Biological control of pests in Ukraine: legacy from the past and challenges for the future

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Abstract

Biological control has a long and rich history in Ukraine which is closely linked with other countries of the Former Soviet Union, and some relevant studies from these countries are also included in this review. The use of natural enemies against Ukrainian insect pests demonstrates that biological control approaches have enjoyed a degree of success. The release of *Aphelinus mali* in a classical biological control programme against the woolly apple aphid, *Eriosoma lanigerum*, was successful. In contrast, the release of the hemipterans, *Perillus bioculatus* and *Podisus maculiventris*, against the Colorado potato beetle, *Leptinotarsa decemlineata*, was not successful, and studies in the use of these hemipterans as biological control agents continue. Conservation biological control is practised in apple and cereal cropping systems resulting in a number of predators and parasitoids being preserved. Augmentation of natural enemies, especially predatory mites against the spider mite *Tetranychus urticae* in greenhouse cucumber and tomato production, has provided suppression of this pest. Various strains of *Bacillus thuringiensis* are being used inundatively against lepidopteran, coleopteran and mosquito pests. A granulovirus has been studied for use against the codling moth (*Cydia pomonella*), and the fungus, *Beauveria bassiana*, is being evaluated against several insect pests. Entomopathogenic nematodes have generated some interest for future use in Ukraine as potential biological control agents against soil-inhabiting pests. Although biological control programmes have been practised for many years, the agricultural sector in Ukraine is moving from a command to a market economy. The latter economy is profit-driven and relies more on chemical pesticide usage. The challenge is to integrate biological control programmes into the market economy.

Keywords: Entomopathogens, Entomophagous insects, Natural enemies, Parasitoids, Pathogens, Predators

Introduction

Ukraine has an extensive and varied history in applied biological control in which natural enemies have been used for the regulation of host population densities [1]. Biological control including classical biological control, augmentation, and conservation, has been practised in Ukraine [2, 3], generally quite successfully. There are a number of reasons for the high interest in biological control including the relatively low labour cost, excellent facilities for mass rearing of beneficial organisms, the existence of qualified experts, recognition by local and state officials of the importance of biological control and integrated pest management (IPM) approaches, and the high cost of imported synthetic pesticides. Moreover, increasing awareness among the public about the negative impact of chemical pesticides on the environment and human health creates a favourable atmosphere for the further development of biocontrol programmes.

Ukraine is uniquely positioned to produce biocontrol products including entomophagous insects and entomopathogenic microorganisms at a local level at a much lower cost than for importing them. In this paper, the past and present status of biological control programmes in

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Ukraine and, to some extent, relevant publications from other countries of the Former Soviet Union as well as from Europe and North America are reviewed. However, some of the most extensively researched and utilized biological control agents are Trichogramma spp. which are only briefly covered in this review. A future review will cover these important biological control agents.

As we look to the future, the use of biological control technology, in large part, will be tied to the shift in the Ukrainian agricultural economy. This aspect is also covered in this review.

**Historical overview**

The first documentation of successful biological control in the field was in the 19th century in Ukraine when Ilya Metchnikoff, Professor of Microbiology at the University of Odessa (southern Ukraine), demonstrated that the green muscardine fungus, *Metarhizium anisopliae* (Metschn.) Sorokin (Hypocreales, Clavicipitaceae), could be used to control the wheat cockchafer, *Anisoplia austriaca* (Herbst) (Col., Scarabaeidae) [4]. Using beer mash as a medium, he produced the fungus, mixed the conidia with soil and placed healthy larvae of the wheat cockchafer into the soil–conidia mixture. Ten days later, he evaluated his experiment and found that eight out of nine larvae had died from a fungal infection. Thus, he is credited with establishing the concept that economically important insects could be intentionally killed by a microorganism. Subsequently, he conducted a similar experiment against another pest, the sugar beet curculio, *Cleonus punctiventris* (Germar) [4]. Shortly thereafter, I. M. Krasiltschik [5] at the University of Odessa utilized Metchnikoff's methods and established a special laboratory to produce conidia of this fungus on a large scale. *Metarhizium anisopliae* was mass produced in Ukraine for use against larvae of the sugar beet curculio.

In 1913, A. Mokerzetskiy in Crimea and V. Pospelov in the Kiev region studied the possibilities of using *Trichogramma* spp. to control the codling moth, *Cydia pomonella* (L.) (Lep., Tortricidae). In the late 1920s and early 1930s, several entomophagous insects were introduced for classical biological control of exotic pests. In 1930, the Institute of Crop Protection in Leningrad was established and recognized as the main research arm for biological methods for plant protection. In 1936, the Institute of Crop Protection and Phytopathology in Ukraine. With his move, the research focus of the institute shifted to insect pathology and diseases of beneficial insects [8]. Biological control programmes in Ukraine suffered a major set back with the growing use of synthetic organic chemical pesticides in the 1950s. This situation was not unique to Ukraine as many other countries also began to rely heavily on chemical pest control.

In Ukraine, biological control approaches re-emerged in the late 1950s–early 1960s. This was precipitated by the need to find alternatives to chemical pesticides because of the growing public concern about their use. In 1957, the Institute of Plant Protection (formerly the Institute of Entomology and Phytopathology) created the Department of Microbiological Plant Protection. The main research focus of this group was on the white muscardine fungus, *Beauveria bassiana* (Bals.-Criv.) Vuill. (Hypocreales, Clavicipitaceae). Promising results were obtained with this fungus in combination with insecticides to control the Colorado potato beetle, *Leptinotarsa decemlineata* (Say) (Col., Chrysomelidae). Telnaga [9, 10] developed a method to combine reduced amounts of insecticides with *B. bassiana* that gave good results in controlling the Colorado potato beetle, codling moth and other insects. Research conducted by this group resulted in several published articles on the ecological basis for using predators, parasitoids, and pathogens [11–15].

