CANOPY-DWELLING ARTHROPOD RESPONSE TO RYNAXYPYR AND LAMBDA-CYHALOTHRIN TREATMENTS IN MAIZE*

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The technology of grain corn production has recently been continuously changing due to spreading of insecticidal in-crop treatments in Europe. The aim of these interventions is to prevent damage caused by serious lepidopterous pests in maize. We carried out in-maize field experiments using two different active ingredients of insecticides in four consecutive years (2014–2017). A field experiment was conducted to compare the effect of applications of rynaxypyr (ANT) and rynaxypyr + lambda-cyhalothrin (PYR) on the canopy-dwelling arthropod community in commercial maize grain acreage. The effects of both ANT and PYR treatments against *Ostrinia nubilalis* Hübner (Lep.: Crambidae) were tested through four-year field experiments. The quantitative and qualitative assemblages of the perished arthropods and diversity alterations measured by canopy netting and grounded tarpaulins greatly differed in the different insecticide treatments. A significant number of dead arthropods was recorded after PYR treatment. Populations of other natural enemies (Coccinellidae, Chrysopidae, etc.) and endangered species (*Calomobius filum,* Rossi) were also negatively affected. The arthropod community of the examined maize plots was drastically altered by sprayings, which, among other factors, may account for the mass appearance of the other non-target pest organisms (Aphidae: *Rhopalosiphum* spp.*,* Miridae: *Trygonotylus* spp.).

anthranilic diamide, in-crop treatment, non-target arthropods, pyrethroid, side effect

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INTRODUCTION

Maize is one of the most important crops worldwide with an annual cultivation area of more than 178 million ha and an annual harvest of 939 million t of grain. The cropping area within the 28 member states of the European Union (EU) reached 9.61 million ha in 2014 for grain maize and 6.07 million ha for silage maize. The largest maize producers are France, Romania, Germany, Hungary and Italy, where maize is grown in each of these countries on more than 1 million ha (W a s d e , 2018). A substantial part of the world's agriculture production makes maize, which warrants efforts aimed at investigating yield-decreasing factors, as well as at assessing the related biological and economic damage (M e i s s l e et al., 2009).

At present, the most important arthropod pests of maize in Europe are the European corn borer, *Ostrinia nubilalis*, Hbn. (Lep.: Crambidae) and the cotton bollworm, *Helicoverpa armigera*, Hbn. (Lep.: Noctuidae) due to the direct damage and fusarium contamination in yield (C a t a r i n o et al., 2016; C h e h r i , G o d i n i , 2017). In the inflicted areas, *O. nubilalis* occurs in a large proportion (20–40%) in Hungary hence causing an estimated yield loss of 5–30% in the absence of preventive measures (M e i s s l e et al.*,* 2009). *H. armigera* poses problems typically in the central and southern counties (K e s z t h e l y i et al.*,* 2013).

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Table 1. Places of experimental areas and cultivated maize hybrids in four years (2014-17)

Year	Settlement	Geographical coordinates (GPS)	Cultivated maize hybrid	Maturity group
2014	Bodrog	46°30'30.09"N 17°38'08.73"E	PR38A79	FAO 310
2015	Kaposmérő	46°23'41.03"N 17°42'12.78"E	NK Altius	FAO 320
2016	Kaposmérő	46°22'39.10"N 17°42'19.94"E	PR38A24	FAO 380
2017	Kaposvár	46°22'58.96"N 17°44'50.43"E	SY Photon	FAO 350

In this study, the main target of in-crop insecticide treatments were the western corn rootworm, *Diabrotica v. virgifera* LeConte (Col.: Chrysomelidae) adults, as well as *O. nubilalis* and *H. armigera* larvae present in Hungary. The most commonly used active ingredients in spray insecticides were pyrethroids, and lately anthranilic acids. Furthermore, organophosphates and neonicotinoids were also used in minor quantities (Meissle et al., 2009). Pyrethroids are axonic excitoxins, the toxic effects of which are mediated through preventing the closure of the voltage-gated sodium channels in the axonal membranes. Pyrethroids are toxic to beneficial insects such as honey bees and dragonflies (Ray et al., 2000), whereas anthranilic diamides act through ingestion or contact and exert ovicidal and ovi-larvicidal effect on certain species hence offering excellent control means. Their utilisation fosters sustainable agricultural protection (Ioriatti et al., 2009).

