

SHORT COMMUNICATION

# IMPACT OF A SHORT-TIME EXPOSURE TO DIFFERENT TEMPERATURES ON WAKENING TIME OF *VERTIGO ANTIVERTIGO* (DRAPARNAUD, 1801) (GASTROPODA: EUPULMONATA: VERTIGINIDAE)

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**ABSTRACT:** I checked experimentally how a short-time exposure to different temperatures: 1 °C, 24 °C and 34 °C in a high humidity affects waking time of *Vertigo antvertigo* (Draparnaud). The snails were exposed to experimental conditions for 48 hours (each snail in a separate test tube, provided with food and a source of calcium). After that time, all the individuals were transferred to room temperature (24 °C) for counting the time needed for waking. The shortest waking time was noted for snails kept at 1 °C, the longest – for snails kept at room temperature of 24 °C.

**KEY WORDS:** temperature; *Vertigo*; laboratory experiment

Temperature is one of the most important factors which, along with humidity, affect land snail activity (e.g. COOK 2001, CAMERON 2016). Each species reacts individually to different conditions and is active in a specific range of temperatures (e.g. CAMERON 1970, KSIĄŻKIEWICZ-PARULSKA 2017). Though the reaction of land snails to specific temperatures has long attracted attention of researchers (see e.g. WARBURG 1965, CAMERON 1970, KSIĄŻKIEWICZ-PARULSKA 2020), the reaction to temperature changes has remained unexplored. In view of the climate crisis which entails extreme weather events, the knowledge of mollusc reaction to temperature changes is particularly important. Knowledge of species-specific reactions to such changes is needed to predict the effects of global warming on the activity, including reproduction traits.

This study is a contribution towards a better understanding of the effect of short-term temperature fluctuations on the behavioural pattern of *Vertigo antvertigo* (Draparnaud, 1801). The species is a member of the Vertiginidae, a family of minute land-snails. The occurrence of *Vertigo* snails has been altered by

climate changes (POKRYSZKO 2003). For these snails global warming implies habitat quality deterioration and range contraction documented in Quaternary fossils (POKRYSZKO 2003). The direct impact of climate changes on behavioural and physiological traits of *Vertigo* snails remains unknown. The hygrophilous *V. antvertigo* is a uni-habitat species associated with wetlands covered with sedge or reed beds, usually on lake shores or river banks (POKRYSZKO 1990, 2003, MYZYK 2011). It is found either in open habitats or in alder fens but never in habitats completely drying up for a part of the year (POKRYSZKO 1990). Water-dependent habitats, and thus also species which are strictly confined to wetland ecosystems, are particularly vulnerable to the current climate changes (DAWSON et al. 2003). Therefore, *V. antvertigo* may be a good example of a gastropod whose habitat and the species itself is directly threatened by increased temperatures and intense droughts.

Individuals of *V. antvertigo* for the experiments were collected from their natural habitat adjacent to an astatic forest pond (52°25'29.9"N, 16°46'24.1"E) on the 6th of April 2020 (Figs 1–2). The tempera-



Figs 1–2. Site of *Vertigo antvertigo* from which the experimental snails were collected (1), and individual snails on a sedge leaf blade at the same site (2)

tures on the day preceding the collection were 15 °C during the day and –3 °C during the night. On the day of collection the temperature raised to 21 °C (source: <https://www.accuweather.com/>). The snails were collected from leaf blades (live or dead) of *Carex acutiformis*. On the same day each individual was placed in a separate 120 ml tube with food (leaf litter from the locality), a source of calcium (dolomite dust) and water-saturated cotton-wool substrate for two-day acclimation prior to the experiment. The tubes with animals were stored in a humidified transparent plastic container (relative humidity RH ca. 100%) at room temperature (24 °C) and natural photoperiod. During the acclimation period the tubes were sprinkled with water once a day (6 p.m.) to stimulate the activity. Only adult snails with fully developed shell (with apertural armature; 4.0–5.0 whorls; [POKRYSZKO 1990](#)) were used.

Following the acclimation period, on the 8th of April, active snails (actively crawling, having fully extended foot and tentacles) were moved to 15 ml plastic test tubes (12 cm long, inner diameter 14 mm). All the experimental tubes were prepared in the following way: water-saturated cotton wool with shredded plant material on the bottom as a substratum and a 6–7 cm long leaf of *C. acutiformis* for the snails to climb up. The plant material was also a source of food during the experiment. Just before the experiment started and the tubes were closed with plastic plugs, their interior was sprinkled with water to maintain a high humidity level inside the tubes during the experimental period.

Eighty experimental individuals were exposed to experimental conditions for two days (from 8.04.2020, 10 a.m. to 10.04.2020, 10 a.m.). The

snails in the tubes were divided into three groups further exposed to different temperature regimes in climatic rooms: (1) 28 snails exposed to 1 °C, (2) 24 snails exposed to 24 °C (room temperature), (3) 28 snails exposed to 34 °C. The temperatures were chosen to reflect a relatively wide range of temperatures. The temperature change from 1 °C to 24 °C was comparable to the natural thermal regime occurring at the same time in the habitat of *V. antvertigo*. The temperature of 34 °C was found to be survivable for some *Vertigo* snails (see e.g. [KSIĄŻKIEWICZ-PARULSKA 2020](#)), and may be experienced in the wild in the spring/summer period. The transition between the experimental temperatures (from 24 °C to 1 °C and from 24 °C to 34 °C) in the climatic room took about 30–40 minutes. Natural photoperiod was maintained in the course of the experiment.