The Ukrainian government was a primary force in the re-emergence of biocontrol programmes in the 1960s. It issued decrees and orders that encouraged the use of biocontrol agents. A Coordinating Committee on the use of biological methods for plant protection was formed in 1966. This committee consisted of scholars from ten universities and eight research institutions who developed the plans and guidelines for biological control research and its practical application. The biological control approaches became important resulting in the establishment of 13 companies that focused on mass rearing of microbial pathogens, and 268 insectaries for rearing beneficial species, producing parasitoids and predators as well as microbial pathogens of insects and plant diseases in the 1970s. Accordingly, biocontrol agents were used on 1.7 million ha during the 1980s, with the egg parasitoids, *Trichogramma* spp., representing 80% of the agents used. However, the application of synthetic organic chemical pesticides during the 1970s and 1980s was not insignificant with a peak rate of 5.5 kg/ha reported in 1986. Overall, the use of biological control products was considerable during this period, and this lasted until 1991 when the Ukrainian economy started to decline. During a short period beginning in 1991, 40% of the beneficial insectaries were lost. With the reduction in releases of *Trichogramma* parasitoids, outbreaks of lepidopteran...
pests, especially the European corn borer, *Pyrausta nubilalis* (Hu¨bner) [Ostrinia nubilalis] (Lep., Pyralidae), occurred on maize. By 1999, biocontrol products were used on 0.9 million ha and only about 90 small, local beneficial insectaries remained to produce biological control agents. In present day Ukraine, *Trichogramma* spp. are the most commonly used biocontrol agents against a number of lepidopteran pests [16, 17]. They are produced in small, local laboratories on a very limited scale.

### Classical biological control with predators and parasitoids

The development of classical biocontrol has been fostered through the introduction of a number of natural enemies to target exotic pests in Ukraine and Russia (Table 1). Classical biocontrol has been employed extensively since the 1920s, and some programmes have been quite successful. Some 26 natural enemies have been successfully introduced and established within the Former Soviet Union countries against 12 exotic arthropod pests. Generally, one in five introductions has resulted in establishment and successful pest suppression (Table 1) [18–20]. However, there have been several projects where the natural enemies did not become established in the release areas because of poor climatic or habitat adaptation or the absence of a host. For example, this occurred with the introduction of predatory pentatomids against the Colorado potato beetle.

### Woolly apple aphid

The woolly apple aphid, *Eriosoma lanigerum* (Hausmann) (Hem., Aphididae), was first detected in the southern part of the Ukraine’s Crimean Peninsula in the early 1920s on apple trees. Subsequently, it was found in the Odessa region in 1926. A classical biological control programme was initiated in 1930 with the importation of the parasitoid *Aphelinus mali* from South America by Meyer [21], which was released in the Crimean region. The parasitoid kept the woolly apple aphid under effective biological control for nearly 40 years as it proved to be a very efficient parasitoid (giving 95–98% parasitism). It has 6–9 generations per year and high fecundity (100 eggs per female). In 1966, however, apple orchards in the Crimean Peninsula experienced significant increases of this aphid pest with reports of several outbreaks. Only 25–30% of the aphid population was parasitized by the parasitoid. The outbreaks were attributed to the intensive use of pesticides which killed beneficial arthropods. By collecting *A. mali*

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**Table 1** Successfully introduced natural enemy(ies) (i.e. classical biological control agents) and their target pest(s)

<table>
<thead>
<tr>
<th>Date</th>
<th>Natural enemy species (Order, Family)</th>
<th>Exotic pest species (Order, Family)</th>
<th>Region/country</th>
</tr>
</thead>
<tbody>
<tr>
<td>1926</td>
<td><em>Aphelinus mali</em> (Haldeman) (Hym., Aphelinidae)</td>
<td><em>Eriosoma lanigerum</em> (Hausmann) (Hem., Aphididae)</td>
<td>Crimean Ukraine</td>
</tr>
<tr>
<td>1931</td>
<td><em>Rodolia cardinalis</em> (Mulsant) (Col., Coccinellidae)</td>
<td><em>Icerya purchasi</em> Maskell (Hem., Margarodidae)</td>
<td>Georgia</td>
</tr>
<tr>
<td>1933</td>
<td><em>Cryptolaemus montrouzieri</em> Mulsant (Col., Coccinellidae)</td>
<td><em>Pseudococcus gahani</em> Green [P. calceolariae (Maskell)] (Hem., Pseudococcidae), <em>Pulvinaria auranti</em> Cockerell [Chloropulvinaria auranti] (Hem., Coccidae), <em>Pulvinaria floccifera</em> Westwood [Chloropulvinaria floccifera] (Hem., Coccidae)</td>
<td>Crimean Ukraine, Georgia</td>
</tr>
<tr>
<td>1947</td>
<td><em>Lindorus lophanthae</em> (Blaisdell) [Rhyzobius lophanthae] (Col., Coccinellidae)</td>
<td><em>Chrysomphalus dictyospermi</em> (Morgan) (Hem., Diaspididae)</td>
<td>Georgia</td>
</tr>
<tr>
<td>1947</td>
<td><em>Pseudaphycus malinus</em> Gahan (Hym., Encyrtidae)</td>
<td><em>Pseudococcus comstocki</em> (Kuwana) (Hem., Pseudococcidae)</td>
<td>Trans Caucasus and Middle East, Republics of Former Soviet Union</td>
</tr>
<tr>
<td>1960</td>
<td><em>Coccophagus gurneyi</em> Compère (Hym., Aphelelinidae)</td>
<td><em>Pseudococcus gahani</em> [P. calceolariae] (Hem., Pseudococcidae)</td>
<td>Georgia</td>
</tr>
<tr>
<td>1960</td>
<td><em>Encarsia formosa</em> Gahan (Hym., Encyrtidae)</td>
<td><em>Trialeurodes vaporariorum</em> (Westwood) (Hem., Aleyrodidae)</td>
<td>Ukraine</td>
</tr>
<tr>
<td>1962</td>
<td><em>Allotropa burrelli</em> Muesebeck A. convexifrons Muesebeck (Hym., Platygasteridae)</td>
<td><em>Pseudococcus comstocki</em></td>
<td>Caucasus, Middle East, Republics of Former Soviet Union</td>
</tr>
<tr>
<td>1967</td>
<td><em>Macrocentrus ancylovorus</em> Rohwer (Hym., Braconidae)</td>
<td><em>Grapholita molesta</em> (Busck) (Lep., Tortricidae)</td>
<td>Russia, Ukraine</td>
</tr>
</tbody>
</table>
from areas where it still occurred and releasing it into outbreak populations, the woolly apple aphid outbreak populations were reduced to non-damaging levels [22–24].

