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The usage of beneficial insects as a biological control measure in large-scale farming - a case study review on *Trichogramma* spp.

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The usage of beneficial insects as a biological control measure in large-scale farming - a case study review on *Trichogramma* spp.

Abstract: Large scale crops like maize, soybean, wheat and rice have changed the ecosystems worldwide, causing a major impact on global agricultural diversity. Intensive farming includes wide range of synthetic substances which are very often applied irrationally and excessively. Given the prevalence of large-scale farming in world agriculture, it is necessary to begin the transition from conventional crop protection to integrated pest management (IPM) in these agroecosystems. One of the most important components of IPM are biological control measures with augmentative release of commercially available species of the genus *Trichogramma* Westwood, 1833 (Hymentoptera: Trichogrammatidae) as potentially successful and environmentally friendly methods. Besides *Trichogramma,* many other beneficial organisms are constantly being tested as potential biocontrol agents such as *Chrysopa* spp. (Neuroptera: Chrysopidae) and *Orius* spp. (Hemiptera: Anthocoridae). Minimizing the use of chemicals and replacing them with biological plant protection is fully in line with the agriculture development strategy and confirmed to be achievable in practice. It is especially important to apply such tactical decisions in the production of large-scale crops, which, at the same time, represent the biggest polluters of the environment as well.

Key words: beneficial insects; biological control; Trichogramma spp.; large-scale crops; IPM

Uporaba koristnih žuželk kot merilo biotičnega varstva pri kmetovanju na velikih zemljiščih - pregledna raziskava na primeru parazitoidnih os iz rodu *Trichogramma*

Izvleček: Poljščine, kot so koruza, soja, pšenica in riž, ki se gojijo na velikih obdelovalnih zemljiščih, so globalno spremenile ekosisteme in imajo globalno največji vpliv na raznolikost v kmetijstvu. Intenzivno kmetijstvo uporablja širok spekter sintetičnih snovi, ki so pogosto uporabljene neracionalno in v prevelikem obsegu. Zaradi prevladovanja kmetovanja na velikih zemljiščih v svetovnem merilu je potrebno začeti s prehodom iz konvencionalnega varstva kmetijskih rastlin na integrirano zatiranje škodljivih organizmov (IPM) v agroekosistemih. Med najpomembnejšimi komponentami integriranega varstva rastlin so ukrepi biotičnega zatiranja škodljivcev s sproščanjem komercialno dostopnih vrst parazitoidnih os iz rodu *Trichogramma* Westwood, 1833 (Hymentoptera: Trichogrammatidae) kot potencialno učinkovitih in okolju prijaznih metod. Poleg vrst iz rodu *Trichogramma* se v biotičnem varstvu stalno preiskušajo mnogi drugi koristni organizmi, kot so tenčicarice (*Chrysopa* spp., Neuroptera: Chrysopidae) in plenilske stenice iz rodu *Orius* (Hemiptera: Anthocoridae). Zmanjševanje uporabe kemikalij in njihovo nadomeščanje z biotičnim varstvom rastlin je popolnoma v skladu z razvojno strategijo kmetijstva in je potrjeno lahko doseženo v praksi. Še posebej je pomembno uporabiti te metode v velikopovršinski pridelavi kmetijskih rastlin, ki hkrati predstavlja tudi enega izmed največjih onesnaževalcev okolja.

Ključne besede: koristne žuželke; biotični nadzor; *Trichogramma* spp.**;** veliko površinsko gojene kmetijske rastline; IPM

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1 INTRODUCTION

A combination of high commodity crop prices, rising global food demand and technological advances, has transformed the scale of global crop production (Hochman et al., 2014). Certain crops are becoming more prevalent taking into consideration monoculture mainstreaming in agricultural production globally. These large-scale crops which occupy most of the global agricultural area, have already transformed the land use dynamics and changed the ecosystems worldwide, and are still causing major impact on global agricultural diversity. Wheat (*Triticum* spp.), rice (*Oryza* spp.), corn (*Zea* spp.) and soybean (*Glycine* spp.) are prime examples. These four crops alone occupy approximately 50 % of the world's entire agricultural lands, while the remaining crops cover the rest (Ben Ari & Makowski, 2016).

Large scale farming (or intensive farming) is generally defined as highly mechanized and commercialized cropping activities with much greater use of external inputs (Tittonell et al., 2020). It also includes wide range of synthetic substances (fertilizers, fungicides, insecticides, and herbicides). These agrochemicals, used to prevent crop of diseases, manage the weeds and pests and boost plant growth, are very often applied irrationally and excessively. In such conditions side effects often occur. Inadequate use of pesticides can lead to pests' resistance, excessive pesticide residues in food, pollution of agroecosystems and suppression of beneficial organisms. Among all, these chemical agents are toxic for both wildlife and humans. Additionally, agrochemicals are often associated with reduction of populations of birds, amphibians and insects (bees, butterflies) by destroying their food source, contaminating soil and ground waters (Feshchenko, 2019).

