Research Article

Janetta Niemann, Justyna Szwarc*, Jan Bocianowski, Dorota Weigt, Marek Mrówczyński

In-field screening for host plant resistance to *Delia radicum* and *Brevicoryne brassicae* within selected rapeseed cultivars and new interspecific hybrids

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**Abstract:** Rapeseed (*Brassica napus*) can be attacked by a wide range of pests, for example, cabbage root fly (*Delia radicum*) and cabbage aphid (*Brevicoryne brassicae*). One of the best methods of pest management is breeding for insect resistance in rapeseed. Wild genotypes of *Brassicaceae* and rapeseed cultivars can be used as a source of resistance. In 2017, 2018, and 2019, field trials were performed to assess the level of resistance to *D. radicum* and *B. brassicae* within 53 registered rape-seed cultivars and 31 interspecific hybrid combinations originating from the resources of the Department of Genetics and Plant Breeding of Poznań University of Life Sciences (PULS). The level of resistance varied among genotypes and years. Only one hybrid combination and two *B. napus* cultivars maintained high level of resistance in all tested years, i.e., *B. napus* cv. *Jet Neuf* × *B. carinata* – PI 649096, Galileus, and Markolo. The results of this research indicate that resistance to insects is present in *Brassicaceae* family and can be transferred to rapeseed cultivars. The importance of continuous improvement of rapeseed pest resistance and the search for new sources of resistance is discussed; furthermore, plans for future investigations are presented.

**Keywords:** *Brassica napus*, rapeseed, pest resistance, hybrids, cabbage root fly, cabbage aphid

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**1 Introduction**

Rapeseed (*Brassica napus* L. *ssp. oleifera* Metzg.) is one of the three most important sources of vegetable oil in the world. The European Union (EU) was the world leader in rapeseed production in 2017 (22 million tons), followed by Canada (21 million tons), China (13 million tons), India (7.9 million tons), Australia (4.3 million tons), and Ukraine (2.1 million tons) [1]. The greatest producers of rapeseed in the EU are France, Germany, Poland, Romania, Great Britain, the Czech Republic, Hungary, Denmark, and Slovakia, respectively [2,3]. Protection from pests is an essential part of breeding programmes – for example, yield losses caused by pests in Poland can range from 15 to 50% [4]. Moreover, a significant increase in the threat from pests is expected, related both to climatic changes and to agrotechnical simplifications [5,6].

Rapeseed plants in Poland are attacked by a wide range of pests. Among them, two economically important insects can be distinguished – cabbage root fly (*Delia radicum* L.) (Diptera: Anthomyiidae) and cabbage aphid (*Brevicoryne brassicae* L.) (Homoptera: Aphididae). The cabbage aphid is one of the most important and commonly occurring insect pests of rapeseed worldwide [7]. *Brevicoryne brassicae* causes significant yield losses in many crops in the family *Brassicaceae*, including mustards and crucifers. Heavy infestation can result in severe plant damage, causing death of seedlings and young transplants. Symptoms in larger plants include curling and yellowing of leaves, stunting of plants, and deformation of developing heads [8,9].

The cabbage root fly is one of the most important pests of many *Brassica* crops in the temperate regions of Europe and North America. After overwintering as pupae and hatching in early spring, females lay eggs in close proximity to the host plant. Depending on the temperature, eggs hatch in about 4 days [8]. The number of generations varies each year from one to four, depending on climatic conditions [10]. Larvae of *D. radicum* can damage plants by feeding on root tissue, resulting in

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* Corresponding author: Justyna Szwarc, Department of Genetics and Plant Breeding, Poznań University of Life Sciences, Dojazd 11, 60-632, Poznań, Poland, e-mail: justyna.szwarc@up.poznan.pl
Janetta Niemann, Dorota Weigt: Department of Genetics and Plant Breeding, Poznań University of Life Sciences, Dojazd 11, 60-632, Poznań, Poland
Jan Bocianowski: Department of Mathematical and Statistical Methods, Poznań University of Life Sciences, Wojewódzkiego 28, 60-637, Poznań, Poland
Marek Mrówczyński: Institute of Plant Protection – National Research Institute, Władysława Węgorka 20, 60-318, Poznań, Poland

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wilting of leaves or the entire plant and eventually reducing the yield and quality of the crop. Moreover, roots attacked by *D. radicum* are more susceptible to secondary root pathogens, such as *Fusarium* spp. [10,11].

To date, three resistance mechanisms have been recognized in the interaction of *Delia–Brassica* and *Brevicoryne–Brassica*: antixenosis, antibiosis, and tolerance [12]. Antixenosis (non-preference, avoidance) denotes morphological or chemical plant traits that make it unattractive for insects. For example, variation in cabbage leaf colour makes it less attractive to *B. brassicae* [13]. Antibiosis resistance is based on adverse effects of the plant after feeding [14]. Antibiosis does not prevent infestation, but rather causes increased mortality or delayed development of insects. Tolerance means the ability of a plant to reduce inflicted damage. A tolerant host is able to grow and reproduce despite the presence of a high number of insects [12,13]. In contrast to antixenosis and antibiosis, tolerance is independent of the herbivore response but is an adaptive mechanism helping plants to grow normally under biotic stress [15].

