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Spatial Distribution of Cabbage Root Maggot (*Delia radicum*) and Clubroot (*Plasmodiophora brassicae*) in Winter Oilseed Rape Crops in the Czech Republic

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Abstract

Hlavjenka V., Seidenglanz M., Dufek A., Šefrová H. (2017): Spatial distribution of cabbage root maggot (*Delia radicum*) and clubroot (*Plasmodiophora brassicae*) in winter oilseed rape crops in the Czech Republic. Plant Protect. Sci., 53: 159–168.

The amount and spatial distribution of plants afflicted with cabbage root maggot (*Delia radicum*; CRM) and clubroot (*Plasmodiophora brassicae*) in winter oilseed rape crops were assessed in the Olomouc region (Northern Moravia, Czech Republic) over the course of 2012–2014. A total of 16 commercial rape fields were included in the assessments. Plants with tumours showed a significantly lower ($P < 0.001$) level of infestation induced by CRM (24% of plants infested) compared to plants without tumours (37% of plants infested). According to a generalised linear mixed model, plants with thicker hypocotyls are predisposed to significantly higher levels ($P < 0.001$) of root surface damage induced by CRM. The correlation analysis indicates rather weak or intermediate levels of correlation between the two variables (hypocotyls thickness × root surface damage induced by CRM). Both CRM and clubroot symptomatic plants showed a significant tendency for aggregation in rape crops, but not in all cases. Distributions of CRM and clubroot symptomatic plants were either significantly spatially dissociated or not associated in crops. Ovipositing *D. radicum* females showed some tendency to avoid zones with higher number of plants infected by *P. brassicae*. Distributions of CRM and hypocotyl thickness levels were significantly spatially associated in crops in several cases.

Keywords: within-field distribution of pests in crops; spatial associations between pests in crops; Integrated Plant Management; control of insect pests; winter oilseed rape diseases

This paper focuses on two organisms harmful to winter oilseed rape: larvae (maggots) of cabbage root flies (*Delia radicum*) and a pathogen which induces clubroot (*Plasmodiophora brassicae*). The damage induced by them and their importance for growers has substantially increased in the Czech Republic recently (Říčařová *et al.* 2016). Clubroot in brassica crops, caused by the obligate endoparasite *P. brassicae*, is recognised as a serious soilborne disease (Wallenhammar *et al.* 2012; Říčařová *et al.* 2016), usually associated with appreciable yield losses (Wallenhammar 1998; Wallenhammar *et al.* 2012; Říčařová *et al.* 2016). A well known symptom of the damage induced by *P. brassicae* visible on winter oilseed rape plants is their abnormally swollen roots (tumours occur on the main root and even adjacent roots), which cannot then properly supply the above ground plant parts with water and nutrients. Infected plants are poor in growth, wilt and turn yellow. Later (after winter) they have higher susceptibility to lodging (Chytilová & Dušek 2007; Kopecký & Cenklová 2013; Říčařová *et al.* 2016). Cabbage root maggots (CRM) attack the roots and root neck (Košťál 1993; Košťál & Finch 1994). Af-

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flicted places on the roots turn brown. Infested plants can turn yellow on the bottom leaf nodes or, when the level of infestation is high, they show dwarfing or die. Understanding the distribution of pests within crops is a prerequisite for spatially more accurate targeting of an Integrated Pest Management (IPM) strategy (Ferguson *et al.* 1999). Distributions of CRMs and clubroot symptomatic plants should not be random in winter oilseed rape crops. Both the organisms can aggregate into one or more foci in fields. At such places they show significantly higher abundances (Jones *et al.* 1993; Wallenhammar *et al.* 2012; Seidenglanz *et al.* 2013; Říčařová *et al.* 2016).

– The first objective of the paper is to examine if there are some correlations between the levels of infestations induced by the two harmful organisms and some host plant characteristics (hypocotyl thickness, main root length).
– The second objective is to describe the distribution patterns of plants carrying symptoms of damage induced by *D. radicum* respectively *P. brassicae* in winter oilseed rape crops.
– The third objective is to determine if the distributions of plants damaged by the two harmful organisms (symptomatic plants) are spatially associated (or dissociated) in crops.
– The fourth objective is to determine if the distribution of plants showing higher levels of damage induced by *D. radicum* larvae is spatially associated (or dissociated) with the distribution of plants with thicker hypocotyls in crops.
– The fifth objective is to find out if there is a spatial association between the distributions of clubroot symptomatic plants and plants with thicker (or thinner) hypocotyls in crops.

