



## Health Status of Spring Barley Grown on Alfisol as Affected by Catch Crop

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### Authors' contributions

*This work was carried out in collaboration between both authors. Author GL wrote the manuscript, performed analysis of plant health, isolation and identification of fungi, performed the statistical analysis. Author EW designed and carried out the field experiment. Both authors have read and approved the final manuscript.*

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### ABSTRACT

**Aims:** The aim of this study was to evaluate the effect of field pea grown as a catch crop and used for green manure on the health of spring barley grown in cereal crop rotation.

**Study Design:** The experiment was carried out in the randomized block design with four replications.

**Place and Duration of Study:** A field experiment was carried out in the years 2008-2011 at Mochelek (17°51'E; 53°13'N), the Experiment Station of the University of Technology and Life Sciences in Bydgoszcz, Poland.

**Methodology:** The experimental factor was made up by the manner and the time of the catch crop biomass incorporation into the soil: the catch crop plowed-in in autumn; catch crop left as mulch for winter and incorporated in spring; control. The evaluation of the health of roots, stem base and spikes was complemented by mycological analysis.

**Results:** Catch crop has an ambiguous effect on the health of barley. Averaged over the three years it significantly decreased the stem infection with *Fusarium* spp. and *Cochliobolus sativus* and leaf infection with *Puccinia hordei*. On the other hand, the use of cover crop increased leaf infection with *Blumeria graminis* and *Rhynchosporium secalis* as well as spike infestation with *Fusarium* spp. and *C. sativus*. There was no significant effect

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of catch crop on the occurrence of eyespot, sharp eyespot, root rot and net blotch. The pathogens which occurred on diseased roots were predominantly *Gaeumannomyces graminis*. There were also many *Fusarium* and *C. sativus* isolates. The infected stems were most often infested by *Fusarium* spp., *C. sativus* and *Glomerella graminicola*. The pathogens on necrotic spots on barley spikelets were mostly represented by *C. sativus* and less frequently - by *Fusarium* spp.

**Conclusion:** Catch crop decreased the stem infection with *Fusarium* spp. and *C. sativus* and leaf infection with *P. hordei*. It also contributed to the increase in leaf infection with *B. graminis* and *R. secalis* and spike infestation with *Fusarium* spp. and *C. sativus*.

*Keywords:* Catch crop; diseases; field pea; fungi; green manure; health; mulch; spring barley.

## 1. INTRODUCTION

Barley (*Hordeum vulgare* L.) is the fourth major crop grown in Poland [1]. The relatively cost-effective cereals production and a large potential of their use has made cereal crops dominate the crop structure in Poland, accounting for 72.6% of the total crop acreage in 2013. Often such situation forces the cultivation of a cereal crop after itself, which leads to a decrease in the grain yield. Growing cereals after cereal crops, including barley, can intensify disease infection of the roots and stem base [2,3,4]. Its effect on leaf and spike infection is less considerable [5,6,7]. With a large share of cereals in the crop structure, there is an accumulation of pathogens and a decrease in the saprotrophic microorganisms in soil [3], which has an unfavourable effect on an increase and competition of crops towards agrophages [4]. It also leads to an accumulation of unfavourable substances secreted by plants during the growing season and released during the decomposition of their residue [3,8,9].

To limit the grain yield losses, due to inadequate crop rotation, in practise, one often intensifies the chemical plant control and mineral fertilisation. Such measures do not always bring expected results and, additionally, excessive amounts of chemicals in production, accompanied by a failure to comply with the principles of good agricultural practise, can pollute soil and ground waters [5,10]. With that in mind, one searches for such agricultural production intensification methods which would be safe for the environment while maintaining sustainable agricultural development [11]. Besides compliant with the European Union regulations (Directive 2009/128/EC) which provide that the EU member states are obliged to implement the principles of integrated plant protection, it cannot be only limited to the use of chemical plant protection agents. One must use such solutions which will lead to maintaining agrophages at the level which would not be harmful for the crop and will facilitate producing a healthy yield with minimum disturbances of the agricultural ecosystem operation.

The negative effects of growing cereal crops after themselves can be ameliorated through the introduction of catch crop as plants performing phytosanitary functions [2,3,12]. Their special role involves a comprehensive effect on biological, physical and chemical soil properties and limiting nutrients leaching from it. An intensive activity of saprotrophic microorganisms which follows after introducing organic matter to the soil [13,14] results in an antagonistic effect towards to the pathogens, leading to a decrease in the diseases severity on the plants grown thereafter [15]. Those features have made the catch crops become an indispensable component of crop rotations in integrated plant protection.

The aim of this study was to evaluate the effect of field pea grown as a catch crop and used for green manure on the health status of spring barley cultivar, 'Tocada', grown in cereal crop rotation.

## 2. MATERIALS AND METHODS

### 2.1 Field Experiment

The field experiment was carried out over 2008-2011, at Mochelek Experiment Station, in the vicinity of Bydgoszcz, mid-west Poland (17°51' E, 53°13' N). The research was performed in a randomized block design with four replications on light loamy-sand soil (on *Alfisol*). The plot was 250 m<sup>2</sup> in size. Field pea (*Pisum sativum* L.), cultivar 'Wiato', grown as a catch crop, was sown 5-9 August in 2008, 2009 and 2010 after the harvest of winter wheat and used as green manure for the following spring barley. The experiment factor was made up by the manner and time of the catch crop biomass incorporation into soil: A - catch crop plowed-in in autumn - using autumn plowing; B - catch crop left as mulch for winter and incorporated in spring - using disking; C - control - without catch crop. The plants in treatment A were harvested using a sickle bar mower from October 15 to November 3. Later soil in treatments A and C was plowed to the depth of 27 cm. In treatment B field pea was left uncut over winter. In spring the pea biomass was cut and mixed with soil using disk cultivator.