For wakening up all the snails were placed at 24 °C (room temperature). All the tubes were opened and sprinkled with water to stimulate the snails' activity. Then the time to awakening was measured. All these operations were done in the shortest possible time. It was observed that after the two-day exposure to experimental conditions all the snails were inactive.

The wakening time of *V. antvertigo* in three different temperatures was compared with one-way ANOVA randomised version with RndomPro 3.14 ([JADWISZCZAK 2009](#)). Non-parametric methods were used because of the lack of normality. Differences were considered significant at  $p < 0.05$ .

In the course of the experiment four snails died (3 from 1 °C and 1 from 34 °C), therefore the comparisons included results from 25 snails kept in 1 °C, 24 snails from 24 °C and 27 snails from 34 °C ([Table 1](#)). The analyses showed that the wakening times were

Table 1. Wakening time of each individual of *V. antvertigo* exposed to temperatures: 1 °C, 24 °C, and 34 °C as well as mean value and standard deviations (SD) for each experimental option

	Wakening time [sec]		
	1 °C	24 °C	34 °C
1	605	5,546	14,647
2	3,799	6,960	7,680
3	4,128	5,880	10,106
4	243	6,925	6,150
5	605	5,683	3,932
6	1,562	6,967	6,168
7	1,562	5,910	4,960
8	1,880	8,354	4,960
9	1,532	10,244	5,340
10	270	10,244	8,443
11	1,668	10,170	1,800
12	1,570	10,170	8,114
13	1,570	8,522	6,191
14	930	7,320	10,613
15	1,603	7,740	5,797
16	2,953	9,889	5,434
17	1,263	7,473	8,662
18	605	7,473	3,834
19	1,087	6,540	4,750
20	1,900	9,845	4,781
21	1,733	5,948	5,833
22	1,320	6,605	6,252
23	6,315	9,256	4,808
24	1,278	8,780	5,497
25	2,155		7,448
26			5,510
27			5,868
Mean	1,765.5	7,851.8	6,428.8
SD	1,333.9	1,634.5	2,520.0

significantly different between the snails exposed to different temperatures: 1 °C vs. 24 °C ( $F = 204.699$ ,  $p < 0.001$ ), 1 °C vs. 34 °C ( $F = 67.919$ ,  $p < 0.001$ ); 24 °C vs. 34 °C ( $F = 5.564$ ,  $p = 0.024$ ). The earliest wakening was noted for snails kept at 1 °C, the latest for those kept at 24 °C (Table 1, Fig. 3).

The results may suggest that *V. antvertigo*, as a cold-blooded animal, reacts to temperature changes, and the reaction is related to the temperature amplitude. The snails which were kept at 1 °C experienced the greatest temperature amplitude (23 °C) compared to those exposed to 34 °C (10 °C) or those kept at room temperature of 24 °C, which actually did not experience any temperature fluctuation. Temperature deviations from the normal activity range entail behavioural and physiological defence against thermal

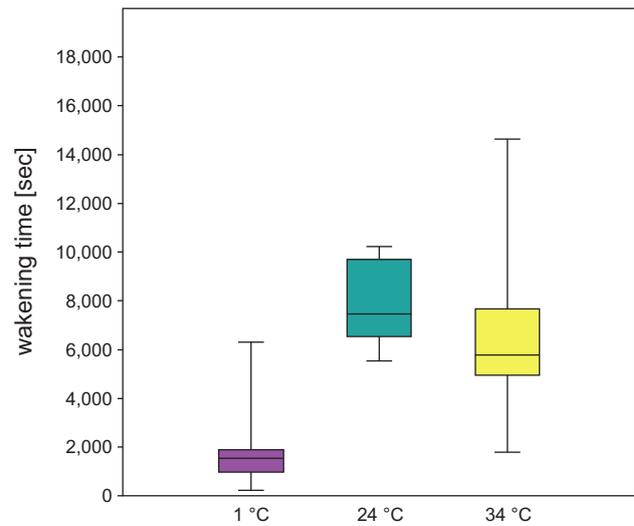


Fig. 3. Box plot showing wakening time of *Vertigo antvertigo* in experimental temperatures: 1 °C (purple box), 24 °C (turquoise box), 34 °C (yellow box). Boxes – quartiles 25–75%; horizontal line inside the box – mean; minimum and maximum values – whiskers

stress. However, the physiological processes of the snail entering into torpor are dynamic and it may take a few days to prepare the organism for this metabolic state (KOSICKA et al. 2020). Therefore, it is likely that *V. antvertigo* in the experiment did not enter torpor, but remained inactive while still ready to resume activity. Such behaviour makes it possible to retain the ability for a quick reactivation when some sporadic temperature fluctuations occur. The delay in entering torpor may save the energy needed to waken the body from dormancy or to prepare the body for torpor. It should be pointed out that the results may have been affected by the relatively abrupt temperature changes as well as air oxygen saturation and humidity variation in the test tubes. The water loss in land snails depends on both temperature and humidity (WARBURG 1965). Thus, it is possible that even a subtle change in any of the two mentioned factors, associated with exposure to different temperatures, may have affected the wakening time as well as induced the inactivity of snails observed after two days temperature exposure (especially at 24 °C). Further studies, including both field observations and laboratory experiments, are needed to understand better the snails' reaction to extreme weather events.

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