

MYCOLOGICAL PURITY OF BROAD BEAN (*Vicia faba* L.) SEEDS IN THE CONDITIONS OF COMPANION PLANTING AND DIFFERENTIATED PROTECTION

Summary

The subject of the study concerns the seeds of broad bean of White Hangdown cultivar derived from the strict field experiment (2010–2012). The experiment consisted of 10 combinations including two alternating rows sowings of broad bean with fennel and coriander, four variants of biological protection using Polyversum WP, Bioczos BR, Biosept 33 SL preparations, and three variants of chemical protection using Vitavax 200 FS, Decis 2.5 EC, Fastac 100 EC, Penncozeb 80 WP. Applied protection variants improved mycological purity of broad bean seeds. The best and comparable results were noted in the combination with the use of seed dressing Vitavax 200 SL, and subsequent triple foliar application of insecticides and fungicides as well as biological dressing Polyversum WP, and four-time spraying with biotechnical preparations. Companion planting of broad beans with fennel and coriander favored seeds colonization by fungi. Especially the neighborhood with fennel enriched the community of fungi present in the seeds of broad beans in *Penicillium* spp. (34.6%). Irrespective of the protection, *Alternaria alternata* (28.6%), *Botrytis cinerea* (12.3%), *Epicoccum purpurascens* (5.1%) and saprobionts of *Penicillium* genus (22.3%), were isolated the most frequently.

Key words: pathogenic fungi, broad bean seeds, biological and chemical protection

CZYSTOŚĆ MIKOLOGICZNA NASION W UPRAWIE WSPÓLRZĘDNEJ BOBU (*Vicia faba* L.) ORAZ W WARUNKACH ZRÓŻNICOWANEJ OCHRONY

Streszczenie

Przedmiotem badań były nasiona bobu odmiany Hangdown Biały pochodzące ze ścisłego doświadczenia polowego (2010–2012 r.). Eksperyment obejmował 10 kombinacji w tym dwa siewy na przemian rzędowe bobu z koprem włoskim oraz kolen-drą siewną, 4 warianty ochrony biologicznej z wykorzystaniem preparatów: Polyversum WP, Bioczos BR, Biosept 33 SL oraz 3 warianty ochrony chemicznej: Vitavax 200 FS, Decis 2,5 EC, Fastac 100 EC, Penncozeb 80 WP. Zastosowane warianty ochrony poprawiały czystość mikologiczną nasion bobu. Najlepsze i porównywalne efekty notowano w kombinacjach z wykorzystaniem zaprawy nasiennej Vitavax 200 SL i późniejszą trzykrotną nalistną aplikacją insektycydów i fungicydu oraz zaprawy biologicznej Polyversum WP i czterokrotnym opryskiwaniem preparatami biotechnicznymi. Uprawy współ-rzędne bobu z koprem włoskim i kolendrą siewną sprzyjały kolonizacji nasion przez grzyby. Szczególnie sąsiedztwo z koprem włoskim wzbogacało zbiorowisko grzybów zasiedlających nasiona bobu w *Penicillium* spp. (34,6%). Niezależnie od ochrony z największą częstotliwością izolowano *Alternaria alternata* (28,6%), *Botrytis cinerea* (12,3%), *Epicoccum purpurascens* (5,1%) oraz saprobionty z rodzaju *Penicillium* (22,3%).