Spring barley, cultivar 'Tocada', was sown 2-4 April over 2009-2011, using a row drill in treatments A and C and a drill equipped with coulters in treatment B. Before sowing, the barley grains were treated with Kinto Duo 080 FS (triticonazole 20g·dm<sup>-3</sup> + prochloraz 60g·dm<sup>-3</sup>). At the stem elongation phase (growth stage (GS) 32-33 according to Zadoks et al. [16] scale) the plants were sprayed with Capalo 337.5 SE (fenpropimorph 200 g·dm<sup>-3</sup> + epoxiconazole 62.5g·dm<sup>-3</sup> + metrafenone 75g·dm<sup>-3</sup>) at 1.5dm<sup>3</sup>·ha<sup>-1</sup>. The weather conditions throughout the research period are shown in Table 1.

**Table 1. Weather conditions in the growing season of spring barley at the experiment site**

| Months     | Temperature (°C) |      |      |           | Precipitation (mm) |       |       |           |
|------------|------------------|------|------|-----------|--------------------|-------|-------|-----------|
|            | 2009             | 2010 | 2011 | 1949-2011 | 2009               | 2010  | 2011  | 1949-2011 |
| March      | 2.4              | 2.4  | 2.2  | 1.9       | 43.7               | 28.6  | 11.7  | 24.5      |
| April      | 9.8              | 7.8  | 10.5 | 7.4       | 0.4                | 33.8  | 13.5  | 27.4      |
| May        | 12.3             | 11.5 | 13.5 | 12.7      | 85.3               | 92.6  | 38.4  | 43.2      |
| June       | 14.5             | 16.7 | 17.7 | 16.3      | 57.4               | 18.1  | 100.8 | 53.7      |
| July       | 18.6             | 21.6 | 17.5 | 18.0      | 118.0              | 107.4 | 132.5 | 73.1      |
| Mean/Total | 11.5             | 12.0 | 12.3 | 11.3      | 304.8              | 280.5 | 296.9 | 221.9     |

### 2.2 Samplings and Measurements

The barley health status was estimated based on the root, stem base, leaves and the spike infestation rate. The occurrence of foot and root rot diseases and their intensity were determined at the milk stage (GS 75-77). The evaluation of health status of roots and stem base infection rate caused by *Rhizoctonia* spp., *Oculimacula* spp., *Fusarium* spp. and *Cochliobolus sativus* (anamorph *Bipolaris sorokiniana*) was carried out using the 0-4 degree scale (0° - healthy, no symptoms, 4° - severe infection). The degrees of infection were converted into the DI (disease index) according to the transformation of Townsend and

Heuberger [17]. The severity of leaf diseases was assessed at growth stages (GS 71-73). This was assessed as the percentage of the two-top-leaves surface with disease symptoms. The occurrence of net blotch (*Pyrenophora teres*), powdery mildew (*Blumeria graminis*), leaf scald (*Rhynchosporium secalis*) and leaf rust (*Puccinia hordei*) was assessed. The infection rate on the spike surface caused by *Fusarium* spp. and *C. sativus* was also evaluated. Each time the health status of 25 randomly chosen plants per plot were assessed.

### 2.3 Statistical Analysis

The results were subjected to two-way analysis of variance, and then combined analysis over the years. The significance of differences between the factor levels were determined with the Tukey test, at the significance level of  $P=0.05$ .

### 2.4 Isolation and Identification of Fungi

The material for the analysis was taken from the stem base and roots with disease symptoms, regardless of the experiment factor. A hundred of 5 mm scraps from diseased stem bases and roots and a hundred scraps from diseased roots were sampled. The material was rinsed for 45 minutes in tap water, disinfected for 15 seconds in 1%  $\text{AgNO}_3$  and then rinsed in sterile distilled water and placed onto acidified PDA (Potato Dextrose Agar) with 50 mg of streptomycin per  $1 \text{ dm}^3$ , in Petri dishes.

To confirm the fungi causing disease symptoms on spikes, the blotting paper assay method [18] has been used. Each time a hundred of barley spikelets with disease lesions were placed on wet blotting paper in Petri dishes. Three sheets of sterilized paper were moistened with sterile distilled water and placed in sterilized plastic Petri dishes. The Petri dishes were incubated at the temperature of  $22 \pm 1^\circ\text{C}$  under alternate cycles of 12 hours near-ultraviolet light (NUV) and darkness. After ten days of incubation the spikelets were checked for the associated fungi.