For most growers, the use of pesticides is an essential form of protection against harmful organisms [16]. However, there has been an increasing emphasis on the use of environmentally friendly methods of pest control. For example, in 2013, the EU restricted the use of certain neonicotinoids, and in 2018, banned three main neonicotinoids (Commission Implementing Regulation [EU] 2018/783, 2018/784, 2018/785). Moreover, Integrated Pest Management, which focuses on reducing the use of pesticides, has become compulsory for all farmers in the EU since 2014 (Directive 2009/128/EC). Therefore, breeding cultivars with resistance to insect pests fits perfectly into the currently applicable requirements and modern environmentally friendly trends [17,18]. The natural genetic variation among the wild relatives of crop species can provide good sources of novel host plant resistance [19].

Wild and related species of the *Brassicaceae* family are proved to be a valuable source of desirable agronomic traits. For example, *Sinapis alba* has been shown to be tolerant to crucifer flea beetle [20]; *B. juncea*, *B. carinata*, and *B. nigra* can be used to transfer blackleg resistance genes [21]; and *B. rapa*, *B. carinata*, and *S. alba* may act as a source of pod shattering resistance [22]. The assessment of the level of resistance within various *Brassicaceae* wild species or *Brassicaeae* hybrids may help identify genotypes with desired traits, which then can be included into rapeseed breeding programmes.

The aim of this research was to determine the range of pest resistance levels among selected rapeseed cultivars and new *Brassica* hybrid combinations obtained from the Department of Genetics and Plant Breeding of Poznań University of Life Sciences (PULS). This study has been conducted to identify the sources of resistance not only in rapeseed cultivars but also in other brassicaceous species. Consequently, this strategy will allow the assessment of the genetic resistance of interspecific *Brassica* hybrids in comparison with the parental forms in the future.

To the best of our knowledge, this is one of the few studies in which in-field comparison of resistance has been made among rapeseed cultivars and interspecific hybrids towards economically important insect pests.

## 2 Materials and methods

### 2.1 Experimental design

The experiment was conducted for three consecutive years (2017, 2018, and 2019) on the testing fields in PULS experimental station Dłoń (51°41′23″N, 17°04′10″E) located 100 km south from Poznań, Poland. The whole experiment was set up in a completely randomized block design with five replications (on the basis of six plants) in each year (*N* = 90), and each single plot size was 10 m² with a 0.30 row distance and a sowing density of 60 seeds/m². The field experiment in Dłoń was conducted on typical heavy soil of quality class III [23]. Agricultural practices were optimal for local agroecological conditions in Dłoń. Plots were harvested using a plot harvester. In crop seasons 2016/2017, 2017/2018, and 2018/2019, the weather conditions were normal for Poland. The seasonal rainfall in Dłoń was 667 mm in 2017, 372 mm in 2018, and 393 mm in 2019, whereas the mean annual temperatures in 2017, 2018, and 2019 were 9.6, 10.8, and 11.1°C, respectively.

### 2.2 Plant material

Seeds of 53 rapeseed cultivars and 31 hybrid combinations were used as the research material (Table 1). All *Brassica* interspecific hybrids were generated in the Department of Genetics and Plant Breeding of PULS with the application of *in vitro* culture of isolated embryos according to the method described by Niemann et al. [24]. In order to obtain interspecific hybrids with genetic
Table 1: List of Brassicaceae hybrids and B. napus cultivars used as the research material

