



Thrips (Thysanoptera) associated with two genetically modified types of linseed (*Linum usitatissimum* L.)

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Abstract The aim of this study was to determine thrips abundance, species composition and seasonal dynamics on two genetically modified types of linseed, i.e., overproducing flavonoids, overproducing glycoside derivatives of phenylpropanoids and the respective non-modified, control plants with concentrations of phenylpropanoids typical for this variety. The experiment was conducted near Wrocław, Poland, in 2011–2013. For the determination of thrips species composition, 10 plants were collected from plots when linseed plants were at the full blooming stage. A sweep net was also used to study thrips on flax. In the three years of the study, 33 species and 2 genera of Thysanoptera were identified on oil flax plants. Irrespective of the type of linseed used, *Thrips angusticeps* was the dominant species. In the three years of the study, the lower numbers of thrips occurred on the genetically modified types of flax, i.e., the ones overproducing flavonoids or overproducing glycoside derivatives of phenylpropanoids than on the non-transformed plants. Fewer species of Thysanoptera were identified on oil flax overproducing flavonoids in comparison with the control plants.

Keywords Genetically modified plants · Oil flax · Thysanoptera · Species composition · Abundance · Phenylpropanoids effect

Introduction

Linseed (flax, *Linum usitatissimum* L.), one of the oldest cultivated crops, continues to be widely grown for oil, fiber and food [40]. In the 2014, the global world linseed production area harvested was 2,600,554 ha. The important cultivation areas of this crop were located in Canada, the traditional leader of flax cultivation (620,800 ha), followed by Kazakhstan (446,000 ha) and Russian Federation (441,475 ha). The largest areas of linseed production within the European Union were located in the UK (15,000 ha), France (10,993 ha), Belgium (10,000 ha) and Spain (9,500 ha). Other smaller producers were Sweden (6560 ha), Germany (4200 ha) and Poland (2386 ha) [11]. A decrease in linseed area harvested is evident in Western European countries since the year 2000. The possible reason of such situation can be removal of fiber flax and linseed subsidies that growers had been receiving to grow this crop. Basically, linseed products might be used in different branches of industry. Flax plant utilization for food, feed and fiber, as well as processing of flaxseed has been recently thoroughly reviewed [49]. Numerous prospective applications of linseed are pointed out in the review of Zuk et al. [62]. Hence, considering such promising and wide spectrum of linseed applications, there is hope that this crop will again become popular and highly prized in the nearest future.

The recognized beneficial properties of linseed are mainly associated with its oil, but a great amount of bioactive phytochemicals remains in the seedcake (the leftovers of the flax seeds after oil extraction). Flax seed

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cake contains phenolic acids, flavonoids and other phenylpropanoids that are known to show a wide range of biological activities and thus have a beneficial influence on human health [27]. The valuable effects of these compounds are mainly due to their antioxidant properties. The beneficial properties of linseed products could clearly be enhanced if the secondary metabolite accumulation in the plant organs could be increased. To increase the level of flavonoids and other phenylpropanoids and their glycoside derivatives, two genetically modified linseed types were generated [7, 61].

Plant phenylpropanoids are a very broad group of biochemical compounds which form secondary metabolites in enzymatic biosynthesis. They include flavonoids, phenolic acids, phenols, lignans and tannins [36]. Flavonoids are a group of secondary plant metabolites important for plant growth and development. They defend plants against various biotic and abiotic stressors, including pathogens and insect pests. Flavonoids can have a negative effect on non-adapted insects or may reduce the nutritive value of their food. They may behave as antifeedants, as digestibility reducers and as toxins [47, 48, 54, 57]. Plants currently bred for increased levels of phenolics might well influence different aspects of insect–plant interactions but as yet we do not have enough data available to evaluate what these effects might be. Studies on these interactions are needed if we are to have a better understanding of how increasing the expression of a group of organic compound-like flavonoids in plants could influence insect ecology.

A few species of thrips are pests of flax and by feeding on this plant they cause damage to both fiber and oil flax. The adult and larvae of *Thrips linarius* and *T. angusticeps* are mentioned as main pests of flax in Europe [4, 14, 8, 16, 17, 59, 60]. Besides the above-mentioned species *T. tabaci* and the predatory *Aeolothrips intermedius* breed on flax [59]. Many other herbivorous species of thrips have been identified in this crop [12, 14, 59]. Most of them are relatively rare and are not known to cause noticeable crop damage. *T. linarius* is a monophagous insect which feeds primarily on flax [8, 14, 59]. In most areas of Europe, this species produces one complete generation [4, 14]. In Romania, a partial second generation has been observed, and in Poland two complete generations [6, 60]. *T. angusticeps* is a polyphagous, widespread and common species in Europe. It prefers flax but feeds on many crops, like sugar beet, onion, peas, narrow-leafed lupin, broad bean, winter wheat and winter rye [10, 21, 41, 44, 51, 52, 58]. *T. angusticeps* has two generations per year in Europe [8, 59]. According to research by Franssen and Huisman [13], in the Netherlands the first is formed by wintering brachypterous specimens feeding and breeding on flax seedlings, the second by the macropterous ones feeding on flowers and fruits. The third

herbivorous species which can breed on flax, *T. tabaci* is a worldwide pest of agricultural and vegetable crops. It causes damage to cucumber [43], onion [39], leek [31], cabbage [25, 34, 42], pea [41], potato and tomato [15]. Besides the direct damage, adults of this pest can transmit tomato spotted wild virus (TSWV) [46]. The number of annual generations of *T. tabaci* depends on climatic conditions. In cold areas, 2–3 generations may be produced. In warmer regions, the number of annual generations is about 6–8 [15]. In the Netherlands, oats thrips (*Stenothrips graminum*) were very frequently collected on flax plants as well [14].