The isolates of fungi were determined according to the species, applying the mycological keys. To confirm the species representation of the *Gaeumannomyces graminis*, *Rhizoctonia*, *Oculimacula* isolates and some *Fusarium* species, an additional polymerase chain reaction (PCR) was performed. For this purpose, we use a species-specific SCAR primers (Sequence Characterized Amplified Region), i.e. NS5/GGT-RP for *G. graminis* [19], Rc2F/R for *R. cerealis* [20], ITS1/GMRS-3 for *R. solani* [21], Ta05F/R for *O. acufiformis* and TyV5F/R for *O. yallundae* [22], JIAF/R for *Gibberella avenacea* (anamorph *F. avenaceum*) [23], Fp82F/R for *Fusarium poae* [24], and Fc01F/R for *Fusarium culmorum* [25]. Total DNA was isolated using the adapted method described by Doyle and Doyle [26]. The amplification reaction was performed with the *Taq* PCR Core Kit (QIAGEN Inc., USA) in a final volume of  $12.5 \mu\text{L}$ , consisting of  $2.5 \mu\text{L}$  of the solution containing the fungal DNA,  $0.25 \mu\text{L}$  dNTP mix,  $2.5 \mu\text{L}$   $5 \times$  buffer Q,  $1.25 \mu\text{L}$   $10 \times$  PCR buffer,  $0.5 \mu\text{L}$  magnesium chloride,  $0.1 \mu\text{L}$  of *Taq* DNA polymerase and  $0.75$  each of the primers. Amplification was carried out in Mastercycler ep gradient thermocycler (Eppendorf, Germany). The PCR products were separated by electrophoresis ( $4 \text{ V/cm}$ ) in 1.4% agarose gels with  $1 \times$  TBE buffer (89mM Tris-borate and 2mM EDTA) and visualized under ultraviolet light following ethidium bromide staining.

### 3. RESULTS

In years 2009-2011 the severity of diseases in spring barley was relatively low. The highest disease index value for all the foot and root rot diseases was observed on the roots of spring barley (Table 2). Less intense fungal infection was noted on stem base. The symptoms were mainly caused by *Fusarium* spp. and *C. sativus*. The eyespot and sharp eyespot symptoms were less frequent.

Catch crop plowing in autumn significantly decreased the stem base infection with *Fusarium* spp. and *C. sativus*. A significant differentiation in the root rot occurrence was observed only in 2011. The lowest disease symptoms were recorded on the mulched plots and the highest for the control. There was no significant effect of the test agent on the occurrence of eyespot and sharp eyespot.

The symptoms which were most frequently observed on barley leaves were net blotch (Fig. 1). The disease severity depended on the catch crop applied only in 2010. The highest net blotch symptoms were noted on the mulched plots and the lowest on the control. Significantly fewer symptoms of powdery mildew, leaf rust and then leaf scald were observed during this research. For the average of the three years, powdery mildew and leaf scald were least intensive in the control, and the catch crop increased the infection rate. The fewest symptoms of leaf rust were recorded on the barley grown on the mulched plots and most in the control, without a catch crop. A significant effect of the catch crop on the occurrence of powdery mildew, leaf scald and leaf rust was observed only for the years average. Variation was not significant, however, in each research year, although the trend was the same as for the years average.

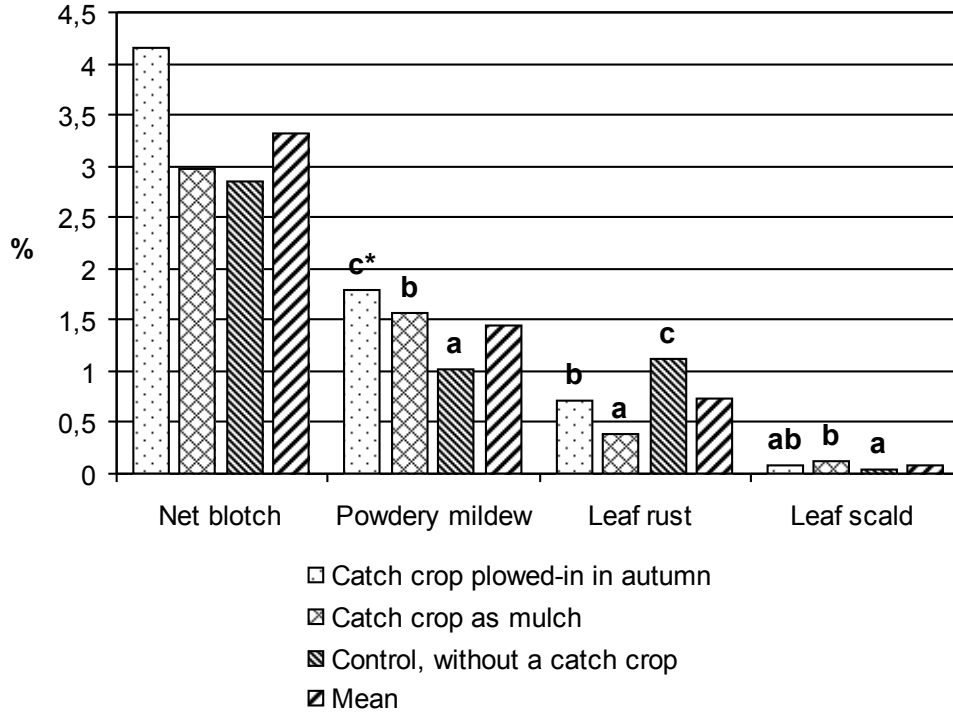
**Table 2. Health status of roots and stem base of spring barley**

