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The effect of copper barriers on pulmonate land snails

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1. Abstract

Solid copper has recently been introduced as a new and environmental-friendly repellent against slugs and snails. Still, no scientific proof is at hand to support this claim. The aim of this thesis is to investigate a possible influence of solid copper has on the behavior of pulmonate land snails and if it has a repellent effect on them, and to identify possible explanations for the origin of this effect.

Quantitative experiments in the form of an escape scenario in which the Portuguese slug (*Arion lusitanicus* (Mabille)) had to overcome barriers of soil, aluminum and different qualities of copper in order to gain food have been conducted in August 2009. Additionally, at the same time a qualitative analysis was done in order to show if the effect is also present in other mollusk species.

The results prove that copper has indeed a significant repellent effect on Portuguese slugs, with a median of the time it takes the snails to overcome the copper barrier more than 23 times as high as that it takes them without a barrier. At the same time the effect seems not to be limited to *Arion lusitanicus* (Mabille) but to be common throughout all mollusks as the qualitative analyses and other scientific work shows (Range et al., 2008).

The nature of the effect is unknown, but could be connected to the uptake of copper into the snails metabolism, an intoxication with copper compounds found in copper patinas, or a couple of less probable explanation, while further scientific research is urgently needed in this field.

Without further knowledge about the nature of the repellent effect it is difficult to say if solid copper can bear comparison with other molluscicides available. However, the formation of toxic copper compounds as copper sulphates on exposed solid copper and the washing of copper particles to the soil through precipitation lead to reasonable doubt about the sensibleness of using this method on a large scale.

2. Introduction

Recently more and more popular science magazines and internet resources propagated solid copper as a new, effective and environmental-friendly method to keep slugs and snails away from garden and crops. It was claimed that this effect could be explained with the mollusks being deterred by electricity when coming in touch with the metal. However, the validity of these assumptions was uncertain as they lacked any scientific proof.

It is the primary objective of the this work to reveal if pulmonate land snails show any change in behavior on touching solid copper, therefore showing its efficacy as a snail repellent barrier.

Secondly, in the case of an actual effect of solid copper on snails, it shall be demonstrated if the thickness of the copper sheet is of any importance, as this would be a relevant matter for the construction of barriers as well as for the comparison of the quality of available commercial solutions.

Finally, the nature of the effect of copper on mollusks is of great importance and a major factor when it comes to the comparability with other mollusk repelling solutions, much depending on the ecological consequences, and therefore shall be discussed here. And some attention shall be directed to the claim that the nature of the effect is an electrical shock.

3. Material and Methods

3.1 Quantitative analysis

These experiments were carried out in order to investigate whether there is a significant difference in the behavior of the Portuguese slug (*Arion lusitanicus* (Mabille)), a terrestrial pulmonate gastropod mollusk, when it crosses different kinds of barriers or not.

The Portuguese slug (*Arion lusitanicus* (Mabille)) was chosen as the ideal test object as it is very common throughout Europe, easy to catch, and experiments with it are not restricted by any laws or regulations. Additionally it is a common garden pest, well known by both private as well as commercial gardeners, and therefore an important aim for any commercial use of copper as an anti-snail product.

The testing area is located in an agricultural area in the community Luftenberg an der Donau in Upper Austria. The experiments were conducted in a laboratory offering constant climate conditions even in a hot summer, set up for this purpose.

The experimental set-up was based on 3 large round plastic bowls (58 cm in diameter, 7.5 cm in height) which were filled with clayey earth to a height of 2 cm. This setup was prepared one week before the start of the first experiment and kept under the same constant climatic conditions. The soil was kept moist over the whole time of the experiments.

The barriers each were made of 4 stripes (5 cm by 35 cm) of the metal, neatly taped together with insulating tape on the bottom side to form a square barrier 5 cm wide, with an outside length of 40 cm and an inside length of 30 cm. The barriers were positioned inside the plastic bowls filled with soil. The outline of the earth barrier was drawn into the soil using a ruler. Lettuce was placed outside the barriers as an incentive for the snails to cross the barrier (Figure 1).

The snails were collected between 7:30 pm and 8:00 pm the day before the experiment was carried out and kept without food in a ventilated plain plastic box under the same climatic conditions as the experiments.

The experiments started at around 7:30 pm by dropping 12 snails inside each barrier at random and ended 4 hours later.

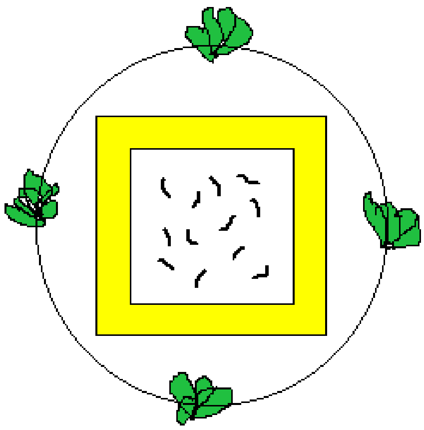

Figure 1 Scheme of the experimental set-up of the quantitative experiments.