MATERIAL AND METHODS

All evaluations were made in winter oilseed rape crops located in the Olomouc region (northern part of Moravia, Czech Republic) in the course of autumns 2012–2014. The selected fields were not trial fields but common commercial fields showing a sufficient level of crop uniformity, regularity in shape (rectangle) and suitable acreage (0.6–30.2 ha). All the fields were under conventional cultivation practices typical for the region. With regard to pesticides, only herbicides were applied to the crops in autumn. Before sampling plants one preemergently applied spraying was made. At all localities the same active ingredients (metazachlor + clomazone, as tankmix)

Table 1. A list of selected fields along with brief descriptions of them. All plant samples were obtained from these fields during the autumns of 2012, 2013, and 2014

Locality (field)	Date of sampling	Growth stage (BBCH)^1	Field acreage (ha)	Soil texture	Crop rotation^2		
					1	2	3
Rapotín	18.10.2012	15–18	1.4	clay loam	barley^w	wheat^w	rape^w
Hrabenov	25.10.2012	15–18	7.5	sandy loam	barley^s	wheat^w	fodder
Hrabová	29.10.2012	18–19	10.5	clay loam	wheat^s	wheat^w	clover
Libina	29.10.2012	18–19	14.7	sandy loam	–	flax	clover
Bludov	12.11.2012	19	24	sandy loam	wheat^s	sugar beet	–
Plinkout	12.11.2012	19	16.2	sandy loam	barley^s	wheat^w	peas
Rapotín Ia	25.10.2013	14	15.7	sandy loam	barley^w	wheat^s	rape^w
Libina	4.11.2013	15–16	10.5	clay loam	barley^s	grain maize	wheat^w
Uničov	4.11.2013	15–16	30.2	sandy loam	barley^s	wheat^w	peas
Rapotín II.	10.12.2013	19	19.2	sandy loam	oat	clover	–
Rapotín Ib	2.12.2013	19	15.7	sandy loam	barley^w	wheat	rape^w
Nový Malín	13.11.2014	16	4	sandy loam	wheat^w	barley^s	grain maize
Zábřeh na Moravě	13.11.2014	14–15	4	clay loam	wheat^w	rape^w	clover
Libina	10.11.2014	16	2	sandy loam	wheat^s	wheat^w	clover
Bludov	10.11.2014	15–16	4	sandy loam	barley^w	rape^w	wheat^s
Rapotín	7.11.2014	12–13	0.6	sandy loam	barley^w	rape^w	barley^w

^{1}BBCH scale was used; ^{2}previous crops in the last three years; ^{w}winter type of crop; ^{s}spring type; conventional tillage with ploughing before sowing was used at all localities (except for Nový Malín = plough-less tillage)

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and the same rates were used. Descriptions of soil types and husbandry operations (varieties, crop rotations, and tillage systems) used on the localities are listed in Table 1. Meteo records for the three seasons originated from an automatic meteo station located in Šumperk (the station is part of a network of stations run by the Czech Hydrometeoterological Institute). The distances between the location of the meteo station and the individual fields varied from 1 km to 10 km. The above ground (+5 cm) and soil (–5 cm) temperatures recorded during the three autumns are illustrated in Figure 1.

In each of the fields the assessment places (AP) were designated (6 × 6 = 36 places in 2012 and 2013, 5 × 5 = 25 places in 2014, located in a rectangular grid pattern within the crop) before sampling plants. Each of the places was characterised by its *x* and *y* coordinates and by its number. Because the fields did not have the same acreages, the distances among the AP in individual grids varied from one to another (Figures 2–4). Ten plants from each AP were collected from every crop (n = 36 × 10 resp. 25 × 10 plants per field). Then the plants were transported to the laboratory in bags, each marked with the AP number. Sampling was carried out once a season in each crop. The times of sampling differed in some cases in one season (Table 1). The prevailing growth stages in the crop were always recorded (Table 1).

At the localities where the occurrence (at least sporadic) of clubroot symptomatic plants was recorded (Table 2) in crops the values of root neck thickness, root length and information on the presence (YES) or absence (NO) of tumours induced by *P. brassicae* were recorded for all plants. These data were then used for comparisons of hypocotyl thickness and root length values in the groups of plants with or without the clubroot symptoms. A total of 2182 plants were included in the calculations. Linear mixed-effects models, with values fitted using the nlme package (Version 3.1-118; Pinheiro *et al.* 2014) in the program R (Version 3.1.2; R Development Core Team 2014) were used for estimating the effects of *P. brassicae* infection symptoms on the root characteristics.

In contrast to the previous assessment, associations between the levels of root surface damage (%) induced by CRM and the levels of root neck thickness and main root length were analysed on plants collected from fields where clubroot symptomatic plants were absent (Table 2). Generalised linear mixed effects models with gamma errors and logarithmic link function, fitted using lme4 (version: see above) were used. In all models, locality (= field) was treated as the random effect. Likelihood-ratio tests and AIC in the model built according to Pinheiro and Bates (2000) were included in the analysis. Plots of presentation of fixed effects and covariates were created with the effects package (Version 3.0-3; Fox 2003) in R (version: see above).

