Barbara BINIAŚ, Janina GOSPODAREK
University of Agriculture in Krakow, Department of Agricultural Environment Protection
Al. Mickiewicza 21, 31-120 Kraków, Poland
e-mail: rrjgospo@cyf-kr.edu.pl

THE EFFECT OF WATER EXTRACT OF CHAMOMILE (*MATRICARIA CHAMOMILLA* L.) ON FEEDING THE PEA LEAF WEEVIL (*SITONA LINEATUS* L.) BEETLES

Summary

The aim of the study was to determine the effects of aquatic extracts of dry matter of chamomile (Matricaria chamomilla L.) in concentrations 2, 5 and 10% and fresh parts of this plant in concentrations 10%, 20% and 30% on feeding the females and males of pea leaf weevil (Sitona lineatus L.*) beetles on broad bean (Vicia faba* L.*) leaves, variety "Bartek". In the study, the 9 laboratory observations at 12-hour intervals were carried out. The leaf surface consumed by adults of Sitona lineatus was measured. The observations were performed in 6 replicates with the division into males and females. Additionally, the palatability index and an absolute deterrence index were calculated. The feeding of pea leaf weevil females was effectively reduced by applying a dried chamomile extract at a 10% concentration. The feeding of S. lineatus males was reduced to a very small degree, mainly by extracts prepared from dried plant matter. Extracts prepared from fresh chamomile matter did not exhibit any visible inhibitory effect on S. lineatus feeding.*
***Key words**: Matricaria chamomilla* L.*, biological control, Sitona lineatus* L.*, plant extracts*

WPŁYW WODNEGO WYCIĄGU Z RUMIANKU POSPOLITEGO (*MATRICARIA CHAMOMILLA* L.) NA ŻEROWANIE CHRZĄSZCZY OPRZĘDZIKA PRĘGOWANEGO (*SITONA LINEATUS* L.)

Streszczenie

Celem badań było określenie wpływu wodnych wyciągów z suszu rumianku pospolitego (Matricaria chamomilla L.) w stężeniach 2, 5 i 10% oraz ze świeżych części rośliny w stężeniach 10, 20 i 30 na żerowanie samic i samców chrząszczy oprzędzika pręgowanego (Sitona lineatus L.*) na liściach bobu (Vicia faba* L.*) odmiany „Bartek". W badaniach przeprowadzono 9 obserwacji laboratoryjnych, w odstępach 12-godzinnych. Mierzono powierzchnię wyżerek zatokowych, powodowanych przez dorosłe osobniki Sitona lineatus. Obserwacje przeprowadzono w 6 powtórzeniach z podziałem na samce i samice. Dodatkowo przeprowadzono analizę wskaźnika smakowitości oraz bezwzględnego współczynnika deterentności. Skuteczne ograniczenie żerowania samic oprzędzika pręgowanego, uzyskano po zastosowaniu wyciągu z suszu rumianku pospolitego w stężeniu 10%. Żerowanie samców S. lineatus było ograniczane w bardzo małym stopniu, przede wszystkim przez wyciągi na bazie suszu. Wyciągi ze świeżej masy rumianku nie wykazały wyraźnego działania hamujacego żerowanie chrząszczy S. lineatus.*
***Słowa kluczowe**: Matricaria chamomilla* L.*, ochrona biologiczna, Sitona lineatus* L.*, wyciągi roślinne*

