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REVIEW

Do the beneficial fungi manage phytosanitary problems in the tea agro-ecosystem?

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Abstract In recent years, Fungal Biocontrol Agents (FBCAs) have played a significant role in the biological control of pests and plant pathogens of several economically important crops, including tea. Although such agents are environment friendly, the risk of their effect on non-target organisms in tea gardens remains elusive. Among those FBCAs commercialized for field application, their impacts on the populations of arthropod pests and the prevalence of pathogens in tea ecosystems are still limited. Some products based on FBCAs have been applied for the control of pests and plant pathogens in tea crop. This review focuses on the biological potential of the fungal entomopathogens and antagonists against prevalent pests and diseases of tea crop, and their practical applications as bio-pesticides to minimize the load of chemical pesticides. The roles of FBCAs in plant growth promotion, their suitability as alternatives of chemical pesticides, challenges in the use of FBCAs, and perspectives are also discussed.

Keywords Entomopathogens · Fungal antagonists · Tea plants · Pests · Diseases · Biological control

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Introduction

Microbial communities are considered as one of the most important components in present agro-ecosystems. Microbes, especially Fungal Biocontrol Agents (FBCAs) present in the soil reservoir, are considered as potential candidates against a wide range of pests and diseases hampering crops (Vega et al. 2009; Lacey et al. 2015). Their formulations have also been used as an alternative to chemical pesticides in the agro-ecosystem for pest and disease management (Castillo Lopez et al. 2014). Recent studies revealed that these FBCAs, in addition to their potential against pests and plant pathogens, may play additional roles in plant growth promotion and rhizosphere colonization, which add their usefulness to Integrated Pest Management (IPM) (Sain and Pandey 2016). The manifold performances of FBCAs provide promising prospects for their multifaceted, indirect, and economic exploitation for sustainable agriculture, for instance, as biofertilizers for plant growth promotion (Fig. 1; Jaber and Enkerli 2016), and as vertically transmitted fungal endophytes (Lefort et al. 2016), as

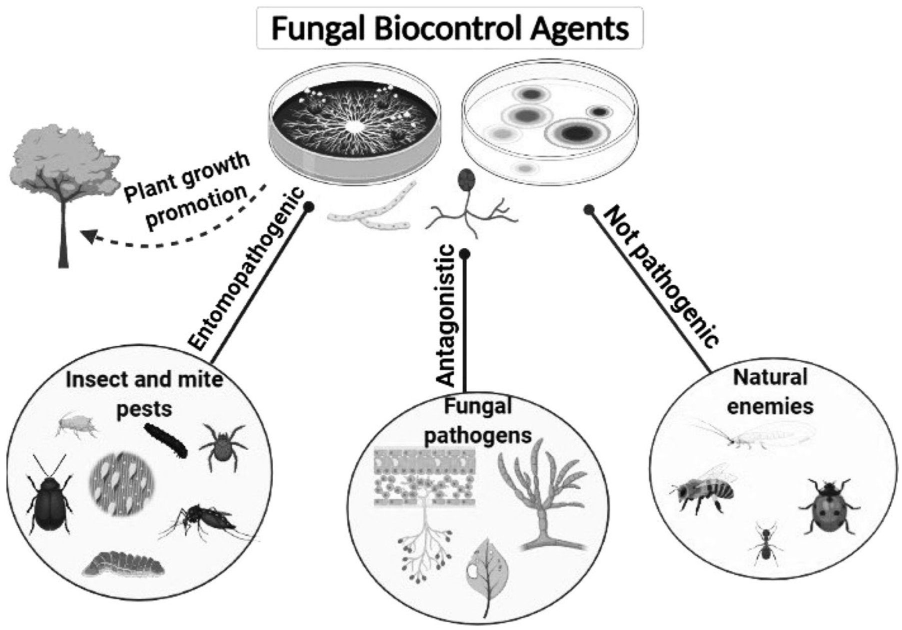

Fig. 1 Graphical image showing manifold performance of FBCAs

well as dual fungal biocontrol agents for pest and disease management (Ownley et al. 2008).

In the biological control system, FBCAs have been used for control of insect pests, mites, and plant diseases (Baron et al. 2019). The viruses, bacteria, and nematodes have also been employed as microbial biocontrol agents for the mitigated damages of arthropod pests and diseases. For example, a nematode, *Steinernema carpocapsae* (Rhabditida: Steinernematidae), has been used for control of caterpillars (Viteri et al. 2018), a simple nucleopolyhedrovirus (SNPV) has been used for control of African bollworm, *Helicoverpa armigera* Hubner (Lepidoptera: Noctuidae) (Valizadeh et al. 2020), and a bacterium, *Serratia marcescens* Bizio, has been used for control of root rot diseases (Dhar et al. 2018). In addition, FBCAs in the phylum Entomophthoromycota include *Conidiobolus, Erynia, Pandora, Zoophthora*, *Neozygites*, *Entomophthora*, and *Entomophaga*. Filamentous FBCAs include the genera *Aspergillus, Beauveria*, *Chaetomium, Metarhizium*, *Cordyceps*, and *Trichoderma* (Sain and Pandey 2016). Latter FBCAs also exist as plant endophytes (Ownley et al. 2008). The FBCAs produce a variety of low-molecular-weight compounds, i.e., secondary metabolites (Figs. 2, 3), accountable for many potentially useful biological activities such as insecticidal (Gao et al. 2020) and fungicidal (Vinale and Sivasithamparam 2020) properties. Despite these recently discovered attributes, attention has been mostly focused on developing the FBCAs as inundative bio-pesticides against the prevalent pests and diseases of economically important crops, including tea (de Faria and Wraight 2007).