Słowa kluczowe: grzyby patogeniczne, nasiona bobu, biologiczna i chemiczna ochrona

1. Introduction

Microbiological purity is a very important quality criterion in the evaluation of the use of seeds as a foodstuff and fodder, as well as seed material [12, 14]. Numerous species of pathogenic and saprotrophic fungi colonizing the surface and inside the seeds are a potential source of endothermic mycotoxins extremely dangerous to the health of human and animal organisms [25, 34, 36, 37]. Toxins such as deoxynivalenol (DON), nivalenol (NIV) and zearalenone (ZEA) produced by certain species of fungi belonging to *Fusarium* genus are reported with the greatest frequency and the largest amounts. These mycotoxins impair the operation of digestive tract, liver, cardiovascular and endocrine system, and cause many acute and chronic diseases in human and animals [33]. Seedborne pathogens impair an ability of germination, can cause rot or decay of seeds, seedlings infection, and the development of infectious diseases in later stages of plant growth [30, 31, 37]. Microbiological purity of seeds is to a high degree affected by plant protection treatments carried out during the growing season. Particularly high efficacy against phytopathogens

transferred with the seeds was demonstrated for synthetic fungicides used for dressing and then foliar application. Unfortunately, they cannot be used in organic crop production systems. An alternative may constitute non-chemical preparations, based on natural substances and microorganisms, however the opinions about their effectiveness are divided and controversial [24]. Some believe that natural plant substances have protective properties comparable to the activity of chemical fungicides [2, 35]. Others in turn, suggest that only their frequent application guarantees the effectiveness [12]. Due to effectiveness and relatively wide range of antifungal activity, Polyversum preparation based on *Pythium oligandrum* fungus has won the recognition in the last decade [3]. Companion planting of different species of plants which may limit the occurrence of pests or weeds are practiced in organic systems of cultivation. There is no reliable information in the available literature on the effect of plants neighborhood (alternating rows crops) on health status. It can be assumed that plant species accurately chosen for companion planting minimize the amount of damage caused by insects, so they can indirectly affect the development of microbial populations colonizing the plants.

It should be remembered that total elimination of protective measures or their low effectiveness deteriorates the quality of seed material and exposes the consumers to mycotoxins presence in basic food products.

These arguments constitute the purposefulness of the study concerning determination of the quantity and quality of fungi communities colonizing the seeds of broad bean of White Hangdown cultivar, planting in companion with fennel and coriander and protected using chemical agents and biopreparations.

2. Materials and methods

The research material in the experiment included the seeds of broad bean of White Hangdown cultivar originated from the strict field experiment conducted in the years 2010–2012 at the Experimental farm of the University of Agriculture in Krakow situated in Prusy. The kind of applied protection was the research factor. The experiment consisted of 10 combinations including two alternating rows sowings of broad bean with fennel and coriander (combinations VI and VII), and four variants of biological protection using Polyversum WP (*Pythium oligandrum*), Bioczos BR (garlic mash in paraffin coating), Biosept 33 SL (extract from grapefruit seed and pulp) preparations (II–V), and three variants of chemical protection using Vitavax 200 FS (carboxin and thiram), Decis 2.5 EC (deltamethrin), Fastac 100 EC (alpha-cypermethrin), Penncozeb 80 WP (mancozeb) (VIII–X). A detailed plan of protection is presented in the scheme below.

I - Control - without protection

II - seeds dressing with Polyversum WP;

III - seeds dressing with Polyversum WP + foliar application 2xBioczos BR, 1xBiosept 33SL;

IV - seeds dressing with Polyversum WP + foliar application 3xBioczos BR, 1xBiosept 33SL;

V - seeds dressing with Polyversum WP + foliar application 4xBioczos BR, 1xBiosept 33SL;

VI - seeds dressing with Polyversum WP + companion planting with coriander;

VII - seeds dressing with Polyversum WP + companion planting with fennel;

VIII - seeds dressing with Vitavax 200 FS;

IX - seeds dressing with Vitavax 200 FS + foliar application 1x Decis 2,5 EC, 1xFastac 100 EC, 1xPenncozeb 80 WP;

X - seeds dressing with Vitavax 200 FS + foliar application 2x Decis 2,5 EC, 1xFastac 100 EC, 1xPenncozeb 80 WP;