| Disease  | Catch crop | 2009   |      | 2010 |      | 2011   |      | Mean   |        |
|--|------------|--------|------|------|------|--------|------|--------|--------|
|  |            | DI     | %    | DI   | %    | DI     | %    | DI     | %      |
| Root rot<br>(complex of fungi)   | A          | 21.0   | 40   | 32.5 | 88   | 25.5ab | 58ab | 26.3   | 62.0   |
|  | B          | 30.5   | 54   | 25.3 | 73   | 19.3 a | 54 a | 25.0   | 60.3   |
|  | C          | 33.3   | 50   | 24.5 | 77   | 33.3 b | 64 b | 30.4   | 63.7   |
|  | Mean       | 28.3   | 48.0 | 27.4 | 79.3 | 26.0   | 58.7 | 27.2   | 62.0   |
| Stem base infected<br>with <i>Fusarium</i> spp.<br>and <i>Cochliobolus sativus</i> | A          | 6.25 a | 24 a | 2.25 | 9    | 18.75  | 53   | 9.08 a | 28.7a  |
|  | B          | 6.00 a | 20 a | 3.50 | 14   | 19.75  | 61   | 9.75ab | 31.7ab |
|  | C          | 9.50 b | 32 b | 3.00 | 12   | 22.00  | 66   | 11.50b | 36.7b  |
|  | Mean       | 7.25   | 25.3 | 2.92 | 11.7 | 20.17  | 60.0 | 10.11  | 32.4   |
| Eyespot<br>( <i>Oculimacula yallundae</i> )  | A          | 3.00   | 8    | 5.50 | 18   | 0.25   | 1    | 2.92   | 9.0    |
|  | B          | 3.50   | 11   | 1.75 | 6    | 0.00   | 0    | 1.75   | 5.7    |
|  | C          | 4.25   | 11   | 2.00 | 7    | 0.50   | 2    | 2.25   | 6.7    |
|  | Mean       | 3.58   | 10.0 | 3.08 | 10.3 | 0.25   | 1.0  | 2.31   | 7.1    |
| Sharp eyespot<br>( <i>Rhizoctonia cerealis</i> )                                   | A          | 3.00   | 11   | 0.75 | 3    | 0.25   | 1    | 1.33   | 5.0    |
|  | B          | 1.75   | 5    | 0.25 | 1    | 1.50   | 3    | 1.17   | 3.0    |
|  | C          | 2.25   | 8    | 0.25 | 1    | 0.00   | 0    | 0.83   | 3.0    |
|  | Mean       | 2.33   | 8.0  | 0.42 | 1.7  | 0.58   | 1.3  | 1.11   | 3.7    |

A - catch crop plowed-in in autumn; B - catch crop as mulch; C - control, without a catch crop; DI - disease index in %; % - per cent of plants/stems with disease symptoms; Means marked with different letters within particular columns are significantly different at P=0.05

On the spring barley spikes, there were observed the symptoms of infestation with genus *Fusarium* and *C. sativus* fungi. For the average of three years the least severe disease symptoms were reported for the control, and the plowing catch crop in autumn increased the

severity of symptoms (Fig. 2). In individual research years the variation was not significant, however, there were similar trends to those for the years average.



**Fig. 1. Percentage of leaf surface with disease symptoms (2008-2011)**

\* Means marked with different letters within particular columns (for particular diseases) are significantly different at  $P=0.05$

The pathogens occurring on necrotic spots on spring barley spikelets were mostly represented by *C. sativus* (72.7%) (Fig. 3). *Fusarium* genus fungi were less frequently isolated (43.3%).

*Gaeumannomyces graminis* was the most common pathogen isolated from infected barley roots, whose share in all the isolates accounted for approximately 39.1% (Table 3). Also a significant number of fungi at the anamorph stage were identified to represent *Fusarium* genus, where *Fusarium culmorum*, *Haematonectria haematococca* (anamorph *F. solani*) and *Gibberella intricans* (anamorph *F. equiseti*) dominated. There were also many *C. sativus* isolates, and much fewer *Rhizoctonia solani*. The fungi isolated from the stem base represented mainly the *Fusarium* genus (56.4%). The isolates included mostly *F. culmorum*, followed by *G. intricans*, *G. avenacea* and *H. haematococca*. Many of the isolates of *C. sativus* and *Glomerella graminicola* were also reported. Clearly less frequently isolated fungi included: *O. yallundae*, *R. cerealis* and *R. solani*. The taxonomy of *O. yallundae*, *R. cerealis*, *R. solani*, *G. avenacea*, *F. culmorum* and *F. poae* was confirmed additionally by applying the PCR method using species-specific SCAR primers.

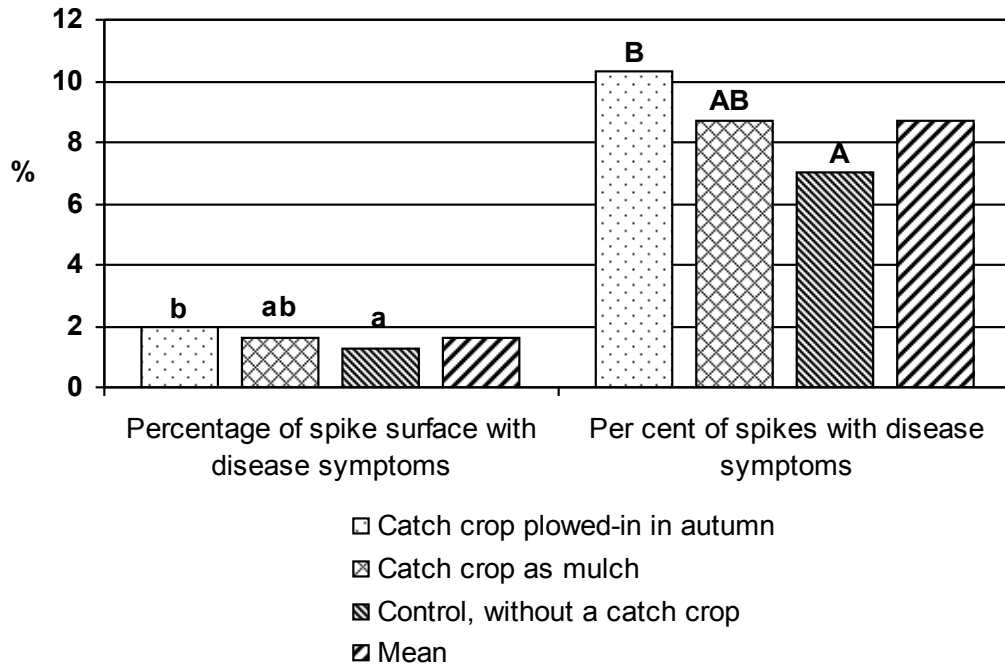


Fig. 2. Spike infection with *Fusarium* spp. and *Cochliobolus sativus* (2008-2011)