In addition, all collected plants (transported from all selected fields regardless of the presence or absence of clubroot symptomatic plants in the crop) were also

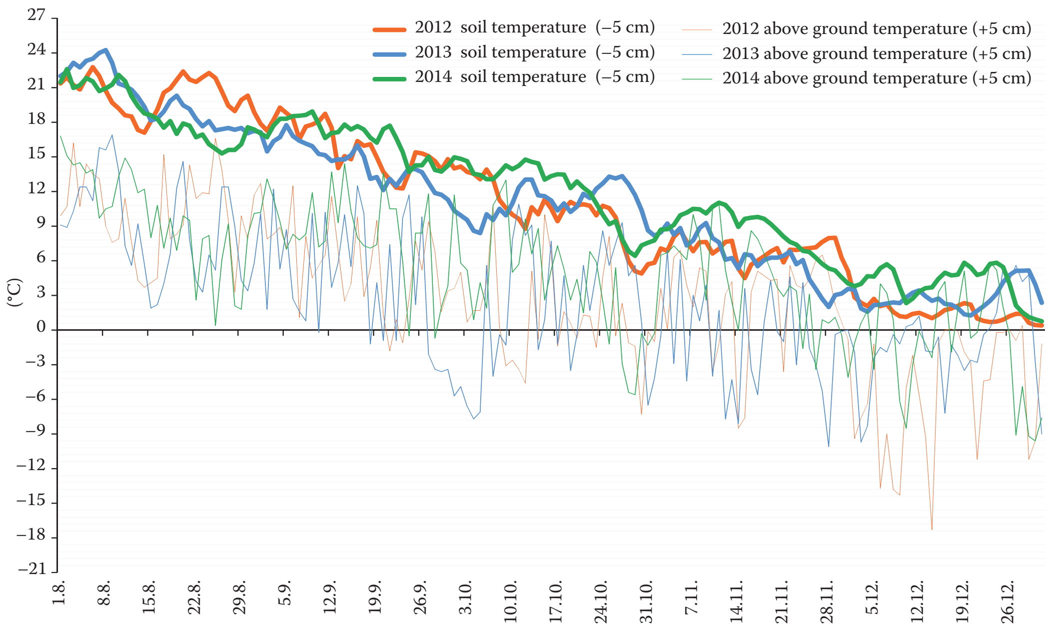

Figure 1. Mean air (5 cm above ground) and soil (5 cm below ground) temperatures recorded for the period 1.8. to 26.12. in seasons 2012, 2013 and 2014 (Meteo station located in Šumperk)

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Table 2. Differences in the levels of CRM and clubroot infestations in the compared crops and the results of correlation analysis (Spearman's correlation coefficient) between the root surface damage induced by CRM and two plant characteristics (hypocotyl thickness, main root length)

Locality (field)	Mean area of root damage induced by CRM (%)	Mean root		Number		r (P) (%)	
		neck thickness (mm)	length (cm)	plants injured by CRM (portion)	clubroot symptomatic plants (%)	hypocotyls thickness (mm) × root surface damage induced by CRM	main root length (cm) × root surface damage induced by CRM
Rapotín	6.91	5.59	11.06	112 (31.11)	15 (4.17)	0.18 (0.000)	0.03 (0.567)
Hrabenov	8.17	8.21	11.37	148 (41.11)	0	0.23 (0.000)	0.14 (0.006)
Hrabová	10.84	7.91	14.29	85 (23.61)	1 (0.27)	0.36 (0.000)	0.26 (0.000)
Libina	16.33	7.48	10.12	136 (37.77)	232 (64.44)	0.08 (0.136)	0.27 (0.000)
Bludov	9.87	9.14	13.75	102 (28.33)	5 (1.38)	0.25 (0.000)	0.25 (0.000)
Plinkout	7.93	7.93	16.07	127 (35.28)	0	0.21 (0.000)	0.12 (0.018)
Rapotín Ia	6.46	4.86	7.98	103 (28.61)	0	0.15 (0.003)	–0.04 (0.424)
Libina	9.61	4.82	8.46	122 (33.88)	0	0.27 (0.000)	0.09 (0.078)
Uničov	12.19	5.68	11.71	185 (51.38)	0	0.27 (0.000)	–0.02 (0.719)
Rapotín II	8.20	5.39	8.94	107 (29.72)	0	0.09 (0.079)	0.02 (0.710)
Rapotín Ib	5.97	3.82	10.82	98 (27.22)	0	0.22 (0.003)	–0.01 (0.891)
Nový Malín	18.82	9.73	10.99	134 (53.60)	43 (17.20)	0.12 (0.078)	0.13 (0.034)
Zábřeh na Moravě	6.25	7.17	10.74	77 (30.80)	0	0.14 (0.027)	0.15 (0.019)
Libina	10.47	7.70	10.14	75 (30.00)	13 (5.20)	0.24 (0.000)	0.05 (0.461)
Bludov	5.30	8.90	8.93	51 (20.40)	119 (47.60)	0.19 (0.003)	0.28 (0.000)
Rapotín	0.90	4.62	9.75	30 (20.00)	0	0.26 (0.000)	0.06 (0.298)

r – Spearman's correlation coefficient; *P* – probability value

used for analysing correlations between the levels of root surface damage (%) induced by CRM and the levels of root neck thickness and main root length. Spearman's correlation coefficients were calculated for this (Statistica v12; 1984–2015).

For analysing spatial distributions of CRM (the distribution of plants with different levels of root surface damage induced by CRM) and clubroot symptomatic plants (plants with tumours on roots) in crops, Spatial Analysis by Distance IndicEs (SADIE) was used (Perry 1995, 1996). For CRM, as baseline information, the mean levels of root surface damage (%) per AP (10 plants per AP assessed; 36 or 25 AP per field) were used. For clubroot, the numbers of clubroot symptomatic plants per AP were used for the same purposes. In addition the distributions of hypocotyl thickness levels (average values stated for 10 plants per AP used in calculations) were determined. For all crops aggregation indices I_a were calculated for both of the harmful organisms and for the hypocotyl thickness values. In the case of non-random distribution of the values in crops, with their significant tendency to aggregate in clusters (patch or gap clusters) the value of I_a is higher than unity ($I_a > 1$ for $P < 0.05$). For the calculations SadieShell software Version 2.0 was used (Perry 1995, 1996, 1998; Ferguson *et al.* 2006). For graphic visualisation of SADIE results Statistica v12 software (1984–2015) was used.

