Scientific Papers. Series A. Agronomy, Vol. LXVIII, No. 2, 2025
ISSN 2285-5785; ISSN CD-ROM 2285-5793; ISSN Online 2285-5807; ISSN-L 2285-5785

COMPARATIVE STUDY OF THE INVASIVE ENTOMOFAUNA ASSOCIATED TO THE SPECIES *Phaseolus vulgaris* L. and *Glycine max* (L.) Merr. ON EXPERIMENTAL PLOTS IN THE "ALEXANDRU CIUBOTARU" NATIONAL BOTANICAL GARDEN (INSTITUTE), REPUBLIC OF MOLDOVA

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Abstract

Soybean and common bean are widely grown as food and fodder plants, but also have high estimated energy potential. They are among the most significant field crops, cultivated annually in the Republic of Moldova. These species are economically feasible, versatile crops, but their major disadvantage is their vulnerability to the impact of invasive entomofauna. The phytosanitary monitoring conducted on the experimental sectors of the Botanical Garden, in the collection of fodder, honey and energy plants, in comparison with the productive sectors in agro-industrial areas, resulted in the estimation of the invasive impact, in 2024, of the most harmful insect species for these crops. The phytosanitary monitoring revealed the presence of a significant complex of parasitic insects, represented by 12 species included in four orders (Coleoptera, Diptera, Hemiptera, Lepidoptera) and nine families. The most harmful and invasive pests for both researched plant species were: Acanthoscelides obtectus Say*., Agrotis spp., Sitonia liniatus L., Aphis fabae Scopoli, Myzus persicae Sulzer., Aphis glycines Matsumara; Vanessa cardui L., Delia platura Meigen. The obtained data are useful for adapting pest control measures to the integrated protection system.*

Key words*: common bean, experimental sectors, parasitic insects, phytosanitary monitoring, soybean.*

INTRODUCTION

The annual climatic instability in the Republic of Moldova and its interconnection with the structure and state of biodiversity in natural and anthropic ecosystems, also connected to global warming, results in the fact that distortions are reported in the quality and composition of flora and fauna, both useful and invasive (Busuioc, 2004; Starodub et al., 2013; Starodub, 2015). This system also includes complexes of parasitic insects, pathogens etc., some of which may be completely out of control, but with their evolution over time can become very dangerous for practically all field and horticultural crops (Ghizdavu et al., 1997; Lazări & Busuioc, 2002). In the Republic of Moldova, the common bean – *Phaseolus vulgaris* L. and soybean – *Glycine max* (L.) Merr. are cultivated annually, for the agri-food and zootechnical sectors, as raw material to produce canned and dried beans and pods, also useful for the chemical and pharmaceutical industries, as well as for the pedo-ecological significance of these plants. Plants of these species possess relative capacities of resistance and tolerance to environmental stressors, species of harmful organisms, which is why they are included in modern agricultural systems, with numerous advantages in optimal crop rotation structures and technologies of usage (Busuioc, 2004; Timuș, 2015; Țugulea & Derjanschi, 2015). The advantages noted are also connected to some disadvantages, which also require efforts to be overcome and, subsequently, to obtain high yields of beans and fodder. Thus, the control measures start with conducting annual and seasonal phyto-sanitary surveys, useful for further specific research on the parasitic impact of invasive organisms, including the analysis of the diversity of harmful insects affecting the crops.

The investigations we initiated are important to ascertain the dynamics of the phenological stages of common bean and soybean plants, the diversity, the parasitic impact of the most dangerous insect species, the local invasive potential, which could affect these local crops, traditionally cultivated both in industrial production associations and in the private sector. All these aspects and elements are necessary to provide the necessary cultivation practice in order to choose and apply the most appropriate control measures to all invasive insect associations in bean and soybean crops, as important links that are included in integrated protection management (Lazări & Busuioc, 2002; Tălmăciu & Tălmăciu, 2014).