Broad bean seeds were harvested in the full maturity stage and stored for two months in a dry room (at a temperature of about 15°C). Laboratory tests included randomly selected 200 pieces of broad bean seeds from each combination. The seeds were surface-disinfected in 50% ethanol, rinsed three times in sterile distilled water, dried on sterile tissue-paper and placed on solidified PDA medium (Potato Dextrose Agar) with chloramphenicol addition in Petri dishes of a diameter of 150 mm. were surface-disinfected in 50% ethanol, rinsed three times in sterile distilled water, dried on sterile tissue-paper and placed of 10 pieces on solidified PDA medium (Potato Dextrose Agar) with chloramphenicol addition in Petri dishes of a diameter of 150 mm. The cultivation was carried out in a climatic chamber for 10 days at 23°C. Emerging fungal colonies were gradually cleaved on agar slants. Then, macroscopic and microscopic observations were performed, which allowed to identify most of fungi species using mycological

keys and monographs [5, 7, 20, 23, 26, 32]. The frequency of particular species or genera occurrence was determined based on the number of certain fungus isolates. Its value was expressed in percentage referring to the total number of isolates (100%) obtained for specified lot of seeds.

3. Results

It was found based on the conducted study, that mycological purity of broad bean seeds varied in years (Tab. 1). In total, 4028 fungal isolates were separated from the seeds for three growing seasons, the highest number 1626 (40.37%) was obtained in 2012, and the lowest, i.e. 954 (23.68%) in 2011. No qualitative differentiation was observed in isolated populations of fungi. 25 species of fungi belonging to 14 genera, and saprotrophic *Penicillium* spp., *Mucor* spp., *Aspergillus* spp. and *Acremonium* spp. were identified. Broad beans seeds were each year colonized with the highest frequency by *A. alternata* species, and its share was 26.1–31.7% (Fig. 1). Also *B. cinerea* was noted in the rank of dominant, but its share was not stable over the years. For example, its occurrence in 2010 was on a level of 8.3%, and it almost doubled in 2012. In turn, the fungi of *Fusarium* genus represented by 8 species, the most abundantly by *Fusarium equiseti*, constituted in 2010 27.2% of isolated fungi population, and in subsequent years their share was 11.8% and 11.1%, respectively. Moreover, broad bean seeds were very abundantly colonized by saprotrophic fungi belonging to *Penicillium* genus (18.3–24.9%) (Fig. 1).