On eight days (10th, 11th, 12th, 13th, 14th, 16th, 29th and 30th of August 2009) the following materials were tested as barriers:

- Soil (distinguished from the soil in the bowl by thin lines pressed into the earth, representing the inner and outer shape of the barrier) (Group 1)
- Aluminum 0.1 mm, AL99.5%, soft (Group 2)
- Copper 0.05 mm, SE-Cu (Cu-HCP) (Group 3)

On four other days (25th, 26th, 27th and 28th of August 2009) the following materials were tested as barriers:

- Copper 0.05 mm, SE-Cu (Cu-HCP) (Group 6)
- Copper 0.1 mm, E-Cu (Cu-ETP) (Group 4)
- Copper Paint (Eckart Metalleffektlack Classica Kupfer) applied on thin plastic sheets (Group 5)

Group	Material	Dates
1	1 Soil	10th, 11th, 12th, 13th, 14th, 16th, 29th, 30th August 2009
2	2 Aluminum 0.1 mm	10th, 11th, 12th, 13th, 14th, 16th, 29th, 30th August 2009
3	3 Copper 0.05 mm	10th, 11th, 12th, 13th, 14th, 16th, 29th, 30th August 2009
4	4 Copper 0.1 mm	25th, 26th, 27th, 28th August 2009
5	5 Copper Paint	25th, 26th, 27th, 28th August 2009
6	3 Copper 0.05 mm	25th, 26th, 27th, 28th August 2009

Table 1 Materials of the 6 groups and the days they were tested on.

The experiments were recorded using a video camera (Canon MV X330i) mounted on a tripod (Manfrotto MA 190 XPROB) and adjusted adequately. During the 4 hours of an experiment the video cassettes (miniDV, 90 minutes with long play) had to be changed twice, which was done within 10 seconds at most.

The following information was gathered:

- Weather condition (1 = sunny, 2 = clouded, 3 = rainy) on the day of collecting and the day of the experiment
- Indoor and outdoor temperature on the day of collecting (7:30 pm) and the day of the experiment (2:00 pm and 7:30 pm outdoors, 2:00 pm and during the whole experiment indoors)
- Indoor humidity (2:00 pm and during the experiments)
- Barometric pressure (from the 16th of August on, at 2:00 pm and 4 times during the experiment, from the 25th of August on always at 7:30 pm at the day of collecting the mollusks)

Temperature and humidity were measured using a Conrad Wetterstation 6802 weather station. The barometric pressure was detected with a Garmin eTrex Vista HCx GPS receiver with a barometric pressure sensor.

After the experiment the snails were released at a site far enough from the collecting area to ensure no snail would be used twice. The animals were handled using clean unpowdered latex gloves at all times.

The videos were dubbed to a computer and saved as AVI-video files (MPEG 4-codec, 720x576 pixels). Each day was analyzed and the following information was gathered for each snail:

- Drop Time (DT): The time the snail was placed inside the barrier.
- Start Movement Time (SM): The time the snail started movement.
- Change Position time (CP): The time the snail moved out of the space it originally occupied when it was dropped.
- Reach Barrier Time (RB): The time the snail touched the barrier.
- Cross Barrier Time (CB): The time the snail left the barrier outbound with its whole body.

From the statistical analysis excluded were animals which

- did not reach the barrier within 60 minutes,
- started to mate during the time of the experiment,
- managed to cross under the border instead over it or
- experienced other problems influencing their behavior.

3.2 Qualitative analysis

In the qualitative analysis it was tested whether different mollusk species showed the same behavior as the Portuguese slug (*Arion lusitanicus* (Mabille)) in the quantitative analysis.

Therefore a small copper plate (0.05 mm Copper, SE-Cu (Cu-HCP), 5 cm by 12 cm) was placed on an isolating glass top situated on a table.

The mollusks were dropped 3 cm before the copper barrier, with lettuce on the other side of the barrier as an incentive to cross (Figure 2). In case the snails decided to go other directions as towards the barrier or did not move at all, the experiment was repeated until that individual reached the barrier.

The behavior of the mollusks upon reaching the barrier was noted down. The following information was gathered:

- Species
- Number of tries until the individual reached the barrier
- Individual showed special behavior (1 = yes, 0 = no)
- Individual crossed barrier (1 = yes, 0 = no)
- Time and date
- Temperature (with a Conrad Wetterstation 6802 weather station)
- Humidity (with a Conrad Wetterstation 6802 weather station)

The following mollusk species were tested in the experiment:

1. *Cepaea hortensis* (Müller), White-lipped snail 12 specimen
2. *Fruticicola fruticum* (Müller) 12 specimen
3. *Arion lusitanicus* (Mabille), Portuguese slug 2 specimen
4. *Arianta arbustorum* (Linnaeus) 1 specimen
5. *Oxyloma elegans* (Risso) 3 specimen

The experiments were recorded using a video camera (Canon MV X330i) mounted on a tripod (Manfrotto MA 190 XPROB). The videos were dubbed to a computer and saved as AVI-video files (MPEG 4-codec, 720x576 pixels).