The introductory data denote the research conducted within the "Alexandru Ciubotaru" National Botanical Garden (Institute) of Moldova State University (NBGI MSU), according to the institutional project and research subprogram (2024-2027). Investigations are conducted annually according to the multilaterally adopted research program, including research on complex biological phytosanitary control, where entomological surveys are also associated, being aimed at establishing insect complexes, especially harmful ones in experimental land plots planted with Fabaceae crops, including common bean and soybean. This program focuses on conducting research, which includes entomological surveys with a phytosanitary focus, on *Phaseolus vulgaris* L. and *Glycine max* (L.) Merr. The goal is to identify the most invasive insect species that pose a threat to the above-mentioned plants, under natural conditions, on experimental plots at NBGI MSU. The main objectives for achieving the proposed goal are: to establish the structure and diversity of invasive insect species affecting in common bean (*Phaseolus vulgaris* L.) and soybean (*Glycine max* (L.) Merr.) crops and to determine the values of major parasitic impact, through comparative analyses of the frequency and abundance indices of pests detected on the experimental plots, during various phenological stages of the plants.

MATERIALS AND METHODS

The research program referred to in the present article was conducted at the "Alexandru Ciubotaru" National Botanical Garden (Institute) of Moldova State University, Chișinău, Republic of Moldova, (NBGI MSU) during the growing season of 2024. The investigations included setting the experiments on the experimental plots of the "Plant Resources" Laboratory, which were planted with a wide range of species, varieties and hybrids of Fabaceae, meant to be cultivated for grains, pods and forage, on which, subsequently, complex study of morpho-taxonomic and bio-ecological characteristics was done. Activities such as demarcation of plots, sowing, soil maintenance and plant care woks were performed from mid-March until August, ending with harvesting and storage of the beans (Figures 1, 2).

A **B**

Figure 1. Experimental plots planted with Fabaceae species: a - soybean, b - common bean, the stage of 5-8 adult leaves (May, 2024, NBGI)

Figure 2. Common beans in the flowering stage (June, 2024, NBGI)

According to ecological characteristics, common bean and soybean seeds are incorporated into the soil later, as compared with other popular crops such as peas, because they need the soil temperature to be on average +7...+10°C, being unable to tolerate lower temperatures. The seeds were planted at a depth of 5-6 cm in the soil, the distance between rows being 70 cm. Further, the cultivation requirements and maintenance measures were observed. Once the plants germinated, visual

phenological observations were made on the emerging organs, besides, periodic methodological maintenance works were performed on the entire experimental sector. Weed removal, soil loosening and irrigation, making periodic phytosanitary records and analyzing plant sensitivity to the impact of drought and heat, which are frequent phenomena in the period May – July, were key activities performed during the growing season of the studied plants. Phenological and complex phytosanitary records were initiated with the appearance of diseases and pests on all mounted Fabaceae forms, including common bean and soybean plants (Figures 3, 4).

A

B

Figure 3. Experimental plots with common bean and soybean (July, 2024, NBGI)

In the dynamics of phenological stages, periodic records were made to visualize the growth and development of plants according to the comparatively estimated potential, by species and plots, according to the guidelines of Beideman, 1974. Phytosanitary records for detecting the diversity of harmful organisms, in particular insect species, were made by taking samples, pictures, field notes, subsequently being analyzed in the laboratory, and determined taxonomically with the help of handbooks for entomological identification (Bei-Bienco, 1962; 1966; Plaviliscicov, 1997). The current methodological guidelines were also used to research and test new chemical and biological products with insecticidal and fungicidal action for the integrated protection management of crops in the Republic of Moldova, 2020-2022.