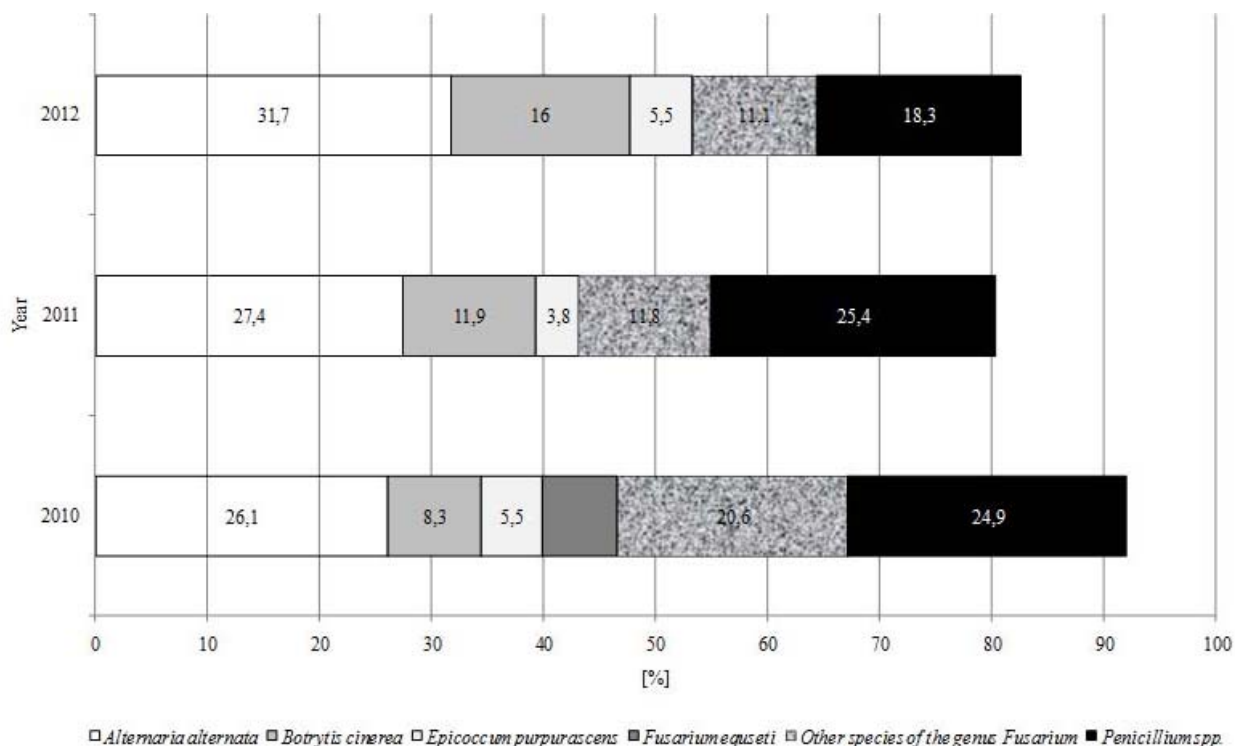
Differentiation in the number of isolated fungal colonies in different variants of protection was found based on mycological analysis of broad bean seeds. The largest number of isolated fungi, an average of 546 isolates, was obtained in lots of seeds from alternating rows crop of broad bean with coriander (VI) and fennel (VII) (Tab. 2). In contrast, an average number of fungal isolates obtained from the seeds of chemical and biological protection variants was identical (348), and also significantly lower than those in plots VI and VII and the control (501). At the same time, it was observed that an increased amount of treatments carried out both using chemical (VIII–X) and biotechnical preparations (III–V), affects the reduction in the number of isolated fungi, and thus also favors an improvement in mycological purity of the seeds (Tab. 2). Moreover, both within the variants of biological and chemical protection, increased level of foliar application of the preparations was accompanied by an increased share of pathogenic species, and the reduced proportion of saprophytes (Fig. 2). In turn, the most balanced ratio of pathogens (58%) to saprophytes (40.85%) was found in the population of fungi isolated from the seeds from companion planting of broad bean with fennel (VII). Single isolates of antagonistic fungi *Trichoderma hamatum* and *Trichoderma harzianum* were noted in all variants of the protection, their share in community of fungi colonizing the seeds remained at a level from 0.5% to 1.7%. Applied protection slightly modified the qualitative composition of fungi colonizing the seeds of broad bean. Pathogenic species such as *Alternaria alternata*, *Botrytis cinerea*, *Epicoccum purpurascens* were noted in the rank of dominants. Community of fungi isolated from the seeds of broad bean grown in alternating rows with fennel was dominated by species belonging to *Penicillium* genus, and their share was up 34.5%.

Table 1. Fungi isolated from broad bean seeds in the years 2010–2012 (number of isolates)

Tab. 1. Grzyby wyizolowane z nasion bobu w latach 2010-2012 (liczba izolatów)