**Colorado potato beetle**

The Colorado potato beetle, *Leptinotarsa decemlineata*, invaded Europe in 1877 from the USA where it fed on a number of solanaceous plants, especially cultivated potato. It spread into Ukraine during 1958–1959 and currently is the key pest of potatoes, often causing as much as 66% reduction in crop yield [25]. This pest is a voracious foliage feeder, has a high fecundity, and has 1–2 generations per year [25]. It has a remarkable ability for developing resistance to a great number of chemical insecticides, including organophosphates and pyrethroids [26, 27]. The development of resistance to pyrethroids, for example, can occur in 8–15 generations [28]. Moreover, because the majority of chemical insecticides used on potatoes are not selective, the pest’s natural enemies are killed, causing further outbreaks.

In its native home in North America, a number of arthropods are known to attack this pest and some of them show good potential as biocontrol agents [29]. Carabid beetles appear to be particularly abundant. In the USA, the adult carabid beetle, *Lebia grandis* Hentz (Col., Carabidae), feeds on the Colorado potato beetle eggs and larvae [30]. Even in the Former Soviet Union, 14 species of carabids and three species of coccinellids are known to feed on Colorado potato beetle [31]. However, their overall effectiveness at keeping this pest under natural biological control is not known.

Gusev [31] indicated that natural enemies can maintain the population of the Colorado potato beetle below the economic threshold level. For example, two predaceous pentatomid bugs, *Perillus bioculatus* (F.) and *Podisus maculiventris* (Say) have had significant impact on the Colorado potato beetle in Canada. Inundative releases of these pre-dators suppressed the beetle density by 62%, reduced defoliation by 86% and increased potato yields by 56% [20].

*Perillus bioculatus* was introduced in 1964 and *Podisus maculiventris* in 1978 from Canada into Ukraine as classical biological control agents against this beetle pest. The introductions provided an opportunity to study these two pentatomid predators under Ukrainian conditions. Studies on the biology, population dynamic and host–pest interaction were initiated with *Perillus bioculatus* in western Ukraine [32–35] which showed that embryonic development continued up to 14 days and the optimal temperature for hatching was 25–30°C. Fecundity also depends on temperature with females laying an average of 350 eggs at 30°C, 214 eggs at 25°C, and 43 eggs at 32°C. The high predatory potential of *P. bioculatus* can be explained by its enormous feeding rate as it feeds on nearly all stages of the beetle. However, feeding on the adult beetle pests appears to be uncommon and restricted primarily to the adult predator. A single nymph predator may consume up to 268 eggs. Survival of *P. bioculatus* instars also depends on the host stage. That is, 72% of the last instars survived when fed only on eggs, 36% on host larvae, and only 24% on adults. Fecundity of the adult predators depends on the host stage they consumed. Thus, predaceous females feeding on beetle eggs, larvae, or adults have a fecundity of 490, 208.5, and 161 eggs per female, respectively. In southwestern Ukraine, this predator has 2–3 generations/year, and its fecundity under natural conditions during the summer approximates to what has been observed in the laboratory. In addition, several studies have shown that the most critical issue for establishment of *P. bioculatus* is overwintering, and specifically its ability to hibernate in colder areas of Ukraine. Up to 70% of the predators can hibernate in an unheated laboratory, whereas in the field, 20–25% could hibernate successfully in the forest litter, and only 5–7% could do so in orchards and gardens [36–41].