1. Introduction

Nowadays, the use of herbaceous plants for natural crop protection is not a widespread practice. However, it may represent a chance for many organic farms where chemical products cannot be applied. The use of plant-derived substances for protection purposes instead of synthetic pesticides is in line with the existing principles of integrated pest management which gives priority to biological methods. Aqueous plant extracts are highly recommendable in this respect due to the ease of preparation, availability of raw materials (many herbaceous plants with potential insecticidal properties are wild weeds), low production costs and the absence of risks associated with their use [7]. Moreover, they do not increase pest resistance, do not require waiting and re-entry periods, do not accumulate in crops and do not cause a phytotoxic effect [5, 14]. The use of plants commonly growing as weeds in meadows and cropland as the basis for producing natural pest control products has twofold benefits. In addition to having a repellent and toxic effect on pests, their number in an agrocenosis is reduced [13]. The wild chamomile (*Matricaria chamomilla* L.) is one example of such a plant. The plant belongs to the family Asteraceae. Due to its strong disinfectant and antibacterial properties, it has long been cultivated as a medicinal plant and used in herbal and traditional medicine [8, 12]. Thanks to its highly complex composition, chamomile essential oil is suitable for a wide spectrum of applications. Chamomile contains an essential oil composed of sesquiterpenes (chamazulene – 1,4-dimethyl-7-ethylazulene, cadinene, myrcene – 7-methyl-3-methylene-1,6-octadiene, spatulenol), β-farnesene, flavonoids (comosin, luteolin – 2-(3,4-dihydroxyphenyl)- 5,7-dihydroxy-4-chromeone, quercetin, patuletin, chrysosplenium), chrysoeriol, hydroxycoumarins (umbelliferone, herniarin – 7-methoxycoumarin, apigenin), ά-bisabolene (6-methyl-(4-methylcyclohex-3enil)-hepta-2,5-dien), choline, bisabolol and sugars, carotenoids, organic acids (salicylic, nicotinic, malic, valeric and tiglic acid), phenolic acids, bitter substances (azulene- isomer naphthalene bicyclo[5,3,0]decapentaene), vitamins and minerals [10]. The main active ingredients include: bisabolol, which has the strongest properties, and chamazulene, β-farnesene and coumarins [17].
The research aimed to determine the effects of aquatic extracts of fresh and dry matter of chamomile (*Matricaria*

chamomilla L.) on feeding the females and males of pea leaf weevil (*Sitona lineatus* L.) beetles on broad bean (*Vicia faba* L.) leaves.

2. Material and methods

The laboratory experiment involved fresh, young leaves of broad bean of Bartek cultivar, collected from the same level below the top of the plant, in order to eliminate an effect of leaf age on the intensity of pest feeding. The collected plant material was treated with aqueous extracts prepared from dry and fresh pieces of chamomile (leaves, flowers and young shoots). In order to prepare the extracts, 2 g, 5 g and 10 g of dry mass of the chamomile were weighed (referred to as the concentration of 2, 5 and 10%), as well as 10 g, 20 g and 30 g of fresh pieces of the plant (referred to as the concentration of 10, 20 and 30%) and then it was poured with 100 ml of cold double-distilled water [15]. The extracts were allowed to stand for 24 hours in the dark at room temperature. Then, the resulting solutions were filtered through a filter paper, and immediately used for the experiment. The leaves of broad bean were immersed for 3 seconds in a suitable extract or double-distilled water (control), then dried at room temperature and placed in Petri dishes lined with moistened filter paper. Then, 2 individuals of pea leaf weevil were placed in each dish, the experiments were conducted separately for males and females. The experiment was performed in 6 replicates. The filter paper in Petri dishes was wetted if necessary during the experiments to prevent drying the leaves. The area of losses caused by the feeding of pea leaf weevil beetles was measured nine times at 12 hour intervals. After 12-, 24-, 36-, 48-, 60-, 72-, 84-, 96-, and 108 hours from the start of the experiment, the surface area of the eaten leaves was measured to investigate changes in feeding intensity of beetles *S. lineatus.*

After conclusion of the conducted observations for pea leaf weevil, the following values were calculated:

• palatability index – as the ratio of the percentage loss of the leaf blade in various combinations to the percentage loss of the leaf blade in the control,

• absolute deterrence index, which included the relationship between the area of leaf consumed by *Sitona* in the analyzed objects and the area of leaf consumed in the control [6]:

$$Adi = [(K–T) : (K+T)] \cdot 100 \quad (1)$$

where: K – the average area of leaf consumed by the pest in the control [mm^2], T – the average area of leaf consumed by the pest in the analyzed object [mm^2].

Statistical analysis of the obtained results was conducted using the Statistica 10.0 PL software. The significance of the differences between means was tested via conducting a single variant variance analysis (the study factor was extract type - prepared from fresh weight or dry weight and at different concentration) and the means were differentiated using the NIR Fisher test at the significance level of $\alpha = 0.05$.