| No. of line | Cross-combination                           | No. of line | Cross-combination                          |
|-------------|----------------------------------------------|-------------|---------------------------------------------|
| H1          | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{rapa ssp. pekinensis} 08\) | H17         | \(B. \text{napus} \times \text{Lisek} \times B. \text{carinata} \text{Dodola}\) |
| H2          | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{rapa ssp. pekinensis} 08\) | H18         | \(B. \text{napus} \times \text{California} \times B. \text{fruticulosa} \text{– PI649097}\) |
| H3          | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{carinata} \text{PI 649091}\) | H19         | \(B. \text{napus} \times \text{Lisek} \times B. \text{fruticulosa} \text{– PI649097}\) |
| H4          | \(B. \text{napus} \times \text{Górcański} \times B. \text{rapa ssp. pekinensis} 08.007574\) | H20         | \(B. \text{napus} \times \text{Lisek} \times B. \text{fruticulosa} \text{– PI649099}\) |
| H5          | \(B. \text{napus} \times \text{Górcański} \times B. \text{rapa ssp. pekinensis} 08.007569\) | H21         | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{carinata} \text{– PI 649094}\) |
| H6          | \(B. \text{napus} \times \text{Górcański} \times B. \text{rapa ssp. Chinensis}\) | H22         | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{carinata} \text{– PI 649096}\) |
| H7          | \(B. \text{napus} \times \text{Lisek} \times S. \text{alba cv. Bamberka}\) | H23         | \(B. \text{napus} \times \text{California} \times B. \text{rapa ssp. pekinensis} 08\) |
|             |                                              |             | \(007574-1\)                               |
| H8          | \(B. \text{napus} \times \text{Lisek} \times B. \text{tournefortii}\) | H24         | \(B. \text{napus} \times \text{California} \times B. \text{rapa ssp. pekinensis} 08\) |
|             |                                              |             | \(007574-2\)                               |
| H9          | \(B. \text{napus} \times \text{Lisek} \times B. \text{rapa Pak Choi 08, 007574}\) | H25         | \(B. \text{napus} \times \text{California} \times B. \text{rapa ssp. pekinensis} 08\) |
|             |                                              |             | \(007574-3\)                               |
| H10         | \(B. \text{napus} \times \text{Lisek} \times B. \text{rapa Pak Choi 08, 007569}\) | H26         | \(B. \text{napus} \times \text{California} \times B. \text{rapa ssp. pekinensis} 08\) |
|             |                                              |             | \(007574-4\)                               |
| H11         | \(B. \text{napus} \times \text{Górcański} \times B. \text{rapa Pak Choi 08, 007574}\) | H27         | \(B. \text{napus} \times \text{Zhongshuang9} \times B. \text{rapa ssp. pekinensis}\) |
|             |                                              |             | \(08 006169\)                               |
| H12         | \(B. \text{napus} \times \text{Jet Neuf} \times B. \text{oleracea var. alboglabra}\) | H28         | \(B. \text{napus} \times \text{MS8 line} \times B. \text{rapa ssp. pekinensis}\) |
|             |                                              |             | \(08 006169-1\)                            |
| H13         | \(B. \text{napus} \times \text{California} \times B. \text{oleracea var.}\) | H29         | \(B. \text{napus} \times \text{MS8 line} \times B. \text{rapa ssp. pekinensis}\) |
|             | \(\text{oleracea var. alboglabra}\)         |             | \(08 006169-2\)                            |
| H14         | \(B. \text{napus} \times \text{Lisek} \times B. \text{oleracea var. alboglabra}\) | H30         | \(B. \text{napus} \times \text{MS8 line} \times B. \text{rapa ssp. pekinensis}\) |
|             |                                              |             | \(08 006169-3\)                            |
| H15         | \(B. \text{napus} \times \text{California} \times S. \text{alba cv. Bamberka}\) | H31         | \(B. \text{napus} \times \text{Zhongshuang9} \times B. \text{rapa ssp. chinensis}\) |
|             |                                              |             | \(08 007574\)                               |

| No. of line | Cultivar name | No. of line | Cultivar name |
|-------------|---------------|-------------|---------------|
| C1          | Amir          | C28         | PX111CL       |
| C2          | Inspirati     | C29         | Anderson      |
| C3          | Bufalo         | C30         | Andromeda     |
| C4          | Atora          | C31         | Arsenal       |
| C5          | Dolar          | C32         | Hybrirock     |
| C6          | Fair           | C33         | Graf          |
| C7          | Fantastik      | C34         | Hary          |
| C8          | Jet Neuf       | C35         | Mickey        |
| C9          | Jupiter        | C36         | 150/47        |
| C10         | Kana           | C37         | Prince        |
| C11         | Azurio         | C38         | Sofia         |
| C12         | Memoris       | C39         | Santana       |
| C13         | Lindora        | C40         | Rubin         |
| C14         | 150/38         | C41         | Monolit       |
| C15         | 150/46         | C42         | Metys         |
| C16         | Walegro        | C43         | Chrobry       |
| C17         | Marita         | C44         | 150/42        |
| C18         | 150/40         | C45         | Kabriiolet    |
| C19         | 150/44         | C46         | Falcon        |
| C20         | Razmus         | C47         | Diger         |
| C21         | Walery         | C48         | Corina        |
| C22         | Aruze          | C49         | Kontakt       |
| C23         | Bazyl          | C50         | Ceres         |
| C24         | Bellinda       | C51         | Gailleus      |
| C25         | Californium    | C52         | Markolo       |
| C26         | Darmor         | C53         | Hewelius      |
| C27         | PR48W26        |             |               |
pest resistance, paternal forms harbouring high level of resistance to *B. brassicae* and *D. radicum* were selected according to the literature data.

All interspecific cross-derived lines were sister-pollinated (five plants were enclosed in one paper bag during flowering) for four generations in order to stabilize the fertility [25]. Morphotypes of plants of the F₅–F₇ generations were compared with the parental lines, as described by Wojciechowski [26]. Analysis of selected morphological traits was performed in order to determine whether the obtained plants resembled the *B. napus* type or the paternal type. The examination was based on (a) leaf colour, (b) presence of trichomes on the lower side of the leaf blade, (c) position of the buds relative to the open flowers, (d) growth habit, (e) type of inflorescence, and (f) flower characteristics (sterile or fertile).