The effect shown by snails on encountering the copper can be described as follows:

- The snail raises its head above the level of its body as if looking around.
- The snail tries to minimize its contact area with the copper.
- The snail retreats to its shell if it has one.

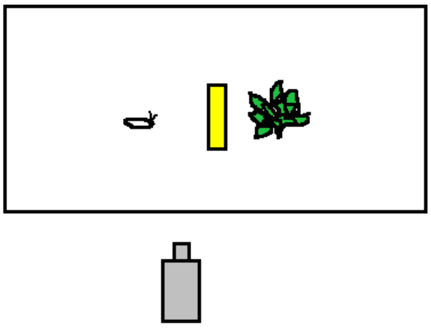

Figure 2 Scheme of the experimental set-up of the qualitative experiments.

3.3 Electrical conduction

This experiment was carried out in order to measure a possible electrical conduction between the snail and a copper surface.

The snails (Portuguese slug, *Arion lusitanicus* (Mabille)) were placed in the middle of copper sheets (0.05 mm, SE-Cu (Cu-HCP)) the size of 11.5 by 20 cm (230 cm^2). The voltage and amperage were measured immediately after dropping the animal and a second time after 3 minutes, always between the metal (about 1 cm away from the animal) and the snail itself (caudal end of the mantle) using a digital multimeter (Voltcraft Digital-Multimeter VC170).

After every use the electrodes of the multimeter and the metal plates were cleaned with pure alcohol in order to remove snail mucus.

3.4 Statistical analysis

Statistics were performed using modules of the statistics package SPSS v. 17.0.

The testing of possible differences in the daily activity was performed using a Kruskal-Wallis H test with significance levels at *0.05, **0.01, ***0.001.

The analysis of differences in the cross-barrier-time of different materials was conducted using the Mann-Whitney U test. The significance levels were corrected for multiple comparisons with the Dunn-Sidak correction (Sokal and Rohlf, 1994). The adapted significance levels for 4 comparisons (Soil, Al 0.1 mm, Cu 0.05 mm) are *0.01274, **0.00251, ***0.00025 and the adapted significance levels for 2 comparisons (Cu 0.05 mm, Cu 0.1 mm, Cu Paint) are *0.02532, **0.00501, ***0.00050.

The effectiveness of copper as a mollusk repellent barrier was demonstrated by comparing the medians of the Cross Barrier Times of soil and copper.

All charts were created with SigmaPlot v. 11.0.

4. Results

4.1 Quantitative analysis

4.1.1 Day-dependent activity

Before comparing the different groups and therefore different materials it was necessary to test whether the eight days (groups 1, 2, 3) and four days (groups 4, 5, 6) of the quantitative analyses could be treated as a whole, or if the groups had to be compared separately for each day.

		Crossing Time			Drop-Reach Barrier Time		
	Groups	**Asymp. Sign.**	**N**	**df**	**Asymp. Sign.**	**N**	**df**
1	**Soil**	,041*	76	7	,043*	76	7
2	**Alu 0,1**	,050	84	7	,040*	84	7
3	**Cu 0,05 123**	,000***	89	7	,299	89	7
4	**Cu 0,1**	,011*	38	3	,738	38	3
5	**Cu Paint**	,993	40	3	,473	40	3
6	**Cu 0,05 456**	,001**	28	3	,331	28	3

Table 2 Significance of differences in the day-dependent activity rates in the same groups using a Kruskal Wallis H test (significance levels at *0.05, **0.01, ***0.001).

The comparison of the Drop-Reach Barrier Time (table 2) showed that only group 1 (soil) and group 2 (aluminum 0.1 mm) showed significant differences in the daily activity (Kruskal Wallis H test, $P < 0.05$, $n = 364$). On comparing the crossing time of the barriers itself, groups 1 (soil), 3 (Cu 0.05 mm), 4 (Cu 0.1 mm) and 6 (Cu 0.05 mm) showed significant differences (Kruskal Wallis H test, $P < 0.05$, $n = 364$).

There are significant discrepancies in the daily performance of the mollusks in the experiment. Therefore it was necessary to analyze each day separately.

4.1.2 Comparison of soil, aluminum and copper

For each day the Cross Barrier Times of the three groups (1 soil, 2 aluminum 0.1 mm, 3 copper 0.05 mm) were compared to each other using a Mann-Whitney U test for non-parametric data.

Date	1 Soil : 2 Alu Asymp. Sig. 2-tailed	1 Soil : 3 Cu 0.05 Asymp. Sig. 2-tailed	2 Alu : 3 Cu 0.05 Asymp. Sig. 2-tailed
10.08.2009	0.00291*	0.00023***	0.00911*
11.08.2009	0.02364*	0.00041**	0.03706
12.08.2009	0.01067	0.00020***	0.00064**
13.08.2009	0.07665	0.00025**	0.00398*
14.08.2009	0.28915	0.00218**	0.24875
16.08.2009	0.13708	0.00135**	0.26986
29.08.2009	0.15040	0.00020***	0.00095**
30.08.2009	0.01238*	0.00025***	0.00234**

Table 3 Significance of differences in the Cross Barrier Time CB for the groups 1, 2 and 3. Significance levels were corrected after Dunn-Sidak for multiple comparison (Sokal and Rohlf, 1994) to 0.01274*, 0.00251**, 0.00025***.