To compare two distributions (distribution of plants showing various levels of damage induced by CRM vs. distribution of clubroot symptomatic plants; distribution of plants showing various levels of damage induced by CRM vs. distribution of plants with various values of hypocotyl thickness; distribution of clubroot symptomatic plants vs. distribution of plants with various values of hypocotyl thickness) in the individual fields, SADIE – Quick Association calculates an overall spatial association index, X (upper case chi), based on the similarity of the local clustering indices from the two distributions – these were calculated earlier as described in the previous paragraph (Perry 1998; Ferguson *et al.* 2006). Values of X are > 0 for distributions that are associated, around zero for distributions positioned at random with respect to one another, and < 0 for distributions that are dissociated. The significance

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of X is tested by comparison with randomisations in which values of the local cluster indices are reassigned among the sample locations (Perry 1998).

RESULTS

The compared winter oilseed rape crops were at various growth stages at the times of sampling (Table 1). The mean levels of root length, root neck thickness, root damage induced by CRM, and clubroot incidence proved different among the fields (Table 2). Plants without tumours had a mean neck thickness of 7.85 mm, and plants with tumours showed insignificantly (P = 0.098) thicker hypocotyls: 8.45 mm (Table 3). Plants without tumours had a mean main root length of 12.13 cm, and plants with tumours showed significantly (P < 0.001) shorter roots: 8.82 cm (Table 3). Overall, 35% of plants were infested with CRM. There was a difference in the proportions of CRM infested plants between the groups of plants with (YES) and without (NO) tumours on their roots. Plants with tumours showed significantly lower (P < 0.001) levels of CRM infestation (24% of plants infested) in comparison with plants without tumours (37% of plants infested; Table 3). According to the generalised linear mixed model, plants with thicker hypocotyls are predisposed to significantly higher levels (P < 0.001) of root surface damage induced by CRM. From the total number of fields compared (16), in 13 cases a significant positive correlation (P < 0.05) between hypocotyl thickness and root surface damage induced by CRM was actually confirmed (Table 2). The absolute values of correlation coefficient *r* indicate rather weaker or intermediate levels of correlation between the two variables. Significant positive correlations (P < 0.05) between root length and root surface damage induced by CRM were also proved in 8 fields (from a total of 16) (Table 2). However, the absolute values of correlation coefficient *r* indicate no or weak correlation between the two variables. These results show some contradiction to the estimates based on the linear mixed model. According to this analysis the plants with longer main roots should be predisposed to significantly lower levels (P < 0.001) of root surface damage induced by CRM.

In seven cases (Rapotín 2012, Hrabenov 2012, Hrabová 2012, Libina 2012, Bludov 2012, Rapotín Ib 2013, Rapotín 2014) the levels of damage induced by CRM were not distributed randomly in crops, the plants were aggregated in clusters there according to the levels of their damage (I_a values higher than unity, P < 0.05; Table 4, Figures 2–4). Distribution of clubroot symptomatic plants (plants with tumours on roots) in only slightly infested crops (Rapotín 2012, Bludov 2012, Libina 2014) was not uniform. Symptomatic plants were significantly aggregated there into relatively small focuses located mostly near the field margins (I_a values higher than unity, P < 0.05). Contrary to that, diameters of gap clusters were substantially larger (Figure 4). In crops with markedly stronger disease incidence (Libina 2012, Nový Malín 2014, Bludov 2014) the symptomatic plants were distributed in a random pattern (Nový Malín 2014, Bludov 2014) or they were almost uniformly distributed throughout the crop (Libina 2012). The results of SADIE also indicate that in seven cases (Rapotín 2012, Libina 2012, Bludov 2012, Plinkout 2012, Libina 2013, Rapotín II 2013, and Rapotín Ib 2013) the levels of hypocotyl thickness were not randomly distributed in the crops. In these crops significant ag-

Table 3. Mean values of root length, hypocotyl thickness and portions of plants infested by CRM in samples collected from crops with a confirmed occurrence of clubroot symptomatic plants. The values are listed separately for groups of plants with (YES) resp. without (NO) tumours (induced by *P. brassicae*) on roots

Clubroot		*N*	Mean	Std. error of mean	Median	Minimum	Maximum	Range
Length of the main root (cm)	NO	1759	12.13	0.08	12	2	23	21
	YES	427	8.82	0.13	9	2	18	16
	Total	2186	11.48	0.07	11	2	23	21
Hypocotyl thickness (mm)	NO	1759	7.85	0.06	8	0.8	18	17.2
	YES	427	8.45	0.12	8	2	18	16
	Total	2186	7.97	0.05	8	0.8	18	17.2
Portion of plants infested by CRM (%)	NO	1759	37	0.01	×	×	×	×
	YES	427	24	0.02	×	×	×	×
	Total	2186	35	0.01	×	×	×	×

doi: 10.17221/181/2015-PPS

Table 4. Distributions of plants with various levels of damage induced by CRM, distributions of clubroot symptomatic plants and distributions of plants according to their hypocotyl thickness (SADIE results: aggregation indices, I_a) and spatial association between their distributions (Quick Association results: overall spatial association indices, X) in crops on the compared fields