### 2.3 Assessment of pest resistance

The assessment of pest resistance was carried out for two insects (*Delia radicum* and *Brevicoryne brassicae*) and consisted of plant damage evaluation. General damage by insects was assessed at the end of the season, in late October 2017, 2018, or in early November 2019 in *D. radicum*. Assessment of pest resistance was carried out for two insect pests (*B. brassicae* and *D. radicum*), as described by Wojciechowski [26]. Analysis of selected morphological traits was performed in order to determine whether the obtained plants resembled the *B. napus* type or the paternal type. The examination was based on (a) leaf colour, (b) presence of trichomes on the lower side of the leaf blade, (c) position of the buds relative to the open flowers, (d) growth habit, (e) type of inflorescence, and (f) flower characteristics (sterile or fertile).

(No pesticides were used on the plots. The average values from six plants were calculated for each replication. In this way, we obtained quantitative trait data with normal distributions.)

### 2.4 Statistical analysis

The normality of the distributions of the studied traits (resistance to *B. brassicae* and resistance to *D. radicum*) was tested using the Shapiro–Wilk normality test [29]. Two-way analyses of variance (ANOVA) with blocks were carried out to determine the effects of year, genotype (cultivars and hybrids, independently), and year × genotype interaction on the variability of resistance to *B. brassicae* and resistance to *D. radicum*). The mean values and standard deviations of the observed traits were calculated for each genotype in all years of study. Fisher’s least significant differences (LSDs) were estimated for individual traits, and on this basis, homogeneous groups were determined. Differences between cultivars and hybrids were tested on the basis of a *t*-test, independently for resistance to *B. brassicae* and resistance to *D. radicum*.

We used the critical significance level equal to 0.05, resulting from a Bonferroni correction. All the analyses were conducted using the GenStat v. 18 statistical software package (VSN International, Hemel Hempstead, UK).

### 3 Results

#### 3.1 Morphology of hybrid plants

The individual interspecific and intergeneric hybrid combinations of F₅–F₇ generations had reasonably uniform

| Scale | Visual symptoms | Plant response |
|-------|-----------------|----------------|
| 1     | Lesions profuse on 100% of the roots and leaf surface | Susceptible |
| 2     | Lesions present on up to 90% of the roots and leaf surface | Susceptible to moderately susceptible |
| 3     | Lesions present on up to 70–75% of the roots and leaf surface | Moderately susceptible |
| 4     | Lesions visible on up to 50% of the roots and leaf surface | Moderately susceptible to moderately resistant |
| 5     | Lesions visible on up to 25% of the roots and leaf surface, little damage | Moderately resistant |
| 6     | Lesions visible on less than 15–20% of the roots and leaf surface | Moderately resistant to resistant |
| 7     | Lesions visible on less than 10% of the roots and leaf surface | Resistant |
| 8     | Lesions visible on less than 5% of the roots and leaf surface | Resistant to highly resistant |
| 9     | No insect damage visible on any analysed part of the plant | Highly resistant |
morphological characteristics. Moreover, plants of all tested lines were very consistent in growth habit. Hybrid plants obtained from crosses between *B. napus* × *B. rapa* genotypes were similar to rapeseed. However, in a small number of cases, some morphological features were similar to those of turnip rape, e.g., lighter leaf colour, trichomes on the lower side of the leaf blade, and turnip rape-like inflorescence. No significant new characteristics, absent in either parent, were reported in the hybrids. All other hybrid plants resembled more paternal morphotypes. Consequently, plants obtained from crosses between *B. napus* × *B. carinata*, *B. juncea*, and *S. alba* genotypes had young leaf surfaces with high trichome density.

3.2 Assessment of pest resistance

The results of the ANOVA indicated that the effects of cultivar, hybrid, and year were significant for both tested traits (resistance to *B. brassicae* and *D. radicum*). The *year* × *genotype* interactions were highly significant for both observed traits for cultivars and hybrids (Table 3).

The mean values of resistance to insect pests for the analysed hybrids and cultivars in the years studied successively, i.e., 2017, 2018, and 2019, are presented in Table 4. In general, the resistance to both pests varied among years. The highest mean level of resistance to *B. brassicae* was observed for cultivars in 2017 (8.991), whereas the lowest in 2018 was also for cultivars (5.513). For *D. radicum*, the highest mean resistance was noticed in 2019 for hybrids (7.153). In contrast, the lowest mean resistance was observed for cultivars in 2017 (4.136).