*Podisus maculiventris* has been introduced not only to Ukraine, but also to Russia, Moldova and some other countries of the Former Soviet Union [42]. This predator can be reared on a number of hosts including *Galleria mellonella* (L.) and *Anagasta kuehniella* (Zeller) [*Ephestia kuehniella*] (Lep., Pyralidae), *Musca domestica* L. (Dipt., Muscidae), and *Sitotroga cerealella* (Olivier) (Lep., Gelechiidae) [43], and on artificial media [44–50]. Furthermore, laboratory colonies of this predator have been maintained successfully for many years [29]. Studies on its field release and establishment have been carried out since 1974. The high efficiency of this predator was observed when it was released in the field at a predator–prey ratio of 1:5. In Moldova, releases of 2nd and 3rd instars of *P. maculiventris* at a predator–prey ratio of 1:10 to 1:30 reduced the numbers of Colorado potato beetle by 60–80% with a significant yield increase to 14 800 kg/ha in experimental treatments versus 6580 kg/ha in the controls [42]. Inundative releases of this predator in southern Russia at a rate of 100 000 predators per hectare provided 91–100% efficacy [51, 52]. Very good results were also obtained in Moldova with field releases of *Perillus bioculatus* in experimental plots of egg plants [aubergines] and potatoes. Despite some successful projects which were conducted in the warm regions of Moldova and southern Russia, attempts to establish *P. bioculatus* and *Podisus maculiventris* as classical biological control agents in the colder areas of Ukraine have proven to be unsuccessful because of limitations in their ability to overwinter and difficulties in getting the agents established in agricultural fields [52]. Currently, the Colorado potato beetle is not under biological control, but studies on its natural enemies are still being conducted to determine whether they can be established in Ukraine. It was shown recently that *P. maculiventris* has a high efficiency against lepidopteran species and can be used in IPM programmes to control pests from that family [53].
Conservation and enhancement

Apple orchards

A diverse community of arthropod pests is associated with apple orchards. Fifty economically important pests of apple occur in orchards within the different regions in Ukraine. Tortricid moths including *Archips* spp., *Pandemis cerasana* (Hübner), *Adoxophyes orana* (Fischer von Rösslerstamm), *Hedyia nubicera* (Haworth) [H. dimidiaalba Retzius], *Anylis ochhatana* Denis & Schiffermüller and codling moth (*Cydia pomonella*), aphids, spider mites, apple blossom weevil (*Anthonomus pomorum* (L.); Col., Curculionidae) and apple sawfly (*Haplocampa testudinea* (Klug); Hym., Tenthredinidae) are the most common pests [54]. Most pest species are sensitive to insecticides and can be eliminated with a single pesticide application. Other species, especially the codling moth and the apple blossom weevil, have developed resistance to organophosphates and pyrethroid pesticides [55]. Fortunately, apple orchards are excellent habitats that provide refuge for a wide range of natural enemies, including parasitoids, microbial pathogens and nematodes [56]. In Ukraine, 1110 species of entomophagous insects, including about 600 species of parasitoids and 500 predators, have been recorded from unsprayed apple orchards [57, 58]. These natural enemies can play an important role in reducing pest numbers in the orchards [59–61].

Research has shown that in apple orchards not treated with pesticides, predatory mites attain high populations and can control the phytophagous mite populations below their economic injury level. However, the application of insecticides in these same orchards resulted in phytophagous spider mite numbers tripling, and under this situation, the predatory mites cannot control them [59–61].

Parasitoids are the largest and most important group of natural enemies of apple pests. Most of the parasitoids belong to the Hymenoptera. Some 50 parasitoids, mainly Hymenoptera and a few Diptera, attack various stages of the tortricid moths in temperate Europe and 30 species have been recorded in Ukraine [60, 61]. They can have significant impacts on the pest resulting in population reduction.

One of the most damaging pests of apples is the codling moth. Parasitoid research on this moth has been undertaken primarily during the past 40 years. Twenty-six entomophagous insects of codling moth have been recorded from Ukrainian orchards and the level of parasitism by various parasitoids varies from 3.5 to 9.3%. The most common parasitoids of the codling moth are *Trichogramma pomonellae* (Schnabl & Mokrzecki) (Dipt., Tachinidae), *Microdus rufipes* Nees [Bassus rufipes] and *Ascochaster quadridentatus* Wesmael (Hym., Braconidae), *Pristomerus vulnerator* (Panzer), *Pimpla turionella* L. [P. contemplator] (O.F. Müller) and *Trichogramma neecator* (Rossi) (Hym., Ichneumonidae), and *Trichogramma embryophagum* (Hartig) (Hym., Trichogrammatidae). Data about the biology, host range and potential for their use against the codling moth have been obtained [54–56].

Eighty species of parasitoids belonging to the Hymenoptera and 16 species belonging to the Diptera have been recorded from the apple moth, *Yponomeuta malinellus* Zeller (Lep., Yponomeutidae) and 56 species are parasitoids of the gypsy moth, *Lymantria dispar* (L.) (Lep., Lymantriidae) [55]. For the gypsy moth, the egg parasitoid, *Anastatus japonicus* Ashmead (Hym., Eupelmidae) has been recorded, while the larval parasitoids include *Apanteles liparidis* (Bouché) [Glyphaptapentes liparidis], *A. solitarius* (Ratzburg) and *A. porthealia* (Muesebeck) [Glyphaptapentes porthealia] (Hym., Braconidae). In addition, more than 100 natural enemies of the tent caterpillar, *Malacosoma neostrusia* (L.) (Lep., Lasiocampidae), have been identified including *Telenomus leeviusculus* (Ratzburg) (Hym., Scelionidae), *Ooencyrtus tardus* (Ratzburg) (Hym., Encyrtidae), and *A. liparidis* [3].

A conservation and enhancement programme showed that flowering plants attracted a large number of natural enemies of foliage-feeding lepidopteran pests. Thus, flowering plants enhanced the numbers of *Phryxe vulgaris* (Fallén) and *Compsilura concinnata* (Meigen) (Dipt., Tachinidae), and *Apechhis compuncctor* (L.) and *Pimpla instigator* (F.) [P. hypochondriaca] (Retzius)] (Hym., Ichneumonidae). Tkachev reported that between 52 and 90% of *Aporia crataegi* (L.) (Lep., Pieridae) larvae were attacked by these parasitoids [3, 55].