As the volume and production of agriculture continue to grow to new global records, so does the environmental awareness of societies. There is an increasingly pronounced demand for production of high-quality food without pesticide residues and other toxic substances. There is also an ecological issue underlining the use of renewable energy sources and preservation of natural resources and environment. Modern trend of sustainable agricultural production imposes the need to change technological process of production with the application of techniques that pollute the environment less and contribute to the health security in general. Numerous studies and social initiatives are calling for conversion to more sustainable agricultural practices due to their favorable effect on ecosystems, biodiversity and human health (Siebrecht, 2020).

Given the prevalence of large-scale farming in world agriculture, it is necessary to begin the transition from conventional crop protection to integrated pest management (hereinafter: IPM). IPM is based on extremely controlled and justified use of chemical agents with the emphasis on the use of alternative ways for pest control, like biological control measures.

1.1 BIOLOGICAL CONTROL MEASURES

One of the most important components of IPM are biological control measures. Biological control or biocontrol is defined as a set of methods significant for pest control (insects, mites, weeds and plant diseases etc.) using their natural enemies. It relies on predation, parasitism, herbivory, or other natural mechanism, but also involves an active human influence (Flint & Dreistadt, 1998). Since the existing form of natural balance between pests and beneficial organisms is usually insufficient to achieve expected results in intensive farming, biological control requires the manipulation of beneficial insects by people to reduce the population of agricultural pests (Raspudić et al., 1999). In a strict ecological sense, applied biological control can be considered as a strategy to restore functional biodiversity in agroecosystems by adding missing entomophagous insects through classical and/or augmentative biocontrol techniques, but it can also be considered as a way of enhancing naturally occurring predators and parasitoids through conservation and habitat management (Altieri, 1994). Biological control is a self-sustaining strategy through which farmers rely on pest control through ecological services provided by restored functional biodiversity, thus avoiding dependence on costly pesticides (Polanczyk & Pratissoli, 2009).

The aim of this paper is to emphasize wide possibility of implementation of beneficial insects in large crop farming, to point out their great potential in agricultural practice and to discuss their high diversity worldwide.

1.2 TRADITIONAL KNOWLEDGE AND HISTORY OF BIOLOGICAL CONTROL

The use of natural enemies to reduce the impact of pests has a long history. There are antecedent historical events that trace the evolution of some of the fundamental concepts in the development of biological control, and several of these events show the remarkable and perceptive insight of man into the workings of nature (Den Bosch et al., 1982). The first description of use of biological control dates from around 300 AD, when predatory ants were used for control of pests in citrus orchards in China, a method which is still used today in Asia. In the 1750s, the British and French transported mynah birds from India to Mauritius to control locusts. Early applied biological control programs began under the USDA's Department and later Bureau of Entomology established in 1881 (Polanczyk & Pratissoli, 2009).

The first introduction of an exotic braconid wasp parasite, *Cotesia glomerata* (Linnaeus, 1758) (Hymenoptera: Braconidae), against the imported cabbageworm, *Pieris rapae* (Linnaeus, 1758) (Lepidoptera: Pieridae), into the United States occurred in 1883 and the introduction of the famous predaceous vedalia beetle *Rodolia cardinalis* (Mulsant, 1850) (Coleoptera: Coccinellidae) to control the cottony cushion scale *Icerya purchasi* Maskell, 1878 (Hemiptera: Margarodidae), followed in 1888 (Clausen 1978). The Department's first large-scale biological control program did not begin until 1905 and involved explorations in Europe and Japan for natural enemies of the gypsy moth *Lymantria díspar* Linnaeus, 1758 (Lepidoptera: Lymantriidae), and browntail moth, *Euproctis chrysorrhoea* Linnaeus, 1758 (Lepidoptera: Lymantriidae), introduced into New England (Vail et al., 2001). It must be pointed out that from 1930 to 1940 there was a peak in biological control activity in the world with 57 different natural enemies established at various places, but World War II caused a sharp drop in biological control activity, and it did not regain popularity after war due to production of relatively inexpensive synthetic organic insecticides (Polanczyk & Pratissoli, 2009).

Today, there are hundreds of biological control products commercially available for pest control, but not all are sufficiently effective for large-scale farming. One of the potentially most successful and environmentally friendly methods for biological control of pests is the augmentative release of commercially available species of the genus *Trichogramma* Westwood, 1833 (Hymentoptera: Trichogrammatidae) (Stouthamer, 1993). These organisms have been identified and successfully used for inundative biological control of lepidopteran pests for more than 120 years (Smith, 1996; Van Lenteren, 2000). *Trichogramma* are egg parasitoids that attack more than 200 lepidopteran host species, including pest groups of borers, webworms, loopers, leafworms, fruitworms, cutworms, bollworms and armyworms (Knutson, 1998).