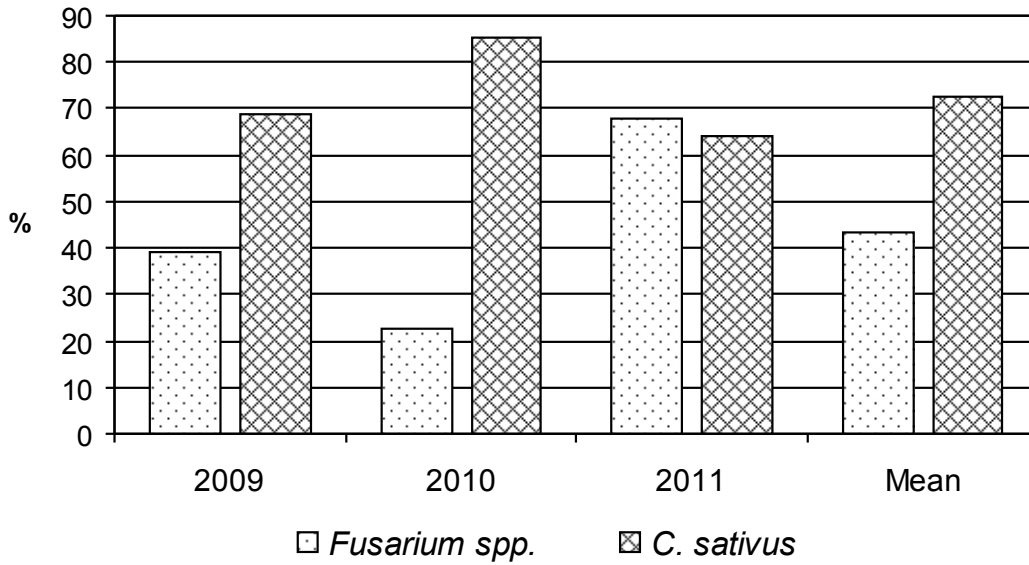


Fig. 3. Per cent of spikelets with disease symptoms colonized by *Fusarium* spp. and *Cochliobolus sativus* - blotting paper assay