Besides, about 20% of the maize fields are treated with insecticides in Hungary. The arthropod communities are diverse in maize, as reported by bibliographical data (Duelli et al., 1989; Pereira et al., 2005; Duan et al., 2006; Griffiths et al., 2006; Rose, Dively, 2007; Elbert et al., 2008). One of the most examined areas are the effects of in-crop insecticide treatments (Chauzat et al., 2009; Maini et al., 2010) and seed dressing (Girolami et al., 2009; Marzaro et al., 2011; Sgolastra et al., 2012) on honey bee communities. The adverse effect of pesticides on non-target organisms and the environment is the issue of great social concern. These non-target arthropods can become vulnerable ecological elements due to the spectrum and duration of the applied insecticide (Stanley et al., 2016).

The aim of this study is to highlight side-effects of insecticides in-crop treatment in maize, focusing on the proportion of non-target arthropod species, which was elicited by the sprayed insecticides. Therefore, our results may give impetus to efforts made in order to explore integrated pest management means in combating serious maize pests.

MATERIAL AND METHODS

Comprehensive experiments were carried out to investigate the involvement of arthropod assemblages in maize stock affected by different insecticide treatments in four consecutive years (2014–2017). Experimental plots under plots of investigations were located in four different grain maize fields in Somogy County, Hungary (Table 1). In these experimental areas maize was cultivated in consecutive years via monoculture. Soil disinfection (tefluthrin 15 kg ha^{-1}) to control *D. v. virgifera* was implemented in the designated areas. Other insecticides were not applied in these areas except the experimental treatment itself.

In order to simulate the effect of the treatment on large plots, extended experimental areas (15 ha) were selected and divided into three smaller plots (4.5 ha each) situated side by side (Lucza, Ripka, 2004).

Flights of *O. nubilalis* were monitored by the Vector T100 light trap in the duration of the flight period of this pest and in the course of the maize growing season. The timing of the in-crop insecticide treatment (T) was set 6–7 days after the flight peak (Pepper, Carruth, 1945) based on the flight peak calculated by the trapping data. The insecticide treatments were implemented using a JD-4710 auto-propelled sprayer.

In order to compare the effect of applications of rynaxypyr, which belongs to anthranilic diamides (ANT), field experiments were conducted: (100 g l^{-1} rynaxypyr; 125 ml ha^{-1} + 90% isodecyl alcohol ethoxylate; 500 ml ha^{-1} + 400 l ha^{-1} water) and double insecticide composition chemicals containing rynaxypyr + lambda-cyhalothrin (pyrethroid) (PYR) (200 g l^{-1} rynaxypyr; 125 ml ha^{-1} + 50 g l^{-1} lambda-cyhalothrin; 0.25 l ha^{-1} + 90% isodecyl alcohol ethoxylate; 500 ml ha^{-1} + 400 l ha^{-1} water) were sprayed on the arthropod community in a commercial maize grain acreage. Insect collections prior to (BEF. TRET. = before treatment) and following the treatments were carried out in two zones corresponding to different insecticide treatments and a third control area was left untreated (UNT).

The different surveys were always timed to the prevailing insecticide treatments (T) (06.07.2014, 30.06.2015, 05.07.2016, 04.07.2017). The specific plant damage types (1 – perforated upper leaves, 2 – bored stalks, 3 – broken tassels and stalks, 4 – masticated ears) were observed so as to assess the effect of different chemical treatments on target-insects (*O. nubilalis, H. armigera*). The above-mentioned damage types of the pests were estimated at two differing times in four experimental years (T + 8 and 14 days before harvesting).

Table 2. Schneider-Orelli's mortality values and their differences in insecticides treated parcels in four consecutive years

	2014	2015	2016	2017
ANT	6.961	6.562	10.100	22.105
PYR	48.639	76.944	27.272	68.315
PYR/ANT	6.987	11.725	2.700	3.090

ANT = rynaxypyr, PYR = rynaxypyr + lambda-cyhalothrin

Several techniques were used to collect the highest possible number of species including the use of boarded tarpaulins as well as insect sweeping nets. Insect nettings were implemented in maize parcels before (T – 7) and after the treatment (T + 1) for observing assemblages and alterations triggered by sprayings of canopy-dwelling arthropod communities. The 3 × 20 net strikes were done in each parcel. The captured specimens were killed by ethyl-acetate and subsequently poured onto small patches of cotton wool placed at the bottom of small glass vials.