Several predacious mites have been recorded attacking the European red mite, *Panonychus ulmi* (Koch), the two-spotted spider mite *Tetranychus urticae* Koch, and *Amphitetranychus viennensis* (Zacher) (Acari, Tetranychidae) [62]. Of 100 phytoseiid predators, there are 30 species in the apple orchards of which 5–6 species are most common. Predatory mites play an important role in decreasing phytophagous mite populations. Some excellent laboratory and field studies have been conducted on *Typhlodromus abbrans* Oudemans [Kampimodromus abbrans] (Acari, Phytoseiidae) as a predator of the apple rust mite (*Aculus schlechtendali* (Nalepa); Acari, Eriophyidae). These studies described the basic biology, distribution, ecology, feeding behaviour, and fluctuations in populations of the phytoseiid in apple orchards. The population of the phytophagous mites can be controlled at or near the economic threshold level when the ratio between predators such as *Typhlodromus* spp., *Amblyseius* spp. and *Metaseiulus* spp. and the prey is 1:50 [62–64].

Cereals

Cereal crops are attacked by several pest species such as cereal flies (Dipt., Cecidomyiidae, Chloropidae), white grubs (Col., Scarabaeidae), click beetles (Col., Elateridae), cutworms (Lep., Noctuidae), flea beetles (Col., Chrysomelidae), stink bugs (Hem., Scutelleridae), thrips

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(Thysan., Phleothripidae) and aphids (Hem., Aphididae). In examining the aphid pests, there are at least 12 species that attack cereals with the most common ones being *Stobion avenae* (F.), *Schizaphis graminum* (Rondani), *Brachycorpus noxius* Mordvilko [Diuraphis noxia] and *Rhopalosiphum padi* (L.). In Ukraine, aphids may have 10–15 generations during the growing season, and during outbreak years they can reduce yield by 10–15%. The economic threshold for aphids is 10–15 individuals per plant. Fortunately, aphids are hosts to a wide range of natural enemies in the Former Soviet Union. More than 70 specialized and 60 polyphagous species have been reported to feed on aphids [65–69]. The main natural enemies attacking aphids are given in Table 2.

In addition, 16 species of spiders in the families Araneidae, Salticidae, and Linyphiidae, and seven species of Entomophthorales fungi are known to attack aphids. If the ratio between aphids and predators or parasitoids is at 20:1 or if 50% of the aphids in a colony are infected by fungi, there is no need to apply chemical insecticides. To enhance aphid predators and parasitoids, it is necessary to use ‘wild insectaries’ which provide plant diversity. These wild insectaries, for example planting rape with winter wheat, significantly increased the number of aphids parasitized [3].

### Augmentation in greenhouses

Greenhouses provide an excellent environment for inoculative or inundative releases of predators or parasitoids. Accordingly, biocontrol of pests in greenhouses was initiated in 1963 when the predatory phytoseiid mite, *Phytoseiulus persimilis* Athias-Henriot (Acari, Phytoseiidae) from Canada was inoculatively introduced to control the spider mite, *Tetranychus urticae*, in cucumber and tomatoes [70–72] providing season long control. These results encouraged both fundamental and applied research on *P. persimilis* for intensive use in greenhouses. Fundamental research included the taxonomy [73, 74], functional morphology, ecology (with particular reference to levels of predation, fecundity, population dynamics and growth), inter-specific interactions, and geographical distribution [74, 75].

On the applied aspect, a number of studies have been conducted on rearing and to develop practical recommendations for releasing *P. persimilis* in greenhouses against the spider mite. Greenhouse and laboratory studies have covered daily activity, fecundity, behaviour, feeding on different diets, and fluctuations in population densities [76–88]. Since the first introduction of this predatory mite, different types of biocontrol agents, notably *Encarsia formosa* Gahan (Hym., Aphelinidae) for control of *Trialeurodes vaporariorum* (Westwood) (Hem., Aleyrodidae), have been studied, and biocontrol and IPM programmes have become common practice in Ukrainian greenhouses. Moreover, a number of published articles on the inoculative and inundative use of natural enemies in greenhouses demonstrate their efficacy against key pests such as aphids and whiteflies [89–101]. In most cases, the natural enemies are either available commercially or produced in small insectaries and microbiological farms which are associated with the greenhouses and used for augmentative releases (Table 3).

Unfortunately, during the past 10 years, the use of biocontrol agents in greenhouses has decreased significantly in Ukraine. In part, the reduction in biological control programmes has been due to the increasing cost of heating greenhouses, which has resulted in a decline in vegetable production. Since the agricultural sector lost the subsidies that helped alleviate the high cost of energy for heating the greenhouses, the profit from this type of production has been low or non-existent. Nonetheless, the total area of greenhouses still in production is large, consisting of 3000 ha, and private rather than public ownership may bring the possibility of reintroducing...
biocontrol programmes into greenhouse production schemes [102].

**IPM in greenhouses**

As discussed above, biological control agents are often used in greenhouses, but usually more than one control tactic is needed to control a pest. For example, several studies have shown the high resistance of phytoseiids to insecticides [103, 104]. Such compatibility may lead to reduced pesticide usage with increased predator activity. Table 4 shows how various control tactics are integrated in cucumber and tomato production.