**Table 3. Fungi occurring on spring barley roots and stem bases with disease symptoms**

| Taxon   | Roots     |           |           |              | Stem base |           |           |              |
|---|-----------|-----------|-----------|--------------|-----------|-----------|-----------|--------------|
|   | 2009      | 2010      | 2011      | Mean (Total) | 2009      | 2010      | 2011      | Mean (Total) |
| <i>Alternaria alternata</i> (Fries.) Keiss.   | 6.2 (5)   | 4.5 (3)   | 5.3 (4)   | 5.3 (12)     | –         | –         | –         | –            |
| <i>Cochliobolus sativus</i> (S. Ito & Kurib.) Drechsler ex Dastur                   | 2.5 (2)   | 10.3 (7)  | 10.6 (8)  | 7.8 (17)     | 9.7 (9)   | 11.2 (11) | 22.2 (21) | 14.4 (41)    |
| <i>Cladosporium herbarum</i> (Pers.) Link. ex Fr.                                   | –         | 1.5 (1)   | 1.3 (1)   | 0.9 (2)      | –         | 1.0 (1)   | 1.1 (1)   | 0.7 (2)      |
| <i>Clonostachys rosea</i> f. <i>catenulata</i> (J.C. Gilman & E.V. Abbott) Schroers | –         | 3.0 (2)   | 1.3 (1)   | 1.4 (3)      | –         | 1.0 (1)   | –         | 0.3 (1)      |
| <i>Clonostachys rosea</i> (Link) Schroers, Samuels, Seifert & W. Gams               | –         | –         | –         | –            | 1.1 (1)   | –         | 1.1 (1)   | 0.7 (2)      |
| <i>Epicoccum nigrum</i> Link  | 1.2 (1)   | –         | 1.3 (1)   | 0.8 (2)      | –         | –         | –         | –            |
| <i>Fusarium culmorum</i> (W.G. Smith) Sacc*   | 3.7 (3)   | 7.5 (5)   | 10.6 (8)  | 7.1 (16)     | 11.8 (11) | 23.6 (23) | 26.5 (25) | 20.6 (59)    |
| <i>Fusarium oxysporum</i> Schlecht.   | 1.2 (1)   | 6.0 (4)   | 3.9 (3)   | 3.7 (8)      | –         | 1.0 (1)   | –         | 0.3 (1)      |
| <i>Fusarium poae</i> (Peck.) Wollenw.*  | –         | 4.5 (3)   | 2.6 (2)   | 2.4 (5)      | –         | 1.0 (1)   | 1.1 (1)   | 0.7 (2)      |
| <i>Gaeumannomyces graminis</i> (Sacc.) Arx et Olivier*                              | 53.1 (43) | 31.2 (21) | 33.0 (25) | 39.1 (89)    | –         | –         | –         | –            |
| <i>Gibberella avenacea</i> R.J. Cook *  | 2.5 (2)   | 3.0 (2)   | 2.6 (2)   | 2.7 (6)      | 16.1 (15) | 6.1 (6)   | 10.6 (10) | 10.9 (31)    |
| <i>Gibberella intricans</i> Wollenw.  | 2.5 (2)   | 6.0 (4)   | 6.6 (5)   | 5.0 (11)     | 9.7 (9)   | 17.4 (17) | 16.0 (15) | 14.4 (41)    |
| <i>Gibberella tricineta</i> El-Gholl, McRitchie, Schoult. & Ridings                 | –         | –         | –         | –            | 1.1 (1)   | –         | –         | 0.4 (1)      |
| <i>Glomerella graminicola</i> D.J. Politis  | –         | –         | –         | –            | 26.8 (25) | 7.1 (7)   | 5.3 (5)   | 13.1 (37)    |
| <i>Haematonectria haematococca</i> (Berk. & Broome) Samuels & Rossman               | 2.5 (2)   | 9.0 (6)   | 5.3 (4)   | 5.6 (12)     | 7.5 (7)   | 13.4 (13) | 6.4 (6)   | 9.1 (26)     |
| <i>Microdochium bolleyi</i> (R. Sprague) de Hoog & Herm.-Nijh.                      | –         | –         | –         | –            | 1.1 (1)   | 1.0 (1)   | –         | 0.7 (2)      |
| <i>Mucor</i> spp.   | –         | 1.5 (1)   | –         | 0.5 (1)      | 1.1 (1)   | 3.1 (3)   | 1.1 (1)   | 1.8 (5)      |
| <i>Oculimacula yallundae</i> (Wallwork & Spooner) Crous & W. Gams*                  | –         | –         | –         | –            | 1.1 (1)   | 2.0 (2)   | –         | 1.0 (3)      |
| <i>Penicillium</i> spp.   | 6.2 (5)   | 1.5 (1)   | 2.6 (2)   | 3.4 (8)      | –         | 2.0 (2)   | 1.1 (1)   | 1.0 (3)      |
| <i>Periconia macrospinoso</i> Lefebvre et Johnson Lefebvre et Johnson               | 4.9 (4)   | 1.5 (1)   | 2.6 (2)   | 3.0 (7)      | –         | –         | –         | –            |
| <i>Rhizoctonia cerealis</i> van der Hoeven*   | –         | –         | –         | –            | 3.2 (3)   | 1.0 (1)   | 1.1 (1)   | 1.8 (5)      |
| <i>Rhizoctonia solani</i> Kühn*   | –         | 1.5 (1)   | 2.6 (2)   | 1.4 (3)      | 4.3 (4)   | 1.0 (1)   | 2.1 (2)   | 2.5 (7)      |
| <i>Sarocladium strictum</i> (W. Gams) Summerb.                                      | –         | 1.5 (1)   | –         | 0.5 (1)      | –         | –         | –         | –            |
| <i>Trichoderma harzianum</i> Rifai  | –         | 1.5 (1)   | 1.3 (1)   | 0.9 (2)      | –         | –         | –         | –            |
| <i>Trichoderma koningii</i> Oudem.  | 6.2 (5)   | 3.0 (2)   | 2.6 (2)   | 3.9 (9)      | 1.1 (1)   | 2.0 (2)   | –         | 1.0 (3)      |
| <i>Trichoderma polysporum</i> (Link ex Pers.) Rifai                                 | 1.2 (1)   | 1.5 (1)   | 1.3 (1)   | 1.3 (3)      | –         | 1.0 (1)   | –         | 0.3 (1)      |
| <i>Trichoderma viride</i> Pers. ex Gray   | 1.2 (1)   | –         | 1.3 (1)   | 0.8 (2)      | 3.2 (3)   | 4.1 (4)   | 3.2 (3)   | 3.5 (10)     |
| Non-sporulating mycelia   | 4.9 (4)   | –         | 1.3 (1)   | 2.1 (5)      | 1.1 (1)   | –         | 1.1 (1)   | 0.7 (2)      |
| Total number of isolates  | 81        | 67        | 76        | 224          | 93        | 98        | 94        | 285          |

\* - The taxonomy of species was confirmed by applying the PCR method using species-specific SCAR primers; In bracket number of isolates



#### 4. DISCUSSION

The relatively great intensity of root rot symptoms could have been due to winter wheat acting as a preceding crop for barley. Barley cultivation after wheat increases the probability of fungal root infections, as indicated by previous reports [3]. *Gaeumannomyces graminis* was isolated from infected barley roots, which is considered to be the main cause of wheat root rot. A short time of cultivation of non-cereal crop in the catch crop is not sufficient to reduce the population of this pathogen in soil significantly, which was confirmed in our study. Other pathogens isolated from barley, including polyphagous fungi capable of attacking not only cereals, considerably reduce the importance of the sequence of crops.