3. Results

A statistical analysis of the obtained results showed that the feeding activity of pea leaf weevil females was to a greater extent reduced by the extracts prepared from dried plants than by those prepared from fresh plant parts. The greatest, i.e. 5-fold reduction in female feeding was achieved by applying an extract at a concentration of 10% (after 108 hours of experiment). The extracts prepared from fresh plant parts reduced female feeding to a small degree. Frequently, those differences were not statistically proven.

The surface area of the ingested broad bean leaves was much smaller for *S. lineatus* males than for females. The effects of the extracts persisted only until the 36th hour of the experiment. As was the case with females, the extracts prepared from dried plant matter were also more effective with males.

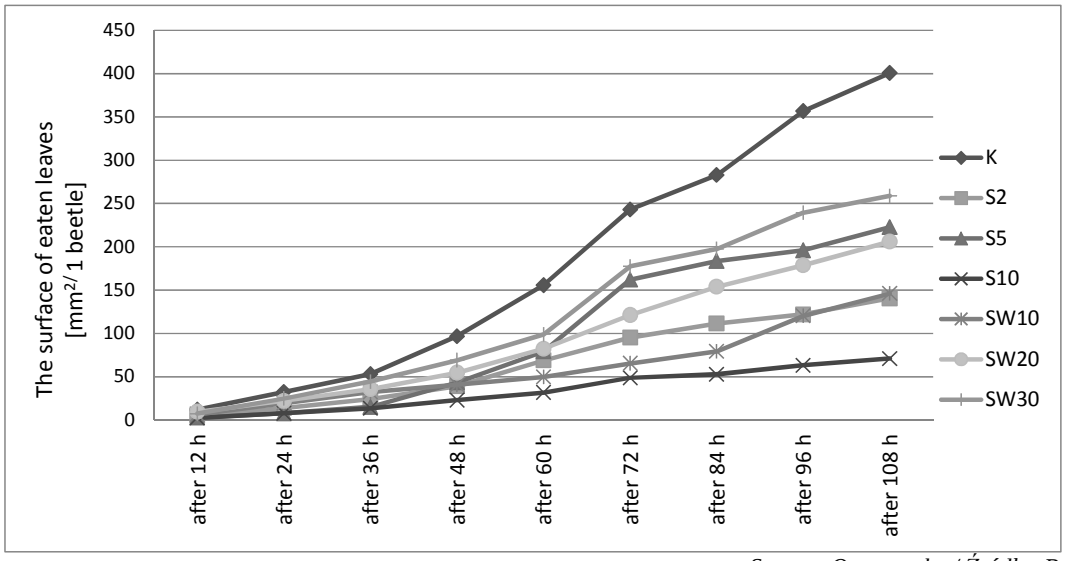

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Fig. 1. The dynamics of female *Sitona lineatus* L. beetles feeding on broad bean leaves, in cycle of 9 observations at 12-hour intervals, after application of the aqueous extract of dried chamomile in concentrations of 2 (S2), 5 (S5) and 10% (S10), and parts of fresh plants in concentrations of 10 (SW10), 20 (SW20) and 30% (SW30) compared to the control (K)
Rys. 1. Dynamika żerowania samic Sitona lineatus L. *na liściach bobu, w cyklu 9 obserwacji w odstępach 12-godzinnych, po aplikacji wodnego wyciągu z suszu rumianku pospolitego w stężeniach 2% (S2), 5% (S5) i 10% (S10) oraz świeżych fragmentów tej rośliny w stężeniach 10% (SW10), 20% (SW20) oraz 30% (SW30) w odniesieniu do obiektu kontrolnego (K)*

Table 1. Results of the statistical analysis of female *Sitona lineatus* beetles feeding on broad bean leaves, in the cycle of 9 observations at 12-hour intervals, after application of the aqueous extract from dried and fresh parts of chamomile. See figure 1 for explanations

Tab. 1. Wyniki analizy statystycznej żerowania samic Sitona lineatus na liściach bobu w cyklu 9 obserwacji w odstępach 12 godzinnych, po aplikacji wodnych wyciągów z suszu i świeżych części rumianku pospolitego. Oznaczenia jak na rys. 1