The aim of this study was to determine thrips abundance, species composition and seasonal dynamics on two genetically modified types of linseed, i.e., overproducing flavonoids and overproducing glycoside derivatives of phenylpropanoids. It was expected that overproduction of the secondary metabolites can influence the thrips populations.

Materials and methods

Two genetically modified types of *Linum usitatissimum* (cv. Linola) were used: type A—overproducing flavonoids (in tables and figures marked as A) [61], type B—overproducing glycoside derivatives of phenylpropanoids (B) [7] and the respective non-modified, control plants cv. Linola containing concentrations typical for this variety of phenylpropanoids (type C) (C). The experiment was conducted at the Experimental Research Station in Pawłowice, near Wrocław, Lower Silesia, Poland (51°1737'N, 17°1176'E) during three growing seasons (2011–2013). Plants were grown on 15 m² plots (10 m × 1.5 m) of sandy soil. The 0.3 m wide space between the experimental plots was maintained mechanically as bare soil, and the plots were weeded regularly. The experiment was designed as a split-plot with four replicates for each genetically modified type and for control cv. Linola of *L. usitatissimum*. Six hundred seeds per 1 m² were sown in each treatment.

Two different methods of thrips collection were used. For the determination of thrips species composition, 10 plants were collected from the central part of each plot (40 plants for treatment) when oil flax plants were at the full blooming stage (BBCH 65). In the laboratory, plants were shaken over a sheet of white paper. Larvae and adults were counted, preserved in 75 % ethyl alcohol and then identified. Every second week thrips were also collected by a sweep net (38 cm in diameter). A sample consisted of 10 180° sweeps taken in the middle of each plot on each sampling date. Samples were placed individually into cardboard containers with a paper towel saturated with

95 % ethyl acetate. Samples were taken to the laboratory, where insects were hand separated from plant material and placed in vials containing 75 % ethyl alcohol. Samples were later sorted, and specimens were counted and identified.

The sex ratio is the percentage of females in a population. It was calculated using the standard formula:

$$Sr = f : (m + f) \times 100,$$

where Sr —sex ratio, f —number of females, m —number of males [55].

The numbers of thrips found using three different sampling methods in different treatments of the experiment were compared using the analysis of variance (ANOVA) followed by Tukey's HSD (post hoc). Statistical significance was evaluated at $P \leq 0.05$. For statistical analysis, Statistica software, version 12 PL, was chosen (StatSoft Inc. Poland [50]).

Results and discussion

Abundance and species composition: sweep net collection

In 2011, 6 collections by a sweep net were done. A total of 646 thrips belonging to 18 species were identified in the collected material from all the treatments (Table 1). A significantly lower number of thrips was found in type B oil flax (overproducing glycoside derivatives of phenylpropanoids) (72 individuals) in comparison with non-modified plants (367) ($F = 4.35$, $df = 2$, $P = 0.0176$). The lowest number of species was also determined in type B (6), while in type A (12) and in control plants (17). In each treatment, the most numerous species were *Aeolothrips intermedius* and *Thrips angusticeps*. The first species made up from 40.6 % (type A) to 66.7 % (B) of all identified thrips and was especially abundant in the non-modified cultivar. The second species made up from 13.9 % (B) to 26.6 % (A) of all collected thrips.

In 2012, in 6 sweep net collections a total of 6203 thrips belonging to 23 species were found (Table 2). This was the highest number of determined Thysanoptera during the three years of the study. No significant differences in the number of recorded insects were found between treatments ($F = 0.15$, $df = 2$, $P = 0.855$). From 15 (type A, type C) to 19 (type B), species were identified. In each treatment, the most numerous species was *T. angusticeps*, making up from 66.9 % (type B, type C) to 71 % (type A) of all collected thrips. Two additional species were also abundant in all the studied treatments, i.e., *A. intermedius* and *T. tabaci*. The percentage of *A. intermedius* in the total

number of thrips recorded in each treatment was similar and fluctuated from 14 to 17.7 %. *T. tabaci* made up from 13 to 16 % of all Thysanoptera. Other identified species occurred in much lower numbers in the collected material and not in each treatment.

