Sveriges lantbruksuniversitet
Swedish University of Agricultural Sciences

Faculty of Landscape Architecture, Horticulture
and Crop Production Science

Control strategy of a pest slug, *Arion lusitanicus*, by using non-toxic Ferramol

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Degree Project • 15 credits
Alnarp 2017

Control strategy of a pest slug, *Arion lusitanicus*, by using non-toxic Ferramol

Kontrollstrategi för spansk skogssnigel, *Arion lusitanicus*, med hjälp av non-toxisk Ferramol

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Credits: 15 HEC
Project level: G2E
Course Title: Degree Project for the Horticultural Management Programme
Course Code: EX0365
Subject: Biology

Place of Publication: Alnarp
Year of Publication: 2017
Cover Art: https://desinsectador.files.wordpress.com/2013/09/arion
Online Publication: http://stud.epsilon.slu.se

Keywords: *Arion lusitanicus*, Iberian slug, mollusk, biological control, life cycle, Ferramol, *Pterostichus melanarius.*

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ACKNOWLEDGEMENTS

This bachelor thesis was performed in the frame of Erasmus student exchange between Swedish University of Agricultural Sciences and Universidad Politécnica de Madrid. Thanks for the opportunity given to me.

I would like to thank my thesis supervisor Lotta Nordmark of the Faculty of Landscape Architecture, Horticulture and Crop Production Science, for making this study possible. In addition, to thank their patience, time and dedication.

I am grateful to Birgitta Svensson and the staff of the farm Höjebromölla bärodling for helping and providing the necessary material to carry out this study.

Finally, a special thanks to my family for providing me with unfailing support and continuous encouragement throughout.

Patricia Galán.

ABSTRACT

Arion lusitanicus has become one of the major pests of Sweden, both in agriculture and gardening. Its high capacity to feed and reproduce makes this slug a difficult pest to control. Molluscicides have been used as control agents, but they have harmful effects on crops. As alternative, a non-toxic molluscicide, Ferramol, which acts by inhibiting the slug feed, were tested in this study. The efficacy of Ferramol ($FePO_4$) in a strawberry field, located in the periphery of Lund, Sweden was studied. The application was adjusted to the lifecycle of *A. lusitanicus*. In the spring when slugs emerged, Ferramol was applied to control the attack on the crop plant, strawberry. For a month and half the effectiveness of different doses of this product was followed, obtaining clear results as a decrease of the number of slugs. Doses of 2.5 g/m², 5 g/m² and 2 * 2.5 g/m² were equally efficient and the beetle fed on the slug eggs that have survived Ferramol, will be introduced in late spring early summer. To reach even more efficient *A. lusitanicus* control, a study using Ferramol in combination with a natural predator, a carabid beetle *Pterostichus melanarius*, is suggested.

Arion lusitanicus se ha convertido en una de las plagas más importantes de Suecia, tanto en agricultura como en jardinería. Su apetito voraz y gran capacidad reproductiva hacen de esta babosa una plaga difícil de controlar. Los molusquicidas se han utilizado como método de control, pero tienen efectos nocivos para los cultivos. Como alternativa se ha probado en este estudio un molisquicida no toxico, Ferramol, que actúa inhibiendo la alimentación de las babosas. Se estudió la eficacia de Ferramol ($FePO_4$) en un cultivo de fresas situado en la periferia de Lund, Suecia. La aplicación se ajustó al ciclo de vida de *A. Lusitanicus*. En primavera, durante la emergencia de las babosas, Ferramol fue aplicado para controlar el ataque al cultivo de fresas. Durante mes y medio se estudió la efectividad a diferentes dosis de este producto obteniendo claros resultados en la disminución del número de babosas. Las dosis de 2,5g/m², 5g/m² y 2*2,5g/m² son igual de eficientes. A finales de primavera principios de verano se introducirá el escarabajo que se alimentará de los huevos de las babosas que hayan sobrevivido al Ferramol. Para llegar a un control aun más eficaz se sugiere un estudio con Ferramol en combinación con un depredador natural, un escarabajo carabideo *Pterostichus* melanarius.

Arion lusitanicus har under senare tid framträtt som ett utav de främsta skadedjuren i Sverige, både inom jordbruk- och trädgårdsnäring. Snigelns förmåga att inta stora mängder föda och reproducera sig snabbt gör den svår att kontrollera. Tidigare har bekämpningsmedel mot blötdjur använts, men detta ger en ogynnsam effekt på grödor. Som ett alternativ testades ett giftfritt bekämpningsmedel mot blötdjur, Ferramol, som hindrar snigeln från att fortsätta äta. Ferramolets ($FePO_4$) effektivitet studerades på en jordgubbsplantering utanför Lund, Sverige. Appliceringen anpassades till *A. lusitanicus* livscykel. När snigeln framträdde på våren applicerades Ferramol för att stävja attacken på jordgubbsplantorna. Under en och en halv månad följdes effektiviteten av olika doser av detta medel. Resultaten visade sig tydliga genom att en minskning av antalet sniglar åstadkoms. Detta resultat var oberoende den dos av medel som användes. Doser om 2,5 g/m², 5 g/m² och 2*2,5 g/m² visade sig lika effektiva. För att nå även bättre kontroll över *A. lusitanicus* förordas en fortsatt studie med Ferramol i kombination av biologisk bekämpning, genom en naturlig fiende, skalbaggen *Pterostichus melanarius.*

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INTRODUCTION

The slug *Arion lusitanicus* Mabille 1868 (synonym *Arion vulgaris* Moquin-Tandon 1855), have become an important agricultural pest in central and Northern Europe in the last decades (Fischer and Reischütz, 1998; Kozlowski, 2000; Grimm, 2001; Paill et al, 2002). *A. lusitanicus* is native at the Iberian Peninsula and has extended quickly, arriving in Sweden around the 1990s (Kozlowski, 2007). This pest is inflicting strong damage to garden and horticultural plants as well as cultivated crops.