In our study, *Oculimacula* spp. and *Rhizoctonia* spp. were not the predominant pathogens isolated from the stem base in barley. It is stated that the DNA content of these pathogens in cereal tissues increases with the development of plants [27,28]. Perhaps that is the reason why in barley, with a much shorter growing season than in winter cereals, there were few symptoms, as evidenced by the low intensity of eyespot and sharp eyespot. *Oculimacula yallundae* and *R. cerealis* are characterized by slow mycelium growth and it probably did not manage to strike a stronger degree of plant tissues [28,29]. A poor severity of the disease and the fact that these pathogens are capable of surviving for some time in soil meant that there was no significant effect of the catch crop on the fungi occurrence. The length of the growing season plays a lesser role in the rate of infection of cereals caused by *Fusarium* spp., which are characterized by a faster linear growth of mycelium, which must have been why there were observed more symptoms of infection by *Fusarium* spp. and *C. sativus*. Although the latter species is characterized by a slow growth, nevertheless, in Poland it is considered one of the main pathogens of spring barley [30]. The stem base infection with *Fusarium* spp. and *C. sativus* was significantly reduced by the cultivation of pea as the catch crop. Similarly previous studies have reported that the cultivation of catch crop can reduce the intensification of symptoms caused by these pathogens [2]. Different results, regarding the importance of catch crop, were reported by Gleń et al. [31], investigating spring wheat with bean as a catch crop. They claim that non-tillage cultivation with mulch significantly increases root rot caused by *Fusarium* spp.; in general, they report that the phytosanitary condition of cultivation with mulch was slightly better, as compared with traditional cultivation with autumn plowing.

The increase in the occurrence of powdery mildew, leaf scald and spike infection with *Fusarium* spp. and *C. sativus* in barley grown after the catch crop, as compared with control, could have been a result of the increased density of sowing. Barley cultivated in soil after pea made the stems and spikes closer to each other, which greatly facilitates the spread of pathogens that cause those diseases, as reported by Walters et al. [7]. According to Jensen and Munk [32], the use of catch crop may increase the infection due to the accumulation of nitrogen in soil. Plowed biomass of field pea contains a lot of nitrogen, which can later be used by barley as indicated by Porta-Puglia and Aragona [33]. According to Newton et al. [34] and Walters et al. [7], barley heavily fertilized with nitrogen is more susceptible to infections caused by *B. graminis* and *R. secalis*. A higher plant density may also indirectly contribute to the severity of disease, including changes in the sowing microclimate. Usually, the temperature in the dense sowing is more uniform, there is a relatively higher humidity, and water remains on the leaves for a longer time, which greatly favours the fungal infection of plants by fungal pathogens [7,35].

The opposite results were obtained for leaf rust, for which most of the symptoms were noted in barley grown without catch crop. However, the first symptoms of the disease appeared

late, when a large surface of the leaf was struck by other pathogens. *Puccinia hordei*, a biotrophic fungal pathogen, mainly affects the places where tissues are healthy, allowing a rapid development of the pathogen [7,36]. Due to the lower surface of the leaves, which could be infected, the development of the pathogen was significantly reduced. A similar trend is also observed for infection caused by *B. graminis* [36], however, in our experiment the pathogen occurred much earlier, before the development of other diseases symptoms. According to Walters et al. [7] and Bingham et al. [35], a high density may also hinder the spread of spores of *P. hordei* since barley grown after pea observed a decrease in the severity of rust.

## 5. CONCLUSION

Catch crop has an ambiguous effect on the health of barley. Although it significantly decreased the stem base infection with *Fusarium* spp. and *Cochliobolus sativus* and leaf infection with *P. hordei*, it also increased the leaf infection with *B. graminis* and *R. secalis* and spike infestation with *Fusarium* spp. and *C. sativus*.

The pathogenic fungi isolated from spring barley roots were mostly represented by *Gaeumannomyces graminis* and from stem bases - with *Fusarium* spp., especially *F. culmorum*. There were also found many *C. sativus* and *Glomerella graminicola* isolates.

The pathogens occurring on necrotic spots on spikelets in spring barley were mostly represented by *C. sativus* and less frequently by *Fusarium* spp.

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## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

1. Anonymous. The use of land and sown area in; 2013. GUS Warszawa. Accessed 25 March 2014.  
Available: [http://www.stat.gov.pl/gus/5840\\_1535\\_PLK\\_HTML.htm](http://www.stat.gov.pl/gus/5840_1535_PLK_HTML.htm); Polish.
2. Lemańczyk G, Skinder Z, Sadowski C. Impact of stubble intercrop and organic fertilisation on the health status of spring barley culm base. EJPau, Ser. Agronomy. 2001;4:2. Available Online: <http://www.ejpau.media.pl/volume4/issue2/agronomy/art-07.html>
3. Lemańczyk G, Wilczewski E. Effects of intercrop plants in stubble and organic fertilization on the health of roots of spring barley in cereal crop rotations. Phytopathol Pol. 2006;40:7-19.
4. Kurowski TP, Adamiak E. Occurrence of stem base diseases of four cereal species grown in long-term monocultures. Pol J Nat Sci. 2007;22(4):574-583.
5. Cameron KC, Di HJ, McLaren RG. Is soil an appropriate dumping ground for our wastes? Aust J Soil Res. 1997;35:995-1035.