In 2013, as in previous years, 6 collections by a sweep net were done. A total of 4495 thrips belonging to 22 species and 2 genera were identified in the collected material from all the treatments (Table 3). This year, significantly more Thysanoptera were found on control plants (1817) in comparison with type A plants (1192) ($F = 6.4$, $df = 2$, $P = 0.0029$). Irrespective of the number of collected insects in each treatment, the number of identified species was almost the same (16 or 17). As in the previous year, *T. angusticeps* was the dominant species in each treatment, making up about half of all identified Thysanoptera (from 44.9 to 56 %). The second most numerous species was *A. intermedius*, comprising 24 % of insects from all treatments (A—16.5 %, B—20.8 %, C—4 %). The abundance of this species was almost three times as high on plots of non-modified plants as on type A. *T. tabaci* was also numerous and occurred in similar numbers in the three studied treatments.

Abundance and species composition: shaking method

Much fewer Thysanoptera were collected by shaking plants over a sheet of white paper than by using a sweep net. In the three years of our study, no significant differences in numbers of thrips were found between treatments. In 2011, only 40 thrips belonging to 6 species were collected by this method from all the treatments of plants which were at the full blooming stage (BBCH 65) (Table 4). Only 3 species were identified on type A, 6 species on type B and 4 on control plants. In each treatment, *T. tabaci* was the dominant species, making up about 50 % of all identified Thysanoptera. The second most numerous species was *T. angusticeps*, comprising 25 % of all insects.

In 2012, a total of 127 thrips were found in the collected material. Among them 5 species were identified, 3 or 4 in each treatment (Table 4). Insects occurred both in the adult and the larval stages. This year, *T. angusticeps* was the distinct dominant, and made up 78.3 % of all found Thysanoptera on type A oil flax plants, 69.5 % on type B and 75.6 % on type C plants. *T. tabaci* was also abundant: 17.4 % in type A, 22 % in type B and 13.6 % in control plants.

In 2013, a total of 88 thrips were found using the shaking method (Table 4). In the collected material, only 2 or 3 species were identified in the treatment. As in the previous year, *T. angusticeps* was the distinct dominant and constituted from 82.4 % (type B) to 93.1 % (type A) of all

Table 1 Species composition of thrips collected by sweep net in 2011

Species	A ^a		B		C		Total	%
	N ^b	%	N	%	N	%		
<i>Aeolothrips intermedius</i> Bagnall, 1934	84	40.6	48	66.7	221	60.2	353	54.6
<i>Thrips angusticeps</i> Uzel, 1895	55	26.6	10	13.9	72	19.6	137	21.2
<i>Thrips tabaci</i> Lindeman, 1889	30	14.5	2	2.8	12	3.3	44	6.8
<i>Frankliniella intonsa</i> (Trybom, 1895)	9	4.3	2	2.8	19	5.2	30	4.6
<i>Frankliniella tenuicornis</i> Uzel, 1895	12	5.8	6	8.3	8	2.1	26	4.0
<i>Chirothrips manicatus</i> Haliday, 1836	3	1.4			13	3.5	16	2.5
<i>Limothrips denticornis</i> Haliday, 1836	6	2.9	4	5.6	6	1.6	16	2.5
<i>Haplothrips aculeatus</i> Fabricius, 1803	2	1.0			4	1.1	6	0.9
<i>Thrips atratus</i> Haliday, 1836	2	1.0			3	0.8	5	0.8
<i>Anaphothrips obscurus</i> (Muller, 1776)	2	1.0					2	0.3
<i>Haplothrips setiger</i> Priesner, 1921	1	0.5			1	0.3	2	0.3
<i>Limothrips cerealium</i> Haliday, 1836	1	0.5			1	0.3	2	0.3
<i>Thrips major</i> Uzel, 1895					2	0.5	2	0.3
<i>Chirothrips hamatus</i> Trybom, 1895					1	0.3	1	0.2
<i>Odontothrips loti</i> (Haliday, 1852)					1	0.3	1	0.2
<i>Thrips fuscipennis</i> Haliday, 1836					1	0.3	1	0.2
<i>Thrips physapus</i> Linnaeus, 1758					1	0.3	1	0.2
<i>Thrips trehernei</i> Priesner, 1927					1	0.3	1	0.2
Total	207ab ^c	100	72b	100	367a	100	646	100
Number of species	12		6		17		18	

Superscript 'a' indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed, superscript 'b' indicates number of thrips and superscript 'c' indicates significant differences in total numbers of thrips

Different small letters next to total numbers of thrips indicate significant differences between the treatments

identified Thysanoptera. Single specimens of *T. tabaci* were also recorded in samples from each treatment.