Sweden's climate is affected by the country's location in the border zone between Arctic and warmer air masses as well as its proximity to the Atlantic, with its warm Gulf Stream (The official gateway of Sweden, [www.sweden.se]). Atlantic low-pressure areas often blow in warmth and precipitation from the southwest. The weather is changeable; a few hours of rain are often followed by sunlight and wind the next day and then new rainfall. Another type of weather, however, creates a more contrasting climate: high pressure zones to the east, which create stable, dry, sunny weather. This high pressure leads to hot spells in summer and cold ones in winter. Fall and winter arrive early in the northern interior, while the southern coastal areas enjoy long, mild fall weather.

Nevertheless, these characteristics are being affected by the climate change. According to the report of the IPCC (2007), increase of the air and ocean temperature will cause warmer days and nights, decreasing the probability of frosts and increasing insect pests attack. Also, there is foreseen an increase of the intense rainfalls, and tropical cyclones that will damage many crops.

According to Slotsbo (2010) and Grimm (2001) the biology of *A. lusitanicus* has an annual life cycle with eggs, larvae or adults (Fig. 1-2). In the end of July *A. lusitanicus* begins copulation and three weeks later it lays eggs (one *A. lusitanicus* can lay over 400 eggs). In general, the first offspring hatch in late summer, but this depends of the ambient temperature. *A. lusitanicus* overwinter primarily as juvenile, mature slugs rarely survive the winter. In November when temperature falls below 3°C slugs begin to migrate towards their winter shelters. Their shelters can be holes in the soil, litter, composts and so on. During the winter when the air temperature increases, there will be periods were the slugs will become active and move out of there winter shelters to forage. Juveniles begin to leave the winter shelters at the end of February, and most of them leave their shelters by the end of March, though depending on the ambient temperature (Paill, 2004). They also found that depending on the ambient humidity and temperature soil, the life cycle could change.

Fig.1. Adult of *Arion lusitanicus* and cluster eggs([google.es/imghp]).

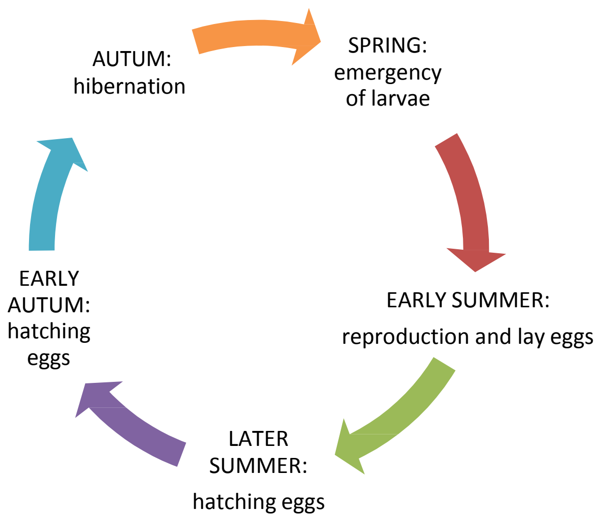

Fig.2. Life cycle of *Arion lusitanicus* (Galan).

In agriculture, the frequently used control strategy of slugs is the application of pesticides (South, 1992; Barker, 2002; Iglesias et al., 2002). However, the effect is often variable, and they may affect other organisms, such as natural enemies, harmfully (Langan et al., 2004). There are other control strategies used in agriculture such as electrified fences (Laznik et al. 2011) or birch tar oil (BTO) applied on fences or as a spray directly on slugs (Lindqvist et al. 2010). Nevertheless, these strategies have a short period of action, increasing the cost and the work required for their control.

Regulation by natural predators is considered to be decisive in sustainable agricultural systems for preventing pest. Carabid beetles are species wealthy and rich in arable habitats global and they are important natural predator of slugs (Kromp, 1999; Fig. 3). Some studies have demonstrated that the carabid beetle *Pterostichus melanarius* (Illiger) is a high-quality predator of eggs and juveniles of the pest *Arion lusitanicus* (Oberholzer and Frank, 2003; Hatteland et al., 2010). However, adults of the Iberian slug secrete sticky mucus when they feel at risk which can irritate the enemies and may cause immunity to attack from many predators (Schroeder et al. 1999).

In general terms, carabid beetles are species that usually lay their egg singly (Lövei and Sunderland, 1996; Matalin, 2006). The activity of most typical groups peaks either in the spring or the autumn. This crest is flexible in many groups, but frequently coincides with the reproductive period of the Iberian slug (Makarov, 1994). *P. melanarius* is a European-Siberian carabid beetle with a nocturnal activity until August and diurnal after this (Desender et al. 1985), but this can change depending by the temperature, light intensity and moisture (Thiele, 1977; Jones, 1979; Matalin, 2006). The life cycle of the carabid beetle in Southern Sweden is characterized by a dynamic activity from the middle of May until the end of September, also the reproductive stage can last for 100 days, from June to September (Wallin, 1987; Paill, 2004; Fig. 4). In other words, this beetle possesses a changeable life cycle that depend on the temperature. In cold winters, under bad conditions, the beetle has biannual cycle life with hibernation at the larval phase, determines the two years development of individuals, and the next year wintering at the adult phase. Likewise, in the populations as a total, the young generation emerges every year, because the emergence happens alternatively in each subpopulation (Matalin, 2006).