Three tarpaulins (0.7 × 2 m) were grounded (T – 1) in two repetitions in each parcel in order to spot arthropod assemblages killed by insecticide treatments. Arthropods dropped onto tarpaulins were collected exactly one day (T + 1) after in-crop sprayings. Collected specimens were preserved in 98% ethanol and the species as well as the genera were identified by following the monograph of Majerus, Kearns (1989) and the Fauna Hungariae collection (Somfai, 1959; Kaszab, 1962; Szalay, 1968; Szelegiewicz, 1977).

The arthropod mortality counts measured by tarpaulins were corrected by using the Schneider-Orelli (1947) formula:

$S = (T – C)/(100 – C) × 100$

where:

T = responded % in treated

C = responded % in control

The data originating from insect nettings were used to study the richness of species (number of species) and to calculate diversity indices (Menhinick (M), Margalef (MR) indices and Whitaker diversity curve) (Whittaker, 1972) as well as the number of specimens (Tothmeresz, 1995; Collwell, 2009). The effects of different chemical treatments on canopy-dwelling arthropod communities were statistically analysed by one-way ANOVA using the SPSS 11.5 software. Mean values were separated by using the Tukey (HSD) test, at $P \leq 0.05$. Statistical calculations were carried out using MS Excel 2007 and the diversity values were determined by the NuCosa 1.05 software package (Tothmeresz, 1993).

RESULTS

In untreated parcels (UNT) a significantly higher number of perforated upper leaves (primary leaf mas-

