Trap type and positioning: how to trap Savi's pine voles using the tunnel system

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Running title: How to trap Savi's pine vole.

Abstract

Savi's pine vole, *Microtus savii*, is the most widespread Italian vole species, an important rodent pest of agriculture, and yet one of the least studied species. One of the reasons for this gap in knowledge is that members of this species are quite difficult to capture with standard trapping procedures, being fossorial and rarely active above ground. For this reason, we developed a protocol that maximizes trapping success. This method requires the identification of the active tunnel holes and the placement of the traps directly in front of the exits. We also compared capture and recapture rates of Savi's pine voles in three different trap types: INRA, Longworth and Ugglan. If properly equipped with food and nesting material, INRA, Longworth and Ugglan traps showed similar capture rates, but the recapture rate of Ugglan traps was the highest of the three kinds of traps. These results, in combination with the species´ fossorial and social habits, lead us to conclude that Ugglan traps are the best suited for studies on Savi´s pine voles. Our results may have implications for planning and implementing management strategies based on traps rather than rodenticides, as well as field studies on other fossorial small mammals.

Introduction

Population-monitoring techniques to assess the density, ecology and distribution of small mammals in the wild are largely based on live trapping (Flowerdew et al. 2004). When planning field studies, attention should be devoted to the selection of the most suitable type of trap in relation to the aims of the study. In fact, good capture rates are the first step for the success of a study because they provide a representative sample of the examined population.

Considering studies on the relationship between species and habitats, the most common topics of interest address the composition of communities and the population dynamics of species; both require an efficient live-trapping protocol. In the first case, the aim is to trap the largest possible number of species, and it is commonly accepted that the most efficient solution is to use multiple trap types (Anthony et al. 2005; Dizney et al. 2008). In the second case, the focus is directed on one or a few target species, and high capture and recapture rates are required for investigating long-term demographic trends. Therefore, it may be useful to use a single type of trap with higher efficiency and lower mortality rates. However, when studying complex communities that include semi-fossorial and fossorial species, more effective systems for above-ground-active species are used, whereas little is known about capturing markedly fossorial species.

Savi's pine vole, *Microtus savii* (de Selys-Longchamps 1838), inhabits fallow fields, cereal fields, ecotonal areas, orchards and forage crops, and it is both the most widespread Italian vole species and one of the least studied from a behavioural and ecological perspective (Bertolino et al. 2015a; Ranchelli et al. 2016). This problem is mainly because its fossorial habits make it extremely difficult to observe in the wild, as well as to capture (Ranchelli et al. 2016). Savi´s pine voles are strictly fossorial, and they spend most of their time in underground tunnels, which they leave mainly to forage (Contreras and McNab 1990). This species is of great interest because it inhabits agricultural areas, and plays an important role in the trophic and ecological dynamics of many habitats. It is also a prey species for many raptors and small- and medium-sized mammalian predators (Amori et al. 2002; Capizzi and Santini 2007; Magrini et al. 2009). Increasing knowledge regarding the behaviour and dynamics of Savi's pine voles would prove especially important from a management perspective. Savi´s pine vole, in fact, is considered a pest species because of the damage it causes to crops when densities are high (Capizzi et al. 2014). During winter, when food availability is low, Savi's pine voles feed on plant roots. They may cause extensive debarking, especially to peach, apple, cherry and citrus orchards, as well as extensive damage to artichokes and potatoes (Capizzi and Santini 2007; Bertolino et al. 2015b; Ranchelli et al. 2016).

Field studies on Savi´s pine voles have employed either signs of presence (Bertolino et al. 2015b), pitfall or snap traps (Osella and Contoli 1986; Cagnin and Grasso 1999). Live traps, commonly employed in population dynamics studies, have been used only occasionally and for short periods. Further, information on the cost effectiveness of different trap types is not available for this species. In this article, we present the trapping method that proved most effective in capturing this extremely elusive species using the tunnel system dug by the voles.

Materials and methods

In 2013, we started a live-trapping study on Savi's pine vole in agroecosystems. A recent taxonomic revision split Savi's pine to three allopatric species (Bezerra et al. 2016). Our study focused on populations in north-central Italy, which belong to *Microtus savii s. str.*.

A first pilot trial was conducted in orchards (kiwi, peach, cherry and apricot) in region of Emilia-Romagna (Italy). Each orchard was approximately one hectare, and this species was known to be present due to the high density of holes in the ground. For each orchard, we established a trapping grid of one hectare with 40 Ugglan and 40 Longworth traps, laid on the ground, baited with peanut butter, carrots, apples and oatmeal on the bottom surface of the trap. We trapped for 10 consecutive nights in each plot of the 4 orchards, for a total of 40 trapping nights. However, our overall catch was of only two Savi´s pine voles.

We therefore started a new preliminary study in an apple orchard (42.327463 N, 11.977942 E) the following year, to determine the most effective protocol for trapping Savi's pine voles. We gathered information on the positioning and capture efficiency of three types of traps commonly used for small mammals weighing 7 to 40 grams: INRA (Institut National de la Recherche Agronomique), Longworth and Ugglan. Since we registered high mortality rates for INRA traps – with 4 dead individuals out of 15 trapped animals in two nights – we decided to activate them only during the day.