Fig.3. Adults of *Pterostichus melanarius* ([google.es/imghp]).

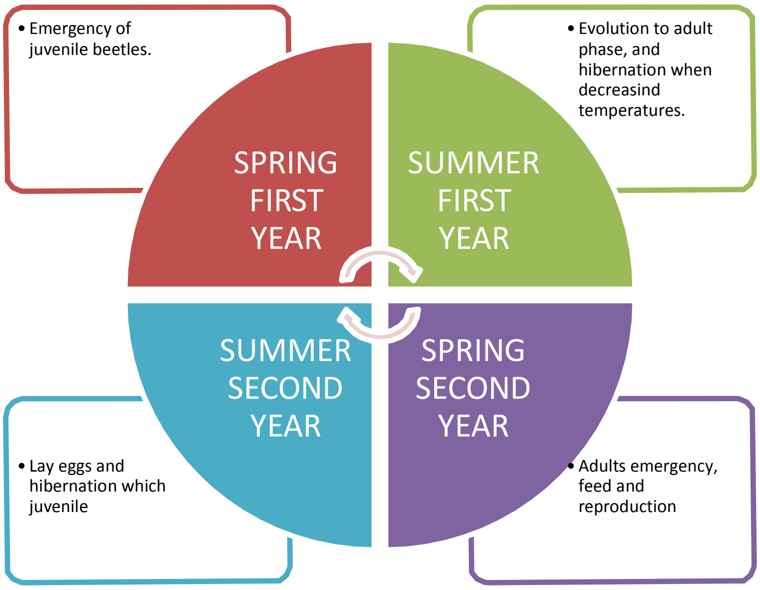

Fig.4. Life cycle of *Pterostichus melanarius* (Galan)

A product called Ferramol which attracts molluscs and acts as molluscicide is currently allowed in organic agriculture. It is a compound whose active ingredient is iron, supplied as ferric phosphate III ($FePO_4$). It inhibits the feed of the slugs and snails; ingestion of the product makes slugs quickly stop eating, to return to their places and die. This leads to an immediate protection of cultivated plants because the ingestion of the product does not provoke the dehydration of the molluscs and does not leave remains of dead molluscs in plants. The excess of the product is reused by plants; thanks to the action of the microorganisms of the soil, the ferric phosphate III transforms to iron and phosphorus, essential nutrients for plants. The molecule perfectly resists moisture and water and is not toxic to humans and animals (De Liñán Carral, 2010).

Although it should be considered, that Ferramol is not a selective product for Iberian slug, but acts on slugs and snails in general.

The purpose of the project was to find out if it is possible to reduce the damage of the pest slug *Arion lusitanicus* to the agricultural crops by applying Ferramol on the slug in juvenile and adult stages during the spring.

MATERIALS AND METHODS

The experimental part of this report was conducted on a strawberry field located in the periphery of Lund, south of Sweden with a latitude N 55° 40′ 51′′and a longitude E 13° 11′ 17′′ (Fig. 5-6). In addition, the meteorological data were obtained from Monthly Journal of the Swedish Meteorological and Hydrological Institute (SMHI); the meteorological station was situated to latitude N 56° 09′ 30′′ and longitude E 13° 23′ 30′′.

Fig.5. Location map ([maps.google.se])

The strawberry field was divided into 16 plots. Not all plots had the same size, eight of them were composed of four 15 m long row beds (crop area 60m²) and each row bed was formed by two lines of strawberries. The other eight plots were formed by two 25 m long row beds (crop area 50m²). The field experiment was surrounded by two blind rows of strawberry.

Fig.6. Detail map farm ([maps.google.se])

Ferramol (W. Neudorff GmbH KG An der Mǘhle 3, D-31860 Emmerthal, Germany) was used for the experiment at the rate of 2.5 g/m², 5 g/m², and double of 2.5 g/m². To compare if there were differences between the treatments, the field was divided into four horizontal blocks of four plots each. One plot was left for control, without Ferramol treatment, and the other 3 plots had different doses of Ferramol (Fig. 7-8). The position of the treatments was performed randomly. To count the number of slugs, 25 x 25 cm boxes were used, one per square, with lateral openings where slugs could enter. Beer was used as attractant.

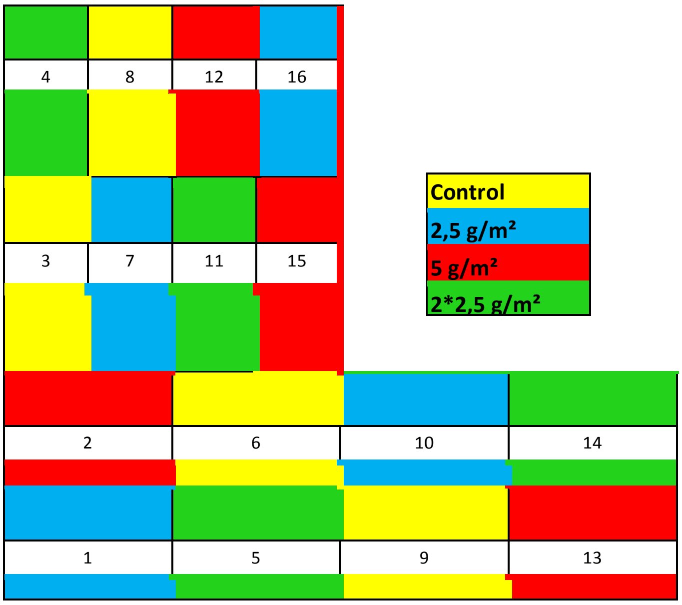

Fig.7. Strawberry field schema and distribution of the treatments (Galan).

