and litter size varied from 2 to 8 pups (Figures 2 and 3). The most intensive breeding occurred in April, during which 7 of the 32 births occurred. One pair of adult voles housed in a cage (60 cm x 60 cm x 60 cm) had 4 births during 6 months, and the numbers of individuals reached 30 via the cross breeding of offspring. The duration of pregnancy was estimated to be 20 days in 5 births and 21 days in 2 births. It was also determined that females had a minimum of 1 and a maximum of 6 births during the breeding season. The interval between 2 successive births was between 21 and 65 days (Table 1). The sex ratio of the 146 pups was 58:42 (male to female).

Postnatal development: Newborns were blind, naked, and dark pinkish. Their ears were closed with skin, and both the fore and hind feet were not open, but there were semi-rigid nails 0.5-1 mm in length on each of the claws. Even though they were naked, a few white

vibrissae 2-3 mm in length occurred on both sides of the rostrum. The mouth, a small hole at the anterior tip of the rostrum, was almost closed at birth. Newborns did not move during the first 2 days of life. The average weight of the newborns was about 2.3 g and total length was 44.1 mm, tail length 7.6 mm, hind foot length 6.3 mm, and ear length 1.6 mm (Table 2). The dorsal color of the newborns became blackish after the second day, and then the abdominal region became blackish. The lower and upper incisors appeared after 4 and 5 days, respectively. The dorsal color turned to light brownish after 4 days, and the whole body was covered with hair by 9 days; dorsal fur was brownish and ventral fur was grayish-whitish. The fore feet first opened on day 6, young opened their eyes between days 7 and 9, followed by the hind feet and ears within 13 days. These young, within 13 days, still suckling, started to move freely in the cage. They stopped suckling by day 17 when total body length and weight were 102.7 cm and 12.9 g,

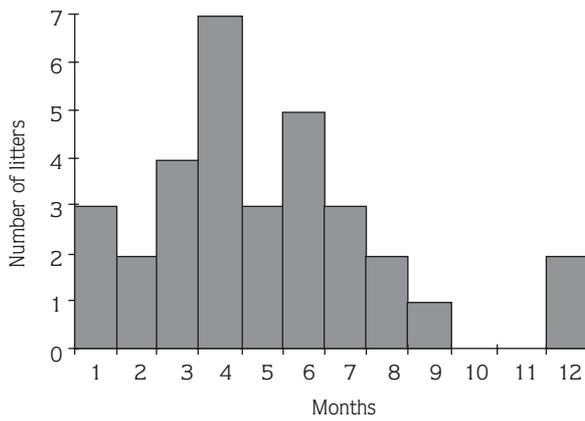


Figure 2. Monthly distribution of litters of 13 *M. rossiaemeridionalis* specimens (n = 32) throughout the reproduction period.

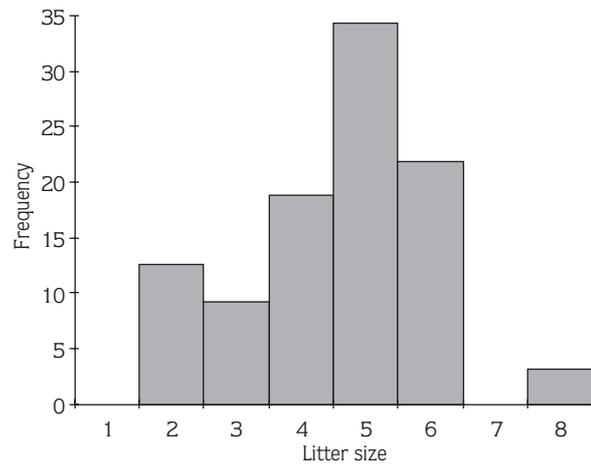


Figure 3. Frequency of the litter size for 32 births observed in captivity.

Table 1. The intervals (days) between previous and successive births.

Approximate interval between births (days)	Number of births	Frequency
21-25	9	47.4
26-30	1	5.3
31-35	1	5.3
36-40	2	10.5
41-45	2	10.5
46-50	1	5.3
51-65	3	15.8

Table 2. Increases in size of external characteristics (mm) and weights (g) between birth and weaning (n = number of individuals, L = lengths).