In the three years of the study, 33 species and 2 genera of Thysanoptera occurring on oil flax were collected using two methods of collection, sweep net and shaking plants over a sheet of white paper. Three of them were the most numerous: *T. angusticeps*, *T. tabaci* and *A. intermedius*. In terms of the feeding group, two first mentioned species are phytophagous, and *A. intermedius* is zoophagous. *T. angusticeps* was the dominant species irrespective of the type of oil flax used. It prefers flax but feeds on many other crops, like sugar beet, onion, peas, narrow-leafed lupin, broad bean, winter wheat and winter rye [10, 21, 41, 44, 51, 52, 58]. *T. tabaci*, which was the second important phytophagous species on oil flax, is an important pest of onion, other onion relatives, and several crops in most parts of the world [15, 25, 31, 34, 39, 41–43]. *Thrips linarius*, mentioned in the literature as a main pest of flax in Europe [4, 8, 16, 17, 59, 60], was not identified in our trials. This species is a monophagous insect which feeds primarily on flax. Flax plants were not grown for many years in the region of our trials. Therefore, we suppose that the prolonged absence of the host plant in this area was the main reason for the lack of *T. linarius* in the collected entomological material. This species was the only pest

observed by Kucharczyk [28] on flax crops in the Lublin Upland (southeastern Poland).

The zoophagous *A. intermedius*, commonly occurring in our trials, was reported as a predator of 44 species of the order Thysanoptera [45]. Predatory larvae and adults feed mainly on the larvae of other thrips, on aphids and also on the larvae and eggs of other insects. Moreover, adults of *A. intermedius* can also feed on pollen [5]. In Poland, the presence of this species has been recorded in the flowers of many plant species, including herbs [29]. In the same location, it was found as an abundant species on Andean lupin, narrow-leafed lupin and *Triticum durum* [20–22]. In the sweep net collection, this species was always much more numerous in samples from control plants than from type A plants.

Sex ratio of the most numerous species

Taking into account the three years of our study and the three types of oil flax, the calculated mean sex ratio of *Thrips angusticeps* collected by the sweep net method was 86.3 % and ranged from 74.2 to 96.4 % (Table 5). Similar values of this ratio were calculated for each treatment. In winter rye, the sex ratio of *T. angusticeps* reached 96.8 % [51].

Table 2 Species composition of thrips collected by sweep net in 2012

Species	A ^a		B		C		Total	%
	N ^b	%	N	%	N	%		
<i>Thrips angusticeps</i> Uzel, 1895	1653	70.5	1370	66.8	1213	66.9	4236	68.3
<i>Aeolothrips intermedius</i> Bagnall, 1934	326	13.8	292	14.3	321	17.6	939	15.1
<i>Thrips tabaci</i> Lindeman, 1889	312	13.2	328	16.0	236	13.0	876	14.1
<i>Frankliniella intonsa</i> (Trybom, 1895)	8	0.3	17	0.8	5	0.3	30	0.5
<i>Frankliniella tenuicornis</i> Uzel, 1895	14	0.8	7	0.3	7	0.4	28	0.5
<i>Anaphothrips obscurus</i> (Muller, 1776)	7	0.3	5	0.2	6	0.3	18	0.3
<i>Haplothrips aculeatus</i> Fabricius, 1803	4	0.2	3	0.1	8	0.4	15	0.2
<i>Chirothrips manicatus</i> Haliday, 1836	2	0.1	4	0.2	5	0.3	11	0.2
<i>Thrips atratus</i> Haliday, 1836	6	0.3	4	0.2	1	0.1	11	0.2
<i>Limothrips denticornis</i> Haliday, 1836	3	0.1	5	0.2	1	0.1	9	0.1
<i>Stenothrips graminum</i> Uzel, 1895	2	0.1			4	0.2	6	0.1
<i>Thrips major</i> Uzel, 1895	2	0.1	2	0.1			4	0.1
<i>Thrips</i> spp.			3	0.1	1	0.1	4	0.1
<i>Thrips flavus</i> Schrank, 1776			1	0.1	2	0.1	3	0.1
<i>Haplothrips setiger</i> Priesner, 1921			2	0.1			2	0.1
<i>Neohydatothrips gracilicornis</i> (Williams, 1916)	2	0.1					2	0.1
<i>Thrips physapus</i> Linnaeus, 1758			1	0.1	1	0.1	2	0.1
<i>Cephalothrips monilicornis</i> (O. M. Reuter, 1880)			1	0.1	1	0.1	2	0.1
<i>Haplothrips angusticornis</i> Priesner, 1921	1	0.1					1	0.1
<i>Haplothrips propinquus</i> Bagnall, 1933			1	0.1			1	0.1
<i>Odonthrips loti</i> (Haliday, 1852)			1	0.1			1	0.1
<i>Oxythrips ajugae</i> Uzel, 1895	1	0.1					1	0.1
<i>Thrips fuscipennis</i> Haliday, 1836			1	0.1			1	0.1
Total	2343a ^c	100	2048a	100	1812a	100	6203	100
Number of species	15		19		15		23	

Superscript 'a' indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed, superscript 'b' indicates number of thrips and superscript 'c' indicates significant differences in total numbers of thrips

Different small letters next to total numbers of thrips indicate significant differences between the treatments

The population of *Thrips tabaci* was mainly represented by females. In 2011, in each treatment sex ratio reached 100 %. In 2012, in samples collected from type A and type B oil flax, only single males were found. More males occurred on plots of control plants, and the sex ratio was 83.1 %. In 2013, again only females were identified in type B and type C combinations, while in the type A combination few males occurred. In this combination, the sex ratio reached 92 %. Vasiliu-Oromulu [55] from mountainous meadows, Šmatas [51], Šmatas et al. [52] from winter rye and winter wheat also identified mainly females of *T. tabaci*. In the Netherlands, the male/female ratio on leek was 1/26 [56]. There are two reproductive modes in *T. tabaci*: thelytoky (asexual reproduction) and arrhenotoky (sexual reproduction) [24]. In thelytokous populations only females exist, and in arrhenotokous, both males and females. These two reproductive modes may coexist in populations collected in the field [38]. However, they differ

ecologically and genetically, also in terms of different host preferences [33].