In addition, to compare the results between the treatments, it was necessary to use a statistical program. In this case the data were analyzed with ANOVA and Tukey's test. ANOVA or analysis of variance is a collection of statistical models, and their associated procedures, in which the observed variance is partitioned into components due to different sources of variation. Tukey's test is a single-step multiple comparison procedure and statistical test generally used in combination with an ANOVA to find which means are significantly different from another.

The experiment started on April 26th, 2010. Ferramol was spread in the field and the beer traps were put inside the boxes between strawberry plants. A total of 150 g/m^2 of Ferramol was used on the plots 15 m long, and 125 g/m^2 on plots 25 m long of the doses 2,5g/m^2. For the doses of 5 g/m^2, 300 g/m^2 were added on 15 m long plots and 250 g/m^2 on 25 m long plots. After 24 hours, the number of slugs in each box was counted. More doses of Ferramol were not added until May 10[th], when a double of 2.5 g /m^2 was added. The traps were replaced two times per week, coming back 24 hours later because the beer lost the attraction power.

Fig.8. Strawberry bed rows with two rows of strawberry on each bed (Galan).

RESULTS

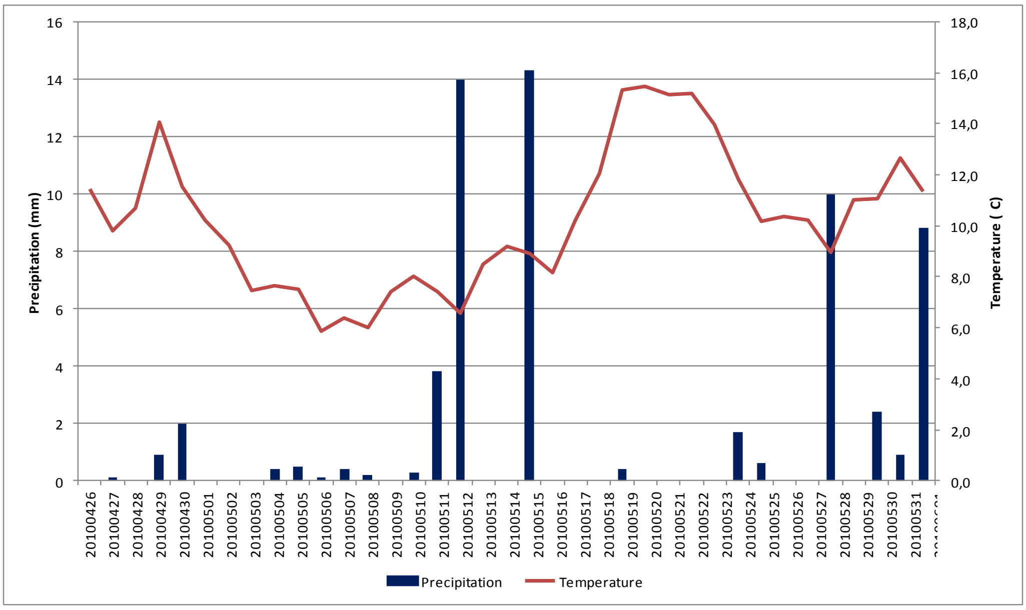

Fig.9. Diagram shows the climate condition during the field treatment: 26 of April to 1 of June 2010.

The results from this experiment were relevant since it could be seen that Ferramol was working properly. But first, it is important to look at the problems while performing the experiment. In some days, it was impossible to quantify the number of slugs in certain boxes, leading to non-applicable values, due to the rough meteorological conditions disturbing the boxes in which the experiment was taking place. The days in which this happened were the 27th of April and the 11th and 28th of May, when the high wind speed moved the boxes from their positions. There were also breaks in the daily data due to heavy rainfall invalidating some boxes as the beer was diluted (Fig. 9).

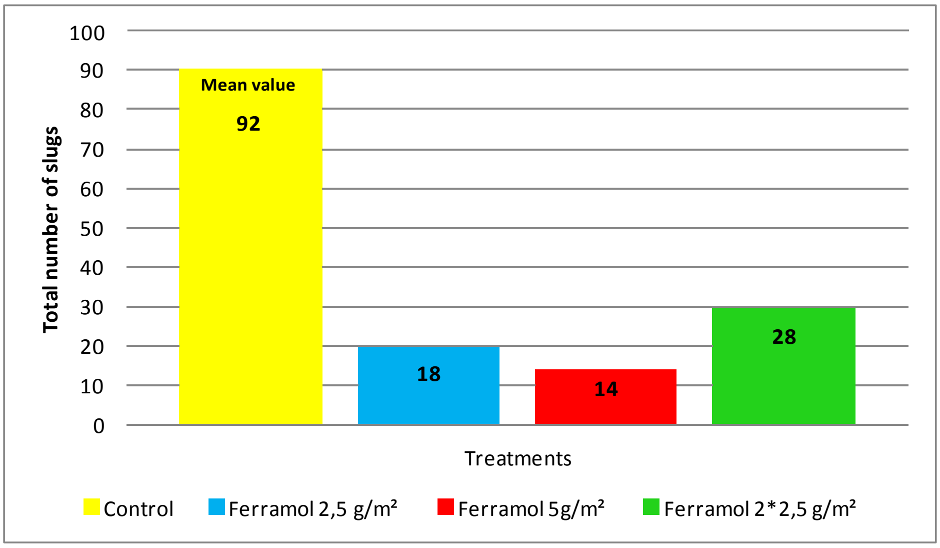

Fig.10. Graph shows the mean value with different treatments and non-treatment control. On the y-axis, the Σ number or total number of slugs that were found, for all the days of the study period (from April 27th to June 1st. The x-axis expresses the treatments: control in yellow, Ferramol 2.5 g/m2 in blue, Ferramol 5 g/m2 in red, and Ferramol 2*2.5 g/m2 in green. (Galán)