Age stages	n	Total L	Tail L	Hind foot L	Ear L	Weight	Days
Birth	64	44.1	7.6	6.3	1.6	2.3	-
Hair growing	62	70.1	14.0	10.9	5.5	6.2	4-7
Eye opening	56	77.4	16.7	13.5	6.2	7.0	9-12
Ear opening	56	86.6	19.0	14.5	7.5	8.5	10-13
Free movements	56	93.0	20.5	15.4	8.6	10.1	13-15
Weaning	56	102.7	22.2	17.1	9.6	12.2	13-17

respectively (Table 2). The dorsal color of the young animals when weaned was almost similar to that of adults. It was also determined that it is impossible to distinguish the sex of the young animals by the external genitalia, but the external genitalia became clearly distinguishable as vagina and penis holes at an age of 24 days. Newborns were measured and weighed up to age 64 days at irregular intervals (1, 2, 3, 4, 5, 7, 9, 11, 13, 15, 18, 24, 30, 37, 40, 45, 53, 64 days). The measurement and weight at these intervals were compared to previous and subsequent days by t-test.

Total length increased until age 45 days, and tail, hind foot, and ear lengths, and weight increased from age 30 to 37 days ($P < 0.05$); then the growth of those characteristics stopped ($P > 0.05$) (Figures 3-7). The external measurements and weight of the young

overlapped with adults after the age of 1.5 months. They were then considered adults. The growth rates of external measurements and weight during the increasing period showed 2 different phases (2 different rates of growth). The first phase was seen mostly during the lactation period, up to roughly 17 days of age, except for hind foot length and weight. The growth rates of total body length, and tail and ear lengths increased up to days 16 and 18. The growth rate of hind foot length was highest during the first week, and the marked fluctuations in the growth rate were observed between 7 and 17 days; then the rate decreased. The growth rate of weight increased up to 24 days of age, and then decreased (Figure 8). During this period, the growth rate was 0.45 g/day up to day 13 and reached 1 g/day between days 18 and 24.

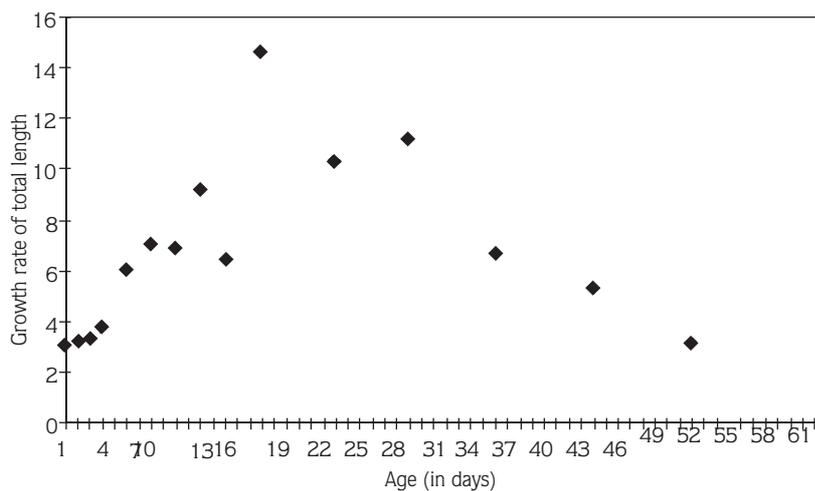


Figure 4. Growth rate of total body length in captivity.

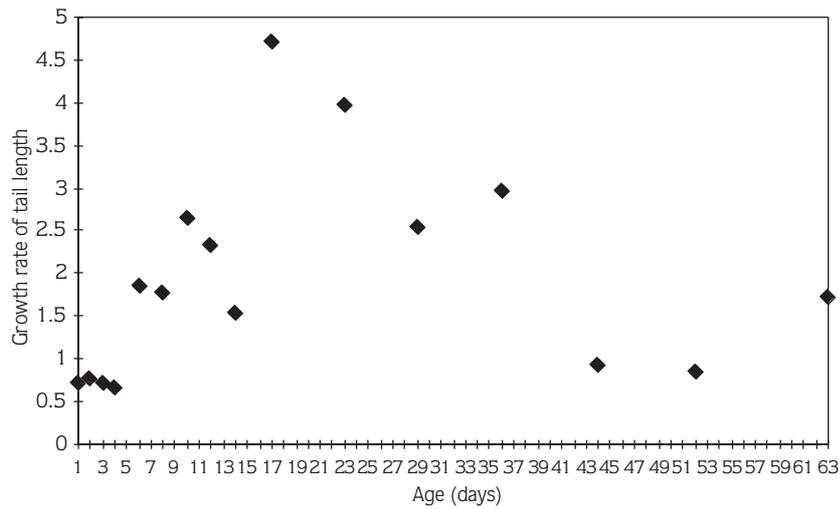


Figure 5. Growth rate of tail length in captivity.