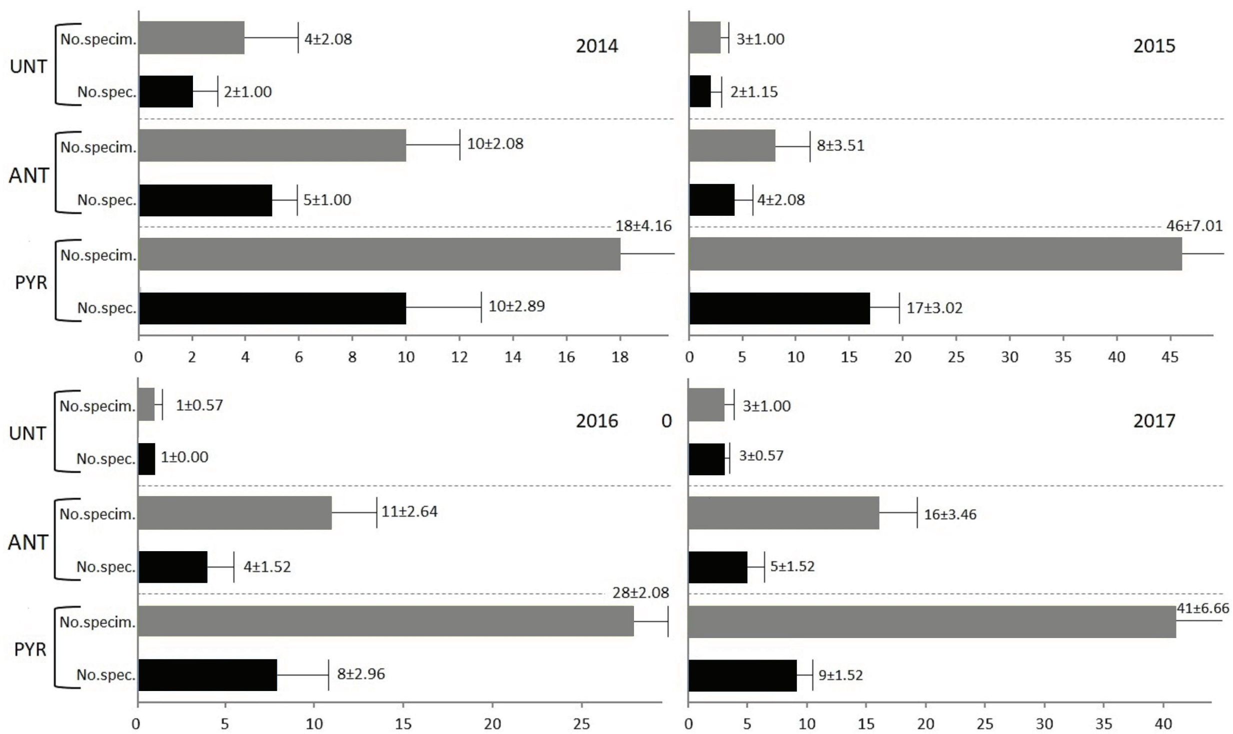

Fig. 1. Mortality of arthropod specimens and species numbers (± standard error) after the different chemical treatments in four consecutive years (measured by grounded tarpaulins)

UNT = untreatment; ANT = treated by anthranilic diamide; PYR = treated by anthranilic diamide and lambda-cyhalothrin

Table 3. Percentage of main canopy dwelling arthropod orders in examined maize parcels in four consecutive years

Years	Treatments	Hymenoptera	Diptera	Coleoptera	Hemiptera: Sternorrhyncha	Hemiptera: Heteroptera	Other insects	Other arthropods
2014	BEF. TRET.	2	4	77	2	5	3	7
	UNT.	2	18	52	9	11	2	6
	ANT.	4	24	40	13	12	1	6
	PYR.	0	0	59	12	29	0	0
2015	BEF. TRET.	2	4	30	31	25	3	5
	UNT.	1	2	44	24	25	2	2
	ANT.	2	4	26	40	20	4	4
	PYR.	0	0	0	54	46	0	0
2016	BEF. TRET.	2	7	25	30	18	8	10
	UNT.	2	4	45	22	16	6	5
	ANT.	3	5	42	15	21	8	6
	PYR.	0	0	71	5	24	0	0
2017	BEF. TRET.	8	10	42	11	21	3	5
	UNT.	6	4	66	2	17	4	1
	ANT.	0	6	69	2	17	6	0
	PYR.	0	0	73	0	27	0	0

BEF.TRET. = before treatment; UNT = untreatment; ANT = treated by anthranilic diamide; PYR = treated by anthranilic diamide and lambda-cyhalothrin

tication), broken tassels and stalks, and total number of plants damaged by *O. nubilalis* were observed compared to ANT and PYR (uniformly $P = 0.000$). The relationship between spraying and the damage caused by *H. armigera* could not be justified when analysing ears in the maturity period of maize ($P = 0.071$). No significant difference was observed either between two chemical treatments in terms of the percentage of damage types mentioned above ($P = 0.065$).

The mortality rate of arthropods (species and specimens) due to in-crop insecticide treatments in the four examined years is shown in Fig. 1. The effect of different chemical sprayings on the numbers of both species and specimens is clearly observable. The effect of PYR on maize arthropod communities is particularly remarkable. Several important maize pests were suppressed by these pyrethroid treatments, e.g. *Diabrotica v. virgifera* LeConte, *Ostrinia nubilalis* Hbn., *Glischrochilus quadrisignatus* Say, *Phyllotreta vittula* Redt., *Trigonotylus* spp., *Rhopalosiphum* spp. In addition, several useful (*Chrysoperla* spp., Coccinelidae: *Coccinella septempunctata* L., *Halyzia sedecimguttata* L, *Harmonia axyridis* Pallas, *Vibidia duodecimguttata* Poda) and in one case a vulnerable (Cerambycidae: *Calamobius filum* Rossi) species were sacrificed for this study. The different effect of two chemical treatments on devastated arthropod groups was confirmed by statistical analysis ($P = 0.014$). However, the impact of the applied insecticides of ANT is perceived as an estimation of absolute values. However, these distinctions could not be statistically proven ($P = 0.065$). The perished arthropods were mostly different larvae in these parcels.

The mortalities corrected by using Schneider-Orelli formula are shown in Table 2. The higher arthropod mortality triggered by PYR is evident. The recorded mortality values are 2–11-times higher in the case of PYR than when ANT (anthranilic diamide) was used. There is a particularly big difference in 2015, which can be explained by the low arthropod specimens' number in the examined maize culture. Thus, the present species appeared to be more vulnerable to the impact of the chemicals.