The obtained data showed that the level of pest resistance varied between cultivars and hybrids. Compared to the analysed cultivars, the mean resistance of hybrid plants was higher in all tested years for *D. radicum*. For *B. brassicae*, the mean resistance of hybrids was higher only in 2018. The difference in resistance to *B. brassicae* among cultivars and hybrids in 2019 was not statistically significant (Table 5).

More detailed results are presented in Tables 5 and 6. The conducted analyses showed significant differences between the tested plants. Moreover, the collected data allowed us to distinguish a group of genotypes with the highest resistance to pests (belonging to group α) in tested years for both hybrids and cultivars. Within those plants, we found individuals that belonged to statistically the best group for all three successive years (Table 7). Four hybrids (e.g., *B. napus* cv. Górczański × *B. rapa* Pak Choi 08, 007574) and 27 cultivars (e.g., Inspirati) maintained the high level of resistance to *B. brassicae* during the tested years. However, only five hybrids (e.g., *B. napus* cv. Jet Neuf × *B. carinata* PI 649091) and two rapeseed cultivars (Galileus and Markolo) maintained the high level of resistance to *D. radicum*. Among the tested plant genotypes, only one hybrid and two cultivars remained resistant for both pests in three years, i.e., *B. napus* cv. Jet Neuf × *B. carinata* – PI 649096, Galileus, and Markolo.

### Table 3: Mean squares (m.s.) from two-way analysis of variance for *Brevicoryne brassicae* and *Delia radicum* (hybrid and cultivar resistance) (N = 90)

| Source of variation | *Brevicoryne brassicae* | *Delia radicum* |
|---------------------|-------------------------|-----------------|
|                     | d.f. | m.s. | p-Value | d.f. | m.s. | p-Value |
| **Hybrids**         |      |      |         |      |      |         |
| Block               | 4    | 0.73 | <0.001  | 4    | 1.27 |         |
| Hybrid              | 30   | 2.7592 | <0.001  | 30   | 20.438 | <0.001 |
| Year                | 2    | 241.1076 | <0.001  | 2    | 18.884 | 0.022  |
| Hybrid × year       | 57   | 3.3161 | <0.001  | 57   | 12.488 | <0.001 |
| Residual            | 425  | 0.5328 |         | 427  | 4.875 |         |
| **Cultivars**       |      |      |         |      |      |         |
| Block               | 4    | 0.91 | <0.001  | 4    | 1.32 |         |
| Cultivar            | 52   | 5.9015 | <0.001  | 52   | 30.982 | <0.001 |
| Year                | 2    | 1074.9311 | <0.001  | 2    | 290.038 | <0.001 |
| Cultivar × year     | 104  | 7.7494 | <0.001  | 104  | 23.986 | <0.001 |
| Residual            | 897  | 0.4831 |         | 632  | 4.339 |         |

d.f. – the number of degrees of freedom.

4 Discussion

As stated before, in recent years, the use of insecticides became partly limited – some chemicals have been withdrawn due to their harmful effects on the environment. It causes many problems for farmers, as the range of effective insecticides is getting narrowed. Moreover, the use of chemicals may not always be successful as insects can develop resistance. For both insects, i.e., *D. radicum* and *B. brassicae*, cases of resistance to certain pesticides have been reported [30–32]. Considering this, host plant resistance might be the future of pest management, as it is one of the most economically feasible and ecologically sustainable options [33]. Several strategies to obtain insect-resistant rapeseed have been already presented [34]. This study has successfully followed two of them: finding the source of resistance within *Brassicaceae*.
species and selecting the insect-resistant rapeseed cultivars among cultivars that have been already registered.

Previous studies showed that wild species of *Brassicaceae* can be a useful source of resistance to *B. brassicae* and *D. radicum*. For example, *B. fruticulosa* and *B. spinescens* have a very high level of resistance to both pests and may be used as research material to find respective Quantitative Trait Loci (QTLs) or as part of a breeding programme [35,36]. Moreover, Dosdall et al. [37] screened many genotypes within *Brassicaceae* and successfully produced *S. alba × B. napus* hybrids that inherited resistance to *Delia* spp. from *S. alba*.

However, according to the literature data, much uncertainty still exists about insect feeding preferences and sources of plant resistance to pests [38]. Despite this, there is a considerable amount of literature comparing the life history traits of adults and larvae of pollen beetles among species of *Brassicaceae* [39–41]. For example, *S. alba* may act as a donor of resistance, which can be successfully introgressed into rapeseed. Moreover, *S. alba* genotypes show resistance to a few other pests of rapeseed: root flies *Delia* spp. [37,42], flea beetle *P. cruciferae* [43,44], and bertha armyworm *Mamestra configurata* [45]. However, based on the in-field screening performed in this study, it is not possible to confirm that the obtained *B. napus × S. alba* hybrid combinations were able to maintain higher level of resistance to *D. radicum* or *B. brassicae* during the three consecutive years of study. Furthermore, review of the literature supports resistance to pollen beetles also in *Eruca sativa* [40] and in *C. abyssinica* [46].