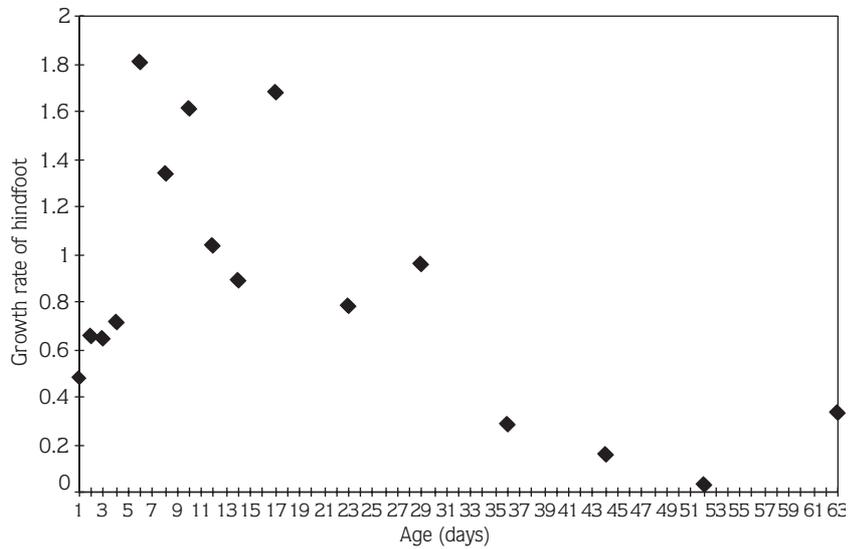


Figure 6. Growth rate of hind foot length in captivity.

In order to determine when sexual maturity was attained, pairs were established in the cages as follows: young weaned female-adult male; young weaned male-adult female. These pairs were observed until the first birth occurred. The first birth was observed in a 55-day-old female; when the pregnancy period is considered, it can be said that this female mated at an age of 35 days. The weight of this female was 22.4 g at mating and the litter size was 4. This first birth was followed by other females mating at ages 40, 42, 60, and 70 days. The weights of these female were 25, 26.2, 27.1, and 31 g, respectively. The first mating was observed in a 42-day-

old male with a weight of 25 g. Other males (n = 6) copulated at age 47, 49, 49, 50, 60, and 70 days, and the weights of these male were 30.1, 27.3, 30.8, 32.3, 33.4, 27.6, and 34.2 g, respectively.

Discussion

Petrov and Ruzic (1982) reported that this vole usually lives in meadows and cultured lands, and its habitat becomes arid towards southeast Europe. According to our findings, this vole's distribution is restricted to humid habitats; it usually occupies rivulet

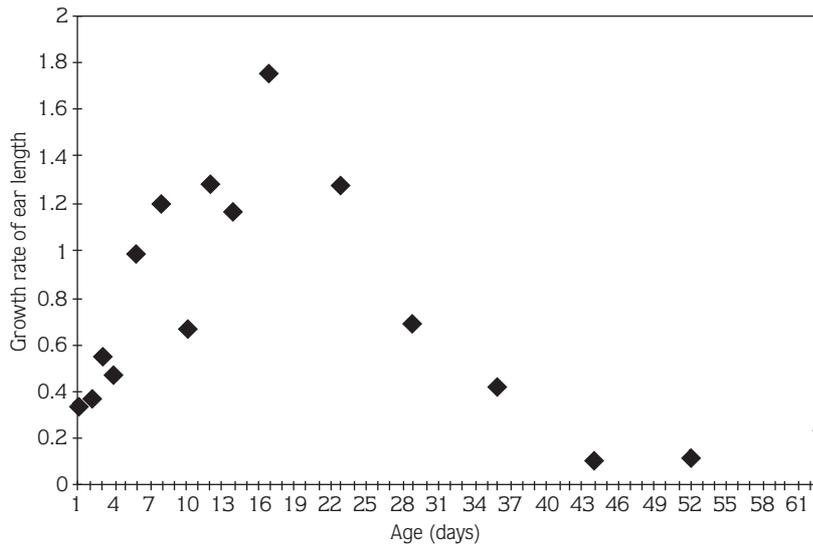


Figure 7. Growth rate of ear length in captivity.