No.	Fungal species	Years			Sum	Percentage share
		2010	2011	2012		
1.	<i>Acremonium glaucum</i> W. Gams	0	0	2	2	0.05
2.	<i>Acremonium</i> spp.	3	0	0	3	0.07
3.	<i>Alternaria alternata</i> (Fr.) Keissler	378	261	515	1154	28.66
4.	<i>Ascochyta fabae</i> Speg.	30	32	41	103	2.56
5.	<i>Aspergillus</i> spp.		13	14	27	0.67
6.	<i>A. versicolor</i> (Vuill.) Tiraboschi	17	0	5	22	0.55
7.	<i>Botrytis cinerea</i> Pers. ex Nozza et Balb.	120	114	261	495	12.29
8.	<i>Cladosporium herbarum</i> (Pers.) Link ex Gray	13	11	19	43	1.07
9.	<i>Colletotrichum coccodes</i> (Wallr.) S. Hughees	2	3	9	14	0.35
10.	<i>Colletotrichum gloeosporioides</i> (Penz.) Sacc.	27	20	24	71	1.77
11.	<i>Epicoccum purpurascens</i> Ehrenb. ex Schlecht	79	36	90	205	5.09
12.	<i>Fusarium avenaceum</i> (Corda ex Fr.) Sacc.	40	20	37	97	2.41
13.	<i>F. culmorum</i> (W.G. Smith) Sacc.	46	16	26	88	2.18
14.	<i>F. equiseti</i> (Corda) Sacc.	96	33	67	196	4.87
15.	<i>F. oxysporum</i> Schlecht	40	25	26	91	2.26
16.	<i>F. poae</i> (Peck) Wollenw.	31	10	7	48	1.19
17.	<i>F. solani</i> (Mart.) Sacc.	11	5	2	18	0.45
18.	<i>F. sporotrichoides</i> Sherbakoff	9	4	12	25	0.62
19.	<i>F. tricinctum</i> (Corda) Saccardo	25	0	4	29	0.72
20.	<i>Mucor</i> spp.	58	40	81	179	4.44
21.	<i>Penicillium</i> spp.	361	242	297	900	22.34
22.	<i>Phoma exiqua</i> Desm.	3	6	11	20	0.50
23.	<i>P. glomerata</i> (Corda) Wollenweber et Hochapfel	4	10	12	26	0.65
24.	<i>Rhizopus nigricans</i> Ehrenberg	12	9	4	24	0.60
25.	<i>Sclerotinia sclerotiorum</i> (Lib.) de Bary	9	5	0	14	0.35
26.	<i>Trichoderma hamatum</i> (Bon.) Bain	4	4	8	16	0.40
27.	<i>Trichoderma harzianum</i> Rifai	3	2	20	25	0.62
28.	<i>Trichothecium roseum</i> Link	20	13	14	47	1.17
29.	<i>Verticillium albo-atrum</i> Renke et Barthold	3	12	7	22	0.55
30.	Non-sporulating cultures	4	8	10	22	0.55
Total		1448	954	1626	4028	100.00
Percentage share		35.95	23.68	40.37	100.00	

Source: own work / Źródło: opracowanie własne



Source: own work / Źródło: opracowanie własne

Fig. 1. The share of dominants in community of fungi isolated from broad bean seeds

Fig. 1. Udział dominantów w zbiorowisku grzybów wyizolowanych z nasion bobu

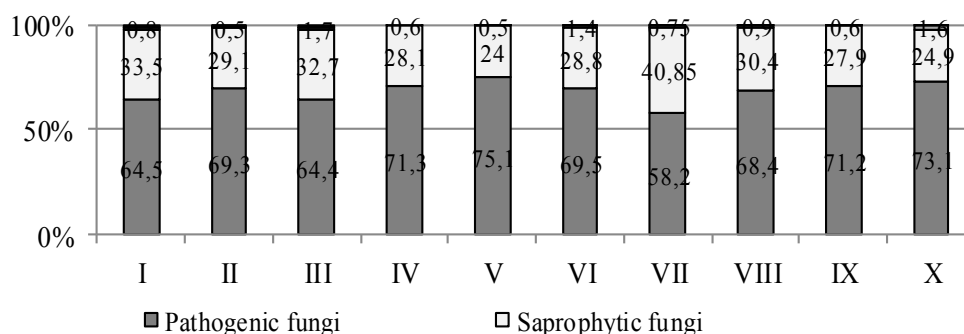
Table 2. The impact of protection on the share of fungi isolated from broad bean seeds [%]