Breeding programmes depending on resistant materials are presently also being applied against *Ceutorhynchus obstrictus* (Marsham) (Coleoptera: Curculionidae). Previous experience in other countries has shown that among the tested *Brassicaceae* species, the white mustard *S. alba* was much less susceptible than rapeseed to *C. obstrictus* damage [47].

These studies confirm our assumption that some of the interspecific or intergeneric hybrids can be successfully used as part of future breeding strategies.

Generally, rapeseed cultivars are not considered a very promising source of resistance to pests, as screenings for resistance within existing varieties rarely bring expected results [38,48,49]. Despite this, we managed to find genotypes within *B. napus* (Galileus and Marcolo), which are moderately or highly resistant to both *B. brassicae* and *D. radicum*. Our observations have shown that in the future more assessments should be performed to verify a greater number of cultivars.

Our research has proven the existence of insect-resistant genotypes among rapeseed cultivars and *Brassicaceae* hybrids. A few genotypes were able to maintain the high level of resistance in the three consecutive years of field experiments, which seems to be very useful in future insect resistance breeding. Observed differences in the infestation level allow us to conclude that the plant response might be conditioned by genotype, which may give a chance to identify resistance genes. Future work should focus on laboratory studies, to determine the genetic basis of resistance, as it may depend on three systems: antixenosis, antibiosis, or tolerance [35]. Moreover, research conducted by Hao et al. [50] showed that aphids have preferential behaviour regarding the host plant. Upper epidermis thickness and trichome length had significant impact on aphids’

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**Table 4:** Mean resistance to *Brevicoryne brassicae* and resistance to *Delia radicum* (and standard deviations) of all investigated *Brassica napus* cultivars and hybrid lines over three years

|                      | 2017         | 2018         | 2019         |
|----------------------|--------------|--------------|--------------|
|                      | Hybrids      | Cultivars    | Hybrids      | Cultivars    | Hybrids      | Cultivars    |
| **Resistance to *Brevicoryne brassicae*** |             |              |              |              |              |              |
| Number of observations | 309          | 530          | 93           | 265          | 117          | 265          |
| Mean                 | 8.803        | 8.991        | 6.28         | 5.513        | 7.692        | 7.57         |
| Standard deviation   | 0.5494       | 0.0968       | 1.913        | 2.326        | 0.6881       | 0.6599       |
| t-Statistic          | -5.96        | 3.13         | 1.65         |              |              |              |
| p-Value              | <0.001       | 0.002        | 0.1          |              |              |              |
| **Resistance to *Delia radicum*** |             |              |              |              |              |              |
| Number of observations | 310          | 265          | 93           | 265          | 118          | 265          |
| Mean                 | 6.697        | 4.136        | 6.581        | 5.804        | 7.153        | 5.362        |
| Standard deviation   | 2.617        | 2.568        | 3.076        | 2.326        | 1.556        | 2.537        |
| t-Statistic          | 11.8         | 2.12         | 8.46         |              |              |              |
| p-Value              | <0.001       | 0.035        | <0.001       |              |              |              |
preference on initial probing, which leads to a conclusion that physical properties of rapeseed leaves may be important for B. brassicae host choice.

The level of plant damage varied over the years of observation. Therefore, it can be concluded that the results of the field trials might have been partly dependent on the weather or other abiotic and biotic stresses [34]. Population dynamics of insects may be affected by parameters such as temperature, humidity, and total rainfall [51,52]. Many factors affect the plant response to insects, which makes it harder to find individuals with true genetically induced resistance to insects.

Currently, insect resistance research is focused on quantitative resistance, as it might provide a more durable effect than pyramiding single resistance genes [34]. Variability of insect-derived damage observed in our study proves the complexity of plant response to pests. This might indicate that the resistance of tested genotypes relies on multiple genes located in QTLs. This type of resistance is usually harder to track, because of its complexity and dependence on environmental factors [53]. This makes quantitative traits difficult to include in breeding programmes. However, a study by Ekuere et al. [54] proves that it is possible to track QTLs conferring resistance to Delia spp. by using linkage analysis. Successful introduction of multigenic resistance to insects in Brassica crops would be a great strategy in pest management.

### Table 5: Mean values and standard deviations (s.d.) for hybrid resistance to Brevicoryne brassicae and resistance to Delia radicum (N = 90)