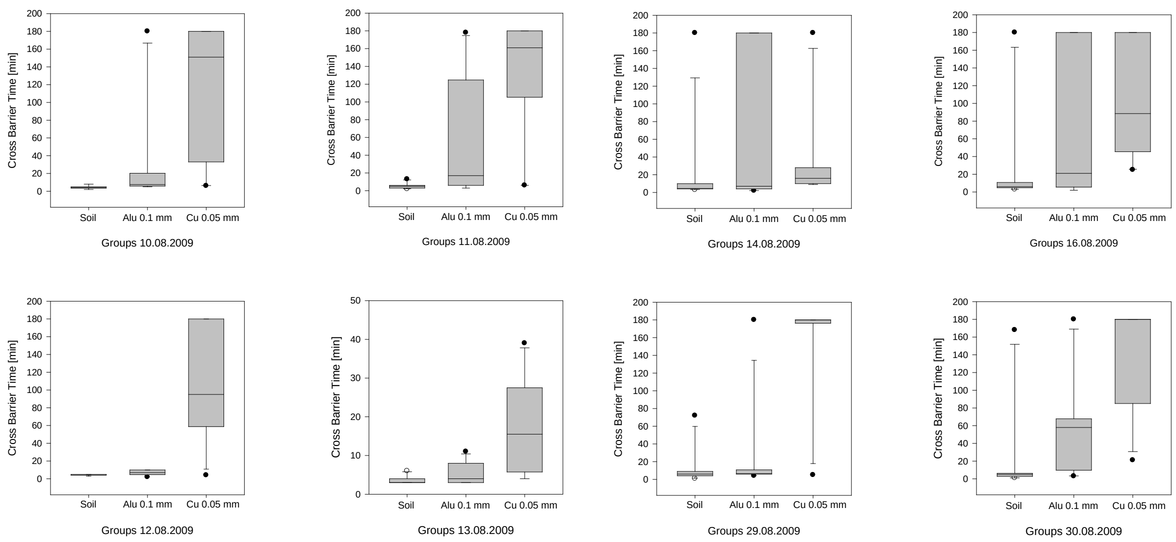

Fig. 3 Box plot graphics of the comparison of soil, aluminum and copper for each of the 8 days.

Concerning the comparison of the Cross Barrier Time of soil and copper the results show a clear result, with 8 out of 8 days showing a highly significant difference (Mann Whitney U test, $P < 0.01$, total $n = 174$) in the time it takes the mollusks to cross the barrier. At the same time the median of the crossing times of copper is 23.65 times higher than that of pure soil (median soil = 4, median copper 0.05 mm = 111), providing a suitable measure for its efficacy.

Date	1 Soil	2 Alu	3 Cu	Ratio Soil : Cu
10.08.2009	4	7	151	37,75
11.08.2009	5	17	161	32,20
12.08.2009	4	7	95	23,75
13.08.2009	3	4	16	5,17
14.08.2009	5	7	16	3,56
16.08.2009	6	21	89	14,75
29.08.2009	6	7	180	30,00
30.08.2009	5	58	180	36,00
TOTAL	**5**	**16**	**111**	**23,65**

Table 4 Medians of the Cross Barrier Time for the groups 1, 2 and 3.

The comparison of the Cross Barrier Time of soil and aluminum did not show a clear trend, with 3 out of 8 days showing a significant difference (Mann Whitney U test, $P < 0.05$, total n = 169).

Comparing the Cross Barrier Time of aluminum and copper did show a trend in difference, with a significant difference on 5 of 8 days (Mann Whitney U test, $P < 0.01$, total n = 172).

4.1.3 Comparison of copper of three different qualities

Comparing 0.05 mm to copper paint did not show any significant results (Mann Whitney U test, $P < 0.05$, total n = 68). Comparing 0.05 mm copper to 0.1 mm copper (Whitney U test, $P < 0.05$, total n = 66) did not show a trend for a significant difference with only one significant result.

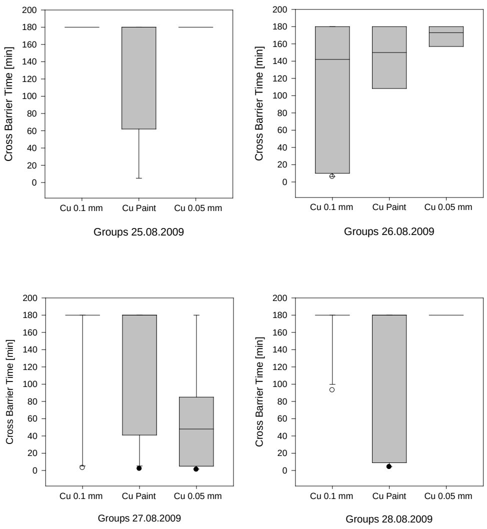

Fig. 4 Box Plot graphics for the Cross Barrier Time of Cu 0.1 mm, Cu Paint and Cu 0.05 mm for each of the 4 days of testing.