As part of the investigations into the extent of damage caused by harmful insects to the studied species, samples of affected organs and soil were collected for subsequent analysis under laboratory conditions. According to the results of entomological surveys, indices of the population *density* (D), *frequency* (F%) and *intensity* (I%) of diseases caused by harmful species were obtained and compared by species, repetitions, plots and phenological phases. To determine the degree of pest attack across different variants and repetitions, plant samples were collected and analyzed in the laboratory. Based on phytopathological research, the frequency of attack **(F %)** and the intensity of development **(I %)** were established. The frequency of attack **(F %)** represents the relative proportion of attacked plants or plant organs **(n)** compared to the total number of analyzed plants or organs **(N)** and is calculated using a standard formula. The intensity of attack **(I %)** represents the extent, as a percentage, to which a plant or organ is affected. The following formula is used to calculate the intensity of attack:

$$I\% = \frac{(n_1 . 1) + (n_2 . 2) + (n_3 . 3) + (n_4 . 4)}{N \times 4},$$

where: n_1, n_2, n_3, n_4 – the number of plants or organs attacked at the respective grade; N – the total number of plants or organs examined; 4 – the maximum grade on the scale used (Îndrumări metodice pentru testarea produselor chimice și biologice de protecție a plantelor în Republica Moldova, 2012).

Scales with varying numbers of grades may be used to assess pest attack intensity. In our experiments, four-grade scale was used, corresponding to certain percentage ranges of the affected areas, as follows: 0 – no visible symptoms; 1 – up to 10% of the leaf blade affected; 2 – 10-25% of the leaf blade affected; 3 – 25-50% of the leaf blade affected; 4 – more than 50% of the leaf blade surface affected.

At the same time, the degree of attack was determined according to the estimation scale after the reaction of soybean and bean plants to

the impact of parasitic insects (Perju, 1995; Starodub, 2015; Timuș, 2016).

A B

Figure 4. A - analysis of soybean roots for diseases; B - visualization of complex diseases in beans (July, 2024)

RESULTS AND DISCUSSIONS

The outbreaks of diseases caused by polyphagous parasitic insect associations specific to common bean and soybean plants growing under the environmental conditions of the Republic of Moldova are primarily caused by unstable environmental factors, creating difficulties in preventing their emergence and invasive impact (Lazăr & Busuioc, 2002; Timuș, 2015).

Not all species of harmful insects detected on these crops cause large economic losses, and in the case of common beans and soybeans grown for grains, the most important issue is the protection of the vegetative organs of the plant. This part of the plants is mostly responsible for the photosynthetic activity during the early stages of the growing season, and the damage caused by insects considerably reduces the productivity of this physiological process, also affecting significantly the subsequent formation of flowers, pods and grains. These associations of harmful insects targeting soybeans and common beans are scrutinized by researchers, and complex phytosanitary surveys are frequently conducted, focusing on entomological aspects to establish the level of resistance and tolerance to the impact of the most harmful insect species and other pests.

Both the analysis of published bibliographic sources and our own research findings indicate that, in the Republic of Moldova, the damage caused by some invasive insect species, along with some pathogen associations to the investigated crops, is of major agro-economic importance. The insects detected annually as potentially invasive pests, found in various phenological phases on common bean and soybean plants, are some common, polyphagous species, attacking plants since the germination stage: *Agriotes* spp. L., fam. Elateridae; bean weevil – *Acanthoscelides obtectus* Say, fam. Chrysomelidae; pea leaf weevil – *Sitona lineatus* L., fam. Curculionidae; turnip moth – *Agrotis segetum* Den. et Schiff, silver Y - *Autographa gamma* L., cotton bollworm – *Helicoverpa armigera* (Hubner), fam. Noctuidae; *Ophiomyia phaseoli* (Tryon) fam. Agromyzidae, silverleaf whitefly – *Bemisia tabaci* Genn. fam. Aleyrodidae, soybean aphid – *Aphis glycines* Matsumura, black bean aphid – *Aphis fabae* Scopoli fam. Aphididae.