In the population of *Aeolothrips intermedius* males occurred in greater numbers in each treatment than in the two previously mentioned species. The average sex ratio of this predatory species was 70.3 % and ranged from 61.5 to 77.1 % (Table 5). In our trials conducted in the same location on Andean lupin, a lower mean sex ratio of *A. intermedius* was determined (45.8 %) [22]. Similarly, a lower ratio (50 %) in an alfalfa crop was reported by Barbuceanu and Vasiliu-Oromulu [2], but it was much higher in mountainous meadows: 77.5 % (shaking method) and 82.3 % (sweep net method) [55].

It was expected that GMO linseed plants overproducing flavonoids or glycoside derivatives of phenylpropanoids can influence the thrips sex ratio of the most numerous species. The results of the presented study do not confirm this hypothesis.

Table 3 Species composition of thrips collected by sweep net in 2013

Species	A ^a		B		C		Total	%
	N ^b	%	N	%	N	%		
<i>Thrips angusticeps</i> Uzel, 1895	668	56.0	766	51.5	815	44.8	2249	50.0
<i>Aeolothrips intermedius</i> Bagnall, 1934	197	16.5	309	20.8	572	31.4	1078	24.0
<i>Thrips tabaci</i> Lindeman, 1889	226	18.9	339	22.7	342	18.8	907	20.2
<i>Limothrips denticornis</i> Haliday, 1836	37	3.1	26	1.7	20	1.1	83	1.8
<i>Chirothrips manicatus</i> Haliday, 1836	19	2.0	15	1.0	14	0.8	48	1.1
<i>Haplothrips aculeatus</i> Fabricius, 1803	12	1.0	9	0.6	6	0.3	27	0.6
<i>Frankliniella tenuicornis</i> Uzel, 1895	9	0.9	1	0.1	16	0.9	26	0.6
<i>Anaphothrips obscurus</i> (Muller, 1776)	1	0.1	2	0.1	8	0.4	11	0.2
<i>Frankliniella intonsa</i> (Trybom, 1895)	5	0.3	3	0.2	3	0.2	11	0.2
<i>Limothrips cerealium</i> Haliday, 1836	3	0.2	2	0.1	6	0.3	11	0.2
<i>Stenothrips graminum</i> Uzel, 1895	6	0.4	2	0.1	3	0.2	11	0.2
<i>Thrips major</i> Uzel, 1895	2	0.1	7	0.5			9	0.2
<i>Chirothrips ambulans</i> Bagnall, 1932					5	0.3	5	0.1
<i>Thrips atratus</i> Haliday, 1836	4	0.2			1	0.1	5	0.1
<i>Haplothrips setiger</i> Priesner, 1921			2	0.1	2	0.1	4	0.1
<i>Thrips physapus</i> Linnaeus, 1758					2	0.1	2	0.1
<i>Aeolothrips fasciatus</i> (Linnaeus, 1758)			1	0.1			1	0.1
<i>Chirothrips hamatus</i> Trybom, 1895	1	0.1					1	0.1
<i>Haplothrips alpester</i> Priesner, 1914	1	0.1					1	0.1
<i>Haplothrips</i> sp.			1	0.1			1	0.1
<i>Odonthrips meliloti</i> Priesner, 1951					1	0.1	1	0.1
<i>Thrips brevicornis</i> Priesner, 1920					1	0.1	1	0.1
<i>Thrips fuscipennis</i> Haliday, 1836	1	0.1					1	0.1
<i>Thrips</i> sp.			1	0.1			1	0.1
Total	1192b ^c	100	1486ab	100	1817a	100	4495	100
Number of species	16		16		17		24	

Superscript 'a' indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed, superscript 'b' indicates number of thrips and superscript 'c' indicates significant differences in total numbers of thrips

Different small letters next to total numbers of thrips indicate significant differences between the treatments

Seasonal changes

The seasonal changes of thrips are presented on the basis of the results achieved by the sweep net collection in 2011 and 2013. Only in both these years differences in thrips numbers between treatments were found. On the other hand, the seasonal changes of these insects in 2012 were similar to those of 2011 and 2013. In 2011, the first single thrips were collected toward the end of May (Fig. 1a). At that time, significantly more insects were found in type A plots than in type B ones. Thrips numbers increased at the end of May and reached the maximum population in the first week of June (BBCH 63–65). Significant differences in the number of thrips at the maximum of their population were found. Significantly more insects occurred on control plants (type C) than on type B plants. In this season, thrips were collected on oil flax plants till mid-July (BBCH 85). The second lower population peak was noted at the

beginning of July (BBCH 81). In the additional two collections done in the third week of June and in the first week of July again more Thysanoptera were found on control plants than on type A and type B ones. As shown in Fig. 1b in the first three collections in 2011 (second half of May and first half of June) among the dominant species on linseed plants this season, *T. angusticeps* or *T. angusticeps* and *A. intermedius* were numerous. In the last three collections (second half of June and first half of July), mainly *A. intermedius* was identified.