Locality (field)	Index of aggregation (I_a)			Index of association (X)		
	CRM	clubroot	hypocotyl thickness	CRM vs. clubroot	CRM vs. hypocotyl thickness	clubroot vs. hypocotyl thickness
Rapotín	**1.38**	**1.41**	**1.64**	0.05 (0.413)	**0.43 (0.005)**	0.25 (0.094)
Hrabenov	**1.33**	×	1.14	×	**0.42 (0.005)**	×
Hrabová	**1.35**	×	1.12	×	**0.63 (< 0.001)**	×
Libina	**1.47**	0.96	**1.61**	**–0.55 (0.992)**	**–0.49 (0.996)**	0.16 (0.204)
Bludov	**1.44**	**1.52**	**1.35**	0.04 (0.421)	0.17 (0.201)	0.22 (0.113)
Plinkout	1.17	×	**1.36**	×	0.33 (0.047)	×
Rapotín Ia	1.04	×	1.08	×	0.14 (0.239)	×
Libina	1.1	×	**1.51**	×	0.20 (0.142)	×
Uničov	0.97	×	1.22	×	0.12 (0.274)	×
Rapotín II	1.19	×	**1.62**	×	–0.07 (0.646)	×
Rapotín Ib	**1.59**	×	**1.76**	×	**0.43 (0.013)**	×
Nový Malín	1.04	0.78	0.8	–0.01 (0.532)	0.14 (0.314)	–0.03 (0.540)
Zábřeh na Moravě	1.13	×	0.83	×	–0.14 (0.731)	×
Libina	1.03	**1.56**	1.27	**–0.53 (0.991)**	**0.598 (0.002)**	–0.38 (0.969)
Bludov	1.06	1.13	0.92	0.08 (0.373)	–0.09(0.648)	0.16 (0.048)
Rapotín	**1.45**	×	0.94	×	**0.51 (0.018)**	×

I_a values higher than unity for $P < 0.05$ indicate that CRM resp. clubroot symptomatic plants resp. plants according to their hypocotyl thickness were significantly aggregated at the field (bold values in the table); X values higher than 0 for $P < 0.025$ indicate that distributions of the compared variables were spatially associated in crops (positive bold values). X values lower than 0 for $P > 0.975$ indicate significant dissociation in their distributions in crops (negative bold values); × – non assessed

gregation of plants in accordance with their hypocotyl thickness to clusters with different shape, acreage and location was recorded (I_a values higher than unity, $P < 0.05$; Table 4).

Distributions of plants damaged by CRM and clubroot symptomatic plants were not spatially associated (or dissociated) in four cases (very low X values). But in two crops (Libina 2012: 64.44 % of