More than a thousand scientific papers have been published on *Trichogramma* and its usage as a biological control agent, making it one of the most researched natural enemies in the world. As a result, they have been widely used in inundative and inoculative biological control programs in more than 30 countries in agricultural crops (e.g. corn, cotton, sugarcane, rice, soybean, fruit trees, vegetables) and natural forests (Knutson, 1998).

1.3 Trichogramma spp. LIFE CYCLE

Trichogramma wasps primarily parasitize eggs of moths and butterflies (Lepidoptera). However, certain species of *Trichogramma* also parasitize eggs of beetles (Coleoptera), flies (Diptera), true bugs (Heteroptera), other wasps (Hymenoptera), lacewings and their relatives (Neuroptera) (Knutson, 1998). The adult female wasp uses chemical and visual clues to locate a host egg (Nordlund et al., 1981). Once a female finds a host egg, she drills a hole through the chorion (eggshell) and inserts two to three eggs into the host egg. Eggs of parasitoid hatch in about 24 hours and the parasite larvae develop very quickly. Larvae develop through three instars. During the third instar, dark melanin granules are deposited on the surface of the egg chorion, causing the host egg to turn black. Larvae than transform to the inactive pupal stage. After about 4-5 days, the adult wasps emerge from the pupae and escape the host egg by chewing a circular hole in the eggshell. The black layer inside the chorion and the exit hole are evidence of parasitism by *Trichogramma* (Ruberson et al., 1993).

2 *Trichogramma* IN MAIZE

Maize (*Zea* spp.) is one of the most abundantly produced cereal in the world. It is grown in every continent except Antarctica (Eckhoff et al., 2003). This large-scale crop has many pests and the European corn borer *Ostrinia nubilalis* Hubner, 1796 (Lepidoptera: Crambidae) is considered the major one worldwide (Mutuura & Monroe, 1970). The egg stages of many corn pests including *O. nubilalis*, are attacked by various species of *Trichogramma* (Ivezić & Trudić, 2021). More than 15 species and strains of native and exotic *Trichogramma* were evaluated in field and laboratory tests to determine those with a high preference for *O. nubilalis* egg clusters or other lepidopteran corn pests (Wang et al., 1999). Several species of *Trichogramma* have been identified as promising biological control agents of *O. nubilalis*, including *T. brassicae* Bezdenko, 1968 and *T. evanescens* Westwood, 1833 (Bigler, 1986; Hassan, 1993). Both species appear to be widespread across Europe and reared in commercial facilities for release as bioagents. In South Europe, these two *Trichogramma* species are considered to be the most abundant *Trichogramma* species in maize (Bohinc et al., 2015; Ivezić et al., 2021).

In order to ensure production of effective parasites, mass rearing facilities developed rearing techniques with stringent quality control procedures. This requires controlled environments, artificial diets and ovipositional substrates, mechanized equipment and operations performed by work units (Knutson, 1998). This process selects the strain of *Trichogramma* with the most efficient ability to fly, locate and parasitize the eggs of a targeted host. After selecting the best strain/colony, *Trichogramma* pupae is used to colonize the crop. Pupae can be programmed to enter a condition of arrested development called diapause. Once in diapause, wasp pupae can be stored for up to 9 months so the large demand for *Trichogramma* during the summer can be met (Bigler, 1994). Cardboard capsules containing host eggs with developing *Trichogramma* are applied to the corn field and can be distributed from the ground or air. Capsules either fall to the ground or are caught in the corn plant. The capsules protect the *Trichogramm*a from predators and weather extremes until the adults emerge from the host egg and escape through tiny holes in the capsules. Released *Trichogramma* are at different developmental stages so that adults emerge from the capsules over several days. This increases the time interval between applications (Knutson, 1998).

Releases of *Trichogramma* are performed manually or mechanically (ground and air) (Li, 1994). Since manual applications have shown as time-consuming process, aerial applications are more acceptable option. One such experiment was performed in Poland, where the use of ultralight aircraft proved to be an effective option (Bzowska-Bakalarz et al., 2020). The results indicate that the low-height aerial application allows precise dosing and satisfactory distribution of bioagents. The efficacy of 60–85 % (depending on the year) of the gyroplane-based spraying operations was comparable with those monitored for ground application. These results show a promising alternative for the application of *Trichogramma*, especially in large-scale crops, because the ground application in intensive agriculture requires too much time and energy (Bzowska-Bakalarz et al., 2020).