| Hybrid | Resistance to Brevicoryne brassicae (9° scale) | Resistance to Delia radicum (9° scale) |
|--------|-----------------------------------------------|---------------------------------------|
|        | 2017 s.d. | 2018 s.d. | 2019 s.d. | 2017 s.d. | 2018 s.d. | 2019 s.d. |
| H1     | 8.8abc 0.42 | 6.333bcd 0.58 | 6.667c 2.31 | 6.9bcde 2.56 | 8.333ab 0.58 | 3.667i 3.79 |
| H2     | 9a 0.00 | 6.333bcd 1.15 | 7.8ab 0.45 | 6.5bcdef 3.38 | 7.333abc 2.89 | 8abc 1.23 |
| H3     | 8.8abc 0.42 | 7abcd 0.00 | 7.333bc 0.58 | 7.6abc 1.58 | 6abc 4.36 | 7.333abc 1.16 |
| H4     | 8.889ab 0.33 | 3i 1.73 | 6.667c 1.53 | 7.3abc 2.41 | 6abc 3.46 | 7.667abc 0.58 |
| H5     | 9a 0.00 | 5.333cde 1.53 | 7.4abc 1.34 | 7.2abcd 2.78 | 7abc 1.73 | 6cdef 2.07 |
| H6     | 9a 0.00 | 3.333hi 2.31 | 7.8ab 0.45 | 7.4abc 2.80 | 3.667cd 3.79 | 7.4abcd 0.55 |
| H7     | 8.8abc 0.42 | 3.667gh 2.89 | 7.333bc 0.58 | 5.8cdef 3.08 | 4bcd 4.36 | 7bcde 1.00 |
| H8     | 8.5bcd 0.71 | 6cdef 1.73 | 8ab 0.00 | 6.1cdef 2.64 | 6abc 2.65 | 8.4a 0.55 |
| H9     | 8.7abcd 0.48 | 5efghi 1.00 | 7.667ab 0.58 | 6.6cdef 2.27 | 4bcd 1.73 | 6defg 2.00 |
| H10    | 8.5bcd 0.85 | 7.333abc 0.58 | 7.8ab 0.45 | 6.5cdef 2.55 | 9a 0.00 | 6.8cdef 1.64 |
| H11    | 8.7abcd 0.48 | 7abcd 0.00 | 8ab 0.00 | 3.5gh 2.92 | 9a 0.00 | 7.8abc 0.84 |
| H12    | 9a 0.00 | 4.333fghi 2.52 | 8ab 0.00 | 6.6cdef 3.37 | 5.667abc 4.04 | 8.333ab 0.58 |
| H13    | 8.9ab 0.32 | 7.333abc 0.58 | 8ab 0.00 | 4.9efg 1.85 | 5.333abc 3.79 | 8.2ab 0.45 |
| H14    | 8.8abc 0.63 | 7.667ab 0.58 | 7.25bc 0.96 | 6.9cde 2.85 | 6.667abc 3.22 | 7.5abcd 1.00 |
| H15    | 8.6abcd 0.70 | 4.333fghi 3.06 | 7.8ab 0.45 | 7.3abc 1.42 | 1d 0.00 | 7.6ab 0.55 |
| H16    | 9a 0.00 | 6.667bcde 0.58 | 7.333bc 0.58 | 6.8bcde 3.55 | 5.667abc 4.04 | 5ghi 1.73 |
| H17    | 8.9ab 0.32 | 6.333bcd 2.08 | 8.2a 0.45 | 7.1abcd 2.18 | 6.667abc 3.22 | 7.8abc 0.84 |
| H18    | 8.7abcd 0.95 | 7abcd 0.00 | 8ab 0.00 | 6.3cdef 1.57 | 5.667abc 4.04 | 7.4abcd 0.89 |
| H19    | 8.4cd 0.70 | 5.333cdeghi 0.58 | 7.6ab 0.55 | 2.3h 1.57 | 6abc 4.36 | 7.2abcd 1.30 |
| H20    | 8.9ab 0.32 | 5.667bcdeghi 1.5 | 7.6ab 0.55 | 8.4ab 0.84 | 5.667abc 4.04 | 7.8abc 0.84 |
| H21    | 9a 0.00 | 6bcd 1.00 | 8ab 0.00 | 7.4abc 2.17 | 6abc 4.36 | 6.8cdef 1.30 |
| H22    | 9a 0.00 | 7.333abc 1.5 | 8ab 0.00 | 7.8abc 2.04 | 8.333ab 0.58 | 7.8abc 0.45 |
| H23    | 7.9e 1.20 | 4.667efghi 2.08 | 7.5abc 0.58 | 4.6fg 1.71 | 5.667abc 4.04 | 7.5abcd 0.58 |
| H24    | 8.3de 1.16 | 7.667ab 0.58 | 8ab 0.00 | 5.2cdef 1.32 | 8.333ab 0.58 | 5.333fghi 1.53 |
| H25    | 8.8abcd 0.63 | 6.333bcd 1.15 | 8ab 0.00 | 6.3cdef 2.16 | 3.667cd 2.89 | 7.8ab 0.45 |
| H26    | 9a 0.00 | 7.667ab 0.58 | 7.333bc 0.58 | 7.7abc 2.58 | 9a 0.00 | 5.667fg 1.53 |
| H27    | 9a 0.00 | 6.667bcde 0.58 | 7.667ab 0.58 | 7.5abc 2.59 | 8.333ab 1.16 | 4hi 2.65 |
| H28    | 9a 0.00 | 9a 0.00 | – – 9a 0.00 | 0.00 | 9a 0.00 | – – |
| H29    | 9a 0.00 | 9a 0.00 | – – 9a 0.00 | 0.00 | 9a 0.00 | – – |
| H30    | 9a 0.00 | 9a 0.00 | – – 9a 0.00 | 0.00 | 9a 0.00 | – – |
| H31    | 9a 0.00 | 6.333bcd 1.15 | 7.4abc 0.89 | 6.1cdef 1.91 | 9a 0.00 | 7.4abcd 0.89 |
| LSD 0.05 | 0.45 | 2.233 | 0.841 | 2.01 | 4.644 | 1.592 |