In 2013, the first sweep net collection was done at the beginning of June. Plants at that time were at the BBCH stage 30–32 (Fig. 2a). The peak of the thrips population was noticed on July 18, when plants were at BBCH stage 62–65. During the population peak significantly more insects were collected on type B plants than on type C ones. The second lower peak of population was observed in mid-July. During this season, thrips were collected on oil

Table 4 Species composition of thrips collected by shaking plants in 2011–2013

Species	A ^a		B		C		Total	%
	N ^b	%	N	%	N	%		
2011								
<i>Thrips tabaci</i> Lindeman, 1889	5	50.0	9	50.0	7	58.3	21	52.5
<i>Thrips angusticeps</i> Uzel, 1895	3	30.0	4	22.2	3	25.0	10	25.0
<i>Aeolothrips intermedius</i> Bagnall, 1934	2	20.0	1	5.6			3	7.5
<i>Frankliniella intonsa</i> (Trybom, 1895)			2	11.1	1	8.3	3	7.5
<i>Thrips atratus</i> Haliday, 1836			1	5.6	1	8.3	2	5.0
<i>Limothrips denticornis</i> Haliday, 1836			1	5.6			1	2.5
Total	10	100	18	100	12	100	40	100
Number of species	3		6		4		6	
2012								
<i>Thrips angusticeps</i> Uzel, 1895	18	78.3	41	69.5	34	75.6	93	73.2
<i>Thrips tabaci</i> Lindeman, 1889	4	17.4	13	22.0	8	13.6	25	19.7
<i>Frankliniella intonsa</i> (Trybom, 1895)	1	4.3	3	5.1	2	4.5	6	4.7
<i>Aeolothrips intermedius</i> Bagnall, 1934			2	3.4			2	1.6
<i>Chirothrips manicatus</i> Haliday, 1836					1	2.3	1	0.8
Total	23	100	59	100	45	100	127	100
Number of species	3		4		4		5	
2013								
<i>Thrips angusticeps</i> Uzel, 1895	27	93.1	28	82.4	22	88.0	77	87.5
<i>Thrips tabaci</i> Lindeman, 1889	1	3.4	6	17.6	2	8.0	9	10.2
<i>Anaphothrips obscurus</i> (Muller, 1776)	1	3.4			1	4.0	2	2.3
Total	29		34	100	25	100	88	100
Number of species	3		2		3		3	

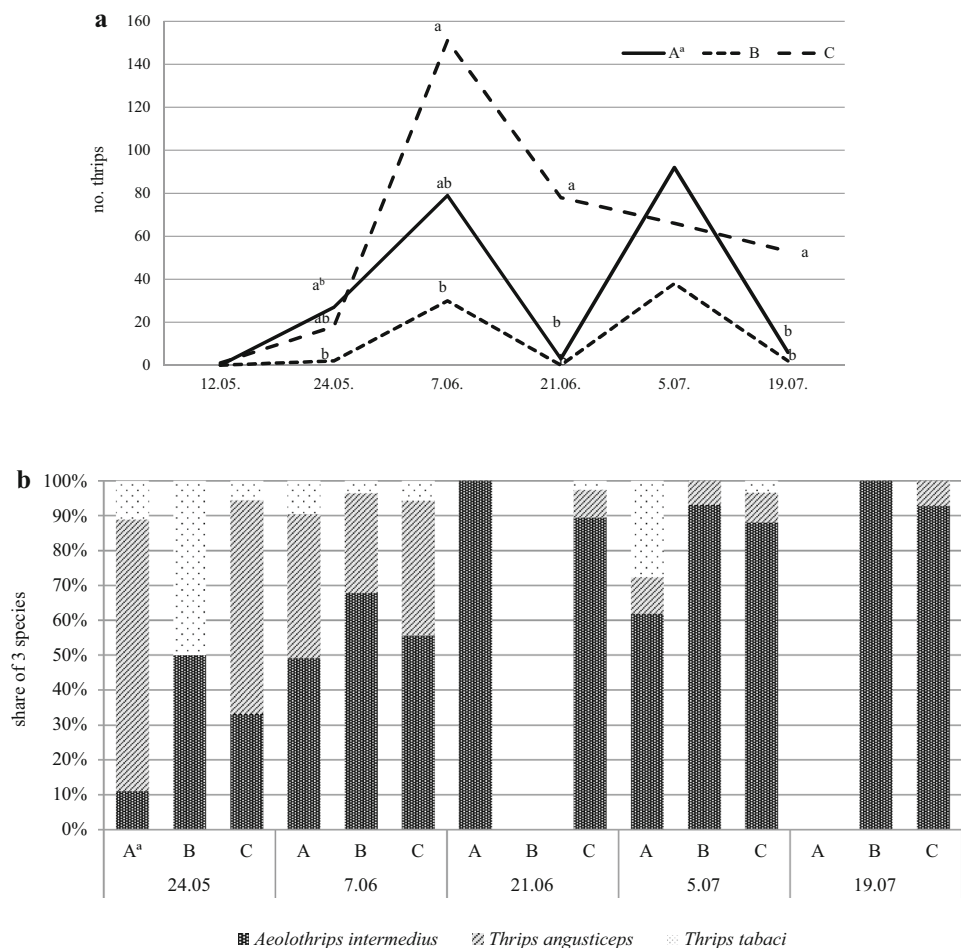
Superscript ‘a’ indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed and superscript ‘b’ indicates number of thrips