Object	after 12 h	after 24 h	after 36 h	after 48 h	after 60 h	after 72 h	after 84 h	after 96 h	after 108 h
K	*c	c	c	b	b	c	cd	c	c
S2	ab	ab	ab	a	a	ab	ab	ab	ab
S5	a	a	a	a	a	abc	bcd	ab	ab
S10	a	a	a	a	a	a	a	a	a
SW10	abc	abc	abc	a	a	ab	ab	ab	ab
SW20	bc	abc	abc	ab	a	abc	abc	ab	ab
SW30	abc	bc	bc	ab	ab	bc	bcd	bc	bc

* Means marked with the same letters in particular time do not differ significantly at α = 0.05

** Średnie oznaczone takimi samymi literami dla danego terminu nie różnią się od siebie istotnie przy α = 0,05*

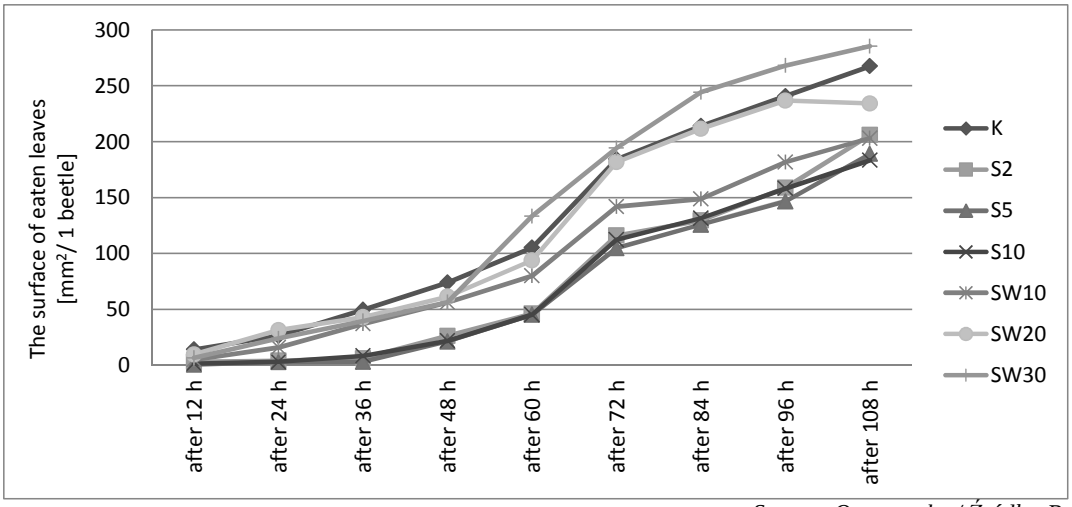

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Fig. 2. The dynamics of male *Sitona lineatus* L. beetles feeding on broad bean leaves, in cycle of 9 observations at 12-hour intervals, after application of the aqueous extract of dried chamomile in concentrations of 2 (S2), 5 (S5) and 10% (S10), and parts of fresh plants in concentrations of 10 (SW10), 20 (SW20) and 30% (SW30) compared to the control (K)

Rys. 2. Dynamika żerowania samców Sitona lineatus L. *na liściach bobu, w cyklu 9 obserwacji w odstępach 12-godzinnych po aplikacji wodnego wyciągu z suszu rumianku pospolitego w stężeniach 2 (S2), 5 (S5) i 10% (S10) oraz świeżych fragmentów tej rośliny w stężeniach 10 (SW10), 20 (SW20) oraz 30% (SW30) w odniesieniu do obiektu kontrolnego (K)*

Table 2. Results of the statistical analysis of male *Sitona lineatus* beetles course feeding on broad bean leaves, in the cycle of 9 observations at 12-hour intervals, after application the aqueous extract from dried and fresh parts of chamomile. See figure 1 for explanations

Tab. 2. Wyniki analizy statystycznej żerowania samców Sitona lineatus na liściach bobu w cyklu 9 obserwacji w odstępach 12-godzinnych, po aplikacji wodnych wyciągów z suszu i świeżych części rumianku pospolitego. Oznaczenia jak na rys. 1