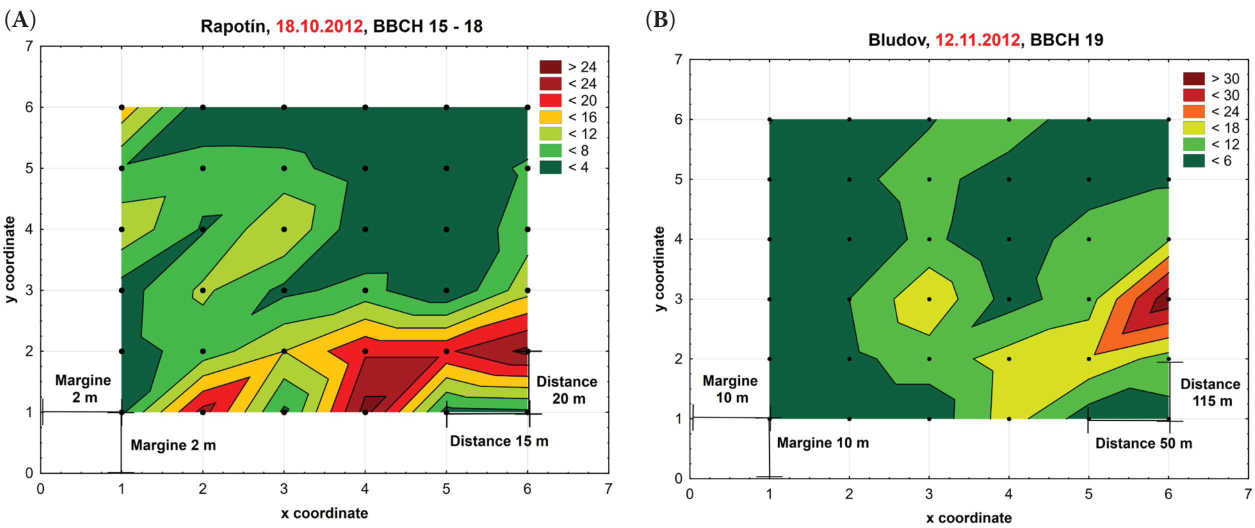
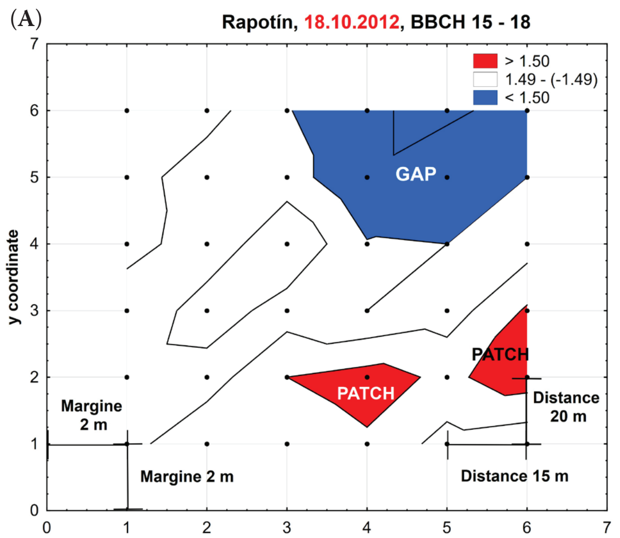
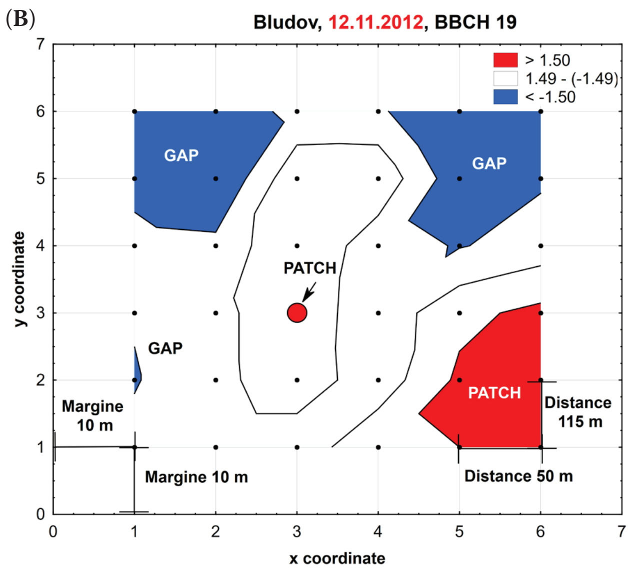


Figure 2. Distribution of winter oilseed rape plants with different levels of root damage induced by CRM (root surface damage, %) within the crop in (**A**) Rapotín 2012 and (**B**) Bludov 2012. The black points mark assessment places (AP) arranged in a rectangular grid (6 × 6 points; 36 sampling places)