It can be seen that the number of slugs is much higher in the control plot than in the rest, suggesting that treatments were working. The difference between the control and the treatments was quite big. However, this difference was less when comparing the different treatments used. The obtained values from ANOVA and Tukey's test were $P \leq 0.05$, meaning that there was no difference between treatments.

Table. 1. Results of Tukey's test. The obtained results show the effectiveness of the treatments used. All values for each treatment were less than 0.01, are very important data, mean the effectiveness rises 99%. (Galán)

Tukey Simultaneous Tests. Response Variable response

All Pairwise Comparisons among Levels of treatment
treatment = Control subtracted from:

Difference Treatment	SE of T-Value	Adjusted P-Value (Control)
Ferramol 2*2.5	-4.696	0.0051**
Ferramol 2.5	-5.429	0.0019**
Ferramol 5.0	-5.723	0.0013**

* Significant **Very significant

Moreover, precipitation is known to be important for the development of the slug, and indeed, it seemed to have some effect on the slug appearance. It can be seen in Figure 11 there are two important points with increasing number of the slugs that came some days after increase of precipitation (11th and 21th of May). Less number of slugs occurred after the lowest precipitation (14th of May). Thus, looking at the relationship between the number of slugs and the precipitation is important. Also, the number of slugs decreased due the excessive moisture in the soil. By looking at the cumulative value, it can be seen how much moisture there is in the soil if no excessive evaporation has taken place.

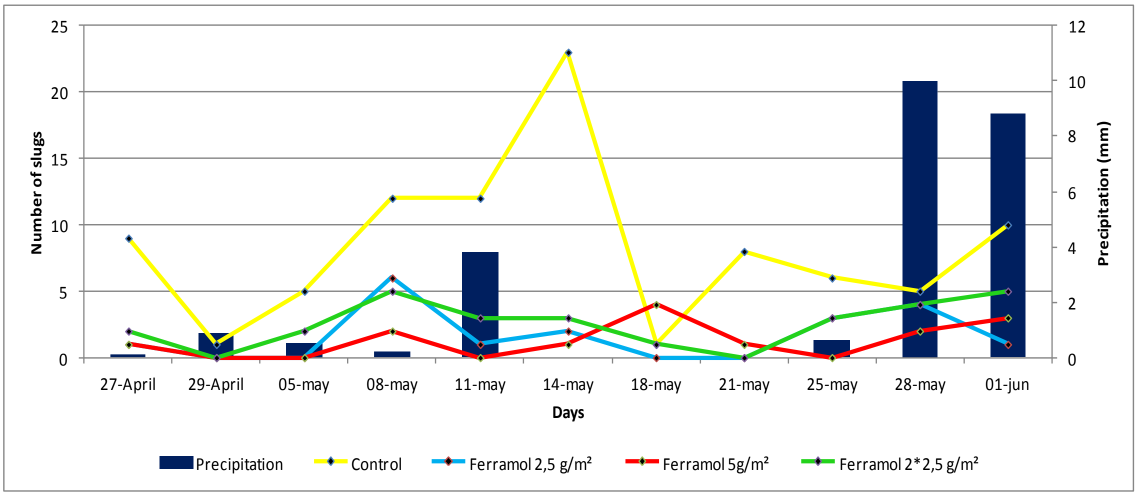

Fig.11. Graph showing the relationship between the number of slugs of the different treatments and the precipitation each day marked. On the primary y-axis (left side) the number of slugs can be seen, and in the secondary y-axis the precipitation in mm can be found. The thin lines represent the number of slugs of each treatment and the colours for each treatment are stated in the bottom part of the graph. Furthermore, the bar represents the precipitation patterns in mm. The period of study goes from the 27th of April to the 1st of June.

In Figure 12 it can be seen how the beginning of the rise of the cumulative precipitation line matches with the beginning of the growing peak for the control treatment, which is the treatment working with no restriction, thus, having free development until reaching the carrying capacity in this ecosystem. Nevertheless, a decrease in the number of slugs was observed that coincided with rising temperatures over 12ºC, suggesting an increased evaporation and less moisture in the soil. Thus, it is important to discuss the validity of this cumulative precipitation, to see the meaning and the possible effect that it has on slug development.