Tab. 2. Wpływ ochrony na udział grzybów wyizolowanych z nasion bobu [%]

Fungal species	Control	Biological protection				Companion planting		Chemical protection			
	I*	II	III	IV	V	VI	VII	VIII	IX	X	
Pathogenic fungi											
<i>Alternaria alternata</i>	26,55	32,08	31,03	34,06	22,60	29,22	26,25	29,43	33,13	26,91	
<i>Ascochyta fabae</i>	2,40	3,23	2,87	2,50	2,26	2,18	2,59	2,63	1,55	3,99	
<i>Botrytis cinerea</i>	11,18	10,78	9,20	10,31	14,41	11,62	10,54	10,54	14,24	14,29	
<i>Colletotrichum coccodes</i>	0,60	0,27	0,00	0,31	0,00	0,18	0,18	0,24	0,62	0,33	
<i>Colletotrichum spp.</i>	2,20	2,96	1,44	1,25	2,26	0,43	1,11	1,44	1,86	3,65	
<i>Epicoccum purpurascens</i>	5,59	3,23	2,30	4,69	10,45	8,35	2,96	3,11	3,10	3,65	
<i>Fusarium avenaceum</i>	2,20	1,08	1,72	1,56	3,11	2,72	2,22	2,63	1,24	5,32	
<i>F. culmorum</i>	1,60	2,43	2,01	1,25	2,54	1,63	1,48	2,15	3,41	3,99	
<i>F. equiseti</i>	4,79	4,85	3,74	2,81	11,02	5,99	4,62	3,59	1,86	4,32	
<i>F. oxysporum</i>	1,20	1,89	4,60	4,06	0,28	1,99	1,29	3,11	3,72	1,33	
<i>F. poae</i>	0,60	1,35	0,86	3,44	0,85	0,91	0,55	2,87	0,62	0,33	
<i>F. solani</i>	0,20	0,00	1,15	0,31	0,85	0,36	0,18	0,48	1,24	0,00	
<i>F. sporotrichoides</i>	1,20	0,81	0,29	0,31	0,85	0,36	0,55	0,96	0,00	0,66	
<i>F. tricinctum</i>	0,20	1,08	0,57	0,94	1,69	0,18	0,74	0,24	0,93	0,99	
Łącznie rodzaj <i>Fusarium</i>	11,98	13,48	14,94	14,69	21,19	14,16	11,65	16,03	13,00	16,94	
<i>Phoma exigua</i>	0,60	0,27	0,29	0,31	0,28	0,36	0,37	0,48	0,93	0,99	
<i>P. glomerata</i>	0,80	0,27	0,57	0,00	0,28	1,09	1,11	0,48	0,93	0,99	
<i>Sclerotinia sclerotiorum</i>	0,20	0,27	0,86	0,00	0,28	0,54	0,18	0,00	0,62	0,00	
<i>Trichothecium roseum</i>	1,60	1,62	0,57	2,19	0,85	0,54	0,92	0,48	1,24	0,99	
<i>Verticillium albo-atrum</i>	0,80	0,81	0,29	0,94	0,28	0,54	0,37	0,48	0,00	0,33	
Total	64,47	69,27	64,37	71,25	75,14	69,51	58,23	68,42	71,21	73,09	
Saprotrophic fungi											
<i>Acremonium glaucum</i>	0,20	0,00	0,00	0,00	0,00	0,18	0,00	0,00	0,00	0,00	
<i>Acremonium spp.</i>	0,20	0,27	0,00	0,00	0,00	0,18	0,00	0,00	0,00	0,00	
<i>Aspergillus spp.</i>	0,40	0,81	0,29	0,00	0,56	0,73	0,37	1,67	0,62	0,99	
<i>A. versicolor</i>	0,40	0,27	0,86	0,63	0,56	0,91	0,37	0,00	0,31	0,66	
<i>Cladosporium herbarum</i>	1,80	2,43	0,57	1,88	1,41	1,27	0,55	0,24	0,93	0,66	
<i>Mucor spp.</i>	3,99	4,58	4,31	8,44	7,06	2,36	4,44	3,59	2,48	2,66	
<i>Penicillium spp.</i>	25,95	19,95	26,15	16,25	14,12	22,87	34,57	23,92	22,91	19,60	
<i>Rhizopus nigricans</i>	0,60	0,81	0,57	0,94	0,28	0,36	0,55	0,96	0,62	0,33	
Total	33,53	29,11	32,76	28,13	24,01	28,86	40,85	30,38	27,86	24,92	
Antagonistic fungi											
<i>Trichoderma hamatum</i>	0,40		0,86	0,31	0,00	0,54	0,37	0,24	0,00	0,66	
<i>Trichoderma harzianum</i>	0,40	0,54	0,86	0,31	0,56	0,91	0,37	0,72	0,62	0,99	
Total	0,80	0,54	1,72	0,63	0,56	1,45	0,74	0,96	0,62	1,66	
Non-sporulating cultures	1,20	1,08	1,15	0,00	0,28	0,18	0,18	0,24	0,31	0,33	
The number of isolates	501	371	348	320	354	551	541	418	323	301	
Percent of all isolates	12,44	9,21	8,64	7,94	8,79	13,68	13,43	10,38	8,01	7,47	
Medium number of isolation cells for protection variant	501					348,25			546		
The mean percentage	12,44					8,64			13,55		