Object	after 12 h	after 24 h	after 36 h	after 48 h	after 60 h	after 72 h	after 84 h	after 96 h	after 108 h
K	*c	b	b	a	a	a	a	a	a
S2	ab	a	a	a	a	a	a	a	a
S5	a	a	a	a	a	a	a	a	a
S10	ab	a	ab	a	a	a	a	a	a
SW10	ab	ab	ab	a	a	a	a	a	a
SW20	bc	b	ab	a	a	a	a	a	a
SW30	abc	ab	ab	a	a	a	a	a	a

* Means marked with the same letters in particular time do not differ significantly at α = 0.05

** Średnie oznaczone takimi samymi literami dla danego terminu nie różnią się od siebie istotnie przy α = 0,05*

All the used extracts had an inhibitory effect on the feeding activity of pea leaf weevil females, which is evidenced by the positive values of the absolute deterrence index (Fig. 3A). The strongest inhibitory effect on the feeding activity of *S. lineatus* females was produced by the dried *M. chamomilla* extract at a concentration of 10%, which achieved an absolute deterrence index Adi = 70. Relatively high values of the absolute deterrence index were also calculated for dried chamomile extracts at a concentration of 2% and extracts from fresh chamomile parts at a concentration of 10%. The palatability index for females was the highest after applying a dried chamomile extract at a concentration of 5% and an extract from fresh *M. chamomilla* parts at a concentration of 30% (Fig. 3 B). This demon-

strates the low effectiveness of those extracts in deterring the feeding of pea leaf weevil females. Moreover, with aqueous extracts from fresh plant parts, the palatability index actually increased as the concentration of the extract increased.

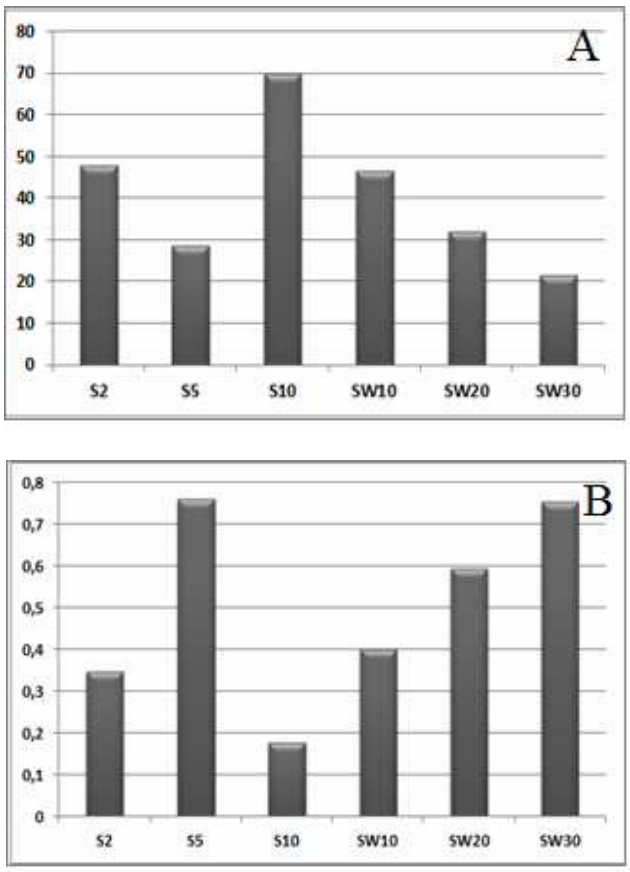

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Fig. 3. Absolute deterrence index (A) of extracts from dry and fresh matter of *Matricaria chamomilla* L. in different concentrations (from 2-10% and 10-30%) for females of *Sitona lineatus* L., calculated according to the formula: Adi = [(K–T) : (K+T)] · 100, where: K – the leaf surface consumed in control [mm^2], T – the leaf surface consumed on analyzed object [mm^2] and palatability index (B), calculated as the ratio of the percentage of the consumed leaf surface on individual objects to the percentage of consumed leaf's surface in control. See figure 1 for explanations