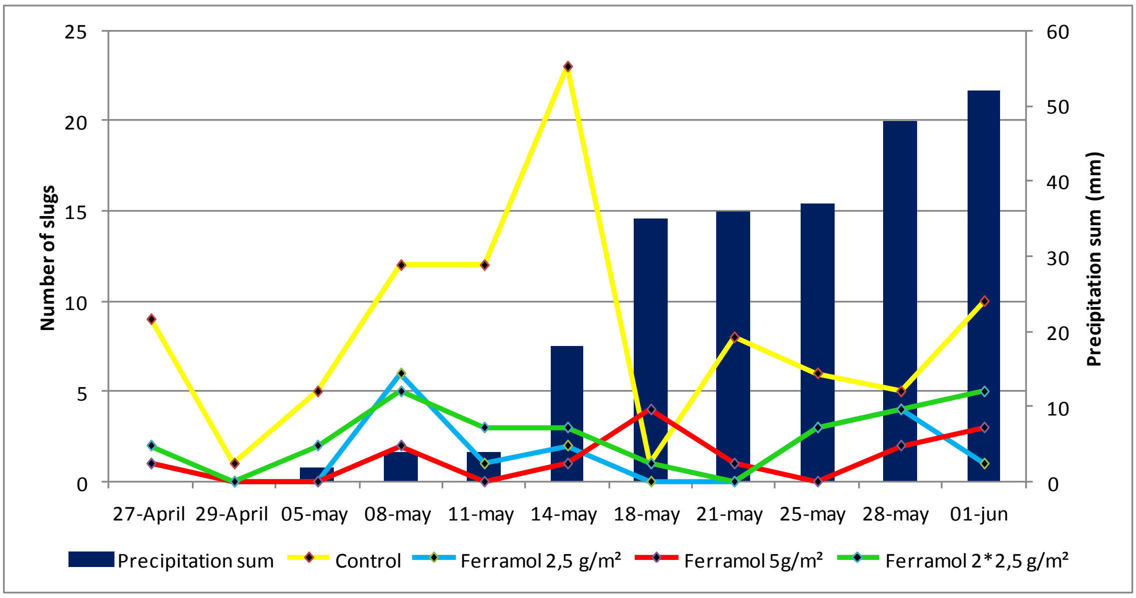

Fig.12. Graph showing the relationship between the number of slugs with different treatments and the total cumulative precipitation. On the primary y-axis (left side) the number of slugs can be seen, and in the secondary y-axis the precipitation in mm can be found. The thin lines represent the number of slugs of each treatment and the colors for each treatment are stated in the bottom part of the graph. Furthermore, the thick line represents the cumulative precipitation patterns in mm. The period of study goes from the 27th of April to the 1st of June.

Also, the relationship between the number of slugs at each plots and the temperature was compared. In Figure 13 it seems that the development of slugs did follow an increasing pattern with increasing temperature, except at temperatures above 12º C coinciding with a decrease in precipitation lowering the number of slugs found. Furthermore, it seems that the optimum temperature for slug development is between 8-10 ºC in this case study.

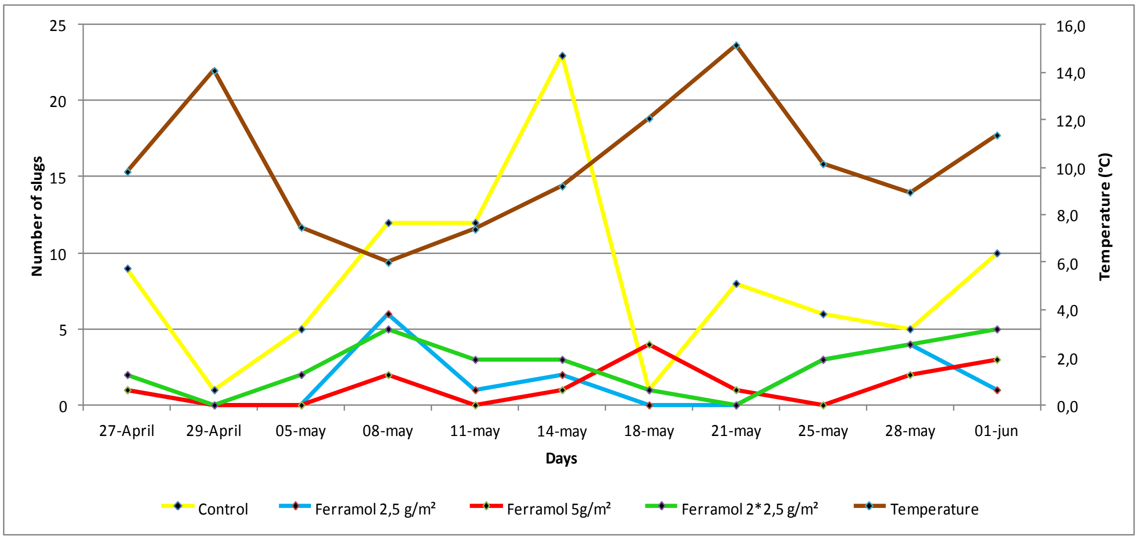

Fig.13. Graph showing the relationship between the number of slugs at the different for treatments and the temperature in ºC. On the primary y-axis (left side) the number of slugs can be seen, and in the secondary y-axis the temperature in ºC be found. The thin lines represent the number of slugs of each treatment and the colours for each treatment are stated in the bottom part of the graph. Furthermore, the thick line represents temperature in ºC. The period of study goes from the 27th of April to the 1st of June.

In addition, the following part of the experiment was to analyze if any of the treatments were better for the control of the slug pest. It can be seen that the best result for pest control, that is, less number of slugs, was with the Ferramol 5 g/m^2 dose. In addition, the graph shows that the treatment of double 2.5 g/m^2 dose of Ferramol is where most slugs have been found (with the exception, of course, of the control plot). Furthermore, the difference between 5 g/m^2 and 2.5 g/m^2 was not high.