I* - Control, II – seeds dressing with Polyversum; III – seeds dressing with Polyversum + 2xBioczos, 1xBiosept; IV - seeds dressing with Polyversum + 3xBioczos, 1xBiosept; V - seeds dressing with Polyversum + 4xBioczos, 1xBiosept; VI – seeds dressing with Polyversum + companion planting with coriander; VII – seeds dressing with Polyversum + companion planting with fennel; VIII - seeds dressing with Vitavax; IX - seeds dressing with Vitavax + 1x Decis, 1xFastac, 1x Penncozeb

Source: own work / Źródło: opracowanie własne



Explanations see Tab. 2 / Objasnienia jak w Tab. 2

Source: own work / Źródło: opracowanie własne

Fig. 2. Percentage of pathogenic and saprotrophic fungi isolated from broad bean seeds depending on the protection

Fig. 2. Procentowy udział grzybów patogenicznych i saprotroficznnych wyosobnionych z nasion bobu w zależności od ochrony

4. Discussion

Regardless of the protection, 4028 fungal isolates were obtained from the seeds of broad bean of White Hangdown cultivar, with the significant advantage (58.23–75.14%) of pathogens represented by 19 species (Tab. 1, Fig. 2). *Alternaria alternata* species was clearly dominating in the communities of fungi isolated from different seed lots. With the highest frequency of 34.06% it was found in the seeds originating from the plot of biological protection (IV), where seeds were dressed with Polyversum WP, 3xBioczos BR + 1xBiosept 33SL (Tab. 2). On the other hand, additional spraying with Bioczoz BR preparation (combination V) contributed to the reduction of *A. alternata* share as much as 11.5%, and noted prevalence of 22.6% was the lowest for the analyzed seed lots. Fungi of *Alternaria* genus, especially *A. alternata*, are ubiquitous plant pathogens associated with different diseases of roots, leaves, stems, and generative organs [15, 18, 25, 28, 37, 38]. However, the most often the diseases symptoms on plants are the result of this species concurrence with other fungi of *Cladosporium* or *Epicoccum* genus. The danger of *A. alternata* presence in the seeds is related to the production of mycotoxins belonging to different chemical groups which demonstrate cytotoxic, teratogenic and mutagenic activity. [21]. The most frequent metabolites of that fungus in the seeds of plants, fruits and vegetables are: alternariol (AOH), monomethyl ether (AME), altenuen (ALT) and tenuazonic acid [4, 21, 25]. Exceptionally dangerous are mycotoxins produced by *Fusarium* genus. A species *F. equiseti* produces many mycotoxins such as fusarenon (FUS), fusarochromanon, diacetoxiscipenol (DAS) and like the *F. culmorum* nivalenol (NIV) and zearalenone (ZEA). Whereas the *F. avenaceum* and *F. oxysporum* are the main producers of moniliformin (MON). The highest number of *F. equiseti* (11.02%), *E. purpurascens* (10.45%) and *B. cinerea* (14.41%) isolates was found in a variant of protection with the maximum number of biological treatments (V) compared to other combinations. However, the latter was recorded with the same frequency in the lot of seeds from chemical protection plots (IX and X). The lowest share of fungi of *Fusarium* genus was observed in seeds from