The researched plants were monitored on the experimental sector of NBGI MSU, divided into plots, in several repetitions, randomly placed, on a natural background, with the presence of soil and terrestrial insect species, under equal environmental and time conditions. Table 1 includes the insect species that caused damage to the monitored plants, differentiated by the development stages of insects which caused damage, affected plant organs and indices of population density (D), intensity (I%) and frequency (F%) of attack. Some species were detected even during the earliest phenological stages of plants, as invasive polyphagous pests found in the soil, which have significantly affected the germinated seedlings, such as: *Agriotes* spp. L., *Acanthoscelides obtectus* Say, turnip moth - *Agrotis segetum* Den. et Schiff. As the plants matured, these pest associated with larvae of moths of the family Noctuidae (*Agrotis segetum* Den. et Schiff, *Autographa gamma* L., *Helicovepra armigera* Hubner), with an attach intensity of 15-30%, varying among the experimental plots. It is significant to note that during the research period, the following invasive species were reported in the monitored plots planted with common bean and soybean, such as; silverleaf whitefly – *Bemisia tabaci* Genn., soybean aphid – *Aphis glycines* Matsumura, black bean aphid – *Aphis fabae* Scopoli, greenfly – *Myzus persicae* Sulzer., painted lady – *Vanessa cardui* L., bean seed fly – *Delia platura* Meigen., which invasively attack the leaves, flowers, pods and grains during their formation. As shown in Table 1, diseases caused specifically by 12 species of

harmful insects of four orders – Coleoptera, Diptera, Hemiptera, Lepidoptera, with polyphagous phytoparasitic specialization, specific to field crops as well as plants from the spontaneous flora were detected as a result of the entophytosanitary monitoring carried out during the growing season of 2024, on the experimental plots of NBGI MSU planted with soybean and common bean. Practically all plants analyzed are subject to pest invasions, which were highlighted during various stages of plant development, starting with plant germination, progressing to serious parasitic impact until the harvest of the grains, the damage expanding to various plant organs and tissues.

Thus, the earliest detected species are beetles of gen. *Agriotes*, bean weevil – *Acanthoscelides obtectus* Say, pea leaf weevil – *Sitona lineatus* L., followed by diverse caterpillars of the species *Agrotis segetum*, Den. et Schiff, *Autographa gamma* L., *Helicoverpa armigera* (Hubner) from fam. Noctuidae. The process of plant colonization, with the appearance of the generative organs, by various species of highly invasive aphids is of particular importance, since these pests seriously affect plants and are vectors of diseases, such as rot, affecting leaves and pods throughout the growing season, such as: black bean aphid - *Aphis fabae* Scopoli, greenfly – *Myzus persicae* Sulzer, soybean aphid – *Aphis glycines* Matsumura. Other significant pests causing severe damage to pods and grains are Pulse pod borer moth, *Etiella zinckenella* Triet., Painted lady – *Vanessa cardui* L., Bean seed fly – *Delia platura* Meigen., associated with species of thrips, bedbugs, wasps etc.

At the same time, during our research, some considerable populations of species of entomophagous insects that hunt on aphids that invade plants were also detected; the most frequent and effective were: *Trichogramma evanescens, Aphelinus flavipes*, associated with various species of ladybugs – *Coccinella* spp., which had an effective biological impact, especially on the regulation of aphid populations.

Table 1. Characteristics of phytosanitary indices of harmful insect species detected in common bean and soybean crops during the 2024 growing season, on experimental plots within the NBGI MSU