Using the information acquired, we carried out the study from April to May 2014 in apple and peach orchards in three regions of Italy: Tuscany, Emilia-Romagna and Piedmont. The study area in Tuscany is located near the town of Foiano della Chiana (hereafter A, coordinates: 43.254467 N, 11.835570 E), and it is an area relatively far away from urban settlements, with an average annual rainfall of 700 mm and mean annual temperatures between +5.8°C and +23.0°C. The only vole species found in this area is Savi's pine vole. The study area in Emilia-Romagna is located near the town of Imola (hereafter B, 44.370779 N, 11.753046 E) in a highly fragmented, predominantly rural area, with an average annual rainfall of 750 mm and mean annual temperatures between +2.6°C and +23.7°C. Savi's pine voles coexist here with common voles (*Microtus arvalis*), which is present at very low densities. The study area in Piedmont is located near the town of Saluzzo (hereafter C, 44.581134 N, 7.497749 E) in an intensive agricultural plain characterized by orchards, with an average annual rainfall of 756 mm and mean annual temperatures between +3.1°C and +22.4°C. We used 120 traps: 40 of each of the three models, INRA, Longworth and Ugglan.

The INRA trap measures 15 x 5 x 5 cm. It has a rectangular housing made of zinc. The internal parts are made of galvanized sheet metal with two axes and a bracket. Animals enter the trap via an oscillating trap door. A plastic rear door allows the extraction of the captured animal (details in Aubry 1950, Girardoux et al. 1998).

The Longworth trap consists of a nesting chamber made of aluminium, measuring $14 \times 6.5 \times 9$ cm, connected to a tunnel ($13 \times 4.5 \times 4.5$ cm), equipped at the other end with a treadle that is triggered by the entrance of the animal (details in Chitty and Kempson 1949).

The Ugglan trap is a multiple-capture wire trap ($24 \times 8 \times 6$ cm), with plastic plates on the bottom and an aluminium lid that covers the sides of the trap, protecting it from rain but allowing partial air circulation through it. A "tramp" tip-plate treadle (gravity controlled) allows the animal to reach the bait. A 5 g counterweight repositions the door, locking the animal in and resetting the trap (details in Lambin and McKinnon 1997).

We placed the traps only in front of active holes, that is, at the exits from the tunnel currently in use (Fig. 1). We placed the traps horizontally, directly inside the first section of the tunnel, so

that no light passed through them. This positioning created an artificial extension of the tunnel itself that left the vole exiting the nest no choice but to enter the trap. If necessary, we dug out some earth to better position the traps. To identify these active holes, on day 0, we closed all the holes found in trapping areas with soil, and we marked each blocked hole with a red stick. After 24 h, we identified the re-opened holes as active holes, and we placed traps only in front of these (EPPO 1975, Tkadlec and Stenseth 2001). This method is commonly used to evaluate the abundance indexes of common voles (Lisická et al. 2007). Bertolino et al. (2015b) recently used it for Savi's pine voles. The three types of traps were set in an alternating pattern to avoid a differential distribution on the ground. Each capture session lasted four days. We equipped traps with hay, to provide thermoregulation and nesting material, as well as apples and sunflower seeds as bait. We replaced the hay and food when a vole was caught in a trap,. To assess recapture rates, we marked all captured individuals by using fur clipping. We checked the traps approximately every 8 h. Because Savi's pine vole has a polyphasic daily activity rhythm (Ranchelli et al. 2016) and because we intended to gather information on night and day capture rates, Ugglan and Longworth traps were left open continuously 24 h/day, whereas INRA traps were de-activated at night to reduce mortality.

Data analysis

Considering our protocol, comparisons of all three trap types was conducted using only diurnal first captures and recaptures. Ugglan and Longworth traps were compared using 24 h trapping data. Chi-square tests were used to compare trapping data, with Yates's continuity correction with two trap types. For comparing the three models of traps, the contribution of each one to a significant Chi-square test was evaluated with the standardized residuals. Statistical analyses were performed using R software (version 3.3.1).

Results

We recorded 73, 39 and 7 capture events of Savi´s pine voles at sites A, B and C, respectively (Tab 1). We grouped the data from the study areas and compared the efficiencies of the different trap types. Considering diurnal first captures only, we found no significant difference in the capture rates of the three trap types ($\chi^2 = 0.95$, P = 0.62). By contrast, recapture rates differed significantly ($\chi^2 = 7.82$, P < 0.02), with Ugglan traps having the highest success (Ugglan standardized residuals z=2.28 significant at P<0.05, INRA and Longworth z<±1.96 not significant). Similarly, 24-h trapping data were comparable between Ugglan and Longworth for first captures (Yates's correction $\chi^2 = 0.69$, P = 0.49), but a higher recapture rate was found for Ugglan traps (Yates's correction $\chi^2 = 5.90$, P < 0.02). Recapture rates were not influenced by few trap-happy animals, since recaptures within single trapping periods increased from 19-20%

of the animals caught in the first day to 63-75% at the last day, with up to 16 different animals recaptured in a day.