broad bean companion planting with fennel (11.65%) and the control plot (11.98%). In other combinations, *Fusarium* genus, the most frequently represented by *F. equiseti*, *F. avenaceum*, *F. culmorum*, and *F. oxysporum*, accounted for 13% to 21.19%. Other pathogens important for legume plants, such as *Ascochyta fabae*, *Colletotrichum* spp., were isolated from broad bean seeds in an amount not exceeding 4%. In addition, even fewer isolates of *Sclerotinia sclerotiorum*, *Phoma exigua*, *P. glomerata*, *T. roseum* and *V. alboatrum* were noted. The available literature lacks information on broad bean seeds colonization by pathogenic fungi. However, the results obtained coincide with mycological analysis of broad bean seeds of White Windsor cultivar previously carried out by the author [10-13]. The above-mentioned fungi species were also isolated from the seeds of other *Fabaceae* plant species such as lupine, lentil, faba bean, pea or bean [1, 8, 9, 16, 17, 19, 22, 27, 29, 37]. Qualitative and quantitative composition of fungi colonizing the seeds is the result of environmental factors and the complex process of interaction between particular species of fungi. In our study, high precipitation recorded in May and June 2010 and 2012, contributed to the greater fungal contamina-

tion of broad bean seeds. Increased humidity especially favored an increased number of dominant fungi isolates, i.e. *A. alternata*, *E. purpurascens*, *F. equiseti* and saprobionts *Penicillium* spp., and *Mucor* spp.

Development of fungi communities colonizing the seed depends on the length of storage period [6]. Immediately after harvest, or after the short-term storage, the share of *A. alternata* species and fungi of *Fusarium* genus can be much higher. In turn, extending the storage of seeds to 1.5 or 2 years promotes colonization of the seeds by saprobionts like *Penicillium* and *Rhizopus*. In our study, mycological analysis of broad bean seeds was conducted after two months of harvest, i.e. after a relatively short storage, hence the high share of *A. alternata* in fungal community. The use of seeds contaminated with fungi as reproductive material can be extremely risky, since it threatens the development of blight diseases of seedlings, shoots, and even later plants dying as a result of fusarium wilt.

5. Conclusions

1. Pathogenic fungi: *Alternaria alternata* – 28.6%, *Botrytis cinerea* – 12.3%, *Epicoccum purpurascens* – 5.09%, *Fusarium equiseti* – 4.9% and saprobionts of *Penicillium* genus – 22.3% were the most frequently isolated from the seeds of broad bean of White Hangdown cultivar.
2. Applied variants of biological and chemical protection in a comparable degree limited the number of fungi isolated from broad bean seeds. Increased number of treatments resulted in an increase in the share of pathogenic fungi and reduction of saprobionts.
3. Companion planting of broad bean with coriander and fennel deteriorates the mycological purity of broad bean seeds. Fennel favors an intensive colonization (34.6%) of the seeds by the *Penicillium* spp. fungi.

6. References

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