### Table 3

**Beneficial organisms used on cucumbers and tomatoes against pests and diseases in the greenhouse**

<table>
<thead>
<tr>
<th>Beneficial organism (Order, Family)</th>
<th>Pest species controlled (Order, Family)</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Phytoseiulus persimilis</strong> Athias-Henriot (Acari, Phytoseiidae)</td>
<td>Tetramychus urticae Koch (Acari, Tetranychidae)</td>
<td>[67, 68, 70, 76, 80–82]</td>
</tr>
<tr>
<td><strong>Aphidoletes aphidimyza</strong> (Rondani) (Dipt., Cecidomyiidae)</td>
<td>Myzus persicae Sulzer, Aphis gossypii Glover (Hem., Aphididae)</td>
<td>[83, 84, 87, 89, 90]</td>
</tr>
<tr>
<td><strong>Encarsia formosa</strong> Gahan (Hym., Aphelinidae), <strong>Macrolophus nubilis</strong> Herrich-Schäffer (Hem., Miridae), <strong>Aschersonia placenta</strong> Berk. &amp; Broome (Hypocreales, Clavicipitaceae), <strong>Lecanicillium lecanii</strong> (Zimm.) Zare &amp; W. Gams (Hypocreales, Clavicipitaceae)</td>
<td>Trialeurodes vaporariorum (Westwood) (Hem., Aleyrodidae)</td>
<td>[3, 86–88, 90, 95–98]</td>
</tr>
<tr>
<td><strong>Trichoderma lignorum</strong> (Tode) Harz [T. viride Pers.] (Hypocreales, Hypocreaceae)</td>
<td>Soil-borne diseases</td>
<td>[98, 99]</td>
</tr>
</tbody>
</table>

### Table 4

**Efficacy of IPM approaches in cucumber and tomato production in the greenhouses as presented by Lisovyi [102]**

<table>
<thead>
<tr>
<th>Crop</th>
<th>Crop protection system</th>
<th>Components of crop protection system</th>
<th>Yield (kg/m²)</th>
<th>Yield addition (kg/m²)</th>
<th>Cost of crop production (US$/m²)</th>
<th>Cost of yield addition (US$/m²)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cucumber</td>
<td>Conventional system¹</td>
<td>Trichoderma spp., Phytoseiulus spp., Amblyseius cucumeris (Oudemans) [Neoseiulus cucumeris] Spraying²: Fungicides 3–5 Insecticides 2–3 Acaricides 2</td>
<td>18.0</td>
<td>–</td>
<td>4.9</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td>IPM with maximum use of biocontrol</td>
<td>Trichoderma spp., Phytoseiulus spp., Macrolophus spp. Spraying²: Fungicides 2</td>
<td>24.0</td>
<td>6</td>
<td>6.5</td>
<td>1.7</td>
</tr>
<tr>
<td>Tomato</td>
<td>Conventional system¹</td>
<td>Trichoderma spp., Amblyseius spp., Aphidius matricariae Haliday Spraying²: Fungicides 3–5 Insecticides 1–2 Acaricides 1–2</td>
<td>24.2</td>
<td>–</td>
<td>7.9</td>
<td>–</td>
</tr>
<tr>
<td></td>
<td>IPM with maximum use of biocontrol</td>
<td>Trichoderma spp., Phytoseiulus spp., Macrolophus spp. Spraying²: Fungicides 3</td>
<td>29.4</td>
<td>5.4</td>
<td>9.6</td>
<td>1.8</td>
</tr>
</tbody>
</table>

¹Conventional system—based predominantly on pesticide use.
²Spray frequency per growing season.

Pathogens in biological control

Various pathogens have been used widely in biological control of pests in Ukraine since the 1990s (Table 5). Historically, insect pathogens have played a significant role in biological control in the country (i.e. Metchnikoff’s experiments). Therefore, it is not surprising that insect pathogens continue to be an attractive alternative to chemical pesticides.

**Codling moth and pathogens**

Codling moth, *Cydia pomonella*, is commonly infected by naturally occurring entomopathogenic bacteria, fungi, etc.
Table 5  Microbiological pesticide products used in Ukraine