Date	4 Cu 0.1 : 6 Cu 0.05	5 Cu Paint : 6 Cu 0.05
	Asymp. Sig. 2-tailed	Asymp. Sig. 2-tailed
25.08.2009	1.00000	0.13163
26.08.2009	0.12778	0.71439
27.08.2009	0.01870*	0.09238
28.08.2009	0.28165	0.12279
GESAMT	0.26764	0.91807

Table 5 Significances of a difference in the in the Cross Barrier Time CB between groups 4 and 6 and 5 and 6. Significance levels corrected after Dunn-Sidak for multiple comparison (Sokal and Rohlf, 1994) to 0.02532*, 0.00501**, 0.00050***.

4.2 Qualitative analysis

Out of a total of 29 pulmonate land snails of 5 different species only one individual of the species *Cepaea hortensis* (Müller) did not show any effect on contact with the copper barrier. (This individual was then tested again, did not approach the barrier on the second try, but touched the barrier on the third try without crossing it, but with showing a change in behavior.)

Out of 28 mollusks which showed a behavioral effect on contact with the copper barrier 24 specimens did not cross the barrier. 4 individuals of the species *Cepaea hortensis* (Müller) showed were deterred but still crossed the barrier.

Spec-No	Species	Total number	SE+CB	SE+NC	NE+CB	NE+NC
1	Cepaea hortensis	11	4	6	1	0
2	Fruticicola fruticum	12	0	12	0	0
3	Arion lusitanicus	2	0	2	0	0
4	Arianta arbustorum	1	0	1	0	0
5	Oxyloma elegans	3	0	3	0	0
	TOTAL	**29**	**4**	**24**	**1**	**0**

Table 6 Number of mollusks which on first encountering the copper barrier showed the effect (SE) and did not show the effect (NE), combined with the mollusks which crossed the barrier (CB) and did not cross the barrier (NC).

4.3 Electrical conduction

On dropping down the mollusks (*Arion lusitanicus* (Mabille)) onto a copper sheet a mean voltage of 156.6 mV was observed, thus no amperage could be detected (standard deviation = 52.38, median = 145, n = 15). After the slugs spent 3 minutes on the copper surface the multimeter showed a mean voltage of 46.0 mV and still no amperage (standard deviation = 22.38, median = 44.5, n = 15) (table 8, page 27).

Therefore no electrical conduction could be proven in this experiment.

5. Discussion

5.1 Quantitative analysis

Study design

Schüder et al., 2003, performed a similar study about the efficacy of different slug repellents, including solid copper. They had chosen the amount of leaf damage caused by the mollusks as the key factor for the comparison of different mollusk repellents. Although it is less time-consuming to record than the drop, start movement, change position, reach barrier and cross barrier times for each snail in a large-scale experiment, this set-up should be treated with great care as the quantity of lettuce eaten by the snail depends on its size and appetite, and on how early in the experiment it had crossed the barrier. Still, in this experiment copper was the third most effective repellent after garlic and ureaformaldehyde, and at the same time showed a mortality rate of 5 % (slugs after 48 hours of exposure, snails after 168 hours of exposure).

In this study the time it takes the snail from encountering the barrier to actually crossing it (Cross Barrier Time CB) is taken as the key variable to compare the efficacy of different barriers as slug repellents. This minimizes the influence of other factors and therefore is an adequate tool for any comparison of possible slug-repellent materials. Therefore an escape scenario had been chosen, in which the snail has nearly no other possibility than to confront the barrier.

It may be said that this experimental design does not reflect natural conditions, but it was to be investigated whether copper is a repellent at all, and not to show its exact efficacy under semi-natural conditions as found in gardens and crops.

Day-dependent activity rates

On testing the possibility of different day-dependent activity rates of the mollusks, significant differences were shown both in the Drop-Reach Barrier Time (RB) and the Cross Barrier Time (CB). How they did occur it can only be speculated about. The animals had been caught, handled and kept under the same conditions and with the same care at all times. Climate conditions in the laboratory also remained constant over the course of the experiments. Therefore it can only be assumed that the condition of the Portuguese slugs when being caught was different and so influenced the experiment. The nature of this difference is still unknown, but could be in connection with fluctuations in the micro-climate or changes in their daily rate of food uptake in the days before collecting. Due to the differences in the daily activity rate the quantitative comparison of soil, aluminum and copper was conducted for each day separately.

Comparison of soil, aluminum and copper

The quantitative analysis showed a clear trend that copper is indeed a strong repellent against mollusks, confirming popular science as well as former studies (Schüder et al., 2003; Hata et al., 1997) and answering the main question of this thesis.

Aluminum, when compared to soil, actually did show significant differences in the Cross Barrier Time on 3 out of 8 days, which cannot be understood as a trend. Therefore a repellent effect of this material could not be proved. This goes along with Schüder et al., 2003, where aluminum also did not show a significant repellent effect against snails and slugs.