6. Mathre DE. Compendium of Barley Diseases. 2nd ed. St. Paul, MN: APS Press; 1997.
7. Walters D, Avrova A, Bingham IJ, Burnett FJ, Fountaine J, Havis ND, Hoad SP, Hughes G, Looseley M, Oxley SJP, Renwick A, Topp CFE, Newton AC. Control of foliar diseases in barley: Towards an integrated approach. *Eur J Plant Pathol.* 2012;133(1):33-73.
8. Shipton PJ. Monoculture and soilborne plant pathogens. *Annu Rev Phytopathol.* 1977;15:387-407.
9. Bais HP, Weir TL, Perry LG, Gilroy S, Vivanco JM. The role of root exudates in rhizosphere interactions with plants and other organisms. *Annu Rev Plant Biol.* 2006;57:233-266.
10. Spalding RF, Exner ME. Occurrence of nitrate in groundwater - A review. *J Environ Qual.* 1993;22:392-402.
11. Doltra J, Olesen JE. The role of catch crops in the ecological intensification of spring cereals in organic farming under Nordic climate. *Eur J Agron.* 2013;44:98-108.
12. Małecka I, Blecharczyk A. Effect of tillage systems, mulches and nitrogen fertilization on spring barley (*Hordeum vulgare*). *Agron Res.* 2008;6(2):517-529.
13. Perucci P, Scarponi L. Effect of different treatments with crop residues on soil phosphatase activity. *Biol Fertil Soils.* 1985;1(2):111-115.
14. Piotrowska A, Wilczewski E. Effects of catch crops cultivated for green manure and mineral nitrogen fertilization on soil enzyme activities and chemical properties. *Geoderma.* 2012;189-190:72-80.
15. Bailey KL, Lazarovits G. Suppressing soil-borne diseases with residue management and organic amendments. *Soil Till Res.* 2003;72(2):169-180.
16. Zadoks JC, Chang TT, Konzak CF. A decimal code for the growth stages of cereals. *Weed Res.* 1974;14(6):415-421.
17. Townsend GR, Heuberger JW. Methods for estimating losses caused by diseases in fungicide experiments. *Plant Dis Rep.* 1943;27(17):340-343.
18. Limonard T. Ecological aspects of seed health testing. Wageningen: International Seed Testing Association; 1968.
19. Fouly HM, Wilkinson HT. Detection of *Gaeumannomyces graminis* varieties using polymerase chain reaction with variety-specific primers. *Plant Dis.* 2000;84(9):947-951.
20. Nicholson P, Parry DW. Development and use of a PCR assay to detect *Rhizoctonia cerealis*, the cause of sharp eyespot in wheat. *Plant Pathol.* 1996;45(5):872-883.
21. Johanson A, Turner HC, McKay GJ, Brown AE. A PCR-based method to distinguish fungi of the rice sheath-blight complex, *Rhizoctonia solani*, *R. oryzae* and *R. oryzae-sativae*. *FEMS Microbiol Lett.* 1998;162(2):289-294.
22. Nicholson P, Rezanoor HN, Simpson DR, Joyce D. Differentiation and quantification of the cereal eyespot fungi *Tapesia yallundae* and *Tapesia acuformis* using a PCR assay. *Plant Pathol.* 1997;46(6):842-856.
23. Turner AS, Lees AK, Rezanoor HN, Nicholson P. Refinement of PCR-detection of *Fusarium avenaceum* and evidence from DNA marker studies for phylogenetic relatedness to *Fusarium tricinctum*. *Plant Pathol.* 1998;47(3):278-288.
24. Parry DW, Nicholson P. Development of a PCR assay to detect *Fusarium poae* in wheat. *Plant Pathol.* 1996;45(2):383-391.
25. Nicholson P, Simpson DR, Weston G, Rezanoor HN, Lees AK, Parry D, Joyce D. Detection and quantification of *Fusarium culmorum* and *Fusarium graminearum* in cereals using PCR assays. *Physiol Mol Plant Pathol.* 1998;53(1):17-37.
26. Doyle JJ, Doyle JL. Isolation of plant DNA from fresh tissue. *Focus.* 1990;12(1):13-15.

27. Nicholson P, Turner AS, Edwards SG, Bateman GL, Morgan LW, Parry DW, Marshall J, Nuttall M. Development of stem-base pathogens on different cultivars of winter wheat determined by quantitative PCR. *Eur J Plant Pathol.* 2002;108(2):163-177.
28. Ray RV, Jenkinson P, Edwards SG. Effects of fungicides on eyespot, caused predominantly by *Oculimacula acuformis* and yield of early-drilled winter wheat. *Crop Prot.* 2004;23(12):1199-1207.
29. Lemańczyk G, Kwaśna H. Effects of sharp eyespot (*Rhizoctonia cerealis*) on yield and grain quality of winter wheat. *Eur J Plant Pathol.* 2013;135(1):187-200.
30. Baturó-Ciesniewska A. Genetic variability and pathogenicity among Polish isolates of *Bipolaris sorokiniana* (*Cochliobolus sativus*) derived from spring barley (*Hordeum vulgare* L.). *J Plant Pathol.* 2011;93(2):291-302.
31. Gleń K, Gorczyca A, Kulig B. Healthiness of spring wheat Zebra and Bryza c.v. depending on applied agrotechnical measures. *Ecological Chemistry and Engineering.* 2007;14(11):1175-1180.
32. Jensen B, Munk L. Nitrogen induced changes in colony density and spore production of *Erysiphe graminis* f. sp. *hordei* on seedlings of six spring barley cultivars. *Plant Pathol.* 1997;46(2):191-202.
33. Porta-Puglia A, Aragona M. Improvement of grain legumes general part: diseases. *Field Crops Res.* 1997;53:17-30.
34. Newton AC, Thomas WTB, Guy DC, Gaunt RE. The interaction of fertiliser treatment with tolerance to powdery mildew in spring barley. *Field Crops Res.* 1998;55:45-56.
35. Bingham IJ, Walters DR, Foulkes MJ, Paveley ND. Crop traits and the tolerance of wheat and barley to foliar disease. *Ann Appl Biol.* 2009;154:159-173.
36. Bingham IJ, Hoad SP, Thomas WTB, Newton AC. Yield response to fungicide of spring barley genotypes differing in disease susceptibility and canopy structure. *Field Crops Res.* 2012;139:9-19.

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