doi: 10.17221/181/2015-PPS





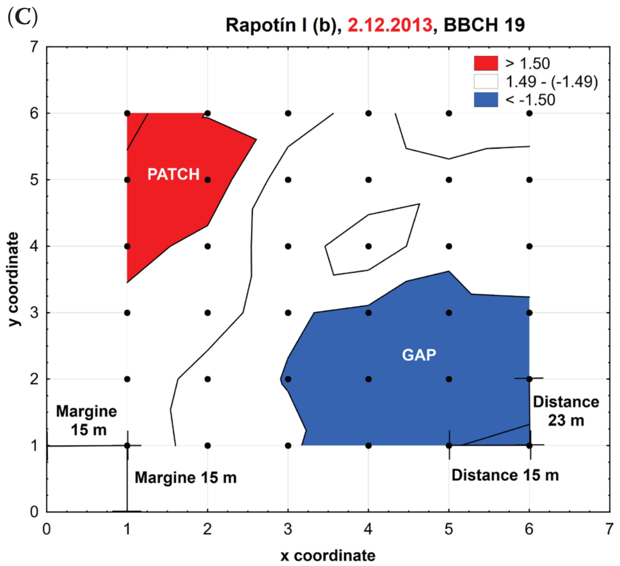


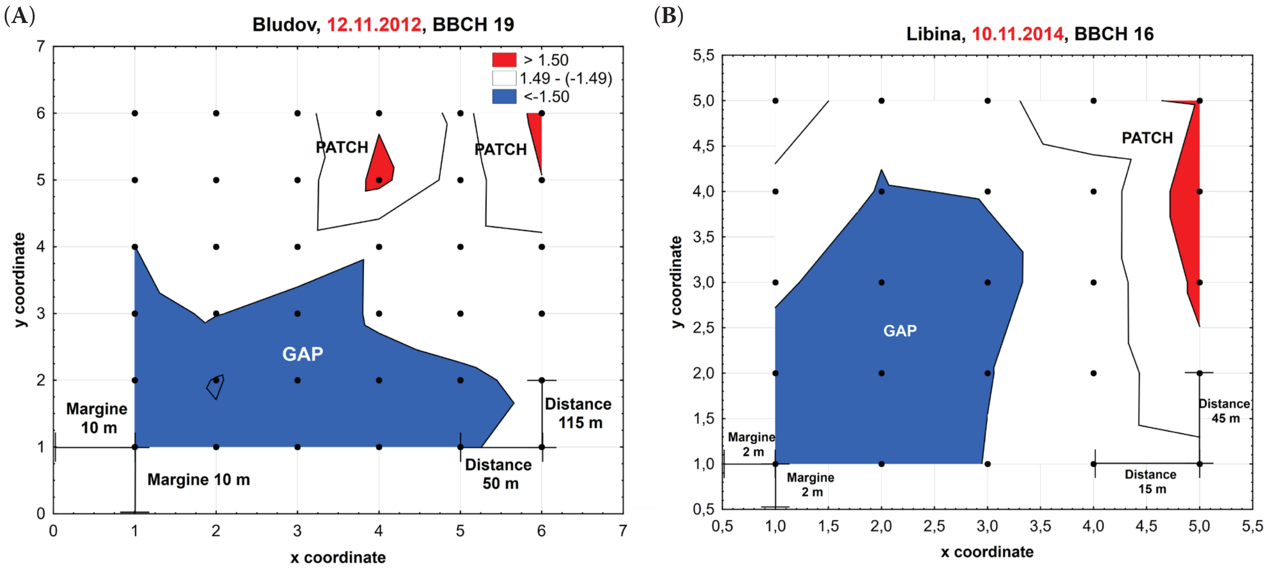
Figure 3. Location of patch (red areas) and gap (blue areas) clusters determined for CRM (on the basis of root surface damage, %) within the crop in (**A**) Rapotín 2012, (**B**) Bludov 2012, and (**C**) Rapotín Ib 2013 (SADIE). The black points mark the AP arranged in a rectangular grid (6 × 6 points; 36 sampling places)

plants showed symptoms of clubroot; Libina 2014: 5.20 % of plants showed symptoms of clubroot) their distributions were significantly dissociated (negative values of X, $P > 0.975$; Table 4). In six cases (Rapotín 2012, Hrabenov 2012, Hrabová 2012, Rapotín Ib 2013, Libina 2014, Rapotín 2014) the distributions of damage levels induced by CRM with the levels of hypocotyl thickness were significantly associated in crops and in one case (Libina 2012) they were significantly dissociated. There was not a significant spatial association (or dissociation) between the distributions of clubroot symptomatic plants and the levels of hypocotyl thickness in crops.

DISCUSSION

Plants with thicker hypocotyls were predisposed to higher levels of root surface damage induced by CRM (generalised linear mixed model), positive correlations between the levels of hypocotyl thickness and the levels of damage induced by CRM proved significant in the most of the crops (in 13 out of 16) and the levels of damage induced by CRM were spatially associated with the distribution of hypocotyl thickness levels in crops in six cases (significant in 6 out of 16 crops). The findings indicate that *D. radicum* (ovipositing females) shows a clear (although not very intensive) tendency for preferring plants with thicker root necks and some tendency for preferring zones with stronger plants (and at the same time avoiding the zones with weak plants) in crops. According to Dosdall *et al.* (1995) there is a correlation between root injury (induced by CRM) and stem diameter. The authors concluded that increases in basal stem diameter resulted in corresponding and statistically significant increases in damage to roots by feeding of CRM for *Brasica rapa* ($r = 0.58$, $P < 0.05$) and

doi: 10.17221/181/2015-PPS



The black points mark the AP arranged in a rectangular grid (6 × 6 points; 36 sampling places)

The black points mark the AP arranged in a rectangular grid (5 × 5 points; 25 sampling places)

Figure 4. Location of patch (red areas) and gap (blue areas) clusters determined for clubroot symptomatic plants (the numbers of plants with tumours on roots per AP were used as the base data for SADIE calculations) within the crop in (**A**) Bludov 2012 and (**B**) Libina 2012 (SADIE)

B. napus (r = 0.61, P < 0.05). *Delia* spp. egg numbers were also positively and significantly correlated with basal stem diameter for *B. rapa* (r = 0.82, P < 0.05) and *B. napus* (r = 0.70, P < 0.05). In general this agrees with our data; we also confirmed a positive correlation but the levels of its intensity were substantially lower in this study. Košťál *et al.* (2000) also emphasised the importance of plant quality for *D. radicum* females searching for sufficient ovipositing places. This is especially related to one step of the female's behaviour close to the substrate surface: stem base circling. At this phase the female, heading down, makes more or less complete circles around the stem base. Plants with thicker hypocotyls are also more attractive for egg-laying *D. radicum* females according to Bittner (2006).

Plants were significantly aggregated in crops in accordance to the levels of their damage induced by CRM in seven cases (2012: Rapotín, Hrabenov, Hrabová, Libina, Bludov; 2013: Rapotín Ib; 2014: Rapotín). A tendency for aggregation of *D. radicum* in crops has also been demonstrated by Jones *et al.* (1993) and Finch and Collier (2000).