Comparison of different qualities of copper

No significant difference could be shown between the 0.05 mm high copper sheets used in comparison to soil and aluminum, and 0.1 mm copper sheets of double the thickness. This leads to two possibilities. Either the difference in thickness between the two copper sheets is not relevant enough, or the repellent effect of copper is simply not dependent on the thickness of the copper surface. The first case would require further experiments, ideally with ultra-thin copper sheets. Nevertheless, as commercial slug tapes, made of very thin copper sheets due to the high price of the material, also seem to work, the second case seems to be more likely.

In the same experimental set-up copper paint was tested against the noted 0.05 mm copper, but did not show a significant difference in the Cross Barrier Time on any of the four days. It can be assumed that copper paint is in fact a repellent against slugs, though it is unknown whether this is due to the copper particles contained, or to a repellent effect of the paint itself. In a study by Range et al., 2008, it was proven that commercial copper paint used for harbor areas had a strong repellent effect on limpets.

The nature of the repellent effect

As the quantitative analysis proves that copper indeed leads to behavioral changes in pulmonate land snails and acts as a strong repellent, it would now be important to know the nature of the phenomenon in order to investigate its suitability for outdoor application.

The only answer offered by popular science so far, the theory of a electrical shock, could not be verified in the experiments.

Therefore a number of possible causes shall be discussed here:

<u>Uptake of toxic copper particles</u>

As shown by previous studies, mollusks take up copper not only through ingestion (Berger et al., 1989), but also directly through the foot, at least in the form of copper compounds (Ryder et al., 1977).

Another study demonstrates the lethal effect of ingested copper on the snail *Theba pisana* (Müller), with an 48-h LD_{50} value of 37.88 µg/snail, leading to an increase in lipid peroxidation and a decline of glutathione in the digestive gland (Radwan et al., 2009; El-Gendy et al., 2009). This leads to the conclusion that copper particles taken up through the foot have a toxic effect on the snail, thus triggering a behavioral change and the avoidance of copper surfaces.

<u>Toxic reaction to the copper patina</u>

Another interesting possibility is the uptake of components of the copper patina through the foot of the snail. As shown by Ryder et al, 1977, the snail's foot is indeed an area of uptake for copper sulphates.

At the same time copper patinas can contain hydrated copper sulphate (posnjakite, $Cu_4SO_4(OH)_6.H_2O$), brochantite ($Cu_4SO_4(OH)_6$), cuprite (Cu_2O) and nantokite ($CuCl$) when formed in urban, rural or coastal areas, even after an exposure of only one month (Watanabe et al., 2001; Watanabe et al., 2006; Watanabe et al., 2009).

A comparison of different copper compounds showed their effectiveness as molluscicides when ingested, with 48-h LD_{50} values of 26.54 µg/snail for copper sulphate, 334.54 µg/snail for copper hydroxide and 582.18 µg/snail for copper oxychloride on *Theba pisana* (Müller) (El-Gendy et al., 2009).

Therefore an appropriate conclusion would be that the snail takes up copper sulphate and/or other copper compounds through the foot on contact with the patina, experiencing its toxic or even lethal effect and ultimately tries to avoid any further contact.

Although this explanation seems to be quite probable at first sight, it has to be taken into account that snails are also deterred by copper surfaces which seem to have not yet formed a patina, which would contradict this theory.

Influence of copper on the molluscan mucus

Snail mucus is a very diverse mixture of different compounds, although the exact composition is yet unknown. (Deyrup-Olsen et al., 1983; Pawlicki et al., 2004) Many mollusk species have to alternate between an adhesive version of their mucus, the glue mucus, and a non-adhesive version, the trail mucus. The first one they use to glue themselves to surfaces, whereas the trail mucus is used to glide along even surfaces. As shown in a study on the role of metals in the mucus copper as well as the other heavy metals zinc, iron and manganese can be found in molluscan mucus in surprisingly high concentrations. (Werneke et al., 2007) The idea is that these metals play an important role in the ability of glue proteins to stiffen the gel, either by catalyzing a chemical reaction or by being part of the binding of the glue proteins itself.

This theory is supported by a recent study showing that metal chelatisation disrupts molluscan gel dynamics, therefore proving that heavy metals play indeed an important role in the gel dynamics (Smith et al., 2008).

Carrying forward this idea, it could be possible that copper ions taken up from the copper sheet influence the gel mechanics of the molluscan mucus, consequently triggering a change in behavior of the mollusk which is losing control over its locomotion ability.

Copper-induced osmotic stress

As shown by Davies et al., 1998, molluscan mucus contains mostly water, which is an important resource for the snail, having great influence on its behavior. Besides the toxic effect of the copper patina it may also be possible that copper chlorides (Rickett et al., 1995), urates (Bernardi et al., 2009) and other compounds present in the patina lead to a dehydration of the mucus. This would subsequently bring great stress to the animal and influence its behavior as it would try to minimize contact with the dehydrating substrate.

Electrostatic charge

Although testing the voltage and amperage of the snails on solid copper did not show any results it could still be possible that the deterring effect is generated by an electrical conduction. Here the first theory would be that there are electric charges between the copper surface and the foot of the snail which could not be detected with a standard multimeter. The second theory includes a small charge created by a potential electrostatic charging of the copper. Still, at the moment no more than speculation can be offered on these possibilities.