DISCUSSION

The year of this study, 2010, was especially cold for slug development since it was one of the coldest and longest winters in the last 20 years. Therefore, temperature was considerably low and rain started very late in comparison to the previous years. Consequently, the results of this experiment might have been compromised by the meteorological conditions, as it was observed with the late increase in slug number. Svensson and Jonsson (2009) found much higher number of individuals compared to this study, obtaining up to 13 slugs in the Ferramol 2.5 g/m² treatment on the 16th of May, whereas this study only reaches 2 slugs on that approximately day for that treatment.

According to climate research conducted by SMHI as future scenarios (Fig. 14 and Fig.15), temperatures and precipitations tend to increase gradually. Conforming to IPCC, the global surface temperatures will be increased in more than 1,5 °C at the end of the 21th century, which could mean more favourable conditions for the development of the slug *A. lusitanicus,* making it more difficult the control of the pest. This is due to the fact that slugs need high humidity conditions on air, moist soils and low-medium temperatures. This has been proven with the increase of number of slugs with approximate 10ºC, whereas with 15ºC and lowering precipitations the number of slugs decreased due to weather limitation (Carmona and Tulli, 2006). Nevertheless, is important to note that the normal life cycle of the slug varies with location. Besides, since in the introduction it can be seen how the temperature and precipitation play an important role on the development and presence of slugs, it is necessary to look at the relationship between the number of slugs and precipitation (Fig. 11).

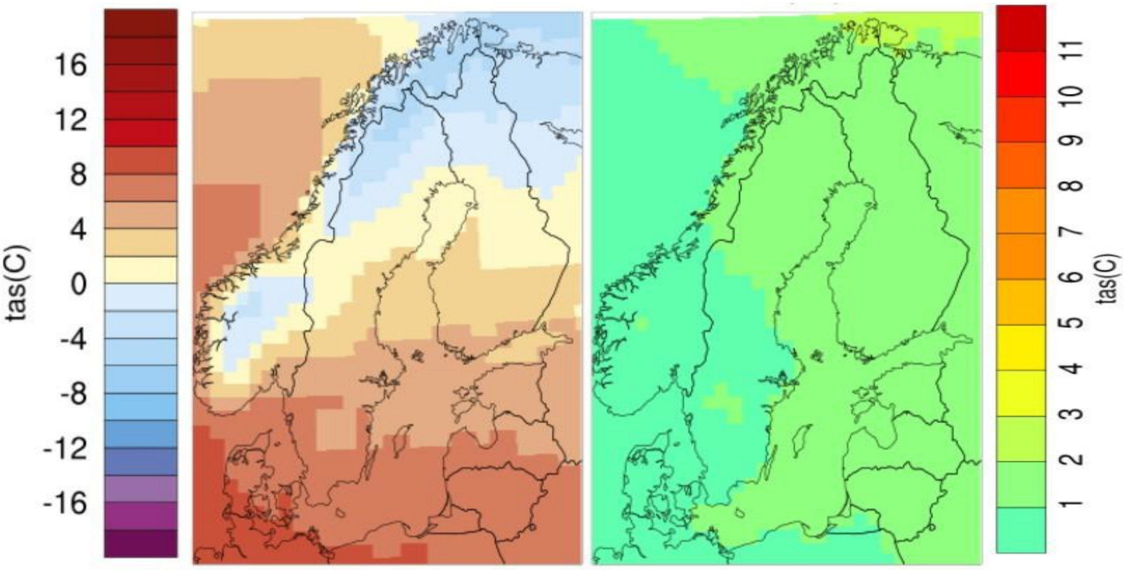

Fig.14. Temperatures in the period 1971-2000 comparing to the climate model RCA3, A1B (2011-2040), that shows the change in ensemble mean. Future scenario from SMHI.

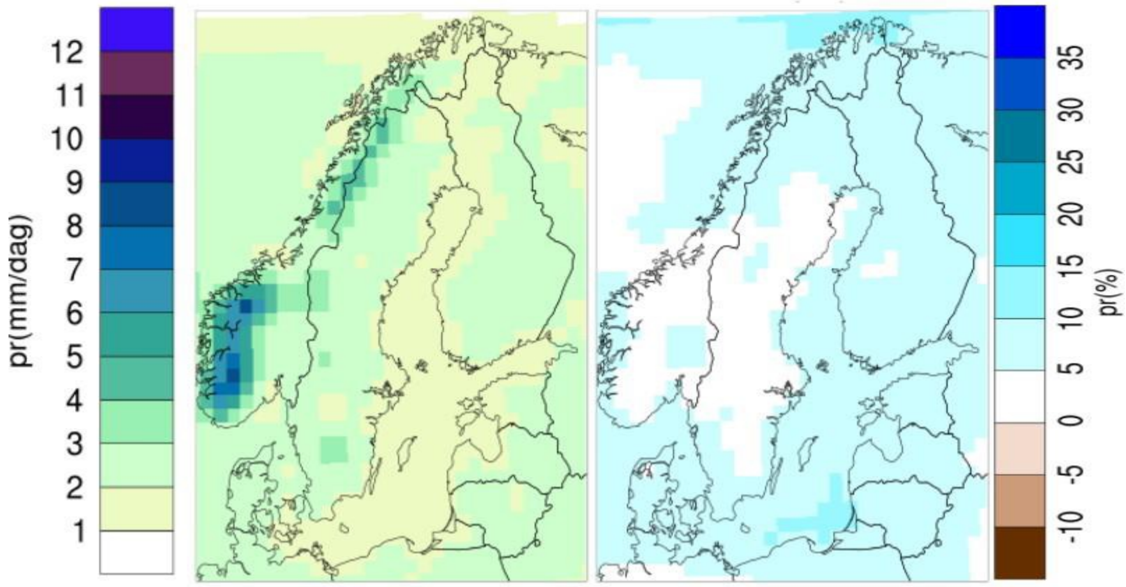

Fig.15. Precipitations in the period 1971-2000 comparing with the climate model RCA3, A1B (2011-2040), that shows the change in ensemble mean. Future scenario of SMHI.