Species name, trophic specialization	Development stage of the insect	Affected plant organs	(D) Density / 100 g soil/m^2 / plant	(F) Frequency %	(I) Intensity %	Nr. of generations per year
Species of polyphagous insects found on common bean and soybean plants						
1. Species of beetles of gen. *Agriotes,* fam. Elateridae ord. Coleoptera	Larvae	They gnaw at the root collar, make cavities in the roots, stems	2-4 larvae/m^2	7-10	5-10	1 in 3-4 years
2. Species of moths of fam. Noctuidae, Ord. Lepidoptera*: Agrotes segetum*, Den. et Schiff, *Autographa gamma* L.	Larvae	Eat first the underground parts, then the aerial parts of the plants	2-3 larvae/m^2	10-15	7-12	1 -2 per year
3. Bean weevil - *Acanthoscelides obtectus* Say, fam. Chrysomelidae , ord. Coleoptera	Larvae, adults	'cut' the root collar, eat young plants	5-7 adults/m^2	12-17	10-15	1 per year
4. Pea leaf weevil - *Sitona lineatus* L. fam. Curculionidae, ord. Coleoptera	Larvae, adults	Eat young plants	5-10 adults/m^2	15-18	12-15	1 per year
5. Cotton bollworm - *Helicoverpa armigera* (Hubner); fam. Noctuidae, ord. Lepidoptera	Larvae	Eat intensively the whole plant	5-12 adults/m^2	15-30	17-25	7 per year
6. Whitefly*, Bemisia tabaci* Gen. ord. Hemiptera, fam. Aleyrodidae	Larvae, adults	Suck the sap from young soybean leaves	1-2 colonies / 10 plants	7-10	5-7	7-8 per year
7. Black bean aphid – *Aphis fabae* Scopoli., fam. Aphididae, ord. Hemiptera	Larvae, adults	Colonies located on the underside of the leaves, severely deform the leaf blade	2-3 colonies /10 plants	10-13	7-10	5 per year
8. Greenfly – *Myzus persicae* Sulzer., fam. Aphididae, ord. Hemiptera	Larvae, adults	Colonize the plant, young leaves	1-2 colonies / 10 plants	5-7	3-5	5-6 per year

Species name, trophic specialization	Development stage of the insect	Affected plant organs	(D) Density / 100 g soil/m^2 /plant	(F) Frequency %	(I) Intensity %	Nr. of generations per year
9. Soybean aphid – *Aphis glycines* Matsumara, fam. Aphididae, ord. Hemiptera	Larvae, adults	Colonize young soybean leaves	3-5 colonies / 10 plants	2-3	1-3	4-5 per year
10. Pulse pod borer moth, *Etiella zinckenella* Triet. – fam. Pyralidae, ord. Lepidoptera	Larvae, adults	Severely affect soybean pods	4-5 affected pods / plant	12-15	7-10	2 per year
11. Painted lady – *Vanessa cardui* L., fam. Nymphalidae, ord. Lepidoptera	Larvae	Consume the leaf blade of soybeans and beans	5-7 larvae in colonies / 10 plants	10-13	7-10	3 per year
12. Bean seed fly – *Delia platura* Meigen., *fam.* Anthomyiidae, ord. Diptera	Larvae	Severely affects bean grains	3-4 affected grains of 10 analyzed	5-10	3-7	3 per year

CONCLUSIONS

The results of the entomological and phytosanitary investigations on soybean and common bean crops, carried out on the experimental plots of NBGI MSU, in 2024, highlighted the range of invasive insect species that caused significant plant damage, resulting in the estimation of the invasive impact indices, the diversity and structure of polyphagous and specific insect communities, compared by crops, phonological stages and plots.

The phytosanitary monitoring revealed the presence of a significant complex of parasitic insects, represented by 12 species included in four orders (Coleoptera, Diptera, Hemiptera, Lepidoptera) and nine families. The most harmful and invasive pests for both researched plant species were: *Agrotis* spp., *Acanthoscelides obtectus* Say, *Sitona lineatus* L., *Aphis fabae* Scopoli, *Aphis glycines* Matsumara; *Myzus persicae* Sulzer., *Etiella zinckenella* Triet*., Vanessa cardui* L., *Delia platura* Meigen. The obtained data are useful for adapting pest control measures to the integrated protection system.

ACKNOWLEDGEMENTS

The research was carried out with the support of the institutional project – Subprogram 010102, MSU "Al. Ciubotaru" NBGI, as well as the subprogram 010701 Institute of Zoology, MSU.

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