Rys. 3. Bezwzględny wskaźnik deterentności (Bwd) (A) wyciągów z suchej i świeżej masy Matricaria chamomilla L *w różnych stężeniach (od 2-10% oraz 10-30%) wobec samic Sitona lineatus L., wyliczony wg wzoru: Bwd = [(K–T): (K+T)] ·100, gdzie: K – średnia powierzchnia wyżerek na liściach w kontroli [mm^2]; T – średnia powierzchnia wyżerek w badanym obiekcie [mm^2] oraz wskaźnik smakowitości (B), wyliczony jako stosunek procentu ubytku powierzchni liścia w poszczególnych obiektach do procentu ubytku powierzchni w kontroli. Oznaczenia jak na rys. 1*

All the used extracts, except for the 30% extract from fresh *M. chamomilla* parts, were characterised by a positive absolute deterrence index with respect to *S. Lineatus* males, although the index values were low (Fig. 4A). For dried plant extracts, the deterrence index grew as the extract concentration increased. A reverse effect was produced by the extracts from fresh plant parts. An extract from fresh plant parts at the highest concentration achieved an absolute deterrence index of about -4, which indicates a stimulating effect on the feeding activity of pea leaf weevil males.

The palatability index achieved a value above 1 in an object with fresh plant extracts concentrated at 30% (Fig. 4B). Furthermore, high values of palatability index were also found for a fresh chamomile extract at a concentration of 20% and for the weakest, 2% concentrated extract from dried chamomile. For pea leaf weevil males, the palatability index values in objects with dried plant extracts decreased as the extract concentration increased. An inverse relationship was observed for extracts from fresh chamomile parts - the palatability index values increased as the extract concentration increased.

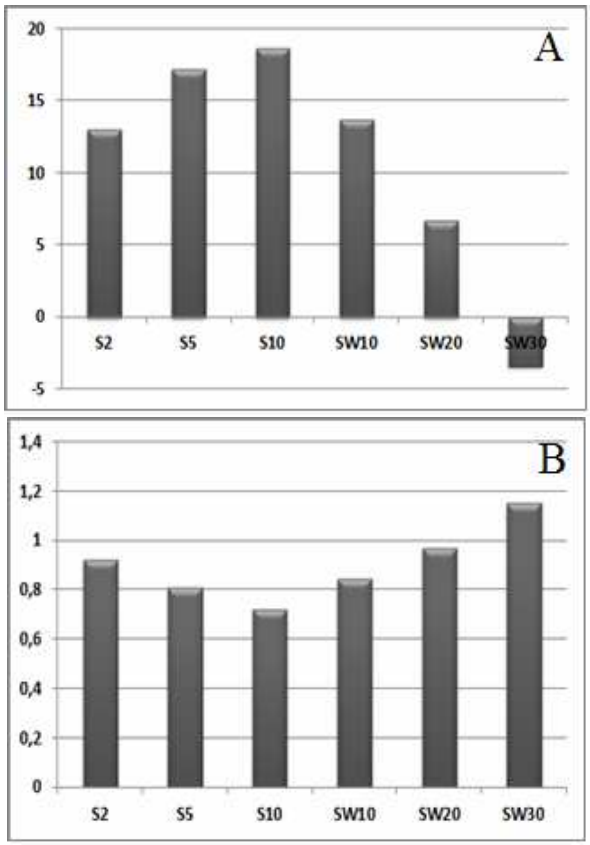

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Fig. 4. Absolute deterrence index (A) of extracts from dry and fresh matter of *Matricaria chamomilla* L. in different concentrations (from 2-10% and 10-30%) for males of *Sitona lineatus* L., calculated according to the formula: Adi = [(K–T) : (K+T)] · 100, where: K – the leaf surface consumed in control [mm^2], T – the leaf surface consumed on analyzed object [mm^2] and palatability index (B), calculated as the ratio of the percentage of the consumed leaf surface on individual objects to the percentage of consumed leaf's surface in control. See figure 1 for explanations