5.2 Qualitative analysis

The qualitative experiment was conducted as it was neither reasonable to include other species than the Portuguese slug (*Arion lusitanicus* (Mabille)) in the quantitative analysis nor was it possible to repeat this analysis for other species in the course of this thesis. Nevertheless, the question remained whether the deterring effect shown was limited to the Portuguese slug. Therefore a behavioral analysis was chosen as the right instrument to test the assumption that the effect is not restricted to one species, but can also be observed in other terrestrial mollusk species.

The species *Cepaea hortensis* (Müller) and *Fruticicola fruticum* (Müller) were chosen as they are both common in Austria, but prefer different habitats than the Portuguese slug. Being at hand at the time of the experiments, 3 representatives of the species *Oxyloma elegans* (Risso) and 1 of the species *Arianta arbustorum* (Linnaeus) were also included in the analysis.

While the other mollusk species showed no special behavior when put on the glass plate and began to move quite soon, one of the Portuguese slugs stayed inactive most of the time. Even when the experimental set-up was changed to include a plastic plate instead of the glass one, assuming that the behavior of the Portuguese slugs may have been affected by that material, this species did not show any better results. Therefore it was not possible to include more than two individuals in the analysis. This still proved was no problem as the efficacy of copper as a repellent barrier to *Arion lusitanicus* (Mabille) had without doubt been proven in the quantitative analysis.

As the results show, out of all snails tested, only one animal of the species *Cepaea hortensis* (Müller), the White-lipped snail, did not exhibit any changes in behavior on first encountering the copper plate and crossed it right away. Still, all white-lipped snails were observed to be swift movers, 4 other animals out of 12 of this species had crossed the barrier. Although clearly being affected by the copper they did not perceive it as an irritation before having crossed the barrier so far that could easily escape to the other side. Hence it was possible that it was the very same with this animal it was decided to test it again. In the third run it actually was affected by the metal surface, not having touched the barrier in the second.

Therefore all 30 snails of 5 species were affected by the copper barrier and did show special behavior, which is a clear sign that this is a common effect throughout all terrestrial mollusk species.

5.3 Usage of solid copper as snail repellent

As the quantitative and qualitative experiments show, solid copper is a strong repellent against the Portuguese slug and other snail species. Still, whether it is a competitive and sensible alternative to traditional repellents or not remains an important question, as from the unknown ecological consequences environmental problems may arise.

The greatest issue at the moment is without doubt the unknown nature of the effect that copper proved to have on snails. As stated in chapter 5.1 there is a wide range of possible causes, and without knowing which one of those is ultimately responsible for this great efficacy of copper as a repellent it is quite dangerous to actually use it. Possible consequences might include the spread of dangerous copper compounds in the environment. The same could also greatly influence the mollusks' fitness when absorbed by the snail, special attention should be given to a possible lethal effects, all posing a considerable threat to endangered mollusk species. Another problem is that copper oxidizes rather fast, and the contents of its patina, a mixture of copper chlorides, copper sulphides and copper carbonates, could easily get into the surrounding environment (Watanabe et al., 2001, Watanabe et al., 2006, Watanabe et al., 2009, Athanasiadis et al., 2009, Afonso et al., 2009). Especially pollutants like nitrogen dioxide (NO_2) and sulfur dioxide (SO_2) have a strong influence on the formation of the patina and its composition (Rickett et al., 1995).

Two effects should also be taken into consideration: First, rain might wash copper particles from an newly exposed copper surface to the ground, this being called the first flush effect (Athanasiadis et al., 2009), with copper run-off being a widespread existing phenomenon (Odnevall Wallinder et al., 2007). Second, the part of the copper barrier in contact with the soils also corrodes, the level of corrosion depending on chloride concentration, oxygen availability in and acidity of the soil and possibly also on other factors (Afonso et al., 2009).

These arguments bring forward another important issue: the impact of copper corrosion on the effectiveness of the barrier as a snail repellent. It was not possible to include that question into this thesis, but a small-scale test with only a couple of Portuguese slugs and pieces of corroded copper, conducted out of curiosity, hinted that there may be no decline in the effectiveness at all. It might even be possible, depending on the nature of the effect of copper on the mollusk metabolism, that the patina could in fact increase the effectiveness of the metal. Still, nothing more should be speculated without proper research.

Another problem, at least at present, is the high price of solid copper, which may disqualify it as the means of choice for private as well as commercial use. Nevertheless, there are some commercial products available, spearheaded by adhesive copper tapes, 3 cm in width and mostly 5 meters in length, intended to be applied to flowerpots and tree trunks. Here certain problem may arise.

First and foremost the narrow width of most of these products, most times 3 cm, could prove to be inadequate for the use against common garden slugs. Especially larger species like the Portuguese slug (*Arion lusitanicus* (Mabille)), reaching lengths of 12 centimeters and more, or the Roman snail (*Helix pomatia* (Linnaeus)) with up to 10 cm, might have less problems overcoming this barrier than a wider one.