Other caterpillar pests of corn, such as the southwestern corn borer *Diatraea grandiosella* Diar, 1911 and American cotton bollworm *Helicoverpa zea* Boddie, 1850, are attacked by native or introduced species of *Trichogramma* (*T. pretiosum* Riley, 1879, *T. deion* Pinto and Oatman, 1986, *T. thalense* Pinto and Oatman, 1985) throughout the world (Manandhar & Wright, 2015). Some companies sell *Trichogramma* for control of these pests, but research to support this usage is lacking. At this point, European corn borer is still the most controlled pest of maize with wasps of the genus *Trichogramma*.

3 *Trichogramma* IN RICE

Rice (*Oryza* spp.) is one of the most important crops in the world, being produced in many locations and under a variety of climatic conditions. Since sizable portions of certain crops are used for purposes other than human consumption, rice is the most important food crop, directly feeding more people than any other crop (Rao et al., 2017). Traditionally, countries in Asia have the largest share in world rice production, but this crop is becoming increasingly important in Africa and Latin America. Meanwhile, the expansion of rice crops poses a challenge for agricultural workers as they constantly face many obstacles, such as pests and diseases (Afifah et al., 2019). Key pests of rice are striped rice stemborer *Chilo suppressalis* Walker 1863 (Lepidoptera: Crambidae), yellow stem borer *Scirpophaga incertulas* Walker 1863 (Lepidoptera: Crambidae), pink stem borer *Sesamia inferens* Walker 1856 (Lepidoptera: Noctuidae), rice leafroller *Cnaphalocrocis medinalis* Guenee, 1854 (Lepidoptera: Crambidae), rice planthopper *Nilaparvata lugens* Stal, 1854 (Hemiptera: Delphacidae) and rice green semilooper *Naranga aenescens* Moore, 1881 (Lepidoptera: Noctuidae) (Tang et al., 2017). Among these, yellow stem borer is considered to be the most important pest of rain-fed lowland and flood-prone rice ecosystems (Barthakur, 2010; Ko et al., 2014). Populations of these pests substantially increased within one decade, therefore, the appropriate, effective, and inexpensive control measures are needed for the continuity of high rice production (Gao et al., 2012). Optimized methods with less environmental impact and high sustainability are in demand, such as releasing biological control agents. As one of the most important natural enemies worldwide, the use of *Trichogramma* wasps in rice fields is the subject of constant research (Afifah et al., 2019). Although *Trichogramma* has been studied for management of key lepidopteran pests in rice, these wasps aren't still commercially used in intensive rice production. Recent findings from China indicate that *Trichogramma* releases may be considered practical for control of striped rice stemborer and rice leafroller. However, it is less clear whether yellow stem borer can also be controlled by *Trichogramma* wasps as less studies have been done on this species so far (Tang et al., 2017). From field surveys conducted in Indian rice fields, there are indicators that yellow stem borer eggs may not be effectively parasitized under natural conditions (Hikjm, 1988; Chakraborty, 2012). On the other hand, more positive results have been reported from a field survey in China showing rather high parasitism rates of yellow stem borer eggs in the range of 46.7 % to 79.1 % (Guo et al., 2002; Samara et al., 2008). In general, there have been a very few attempts to control yellow stem borers by inundative releases of *Trichogramma* (Tang et al., 2017). Positive results were obtained from Indonesia where *T. japonicum* Ashmead, 1904 showed the potential to become candidate for control of white rice stem borer (Yunus, 2018), while promising results were also obtained in Egypt in the control of rice stem borer *Chilo agamemnon* Bleszynski, 1962 (Lepidoptera: Crambidae) due to inundative release of *T. evanescens* (Sherif et al., 2008).

There are rich communities of beneficial insects, spiders, and diseases that attack insect pests of rice, but just a few of them are commercially applied. Certain results indicate that the application of *Trichogramma* wasps shows promising results in the control of rice pests, which has consequently aroused great interest among consumers and rice producers. However, for wider commercial application of these organisms in rice production positive results are lacking, and the use of these organisms is mainly done for the purpose of research.

4 *Trichogramma* IN SOYBEAN

The largest producers of soybean (*Glycine* spp.) in the World are Brazil, the USA, Canada, China and Argentina (https://www.fas.usda.gov/commodities/soybeans). There is a significant variety of insects that may be found in soybean fields at any given time of the season, and among them is a large number of different pests, many of which can cause significant yield losses. As in the above-mentioned crops, different species of the genus *Trichogramma* are present in soybean fields, both as native populations and as introduced biocontrol agents (Bueno et al., 2008). Some of these species are important natural enemies of key pests of soybean production such as sunflower looper *Rachiplusia nu* Guenee, 1852 (Lepidoptera: Noctuidae), velvetbean caterpillar *Anticarsia gemmatalis* Hubner, 1818 (Lepidoptera: Erebidae), soybean budborer *Crocidosema aporema* Walsingham, 1914 (Lepidoptera: Tortricidae) and cotton earworm *Helicoverpa armigera* Hubner, 1808 (Lepidoptera: Noctuidae) (Bortolotto et al., 2015).