Rys. 4. Bezwzględny wskaźnik deterentności (Bwd) (A) wyciągów z suchej i świeżej masy Matricaria chamomilla L *w różnych stężeniach (od 2-10% oraz 10-30%) wobec samców Sitona lineatus L., wyliczony wg wzoru: Bwd = [(K–T): (K+T)] · 100, gdzie: K – średnia powierzchnia wyżerek na liściach w kontroli [mm^2]; T – średnia powierzchnia wyżerek w badanym obiekcie [mm^2] oraz wskaźnik smakowitości (B), wyliczony jako stosunek procentu ubytku powierzchni liścia w poszczególnych obiektach do procentu ubytku powierzchni w kontroli. Oznaczenia jak na rys. 1*

4. Discussion

The experiment demonstrated that the aqueous extracts of dried chamomile may contribute to reducing the feeding of *S. lineatus* males and females. On the other hand, the extracts prepared from fresh chamomile stimulated the feeding in males. So far, no research has been conducted to investigate the effectiveness of wild chamomile and its application in combating chewing pests, such as for example pea leaf weevils. However, a series of studies have been carried out to examine the effect of aqueous extracts from other herbaceous plants on *S. lineatus*. For *S. lineatus* females, an aqueous extract from dried wild chamomile at a concentration of 10%, after a 108-hour observation, contributed to a 5-fold decrease in the eaten leaf surface, whereas the average eaten leaf surface area in this object was only 75 mm^2. By way of comparison, an aqueous extract from the peppermint (*Mentha piperita* L.) and fennel (*Foeniculum vulgare* Mill.) seeds at the same concentrations, after 108 hours, did not reduce the surface area of damage caused to broad bean leaves by females, and the mean surface area of the eaten leaves was 200 mm^2 [3, 4]. In turn, an aqueous wormwood extract (*Artemisia absinthium* L.) at the same concentration after 120 hours following its application reduced the feeding of *S. lineatus* females on pea leaves (*Pisum sativum* L.) by about 1.4 times in comparison to the control [11]. Based on the above, it can be concluded that a 10% dried chamomile extract was far more effective than the extracts from other herbaceous plants mentioned above.

For *S. lineatus* females, the value of the absolute deterrence index reached 70 for 10% dried chamomile extract, 32 for dried peppermint extract at the same concentration [4], 16 for wormwood extract [11], and 19 for fennel seed extract [3]. This confirms the high potential of the above-mentioned extract for deterring the feeding of *S. lineatus* beetles.

The tests revealed visible differences in the feeding of *S. lineatus* males and females. Females ate up to twice as much food as males, and the inhibitory effect of the extracts was much more conspicuous in their case. Similar differences in feeding were recorded in earlier tests [4, 11]. The 30% extract of fresh plant parts had a slightly stimulating, although statistically insignificant effect on the feeding of *S. lineatus* males. A similar stimulating effect on male feeding was achieved by using the 10% extract of fresh wormwood matter [11].

The available literature contains information concerning the use of chamomile extracts to combat other pests, pointing to the high effectiveness of mainly alcoholic extracts. The studies on the effectiveness of aqueous and methanolic chamomile extracts in combating the red flour beetle (*Tribolium castaneum* (Herbst)) revealed 57% mortality after 7 days after the beginning of the observations using alcohol-based extracts. Moreover, the alcoholic extract of the *M. chamomilla* showed high repellency (IR = 0.04) against the beetles under study. On the other hand, the aqueous extract did not increase mortality [9]. Ethanol-based extracts of the wild chamomile in 0.301 mg/ l and 1.517 mg/ l doses were also subject to research to test their effectiveness in combating the mosquito *Culex quinquefasciatus* Say. For a highly concentrated extract, the LC50 value was recorded already after 24 hours after the beginning of the test. Furthermore, the extract displayed high toxicity against *C. quinquefasciatus* larvae and had a strong inhibitory effect on the development of the larvae, reduced the fertility and contributed to developmental defects at the larval, pupal and imago stages [2]. The research findings also suggest the possibility of using aqueous chamomile extracts prepared from dried matter for deterring pea leaf weevil feeding.

5. Conclusions

- The feeding of pea leaf weevil females was effectively reduced by applying a dried chamomile extract at a 10% concentration.
- The feeding of *S. lineatus* males was reduced to a very small degree, mainly by extracts prepared from dried plant matter.
- Extracts prepared from fresh chamomile matter did not exhibit any visible inhibitory effect on *S. lineatus* feeding.

6. References

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