Based on diurnal captures only, mortality rates were similar for Longworth (8.33%) and Ugglan (9.37%) traps (Tab 1). During the preliminary study, we verified a mortality rate of 26.67% for INRA traps when active 24 h/day; this rate decreased to 11.11% when INRA traps were deactivated during the night.

Discussion and conclusions

This is the first study that provides detailed information on trapping procedures for Savi´s pine voles and on how to maximize trapping success. To successfully trap Savi´s pine voles, it is essential to use a protocol that allows the animals to actually enter the traps, of whatever type these may be. In our experience, for the capture of Savi´s pine voles, the trap-positioning protocol may be considered even more important than the trap type itself. Placing the traps only in front of active holes maximizes the trapping success. Savi´s pine voles, in fact, dig numerous tunnels and even more exit holes, but they often leave them rather quickly. Moreover, tunnels can collapse or fall out of use. A noteworthy consequence is that regularly spaced trapping configurations (i.e., grids, transects, or mixed designs, e.g., Flowerdew et al. 2004) are not suitable for this species. Regularly spaced configurations led to an extremely low number of captures in our preliminary study.

Longworth and Ugglan traps are commonly used in field studies on small mammals worldwide (Jacob et al. 2002). They can perform with comparable success (Lambin and MacKinnon 1997) or with very different outcomes, depending on both target species and environmental conditions (Jacob et al. 2002; Ylönen et al. 2003). Anthony et al. (2005) found that Longworth traps were unsuitable for the capture of microtine rodents. For Jacob et al. (2002), it was the Ugglan type that proved much less successful, both for the target species and the environmental conditions in which it was employed. In our case, however, all three models of traps achieved similar capture efficiency for the initial trapping of animals. Despite the fact that the statistical analyses did not show a significant difference between the efficiency of INRA and the other trap types, we encountered several difficulties in their use. These difficulties suggest that they are best suited for studies where animal survival is not fundamental. The high mortality rat of INRA when left active 24 h/day is probably due to their size and to the thermal conductivity of the metal boxes. They are very narrow and for the vole to have space to enter and breathe, these traps can contain little food and even less hay, and they allow very little movement. Little food and the lack of nesting material may cause thermoregulation problems during the night. If stress level are high stress it will naturally be conducive to an increased mortality, especially if the traps are not

frequently checked. To capture live Savi's pine voles while causing as little stress as possible, these traps needed to be checked at least 6 times per day or be deactivated in case of harsh weather conditions. Longworth and INRA traps present a further disadvantage of saturation. Once the first animal is captured, they remain ineffective until they are checked (Andrzejewski et al. 1966). We have also noticed that the efficiency of Longworth traps can decrease in poor weather conditions because mud obstructs the trap trigger mechanism, thereby preventing the closure of the traps.

Ugglan traps show efficiency at first captures and have mortality rates comparable to the Longworth traps. This result is consistent with previous studies in other small mammal species (Lambin and McKinnon 1997). Ugglan traps, as long as they are equipped with nesting material, also present fewer problems of thermal insulation, both in hot and cold weather. Moreover, they have a significantly higher recapture rate. These factors make this model more efficient for studies involving the application of capture and recapture demographic models. The higher recapture rate of Ugglan traps, although resulting from few recaptured individuals, might best be explained by the lower level of stress for captured animals. Lower stress might be due to the fact that the Ugglan traps are not completely closed, while the other two models tested in the study are both completely closed.

These considerations led us to conclude that the most suitable trap type for Savi´s pine vole is the Ugglan trap. It is equally efficient in every season, has the highest recapture rate, and requires little maintenance. Additionally, it is to particularly inexpensive and allows multiple captures of live animals. These factors are all advantageous in the case of a species, which is both social and little known from the behavioural perspective such as Savi´s pine vole.

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Study area	Trap Type	First Captures		Recaptures		Total		Dead (day)	Diurnal mortality rate
		Day	Night	Day	Night	Day	Night		
A	INRA	8	/	6	/	14	/	2	14%
	Longworth	10	8	7	8	17	16	2	12%
	Ugglan	6	4	11	5	17	9	2	12%
B	INRA	3	/	1	/	4	/	0	0%
	Longworth	5	3	1	0	6	3	0	0%
	Ugglan	5	4	8	9	13	13	1	8%
C	INRA	0	/	0	/	0	/	0	0%
	Longworth	1	2	0	0	1	2	0	0%
	Ugglan	2	2	0	0	2	2	0	0%
TOTAL	INRA	11	/	7	/	18	/	2	11%
	Longworth	16	13	8	8	24	21	2	8%
	Ugglan	13	10	19	14	32	24	3	9%

Table 1: First captures and recaptures of Savi´s pine voles during day and night and diurnal mortality rate.

Figure 1. Traps placed in front of active holes, 1. Ugglan 2. Longworth 3. INRA