The graph shows a close relationship between precipitation and the number of found slugs. However, the increase on the emergency of the slugs occurs gradually, that is not according to the same pattern as other studies have found (Svensson and Jonsson, 2009). In this study the increase was direct, while it is seen in the obtained results, that the increase in the slugs population of the control treatment happened some days later according to the rise in temperature. Thus, it is important to study both parameters jointly. Nevertheless, the increase in rainfall was quite late in the duration of the experiment, so perhaps for future studies it would be important to consider a longer time scale in order to see a closer relationship between the slug number and the precipitation. It is also possible that the farmer of the site had wetted the soil due to the lack of precipitation in the previous days, inflicting an increase in the number of slugs, with no change in precipitation.

Furthermore, the effectiveness of the Ferramol was proven, as there was a considerable decrease of the number of slugs between the control squares and the Ferramol treated plots. Although there were apparent differences between the treatments, and the dose of 5g/m² seemed to be the most effective, the statistical Tukey Simultaneous Tests showed that the effectiveness for all the doses was 99% and $P \leq 0.05$. This suggests the suitability of any of the applications. Consequently, an analysis of prices for both types of application needs to be done, as well as an availability study of each application, in order to make a calculative cost to value which treatment is used.

Moreover, the double application of 2.5g/m² was not successful, since the number of slugs found was higher compared to the other treatments, even if being smaller than in the control plot.

A future analysis could focus on the reduction and later disappearance of the slug, since Ferramol only helps to control the slug, or to protect the crop once the slug has started to emerge. One slug lays around 400 eggs, leading to fast reproduction rate. As explained in the introduction, Ferramol acts by inhibiting the feeding of the slug. However, as not all the slugs are going to be affected by the treatment, this would be the right moment to introduce a predator, increasing the control of the pest. To combine Ferramol with the predator *P. melanarius* was necessary to measure the effectiveness of Ferramol under the Swedish Weather condition. After this, it was needed to find the best natural predator adapted to this climate and to operate on the mollusc. The studied option could be the nematode *Phasmarhabditis hermaphrodita,* associated with the bacterium *Moraxella osloensis,* and two carabid beetles *Pterostichus melanarius* and *Poecilus cupreus.*

The nematode has been presented as a good option, since in the laboratory studies it has been shown as a biological control agent for young stages of *Arion lusitanicus*, due to the effects as feeding inhibitor (Grimm, 2001). However, when moving these experiments to the field the nematodes did not survive the following year, because they need high humidity and new nematodes should thus be introduced every year. According with studies in field by Hatteland et al. (2013) the effect of *P. hermaphrodita* did not last long. The application of nematodes had only a moderate effect on the slug and the numbers of *P. hermaphrodita* recovered from soil samples declined sharply about two weeks after application. Furthermore, according to Speiser (2000) nematode treatment is approximately 100 times more expensive than a single application of chemical molluscicides. The nematode treatment is not economically competitive.

Regarding carabid beetles, the choice based on the predatory ability, relying on the studies by Oberholzer and Frank (2002), *P. melanarius* has the potential to decrease slug population in the field by destroying eggs and killing hatched slugs, and *P. cupreus* by destroying only slug eggs. Individual *P. melanarius* appear to have a higher potential to reduce slug population than *P. cupreus*, because they are generally consumed more than *P. cupreus.*

When comparing the lifecycles of carabid beetles and slugs it is obvious that the period of maximum activity of the predator is close to the egg laying period of the slug, which ensures a possible predator-prey relationship. Both beetles have similar life cycles to *P. melanarius* reproduction occurring during spring (as explained in the introduction) and *P. cupreus* in late spring, early summer (Zangger, 1994). So, the *P. melanarius* activity starts at spring, coinciding with the laying period of the *A. lusitanicus*. According to Pianezzola (2013) the egg-laying period, thus, partly overlaps with the activity of P. melanarius, which was still present in late August and the beginning of September.This could provide a very efficient way to decrease the population size of the next generation. Nevertheless, this process needs more long-time research to be able to follow the annual cycle, and more observations are needed to achieve a long-lasting and environmental friendly control method.

CONCLUSION

Ferramol is presented as a good option for controlling the emergence of the slug *Arion lusitanicus,* the doses of 2.5 g/m^2 and 5 g/m^2 being equally efficient and environmentally friendly. The application of the dose is done once on late winter or early spring. More applications are not necessary the rest of the year. For the use of the beetle *Pterostichus melanarius,* first a study of adaptability to the environment should be conducted, in order to ensure that the incorporation does not damage native species and does not break the trophic chain. Once completed the study the beetles could be introduce as species acting on larvae and eggs of *A. lusitanicus.*

When analyzing the suitability of this experiment some limitations were found. First, the methodology should be changed in order to create better plot devices, since some of them were blown by the wind during the very windy days invalidating data collection. This further leads to the conclusion that a longer time and spatial period experiment is needed, to have more accurate results. To decrease the invalidated days by adverse weather conditions, the plot devices should be improved with weight cover putting on boxes or buried.

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