Furthermore, as the qualitative analysis showed, fast moving species like *Cepaea hortensis* (Müller), the white-lipped snail, being able to pass a 5 cm barrier, could perceive the barrier to be a problem only after having already crossed it. Still, further research is needed here.

Then the durability of the copper tape as a whole is directly connected to that of the adhesive. That could presumably be restricted to only a couple of years, depending on local climate.

Another commercial product is the, as it is often called, slug & snail mat, which is made of a black fabric impregnated with copper. Although cheaper and more suitable for large-scale application, this one could prove to be rather harmful to the environment, releasing fine copper particles and copper patina particles into the environment. Here also more research is absolutely necessary.

6. Appendix

6.1 Tables

Day	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
10.08.2009	9	10	11			
11.08.2009	11	10	10			
12.08.2009	11	10	12			
13.08.2009	11	12	12			
14.08.2009	12	11	11			
16.08.2009	10	9	10			
29.08.2009	11	12	12			
30.08.2009	10	10	11			
25.08.2009				5	9	4
26.08.2009				11	8	7
27.08.2009				11	11	11
28.08.2009				11	12	6
Total	**85**	**84**	**89**	**38**	**40**	**28**

Table 7 Number of test objects valid on each day in each group in the quantitative analyses

Try	Cu 0.05 mm 0 min amperage [A]	Cu 0.05 mm 0 min voltage [mV]	Cu 0.05 mm 3 min amperage [A]	Cu 0.05 mm 3 min voltage [mV]
1	0.0	251.4	0.0	48.3
2	0.0	169.3	0.0	44.6
3	0.0	145.0	0.0	13.8
4	0.0	139.1	0.0	26.7
5	0.0	114.4	0.0	57.3
6	0.0	91.4	0.0	45.9
7	0.0	140.1	0.0	31.8
8	0.0	168.2	0.0	38.0
9	0.0	148.8	0.0	84.2
10	0.0	119.8	0.0	44.5
11	0.0	295.4	0.0	100.4
12	0.0	155.5	0.0	36.6
13	0.0	143.9	0.0	40.7
14	0.0	148.7	0.0	55.0
15	0.0	118.2	0.0	21.8
Mean	0,0	156,6	0,0	46,0
Median	0,0	145,0	0,0	44,5
Std. Dev.	0.0	52.38	0.0	22.38

Table 8 The amperage and voltage between Portuguese slug and a copper sheet immediately after dropping it down and after three minutes of exposure to the copper surface.

6.2 Zusammenfassung (abstract in German)

Massives Kupfer wurde als neues und umweltfreundliches Abwehrmittel gegen Schnecken vorgestellt. Trotzdem gibt es keinen wissenschaftlichen Beweis um diese Behauptungen zu unterstützen. Das Ziel der vorliegenden Arbeit ist es aufzuzeigen ob Kupfer wirklich einen Einfluss auf das Verhalten von Landlungenschnecken hat und ob es einen abweisenden Effekt auf sie hat, und Erklärungen für die Ursache des Effekts zu finden.

Quantitative Experimente in der Form eines Ausbruch-Szenarios, in welchem die Spanische Wegschnecke (*Arion lusitanicus* (Mabille)) Erd-, Aluminium- und Kupfer-Barrieren verschiedener Qualität überwinden muss um an Nahrung zu kommen, wurden im August 2009 durchgeführt. Zusätzlich wurde eine qualitative Analyse gemacht um zu zeigen, dass dieser Effekt nicht auf eine Molluskenart beschränkt ist.

Die Ergebnisse zeigen, dass Kupfer wirklich einen signifikanten abweisenden Effekt auf die Spanische Wegschnecke, mit einem Median der Zeit, die die Schnecke zum Überwinden der Barriere braucht, fünfzehn mal so hoch als ohne Barriere. Gleichzeitig scheint der Effekt nicht auf *Arion lusitanicus* (Mabille) beschränkt zu sein, sondern über alle Mollusken hinweg verbreitet zu sein, wie die qualitative Analyse und eine wissenschaftliche Studie (Range et al., 2008) zeigen.

Die Ursache des Effekts ist noch unbekannt, könnte aber in Verbindung mit der Aufnahme von Kupfer in den Metabolismus der Schnecke stehen, mit einer Vergiftung mit Kupferverbindungen die in der Kupfer-Patina vorkommen, oder einigen weniger wahrscheinlichen Erklärungen, weswegen weitere Forschung auf diesem Gebiet dringend notwendig ist.

Ohne Kenntnisse über die Natur des abweisenden Effekts ist es schwierig zu sagen inwieweit Kupfer mit anderen Molliskoiden vergleichbar ist. Aber die Bildung von giftigen Kupferverbindungen wie etwa Kupfer-Sulfaten an exponiertem Kupfer und das Auswaschen von Kupferpartikeln in den Boden durch Niederschlag führen zu berechtigtem Zweifel daran ob es vernünftig ist diese Methode in größerem Umfang anzuwenden.

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6.4 Curriculum Vitae

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