Table 5 Sex ratio index of the most numerous species of thrips collected by sweep net in 2011–2013

Species	A ^a			B			C		
	Male	Female	Sr ^b	Male	Female	Sr	Male	Female	Sr
2011									
<i>Thrips angusticeps</i> Uzel, 1895	2	53	96.4	2	8	80.0	2	70	97.2
<i>Thrips tabaci</i> Lindeman, 1889		30	100		2	100		12	100
<i>Aeolothrips intermedius</i> Bagnall, 1934	29	49	62.8	11	37	77.1	84	134	61.5
2012									
<i>Thrips angusticeps</i> Uzel, 1895	376	1269	77.1	351	1011	74.2	307	896	74.5
<i>Thrips tabaci</i> Lindeman, 1889	1	241	99.6	3	279	98.9	31	152	83.1
<i>Aeolothrips intermedius</i> Bagnall, 1934	44	103	70.1	46	108	70.1	51	133	72.3
2013									
<i>Thrips angusticeps</i> Uzel, 1895	47	612	92.9	65	692	91.4	55	736	93.0
<i>Thrips tabaci</i> Lindeman, 1889	18	207	92.0		338	100		336	100
<i>Aeolothrips intermedius</i> Bagnall, 1934	44	137	75.7	75	201	72.8	156	368	70.2

Superscript ‘a’ indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed and superscript ‘b’ indicates sex ratio of thrips

Fig. 1 Seasonal dynamics of thrips collected by sweep net on different types of linseed plants in 2011. Superscript 'a' indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed and superscript 'b' indicates significant differences in total numbers of thrips. **a** all the thrips. **b** share of three most abundant species



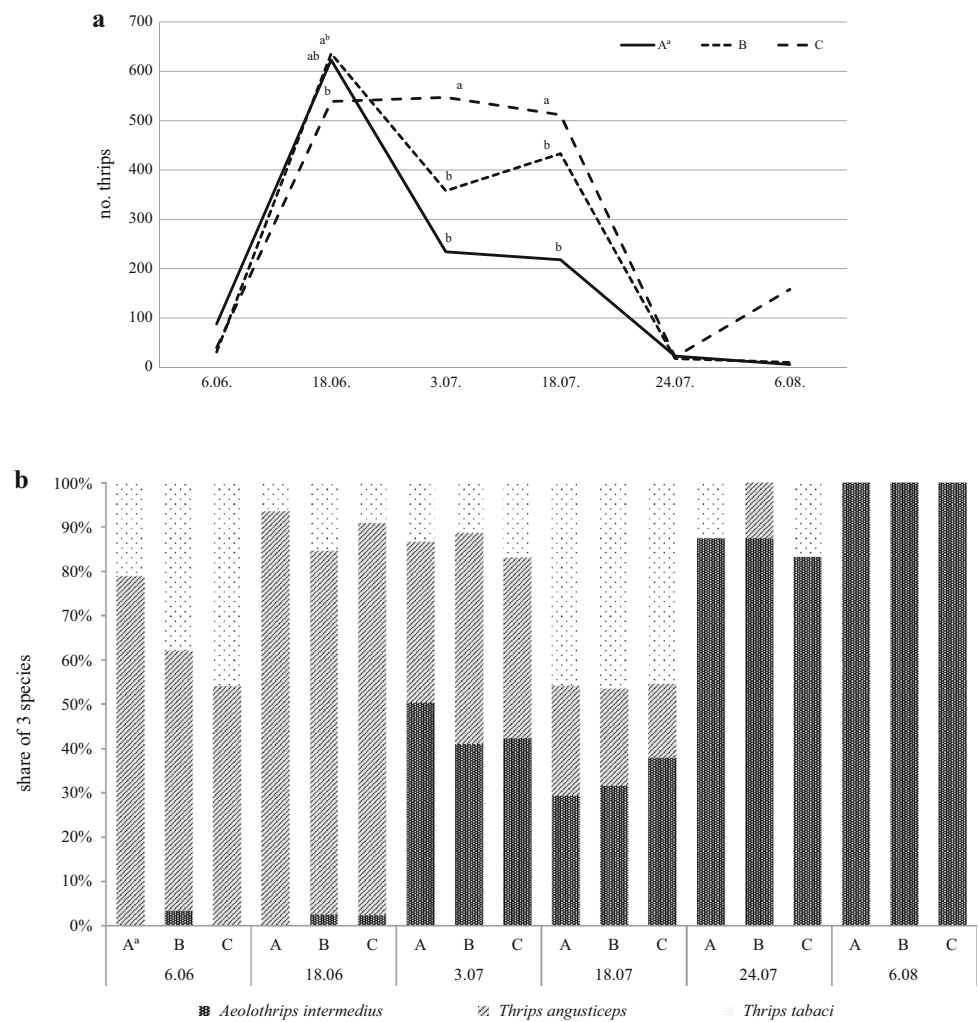
flax till the beginning of August (BBCH 89). At the beginning of July and in mid-July significantly more insects occurred on control plants than on type A and type B plants. In the collections gathered in June, among the three dominant species of Thysanoptera in 2013, *T. angusticeps* was the most abundant (Fig. 2b). In the first collection date also *T. tabaci* occurred in greater percentage. This species was also the most numerous in mid-July constituted almost 50 % of the three dominant species. From the beginning of July, the increase in the *A. intermedium* percentage was observed. It was the main thrips species on linseed of each treatment at the end of July and at the beginning of August.