<table>
<thead>
<tr>
<th>TAXONOMIC GROUP</th>
<th>Genus/Species</th>
<th>ORDER</th>
<th>Family</th>
<th>Target species</th>
<th>Crop</th>
</tr>
</thead>
<tbody>
<tr>
<td>BACTERIA</td>
<td><em>Bacillus thuringiensis</em>&lt;sup&gt;1&lt;/sup&gt; Berliner (Bt)</td>
<td>LEPIDOPTERA</td>
<td>Pyralidae</td>
<td><em>Pyrastha sticticalis</em> (L.) [Loxostege sticticalis]&lt;sup&gt;2, 3&lt;/sup&gt;</td>
<td>Sugar beet, alfalfa, sunflower</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Zophodia convolutella</em> (Denis &amp; Schiffermüller)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Berries, grape</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Mamestra brassicae</em> (L.)&lt;sup&gt;3, 4, 5&lt;/sup&gt;</td>
<td>Vegetables</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Lymantria dispar</em> (L.)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Forestry</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Operophtera brumata</em> (L.)&lt;sup&gt;3&lt;/sup&gt;</td>
<td>Forestry</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Abraxas grossulariata</em> (L.)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Berries, grape</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Pieris rapae</em> (L.)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Cabbage</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Aporia crataegi</em> (L.)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Plutella maculipennis</em> (Curtis) [P. xylostella (L.)]&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Cabbage</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Yponomeutidae</em></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Yponomeuta malinellus</em> Zeller&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Lasiocampidae</em></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Malacosoma neustria</em> (L.)&lt;sup&gt;4&lt;/sup&gt;</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Chrysomelidae</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Leptinotarsa decemlineata</em> (Say)&lt;sup&gt;3, 6&lt;/sup&gt;</td>
<td>Potato, tomato, aubergine</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Curculionidae</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Anthonomus pomorum</em> (L.)&lt;sup&gt;3&lt;/sup&gt;</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Diptera</em></td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Culicidae</em></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Mosquito species</em>&lt;sup&gt;7&lt;/sup&gt;</td>
<td>Aquatic systems</td>
</tr>
<tr>
<td>FUNGI</td>
<td><em>Beauveria bassiana</em> (Bals.-Criv.) Vuille.</td>
<td>LEPIDOPTERA</td>
<td>Tortricidae</td>
<td><em>Cydia pomonella</em> (L.)</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td><em>Coloptera</em></td>
<td></td>
</tr>
</tbody>
</table>
|                 |               |       |        | *Chrysomelidae* | *
|                 |               |       |        | *Leptinotarsa decemlineata* | Potato |
|                 |               |       |        | *Hemiptera* | |
|                 |               |       |        | *Aleyrodidae* | |
|                 |               |       |        | *Trialeurodes spp.* | Cucumber in greenhouse |
|                 |               |       |        | *Trialeurodes vaporariorum* (Westwood) | Cucumber in greenhouse |
|                 |               |       |        | *Tryptaurodes* | |
|                 |               |       |        | *Trialeurodes vaporariorum* | Cucumber in greenhouse |
|                 |               |       |        | *Thysanoptera* | |
|                 |               |       |        | *Thripidae* | |
|                 |               |       |        | *Thrips tabaci* Lindeman | Cucumber in greenhouse |
|                 |               |       |        | *Metarhizium anisopliae* (Metschn.) Sorokin | |
|                 |               |       |        | *Lecanicillium lecanii* (Zimm.) Zare & W. Gams | Field crop |
|                 |               |       |        | *Aschersonia placenta* Berk. & Broome | |
|                 |               |       |        | *Baculoviruses* | |
|                 |               |       |        | *Granulovirus*<sup>8</sup> | |

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viruses, and protozoa. The role of entomopathogens in decreasing codling moth populations and the use of microbial pesticides in apple orchards have been intensely studied [105–111]. In particular, the granulovirus of the codling moth (CpGV) is commonly found in populations and can reduce the fecundity and longevity of the adults. The CpGV was isolated from dead overwintering codling moth larvae collected in Russia in the 1940s. Since that time, its biology, virulence, and diversity have been studied, and it has been evaluated as a bioinsecticide in apple orchards [112–118]. Furthermore, the use of microbial products in apple orchards against the codling moth and other lepidopteran species, separately or in combination with inundative releases of Trichogramma species, has shown high efficacy and IPM programmes have been successfully established in apple orchards [119, 120].

Another insect pathogen that has received a lot of attention for codling moth control is the fungus, Beauveria bassiana. It is one of the most common fungal species infecting codling moth in nature [121–128]. However, its use as a microbial insecticide has been limited because of the lack of humid, warm conditions required for infection. In spite of some limitations of microbial agents, commercially products based on CpGV, Bacillus thuringiensis Berliner and Beauveria bassiana have been developed and these pathogens are registered for use in apple orchards [129–145].

A fourth pathogen that may have use as a bioinsecticide is the entomopathogenic nematode, Steinernema carpocapsae (Weiser) Wouts, Mracek, Gerdin & Bedding (syn. Neoplectana carpocapsae) (Rhabditida, Steinernematidae). It has been recorded from natural populations of the codling moth in many countries of the Former Soviet Union, and there are fragmented data about its natural occurrence in Ukraine [146].

The codling moth is used here as an example of how a fungus, a virus, and a bacterium can be used for pest suppression. In fact, insect pathogens are used inundatively in many cropping systems with bacterial pathogens being the most important in Ukraine for insect suppression (Table 5). At least five varieties of Bacillus thuringiensis are being used against lepidopteran and coleopteran species in various cropping systems and one variety is being applied against mosquito species. Entomopathogenic fungal-based products, such as Beauveria bassiana, Metarhizium anisopliae, and Lecanicillium lecanii (Zimm.) Zare & W. Gams (syn. Verticillium lecanii (Zimm.) Viegas) (Hypocreales, Clavicipitaceae) have also been used in greenhouses, field crops and vegetable crops to control pests. Baculoviruses (nucleopolyhedroviruses and granuloviruses) are not extensively used because of their narrow host range and relatively slow speed of kill, and the technical and economic difficulties in producing them in vitro.

The one exception is the granulovirus of the codling moth which has been produced in vivo and is economical to use.

When using insect pathogens, their virulence must be maintained. In addition, the appropriate application rate for a given pest must be known. In most cases, specific application rates have been developed for each pathogen and insect stage. Microbiological pesticides are usually used against the early larval stages (1st- to 3rd-instar larvae) with rates of 1–5 kg/ha (10⁶–10⁹ Bacillus thuringiensis/ha) for bacterial pathogens and against 2nd- and 3rd-instar larvae with rates of 0.1–0.3 kg or litres per hectare (1×10⁹–3×10¹¹ granules or polyhedral bodies

---

Table 5 (Cont.)