The distribution of canopy-dwelling arthropod communities assessed by the insect net method is shown in Table 3. The dominant insect orders were Coleoptera (in 2014, 2017) and Hemiptera (in 2015, 2016) as seen from the results of insect nettings. Before insecticide treatments, the useful insects were mainly represented by some ladybirds (*Coccinellidae*) (7.64 specimes/10 net strikes) and lacewings (*Chrysoperla* spp.) (4.15 specimens/10 net strikes). Interestingly, the dominant ladybird species was *H. axyridis* (95% of *Coccinellidae*) in this period. Besides the two orders mentioned above, the presence of other arthropod order did not exceed 27% in the years examined. The retrogression of maize canopy-dwelling arthropod community after chemical treatments was confirmed by insect netting. Particularly, the disappearance of useful ladybird species should be emphasized here.

Distinctly remarkable was the proportion of hemipterous species (aphids, phytophagous mirid bugs)

Table 4 Results of arthropods diversity analyses as a function of different treatments

	treatments	No. specim.	No. spec.	Margalef (MR)	Menhinick (M)
2014	BEF. TRET.	152	30	5.971	2.433
	UNT	125	30	6.213	2.683
	ANT	78	28	6.426	3.170
	PYR	74	5	1.160	0.581
2015	BEF. TRET.	74	25	5.808	2.906
	UNT	65	28	6.707	3.472
	ANT	51	27	6.867	3.780
	PYR	13	3	1.169	0.832
2016	BEF. TRET.	115	28	5.901	2.611
	UNT	122	21	4.354	1.901
	ANT	103	24	5.178	2.364
	PYR	48	4	1.033	0.577
2017	BEF. TRET.	48	27	6.974	3.897
	UNT	53	25	6.296	3.934
	ANT	36	20	5.581	3.333
	PYR	11	2	0.834	0.603

No. specim. = total number of specimens; No. spec. = number of species
BEF.TRET. = before treatment, UNT = untreated, ANT = treated with anthranilic diamide, PYR = treated with anthranilic diamide and lambda-cyhalothrin

increasing in the treated parcels. These maize-eating, but not economically significant insects (Aphidae: *Rhopalosiphum padi* L., *R. maydis* Fitch; Miridae: *Trigonotylus ruficornis* Geoffroy, *R. coelestialium* Kirkaldy) were the most dominant canopy-dwelling animals in the examined period of four consecutive years.

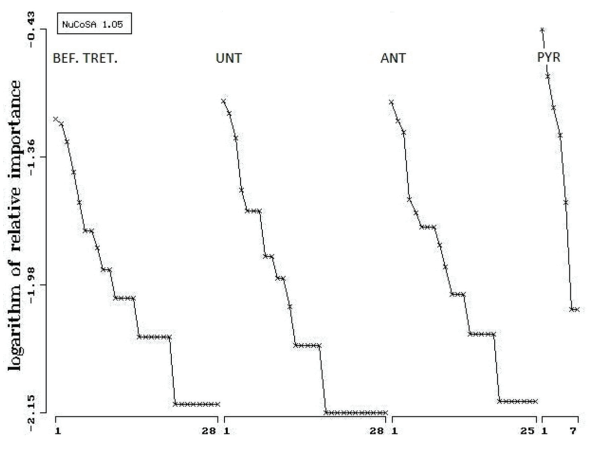

Fig. 2. Whittaker-type dominance-diversity curves of different treatments based on average data of four examined years
BEF.TRET. = before treatment, UNT = untreated, ANT = treated with anthranilic diamide, PYR = treated with anthranilic diamide and lambda-cyhalothrin

The conspicuous increase in the number of chrysomelid beetles was evinced in UNT and PYR parcels in 2016–2017 experimental years ($P = 0.001$). The percentage of attendances of arthropods in UNT and ANT parcels was mostly similar in each of the examined years, whereas the species assemblages of PYR were significantly altered and the number of species greatly decreased. Subsequently, more generations of aphids, the mass attendance chrysomelid beetles as well as the excessively disperse mirid bugs could be observed after this treatment. Other phytophagous and useful organisms completely disappeared or avoided the areas.