In Southern Brazil three species of *Trichogramma* spp. are well known and used for controlling the number of velvetbean caterpillar: *T. pretiosum, T. acacioi* Brun, Moraes and Soares, 1984 and *T. rojasi* Nagaraja and Nagarkatti 1973 (Foerster et al., 2015). In Uruguay, six species of *Trichogramma* have been identified from collections of different crops (Basso et al., 2020). Among the reported species, *T. pretiosum* is the most widely distributed in Uruguay and parasitizing a great number of lepidopteran pests (Basso et al., 1999a; Basso et al., 1999b; Basso & Pintureau, 2004). Given the prevalence, in last decades this species was introduced as biological agent in soybean crops (Basso et al., 2020). The selection of *T. pretiosum* was based on the fact that, in the laboratory, it presented the highest fertility parasitizing eggs of *A. gemmatalis* deposited on soybean plants, when compared to *T. exiguum* Pinto and Platner, 1978 and *T. galloi* Zucchi, 1988, species also present in Uruguay (Basso et al., 2020). In this country a multi-year study was conducted to compare conventional practice with different doses of the egg parasitoid. Although the best results were obtained with the application of chemical insecticides, two releases of *T. pretiosum* by terrestrial methods, 20 days apart, or 4 weekly applications by means of a drone, reached the best results below the thresholds of sanitary intervention, both options with 200,000 parasitoids per hectare (Basso et al., 2020). The application of *T. pretiosum* under the inundative biological control method appears as a real alternative to chemical insecticides for the control of the main lepidopteran pests in soybean crops in Uruguay (Basso et al., 2020). These results showed that biological tool such as egg parasitoids of the genus *Trichogramma* can differentiate and value production of soybean. Beside Latin America, *T. pretiosum* is present in almost all biogeographic regions in the world. With 240 host records in the Americas, it is one of the most commonly collected species, especially in agricultural and other disturbed habitats (Pinto, 1998).

The efforts of large soybean producers to implement alternative ways and eco-friendly methods for pest control management indicate that environmental awareness is constantly growing, but also that future production of large- scale crops should be much more based on the use of beneficial insects and biological control measures in general.

5 *Trichogramma* AS A BIOAGENT IN OTHER CROP SPECIES

Besides in large scale crops, augmentation of *Trichogramma* has been promoted for pest control in cotton, apple, spruce, avocado, tomato and potato production (Olkowski & Zhang, 1990). In Europe, *T. evanescens* is widely used for control of codling moth *Cydia pomonella* Linnaeus, 1758 (Lepidoptera: Tortricidae) in apples (Knutson 1998), but also as an effective tool to decrease population of potato tuber moth *Phthori-*

maea operculllea Zeller, 1873 (Lepidoptera: Gelechiidae) (Saour, 2004). Three *Trichogramma* species, *T. cacoeciae* Marchal, 1927, *T. evanescens*, and *T. principium* Sugonjaev and Sorokina, 1976, are proved as effective candidates in parasitizing potato tumber moth eggs (Saour, 2004). In the USA, parasitism of tomato fruit pests (*H. armigera* or tomato leafminer *Tuta absoluta* Meyrick, 1917 (Lepidoptera: Gelechiidae) by native *T. pretiosum* in tomatoes is considered in the treatment thresholds for these pests with insecticides (Hoffman et al., 1990). Augmentation of *T. pretiosum* is an effective control tactic in Mexico and is a part of the integrated pest program for fresh market tomatoes (Trumble & Alvarado-Rodriguez, 1991). In some countries like China, the Philippines, India, and Taiwan, *T. chilonis* Ishii, 1941 is already being used as a biological control agent in sugarcane plantations. In Indonesia (Grieshop et al., 2014), *T. chilonis* was first developed to address the problem of stem borer in several sugarcane plantations in Java which was later introduced to Lampung after similar problems arose (*Afifah et al., 2019*). The results showed that the release of 150,000 eggs *Trichogramma* spp. per hectare could reduce the population of sugarcane shoot borer *Chilo infuscatellus* Snellen, 1890 (Lepidoptera: Crambidae) while 250,000 eggs are required per hectare to control sugarcane stem borer *C. terrenellus* Pagenstecher, 1900 (Lepidoptera: Crambidae) (Cascone et al., 2015). Besides being able to parasitize *Chilo* spp. *T. chilonis* is also capable of parasitizing *Agrotis* spp. (Lepidoptera: Noctuidae), sugarcane gray borer *Tetramorea schistaceana* Snellen, 1891 (Lepidoptera: Tortricidae), rice leafroller *Cnaphalocrosis medinalis* Guenee, 1854 (Lepidoptera: Crambidae), *H. armigera*, soyabean pod borer *Leguminivora glycinivorella* Obraztsov, 1960 (Lepidoptera: Tortricidae) and beet armyworm *Spodoptera exigua* Hubner, 1808 (Lepidoptera: Noctuidae) (Li-Ying, 1994).