In one case there was a significant spatial dissociation (negative values of X) between the distribution of damage levels induced by CRM and hypocotyl thickness levels. That was recorded in the crop most afflicted by clubroot (Libina 2012). The high proportions of symptomatic plants and their almost uniform distribution (I_a for clubroot symptomatic plants: 0.96) in the crop probably influenced the behaviour of *D. radicum* (and its distribution) in the crop and as well as the levels of hypocotyl thickness (infected plants tended to have thicker root necks maybe due to tissue swelling; Table 3). In the other field heavily infested by clubroot (Bludov 2014) the distributions of CRM and hypocotyl damage levels were also spatially dissociated in the crop, but the value of X was low (–0.09, insignificant). The results indicate that in crops with high clubroot incidence, *D. radicum* preference for stronger plants (r values for CRM × hypocotyl thickness in Libina 2012 resp. Bludov 2014 = 0.08 resp. 0.19) and for zones in the crop with stronger plants (X values for CRM × hypocotyl thickness in Libina 2012 resp. Bludov 2014 is –0.49 resp. –0.09) does not exist or is markedly decreased. Unfortunately, any papers (published results) which could either support or question this hypothesis are not known to us.

From the two fields with a strong incidence of clubroot (Libina 2012, Bludov 2014), a significant dissociation between the distributions of CRM and clubroot symptomatic plants in crops was recorded in only one case (Bludov 2012, the crop most damaged by clubroot). The other confirmed case of significant spatial dissociation between the two distributions occurred in a markedly less afflicted crop in Libina 2014. In the other four crops with confirmed (at least sporadic) occurrence of clubroot (Rapotín 2012, Bludov 2012,

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Nový Malín 2014, and Bludov 2014) the distributions were positioned either at random with respect to one to another (low values of X) or they were insignificantly dissociated (low negative values of X). According to Dosdall *et al*. (1995) there is a positive correlation between the numbers of *D. radicum* eggs per plant and the levels of damage induced by CRM – a higher level of damage induced by CRM in some places is a consequence of more eggs previously being laid there. On the basis of our results *D. radicum* females show a tendency either to avoid the zones with higher portions of plants infested with *P. brassicae* or, at least, not to search for them during the time of egg-laying (practically in all cases the values of X were negative or very close to zero). The results also indicate there could be a spatial niche partitioning between *D. radicum* and *P. brassicae* in rape crops directed: (1) by readily moving *D. radicum* females laying fewer eggs in the zones where higher frequencies of plants infected with *P. brassicae* occur or (2) by higher mortality of CRM on the infected plants. Unfortunately, any published data which could either support or question the hypothesis of *D. radicum* and *P. brassicae* spatial niche partitioning in rape crops is not known to us. Host plant location and recognition by *D. radicum* have been studied in relatively great detail, but more attention has been given to chemical stimuli, above all secondary metabolites of the host plants (Traynier 1967; Finch 1978; Hawkes & Coaker 1979; Nottingham 1988). The role of visual stimuli is also very important, especially in the phase before landing (Prokopy *et al*. 1983; Košťál 1991). In our opinion nobody has studied yet how the chemical or visual stimuli affect the distribution of *D. radicum* adults (or larvae) in large scale plots under field conditions and if *D. radicum* adults can perceive some stimuli emitted by rape plants infected by *P. brassicae* (especially during the early stage of infection).

Clubroot symptomatic plants significantly aggregated into patches in rape crops, but not in all cases. This tendency is clearly apparent in crops with lower frequencies of symptomatic plants (Rapotín 2012, Bludov 2012, Libina 2014). In crops with higher levels of clubroot incidence, the symptomatic plants show a tendency for random (Nový Malín 2014, Bludov 2014) or almost uniform (Libina 2012, practically the whole crop infested at the same level) distribution in crops. Wallenhammar *et al*. (2012) studied the distribution of *P. brassicae* spores (inoculums) in arable soil in Sweden. They demonstrate the patchiness of inoculum distribution even within a small area and emphasise the importance of including an adequate number of subsamples when sampling. They also consider the use of bait plants as the most reliable diagnostic method for assessing soils for the presence of *P. brassicae*, despite the availability of more modern techniques (Faggian & Strelkov 2009). Contrary to Wallenhammar *et al*. (2012) we did not assess the abundance of *P. brassicae* spores in soils, but through the expression of symptoms on the plants (susceptible varieties) always sampled from the all regions (rectangular grids) of the compared fields and through the usage of SADIE (Perry 1995, 1996), our results also indicate the patchiness of *P. brassicae* distribution within fields – but only there where lower levels of disease incidence in crops were recorded. Říčařová *et al*. (2016) also documented a tendency of *P. brassicae* spores (inoculum) and clubroot symptomatic plants to aggregate to one or more focuses in fields. According to the authors this type of distribution is the most frequent in infected arable soils in the Czech Republic.

CONCLUSIONS

(1) There is a positive significant correlation between the levels of damage induced by *D. radicum* larvae on roots and the thickness of root necks under field conditions, but the correlation is not strong.

(2) *D. radicum* larvae and clubroot symptomatic plants show a tendency towards significant aggregation in crops, but not in all cases.

(3) *D. radicum* females show a tendency either to avoid the zones with higher levels of clubroot incidence or, at least, not to search for them in crops.

(4) *D. radicum* (ovipositing females) shows a tendency to prefer the zones with stronger plants (and at the same time avoiding the zones with weak plants) in crops. But this is not significant in all crops. High clubroot incidence in a crop can decrease the preference.

(5) The distribution of clubroot symptomatic plants in crops is not spatially related to the distribution of plants with thicker or weaker root necks in crops.

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