The numbers of arthropod specimens and species were usually the highest before the chemical treatments, and in the untreated parcels, while the smallest value was recorded in pyrethroid-sprayed areas (Table 4). This difference was unequivocally proven also by ANOVA analysis ($P = 0.000$). The species and specimen numbers of ANT treatments were lower than those of UNT, however, the distinctions were not confirmed by statistical examination ($P = 0.062$). The diversity indices (MR, M) followed the same trends. The diversity values are very similar in all examined parcels with the exception of the plot treated with double insecticides composition, PYR. This observation was re-confirmed by Whittaker-type dominance-diversity analysis (Fig. 2), in which the disappearance of rare species can be seen clearly as well as the shrinkage of species spectrums in PYR management. The disparities of the UNT parcels before and after chemical spraying could not be corroborated.

DISCUSSION

The use of in-crop insecticide treatments (ANT and PYR) revealed a significant effect against *O. nubilalis* in a four-year round field experiment. The mid-summer insect damage types (primary leaf mastication, broken tassels and stalks) were effectively decreased by the timed, early summer chemical sprayings in each experimental year. There was no evincible disparity between ANT and PYR regarding efficacy.

In contrast, both ANT and PYR treatments had no effect on the late summer ear mastication caused by *H. armigera*, which can be explained by the absence of pesticide translocation in withering maize plants and the hidden lifestyle of caterpillars (Bennett, 1957; Finlayson, MacCarthy, 1965).

The quantitative and qualitative parameters of the assemblages of the dead arthropods found and diversity alterations measured by canopy netting and grounded tarpaulins differed when different insecticides were applied. The minor absolute differences can be explained by the alteration of the swarming period of some dominant arthropods (e.g. *D. v. virgifera, T. ruficornis, T. coelestialium, Rh. padi*). The indices show a significant reduction of arthropod diversity caused by the pyrethroid effect (PYR), as opposed to anthranilic diamide (ANT), where this regression cannot be observed.

Rynaxypyr is a novel anthranilic diamide insecticide developed for controlling larvae of lepidopteran insect pests, as well as for controlling some species in the orders Coleoptera, Diptera and Hemiptera. It has a favourable toxicological and ecotoxicological profile (Hanning et al., 2009). Rynaxypyr (at recommended application rate 125 ml ha^{-1}) was more effective in the reduction of the larvae of *O. nubilalis* than other insecticides. According to Drobny et al. (2010), this insecticide is not toxic to pollinators and beneficial arthropods. It belongs to a new chemical class with a novel mode of action and is effective against insect populations that have developed resistance to other insecticide groups, thus representing an attractive new tool for integrated pest management programs (Hannig et al., 2009; Audisio et al., 2010).

The diversity of maize arthropod communities have been reduced by the pyrethroid treatment, lambda-cyhalothrin (Gerson, Cohen, 1989; Snodgrass, Scott, 2000). Its advantage is the destruction of pests, for instance imagoes of *D. v. virgifera, O. nubilalis, G. quadrisinatus*, as well as nymphs and adults of mirid bugs (Smith, Stratton, 1986; Inglesfield, 1989; Bommarco et al., 2011). Unfortunately, the effective control of these dangerous pests cannot be realised in practice because of the different times of their flight peaks (Domotor et al., 2009; Keszthelyi, 2010).

CONCLUSION

Recently, the technology of grain maize production has been continuously changing due to the spreading of insecticidal in-crop treatments in Europe. There is relatively little information about the consequences of insecticide spraying on non-target arthropods.

Rynaxypyr was effective for controlling larvae of lepidopteran insect pests, thus it appears that this active ingredient can be successfully applied in IPM programs. Utilisation of this pyrethroid ingredient poses a great danger to beneficial organisms (parasitoids, predators) present in maize fields. The effect of insecticides on these species can unambiguously contribute to the degradation of natural regulators. In addition, the eradication of these positive elements can cause the unexpected accumulation of some pests.

Our study pointed to the utilisation of broad spectrum insecticides (e.g. lambda-cyhalothrin) to become the dominant factor in biotopes of other non-target pest groups (aphids, mirid bugs) to a great extent. The background of this phenomenon can be explained by the vacancy of ecological niches and by the occupation of this vacuum by other pests. This disadvantageous consequence can induce mass appearance of newer plant damages and thus introduce a recurring yield decrease. More worrisome is the destruction of arthropod communities in neighbouring biocoenosis. The isolated populations of vulnerable species (as in our case: *C. filum*) can also suffer in extreme cases.

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