Values with different letters in columns are significantly different.
### Table 6: Mean values and standard deviations (s.d.) for cultivar resistance to *Brevicoryne brassicae* and resistance to *Delia radicum* (*N* = 90)

| Cultivar | Resistance to *Brevicoryne brassicae* (9° scale) | Resistance to *Delia radicum* (9° scale) |
|----------|-----------------------------------------------|------------------------------------------|
|          | 2017 Mean s.d. | 2018 Mean s.d. | 2019 Mean s.d. | 2017 Mean s.d. | 2018 Mean s.d. | 2019 Mean s.d. |
| C1       |                |                |                |                |                |                |
| C2       |                |                |                |                |                |                |
| C3       |                |                |                |                |                |                |
| C4       |                |                |                |                |                |                |
| C5       |                |                |                |                |                |                |
| C6       |                |                |                |                |                |                |
| C7       |                |                |                |                |                |                |
| C8       |                |                |                |                |                |                |
| C9       |                |                |                |                |                |                |
| C10      |                |                |                |                |                |                |
| C11      |                |                |                |                |                |                |
| C12      |                |                |                |                |                |                |
| C13      |                |                |                |                |                |                |
| C14      |                |                |                |                |                |                |
| C15      |                |                |                |                |                |                |
| C16      |                |                |                |                |                |                |
| C17      |                |                |                |                |                |                |
| C18      |                |                |                |                |                |                |
| C19      |                |                |                |                |                |                |
| C20      |                |                |                |                |                |                |
| C21      |                |                |                |                |                |                |
| C22      |                |                |                |                |                |                |
| C23      |                |                |                |                |                |                |
| C24      |                |                |                |                |                |                |
| C25      |                |                |                |                |                |                |
| C26      |                |                |                |                |                |                |
| C27      |                |                |                |                |                |                |
| C28      |                |                |                |                |                |                |
| C29      |                |                |                |                |                |                |
| C30      |                |                |                |                |                |                |
| C31      |                |                |                |                |                |                |
| C32      |                |                |                |                |                |                |
| C33      |                |                |                |                |                |                |
| C34      |                |                |                |                |                |                |
| C35      |                |                |                |                |                |                |
| C36      |                |                |                |                |                |                |
| C37      |                |                |                |                |                |                |
| C38      |                |                |                |                |                |                |
| C39      |                |                |                |                |                |                |
| C40      |                |                |                |                |                |                |
| C41      |                |                |                |                |                |                |
| C42      |                |                |                |                |                |                |
| C43      |                |                |                |                |                |                |
| C44      |                |                |                |                |                |                |
| C45      |                |                |                |                |                |                |
| C46      |                |                |                |                |                |                |
| C47      |                |                |                |                |                |                |
| C48      |                |                |                |                |                |                |
| C49      |                |                |                |                |                |                |
| C50      |                |                |                |                |                |                |
| C51      |                |                |                |                |                |                |
| C52      |                |                |                |                |                |                |
| C53      |                |                |                |                |                |                |
| LSD$_{0.05}$ | 0.085 | 1.61 | 0.76 | 1.65 | 3 | 2.4 |

Values with different letters in columns are significantly different.
Table 7: List of genotypes with high resistance to pests in three successive years

| Brevicoryne brassicae       |
|----------------------------|
| Hybrids | H11*, H13, H18, H22 |
| Cultivars | C2, C3, C4, C6, C7, C8, C9, C12, C13, C16, C18, C31, C33, C34, C35, C36, C39, C41, C43, C44, C47, C48, C49, C50, C51, C52, C53 |

| Delia radicum              |
|----------------------------|
| Hybrids | H3, H4, H17, H20, H22 |
| Cultivars | C51, C52 |

Genotypes resistant to both pests are highlighted in bold font. *Numbers according to Table 1.

In conclusion, we found several sources of resistance to *D. radicum* and *B. brassicae* among the rapeseed cultivars, i.e., Galileus and Marcolo, and interspecific *Brassicaceae* hybrids, i.e., *B. napus* cv. Jet Neuf × *B. carinata* – PI 649096. Some of the genotypes showed high level of resistance over the three successive years of field trials. These genotypes are especially valuable and should be diligently analysed.

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