<table>
<thead>
<tr>
<th>TAXONOMIC GROUP</th>
<th>ORDER</th>
<th>Family</th>
<th>Target species</th>
<th>Crop</th>
</tr>
</thead>
<tbody>
<tr>
<td>Granulovirus (Cont.)</td>
<td>LEPIDOPTERA (Cont.)</td>
<td>Noctuidae</td>
<td>Agrotis segetum (Denis &amp; Schiffermüller)</td>
<td>Vegetables</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Hyphantria cunea</td>
<td>Apple</td>
</tr>
<tr>
<td>Nucleopolyhedrovirus⁹</td>
<td>LEPIDOPTERA</td>
<td>Noctuidae</td>
<td>Mamestra brassicae</td>
<td>Cabbage</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Heliolthis armigera (Hübner)</td>
<td>Tomato</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Lymantriidae</td>
<td>Apple</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Hyphantria cunea</td>
<td>Apple</td>
</tr>
</tbody>
</table>

¹Different varieties of Bt are used against different pests and these are footnoted with each target species.
²Bt var. galleriae.
³Bt var. thuringiensis (includes isolates 5072, 4067).
⁴Bt var. kurstaki.
⁵Bt var. insectus.
⁶Bt var. tenebrionis (includes isolate B-125).
⁷Bt var. israelensis.
⁸The granuloviruses tend to be host specific and each granulovirus was isolated from its respective host.
⁹The nucleopolyhedroviruses tend to be host specific and each nucleopolyhedrovirus was isolated from its respective host.

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per hectare) for viral formulations. Usually 1–2 applications are recommended with 2- to 8-day intervals against each pest generation. Entomopathogenic fungi are used at different rates depending on species. For example, *M. anisopliae* is usually applied at $10^{12}$ conidia/ha, *L. lecanii* at $10^{11}$–$10^{12}$ conidia/ha, and *Beauveria bassiana* at $10^{11}$–$10^{12}$ conidia/ha [3].

**Nematodes**

Entomopathogenic nematodes in the genera *Steinernema* and *Heterorhabditis* (Rhabditida, Steinernematidae and Heterorhabditidae, respectively) represent a most promising group of pathogens that are lethal to many soil-inhabiting insects [147–149]. They are distributed worldwide and have been found on every inhabited continent and many islands [150]. Both genera have been found in Ukraine (T. R. Stefanovska, unpublished data).

Researchers in many countries have shown great interest in using these nematodes for suppression of soil insect pests [151–159]. Since the early 1980s, more than 100 laboratories in over 50 countries have been conducting research on entomopathogenic nematodes [159] including some countries that neighbour Ukraine. For example, Hungary [160, 161], Poland [162], Czech Republic [163], Slovakia [164], and Turkey [165, 166] have research programmes on these nematodes. In Russia, fundamental research on taxonomy, biology, and practical application of entomopathogenic nematodes has been conducted since the late 1960s [167–175]. Ukraine, however, has lagged behind in fundamental and applied studies on these biological control agents. The most studied nematode group associated with insects is the family Mermithidae [176]. Nonetheless, there has been some interest in Ukraine as demonstrated by work on *S. carpocapsae* to control the codling moth larvae [3]. Fragmented data about the natural occurrence of these nematodes and their biology, ecology and host range and the effectiveness of the *S. carpocapsae* Agriotes strain against lepidopteran and coleopteran pests are available [177, 178]. Recently, *Heterorhabditis* spp. were extracted from soils in the Central Forest steppe areas of Ukraine (T. R. Stefanovska, H. K. Kaya, unpublished data).

**Biological control and Ukrainian economy**

Biological control has been practised in the Ukraine for many years, but not consistently. In part, the discontinuous use of biological control has been due to the Ukrainian economy. Currently, the Ukrainian agricultural sector is shifting from a command economy to a market economy. The command economy relied on both biological control agents and chemical pesticides. In this economy, the farm supervisors dictated the use of biological control agents or chemical pesticides depending on the pest population density. Because biological control agents were being used, the growers in the command economy were familiar with the various biological control products. The market economy, however, is profit driven, and the farmers rely primarily on chemical control and, therefore, biological control is used minimally or not at all. Although Ukraine has had a rich past in biological control, the farmers in the market economy need to be educated in biological control and IPM approaches. The state government must develop appropriate policy to encourage the use of biological control and IPM approaches. The consumer must also be educated on biological control, and the problems of chemical pesticide usage.

**Conclusions and prospects**

To further foster biological control in Ukraine, commercially produced natural enemies (predators, parasitoids and pathogens) must be readily available to farmers at a reasonable cost. Previously, the majority of commercial producers of microbial control agents at affordable prices were located outside Ukraine when it was part of the Soviet Union. Following the collapse of the Soviet Union, production of biocontrol products in the countries of the Former Soviet Union declined significantly, and it was not feasible for Ukrainian farmers to buy these products and remain profitable. In some cases, farmers are not aware of biological control agents because they are not commercially available, and companies that sell chemical pesticides often are not interested in introducing biocontrol products. Under these constraints, perhaps the best way to (re)introduce biocontrol products is through local production and demonstration of their successful application in Ukraine. Thus, research must demonstrate that these biological control agents can be efficacious. Issues such as timing of natural enemy release (either inundatively or inoculatively), pest and natural enemy densities, cropping systems, plant diversity, cultural techniques, integration with chemical pesticides, demonstration plots, education programmes for users and consumers, etc., need to be addressed. IPM approaches should use biological control as a cornerstone of control tactics.

Ukraine needs to take all possible steps to reinitiate the ‘era of biocontrol’. It must train scientists who will further advance biological control and take the technology to the commercial companies for production and farmers for implementation. The scientists must then train other individuals (e.g. farm advisors) who will teach farmers about biological control through demonstration plots and workshops on the use and application of biological control agents. Ukraine has the potential to be a major player in organic farming and export of organically grown products to Europe, where the demand for such products is high. Without biological control, the organic market will not flourish.

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