According to many authors, polyphagous species of thrips have two peaks of abundance in their seasonal dynamics [3, 19, 26, 30, 32]. The first, lower peak occurs in spring, and is caused mainly by wintering adult individuals which colonize crops and intensively feed as plants begin to flower. Usually at the end of May and in early June after laying eggs the population of adults decreases. The second, higher peak of abundance is usually recorded in July, and is formed by both the larvae and adults of a new generation.

In the presented trials, thrips occurring in oil flax plants, irrespective of the used genetic types of *Linum usitatissimum*, also had two peaks of abundance. The first, higher one occurred in the first or second ten days of June when plants were at the blooming stage. The second, lower peak of abundance was recorded in the first half of July. At this time, plants were at the ripening stage. Contrary to Franssen and Huisman [13], who observed brachypterous forms of *T. angusticeps* during the earlier stages of flax growth, only macropterous forms were noted in our studies.

There are many examples in the literature that flavonoids can reduce the growth, extend the developmental cycle, interfere with reproduction and decrease survival of many herbivorous insects belonging to different orders other than Thysanoptera [1, 9, 18, 23, 35, 37, 47, 53]. Our trials showed that in 3 out of 6 sampling dates in 2011 significantly fewer thrips occurred on type B, i.e., overproducing glycoside derivatives of phenylpropanoids, than on control, non-modified plants (Fig. 1a). In the same year, in 2 out of 6 sampling dates fewer insects were also found on type A, i.e., overproducing flavonoids than on control ones. In 2013, in 2 sampling dates of 6 fewer Thysanoptera

Fig. 2 Seasonal dynamics of thrips collected by sweep net on different types of linseed plants in 2013. Superscript ‘a’ indicates type of linseed: A—linseed overproducing flavonoids, B—linseed overproducing glycoside derivatives of phenylpropanoids, C—non-modified linseed and superscript ‘b’ indicates significant differences in total numbers of thrips. **a** all the thrips. **b** share of three most abundant species



were recorded in type A oil flax and in 1 case fewer in type B in comparison with control plants (Fig. 2a). Taking into consideration the total number of thrips in the studied treatments, in two years significant differences were found. In 2011, fewer insects were collected on type B and in 2013 on type A in comparison with two other combinations (Tables 1, 3). Additionally, on plants overproducing flavonoids fewer, i.e., 19 species, were identified versus 24 species on control ones. The results of these trials show that the presented genetically modified oil flax plants can reduce the abundance of thrips.

Conclusions

1. In the three years of the study, 33 species and 2 genera of Thysanoptera were identified on linseed plants. Irrespective of the type of oil flax used, *Thrips angusticeps* was the dominant species. Among phytophagous species *T. tabaci* also occurred in greater

numbers. *T. linarius*, which is reported as a main pest of flax in Europe, was not found in our trials. The predatory *Aeolothrips intermedius* was among the most numerous species.

2. Thrips occurring in linseed plants, irrespective of the genetic types of *Linum usitatissimum* used, had two peaks of abundance. The first, and higher, occurred in the first or second ten days of June, when plants were at the blooming stage. The second, lower peak of abundance was recorded in the first half of July. At this time, plants were at the ripening stage.

3. In the most numerous species, like *T. angusticeps* and *T. tabaci*, in each treatment populations were formed mainly by females. In the population of *A. intermedius* males occurred in greater numbers than in the two previously mentioned species and the average sex ratio was 70.3 %.

4. In the three years of the study, the lower numbers of thrips occurred on the genetically modified types of flax, i.e., the ones overproducing flavonoids or

overproducing glycoside derivatives of phenylpropanoids than on the non-transformed plants. Fewer species of Thysanoptera were identified on oil flax overproducing flavonoids in comparison with the control plants.

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