In California, two avocado pests, the omnivorous looper *Sabulodes aegrotata* Guenee, 1857 (Lepidoptera: Geometridae) and the avocado leafroller *Amorbia cuneana* Walsingham, 1879 (Lepidoptera: Tortricidae), can be managed by releasing *T. platneri* Nagarkatti, 1975 in every fourth avocado tree (Olkowski & Zhang, 1990). Large field studies in Canada have shown that two releases, each with 30 million *T. minutum* individuals per acre, resulted in 60 to 80 % egg parasitism of spruce budworm *Choristoneura fumiferana* Clemens, 1865 (Lepidoptera: Tortricidae) in white spruce stands (Olkowski & Zhang, 1990).

The actual rates of release vary considerably, even for the same pest, crop, and country. This range is probably related to the range in dimensional volume of the crop. For example, the total rates of release for *T. brassicae* alone, which is reared from small host eggs against European corn borer in Europe, range from 150,000 to 2.8 million wasps/ha (El-Wakeil et al., 2020). Rates

Table 1: Host and country of origin of the *Trichogramma* species and strains (adapted from Tabone et al., 2010)

Species*	Strain	Host and taxonomy*	Country of Origin
Trichogrammatoidea bactrae Nagaraja, 1979	Bac-1	*Plutella xylostella* Linnaeus, 1758 (Lepidoptera: Plutellidae)	Thailand
Trichogramma bourarachae Pintureau and Babaul, 1988	Bou-1	*Vanessa cardui* Linnaeus, 1785 (Lepidoptera: Nymphalidae)	Morocco
Trichogramma bourarachae	Bou-2	*Helicoverpa armigera* (Lepidoptera: Noctuidae)	Portugal
Trichogramma buesi Voegele, 1982	Bue-1	*Ephestia kuehniella* Zeller, 1879 (Lepidoptera: Pyralidae)	Canada
Trichogramma chilonis	Chi-1	Plutella xylostella	Japan
Trichogramma chilonis	Chi-3	Ephestia kuehniella	Taiwan
Trichogramma dendrolimi Matsumara, 1926	Den-1	*Lobesia botrana* Denis and Schiffermu ller, 1775 (Lepidoptera: Tortricidae)	Italy
Trichogramma evanescens	Eva-1	*Pectinophora gossypiella* Saunders, 1884 (Lepidoptera: Gelechiidae)	Egypt
Trichogramma oleae Voegele and Pointe, 1979	Ole-1	*Prays oleae* Bernard, 1794 (Lepidoptera: Plutellidae)	France
Trichogramma ostriniae	Ost-2	Ephestia kuehniella	Moldova
Trichogramma principium Sugonjaev and Sorokina, 1976	Pri-1	*Earias insulana* Boisduval, 1833 (Lepidoptera: Nolidae)	Syria

*Detailed explanation of the species (author and taxonomy) is provided only for those species which are not mentioned in previous text in several millions of wasps/ha are generally cited in arboreal situations such as forestry, and in fruit or nut orchards, whereas those in agricultural crops such as corn, cotton, and tomato, range from 500 to more than 1 million wasps/ha, with averages of 200,000-600,000 wasps/ha (El-Wakeil et al., 2020). China often reports lower rates than other countries, possibly because of the frequent use of large host eggs (Wang, 2013).

When determining the effectiveness of certain *Trichogramma* species in biological control, it was found that the rate of parasitism does not only depend on the selected *Trichogramma* species, but also on the choice of the appropriate strain. Certain strains within the same species may have different laboratory and field performances, or different preferences towards the same or different hosts. Tabone et al. (2010) analyzed the rate of parasitism of different *Trichogramma* species and strains and pointed out significant differences within the diversity of *Trichogramma* species in certain regions and on different hosts (Table 1.). These results very effectively represent the preference of *Trichogramma* parasitoids for Lepidoptera, both among different species and among different strains within the same *Trichogramma* species.

6 GENERALIST PREDATORS

The scientific community uses all available resources in order to explore the natural potential of entomofauna and introduce less-toxic solutions in pest management programs. Many other beneficial organisms are constantly being tested as potential biocontrol agents such as species of the genus *Chrysopa* spp. (Neuroptera: Chrysopidae) and *Orius* spp. (Hemiptera: Anthocoridae).