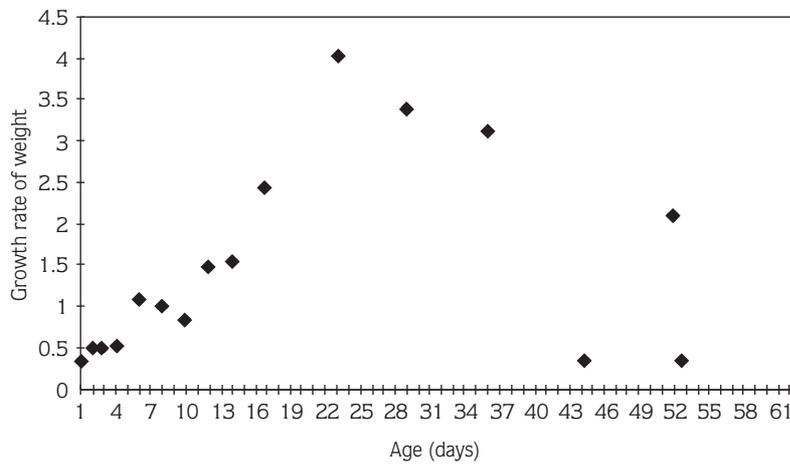


Figure 8. Growth rate of weight in captivity.

sides, marshy grasslands, and humid deciduous forest edges. Mitchell-Jones et al. (1999) stated that *M. rossiaemeridionalis* prefers agricultural lands, windbreaks, meadows, and light forests, and occupies humid habitats in the southern parts of its range. Our findings confirmed that the Turkish population prefers humid habitats, such as meadows and clover fields. Kefelioğlu (1995) also stated that this vole occurred in forest clearings, edges of forest patches in grassy grounds, grain fields, and meadows along river sides, and is distributed between sea level and 2000 m.

Yoccoz et al. (1993), who performed the first eco-biological study on the Finland population of *M.*

rossiaemeridionalis, reported that pregnancy lasted 20-21 days. This finding is consistent with our study. According to Nadeau (1985), a pregnancy period of 20-21 days was characteristic for *Microtus* spp. In support of this presumption, Çolak et al. (1998b) gave the pregnancy period of 21 days for *M. lydius*, which is commonly distributed in western Turkey. A similar pregnancy period was also reported for *M. arvalis*, a sibling species of *M. rossiaemeridionalis* (Frank, 1956). According to Yoccoz et al. (1993), the birth weight of this vole was 2.3 g. This value was found to be similar in Turkish specimens (Table 3); however, birth weights of *M. arvalis* were reported as 1.9 and 2 g by Frank (1956)

Table 3. The mean weights (g) at birth for some *Microtus* spp. distributed in Turkey.

Species	The mean weight at birth	References
<i>M. rossiaemeridionalis</i>	2.3 (Turkey)	Present study
	2.1 (Finland)	Yoccoz et al., 1993
<i>M. arvalis</i>	2	Dakotse and Martinet, 1977
	2	Rychnovsky, 1983
	1.8	Wojciechowska, 1970
	2	Pegel'man and Korabe'nikov, 1972
<i>M. guentheri</i>	2.5	Cohen-Shlagman et al., 1984
<i>M. lydius ankaraensis</i>	3.3	Çolak et al., 1998b
<i>M. socialis</i>	2	Pegel'man and Korabe'nikov, 1972

and Rychnovsk (1983), respectively. These weights of *M. arvalis* were less than those of *M. rossiaemeridionalis* in Turkey. According to Yoccoz et al. (1993), the average litter size was 4.5 for the first birth and 6.1 for successive births. We did not determine a marked difference between the first and successive births of *M. rossiaemeridionalis*. In our observations, a female had 8 newborns in the third birth, but another one produced only 2 in the fifth birth. In general, the average litter size of *M. rossiaemeridionalis* was close to the data given by Yoccoz et al. (1983). According to our study, the sex ratio was 58.2:41.8 (male to female). These findings are also consistent with those published by Yoccoz et al. (1983), who also stated that the sexual maturity of *M. rossiaemeridionalis* took place after 17 days in females and 35 days in males in the Finland population. In contrast, we determined that the earliest sexual maturation occurred at 35 days for females and 42 days for males. Frank (1956) suggested that the sexual maturity of *M. arvalis* was 13 day for females. Martinet (1967) also noted that sexual maturity occurred at 19-22 days for females of *M. arvalis*. According to Frank (1966) and our findings, the sexual maturity of *M. arvalis* takes places earlier than in *M. rossiaemeridionalis*.

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We determined that external characteristics of the young were in the range of adult specimens after 37 days. The growth rate of external characteristics during postnatal development generally increased until weaning and had 2 phases; the first phase was the time between birth and weaning (up until a certain time in weaning, depending on the characteristic), and the second phase was after weaning. In terms of weight we found that the growth rate was 0.45 g/day in the first phase (up to day 13) and 1 g/day in the second phase (between days 18 and 24). Yoccoz et al. (1983) reported that growth rates were 0.5 g/day in the first phase and 1.2 g/day in the second phase. These finding are very similar to our results. In addition, there was a negative correlation between litter size and growth rate. When the litter size increased, postnatal development slowed down. Similar results were also reported for *M. guentheri* by Cohen-Shlagman (1984).

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