Chrysopids commonly known as lacewings, occur in numerous agricultural and horticultural zones of the northern hemisphere. Adults are free-living and usually non-predatory in nature, surviving on nectar and pollen, while three larval stages are highly predatory (Bellows & Fisher, 1999). They are active predators of a wide variety of pests including aphids, chinch bugs, mealybugs, scales, whiteflies, leafhoppers, lepidopterous eggs and larvae, and mites (Principi & Canard, 1984). The efficacy in biological control of aphids as well as other arthropod pests has been recognized for more than 250 years (Dhandapani et al., 2016).

Inundative releases of the common green lacewing *Chrysoperla carnea* Stephens, 1836 (Neuroptera: Chrysopidae) on cotton provided effective results in control of American cotton bollworm *Helicoverpa zea* Boddie, 1850 (Lepidoptera: Noctuidae) and tobacco budworm *Heliotihis virescens* Fabricius, 1777 (Lepidoptera: Noctuidae) (Ridgway & Jones, 1969). Releases of *C. carnea* eggs in field cages at rates of 50,000 and 100,000 per acre can significantly reduce the population of tobacco budworm and increase the yield (Ridgway & Jones, 1969). Green lacewing has also been effective on potato aphid *Macrosiphum euphorbie* Thomas, 1878 (Hemiptera: Aphididae) and buckthorn aphid *Aphis nastrurtii* Kaltenbach, 1843 (Hemiptera: Aphididae) (Capinera, 2001). A species that is very common in corn fields is *Chrysoperla oculata* Ruzicka, 1997. The most suitable prey for *C. oculata* in corn fields is corn leaf aphid *Rhopalosiphum maidis* Fitch, 1856 (Hemiptera: Aphididae) an important pest of corn (Bellows & Fisher, 1999). Many species of the genus *Chrysopa*, such as red-lipped green lacewing *C. rufilabris* Burmeister, 1839, *C. externa* Hagen, 1861 and *C. perla* Linnaeus, 1758 are important predators and are used as biological control agents worldwide. Among the above-mentioned species, most attempts have evaluated the efficacy of *C. carnea* in augmentation releases in the field or in the greenhouses (Ridgway & Mcmuphy, 1984; Nordlund et al., 2001).

In the context of biological control, the genus *Orius* includes several species that have found their place in commercial pest control in agriculture. This genus is represented by very tiny true bugs commonly known as minute pirate bugs and flower bugs (Riudavets & Castane, 1994). They play a key role in the management of various agricultural pests in greenhouse and field environments. They can be found in numerous crops, pastureland and surrounding areas (cotton, soybean, bean, potato, wheat, alfalfa, maize, orchards, other vegetables and ornamental crops), as well as in trees, shrubs, weeds and many wild plants. They prey on thrips, aphids, mites, whiteflies, moths and other tiny arthropods and insect eggs. *Orius* are very effective predators and can thus provide biological pest control in a variety of cropping systems (Brust & Yurchak, 2021).

The species insidious flower bug *Orius insidiosus* Say, 1832 (Hemiptera: Anthocoridea) is one of the most important predators in corn field. The ability of *O. insidiosus* to search, find and destroy European corn borer and corn earworm eggs was investigated in numerous studies. *O. insidiosus* is an important natural enemy of corn earworm in corn, cotton and sorghum. A study conducted in the USA, revealed European corn borer larvae sustain high mortality in field corn and that *O. insidiosus* was the most important predator of these larvae in western Maryland (Brust & Yurchak, 2021). Their population peak coincides with corn pollen-shedding and sulking, during which they feed on second-generation of European corn borer larvae and corn pollen. Therefore, successful biological control of European corn borer larvae by *O. insidiosus* is linked to arthropod prey and corn pollen (Brust & Yurchak, 2021).

Orius insidiosus adults and nymphs are common in soybean fields. Its population dynamics in soybean fields have been linked to thrips population levels and soybean flowering. Nymphs and adults eat soybean aphids in the field. Experimental findings suggest that under certain conditions, *O. insidiosus* can effectively suppress aphid population growth and that they may be key factors influencing aphid population dynamics in soybeans in some areas within the USA. In addition to soybean aphids, soybean thrips are believed to be one of the most important thrips prey of *O. insidiosus* in soybean. It is believed that soybean thrips serve as an important prey resource for *O. insidiosus* in soybeans and may be important in sustaining *O. insidiosus* populations before the arrival of soybean aphids. *O. insidiosus* is known to feed on eggs and first instar of green cloverworm *Hypena scabra* Fabricius, 1789 (Lepidoptera: Erebidae) as well (Brust &Yurchak, 2021).

Rice predators include spiders, ants, some insect families such as Carabidae, plant bugs, amphibians, dragonflies and other beetles and water bugs. However, the most abundant ones are the spiders (Wopereis et al., 2008). Their ability to hunt in a variety of habitats in combination with high abundance, positions spiders as potentially effective biocontrol agents (Symondson et al., 2002). Dispersal by running and ballooning allows spiders to colonize agricultural fields soon after disturbance due to agricultural practices such as ploughing and seed sowing (Radermacher et al., 2020). This applies in particular to agricultural systems with multiple cropping cycles per year and asynchronous planting practice (Marc et al., 1999). In fact, spiders are among the most abundant arthropod predators in rice ecosystems and assumed to contribute to the control of pest species such as plant and leafhoppers (Sigsgaard, 2007). With the ability to capture prey of different feeding guilds, including herbivores and detritivores, spiders may play an important role soon after planting rice fields when herbivore populations still are low (Radermacher et al., 2020). Generalist predators in agricultural systems such as spiders may link aboveground herbivore and belowground detrital systems using prey of both systems (Scheu, 2001; Snyder & Wise, 2001; Wise et al., 2006).

7 CONCLUSIONS

Although the use of beneficial insects may seem simple at first, effective pest biocontrol is determined by many factors. Those factors are adequate selection of parasites and predators, the quality and fitness of used bioproduct, the numbers released and the timing of the release, the release method, complex interactions between the parasite/predator, the target pest, the crop and environmental conditions (Knutson, 1998). However, a fundamental step in the development of any biological control program utilizing beneficial insects is the identification and choice of species and/or strains to use; not all species (or populations) perform equally well, in terms of mass rearing or field dispersal and performance (Ivezić et al., 2018). Therefore, the first step for the implementation of beneficial insects in the program of biological control of pests is the accurate identification and genetic characterization of native species, since autochthonous species are likely to be best adapted to environmental conditions in a specific ecosystem (Whitman & Nordlund, 1994).

In addition to native populations of beneficial insects, biological control also involves the introduction of natural enemies that are not typical of certain areas. The introduction of natural enemies is used when a pest of exotic origin is the target of the biocontrol program (White, 2019). Pests are constantly being imported into countries where they are not native, either accidentally, or in some cases, intentionally. Many of these introductions do not result in establishment or if they do, the organism may not become pest. However, it is not uncommon for some of these introduced organisms to become pests due to a lack of natural enemies to suppress their population number. In these cases, introduction of natural enemies can be highly effective. Once the country of origin of the pest is determined, exploration in the native region can be conducted to search for promising natural enemies (White, 2019). If such enemies are identified, they may be evaluated for potential impact on the pest organism in the native country or alternatively, introduced into the new country for further study. They first need to be placed in a quarantine for one or more generations to be sure that no undesirable species are accidentally imported (diseases, hyperparasitoids etc.). Additional permits are required for interstate shipment and field release (White, 2019).

Besides inundative and inonculative activities for commercial usage of beneficial insects, it is also necessary to preserve the autochthonous residential species populations. Conservation of habitats and preservation of biodiversity as one of the main prerequisites for successful implementation of biological control strategies can be defined as an identification and modification of human influence that allows natural enemies to express their potential to suppress pests (Rechcigl & Rechcigl, 2020). Numerous studies and experience have shown that conserving natural enemies is of tremendous importance in the safe and economical management of insect pests and doing so has to be a major component of a producer's management activities (Rechcigl & Rechcigl, 2020). Likewise, the most important component of biological control is establishing the list of indigenous species of organisms for biological control, which includes only those beneficial species that are indigenous or ubiquitous in specific region. Such an approach has been done in Slovenia and represents an additional safeguard that practically prevents the use of beneficent species that could in any way endanger the common domestic flora and fauna (Trdan et al., 2020).

Over the last decades large scale farming led to increasing production, but also caused substantial environmental degradation as such increases were mostly based on its expansion onto natural areas and greater use of external inputs and other forms of intensified use (IPBES, 2019). Achieving sustainability of agricultural production is one of the key challenges for humanity. Minimizing the use of chemicals and replacing them with biological plant protection is firstly fully in line with the agriculture's development strategy and secondly, confirmed to be achievable in practice. It is especially important to apply such tactical decisions in the production of large-scale crops, which, at the same time, represent the biggest polluters of the environment in general. The use of beneficial insects in biological control is common worldwide but its potential has not been explored for many pests and in many geographic regions. Even in Europe certain countries with very intensive agriculture do not currently use the full potential of these organisms in their agroecological systems. In addition, a recent survey found that very few governmental Extension Services currently provide recommendations for controlling pests with *Trichogramma* spp. or any other beneficial insects (Ivezić & Trudić, 2021). The successful use of augmentative releases of biocontrol agents in pest management programs will depend on a sound and thorough research program, favorable economics, commercial investment, and the development of an extension program to transfer this technology to crop consultants and large scale growers. Mainstreaming agroecology and its biological control alternatives among large scale farmers is urgently needed, but it requires addressing specific questions in research, technology and policy development to support sustainable transitions.

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