Tea (*Camellia sinensis* (L) O. Kuntze) crop is grown primarily in Asia, South America, and Africa. Recently, tea products have increasingly become a primary source of farmers' income, particularly in developing countries. During tea cultivation, several biotic and abiotic stresses hamper with its production (Lehmann-Danzinger 2000). Tea is a perennial crop, and suffers multiple biotic stresses leading to annual yield and quality loss. Among the biotic stresses, arthropod pests and tea diseases are the main threats to the tea production. To fight against these biotic stresses, growers rely upon the applications of chemical pesticides, which have resulted in the development of pesticide resistance (Yamada et al. 2016) and side effects on the tea rhizosphere, as well as effects on human health (Nicolopoulou-stamati et al. 2016). Therefore, bio-pesticides have been considered as an alternative to reduce the load of commercial pesticides in tea crop. Reviews on the potential role of biological control agents in the control of tea pests have already been accomplished (Mamum and Ahmed 2011; Roy and Muraleedharan 2014; Ye et al. 2014). However, these reviews are not critically focused on

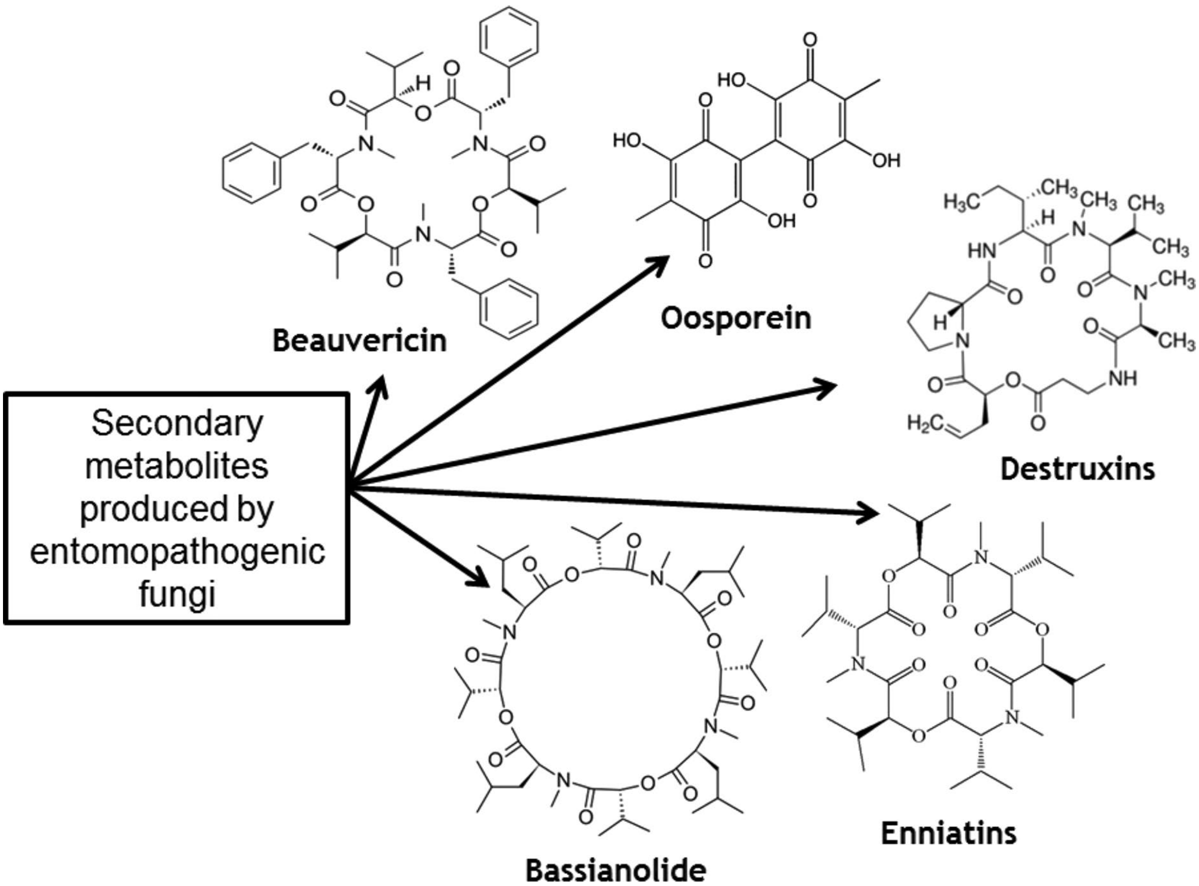

Fig. 2 Basic chemical structures of secondary metabolites of *Beauveria* sp. (beauvericin, oosporein, bassianolide, enniatins) and *Metarhizium* sp. (destruxin)

the FBCAs and their potential applications for pest and disease management in tea crop. Therefore, this review focuses on recent progress in the use of FBCAs for the control of insect pests, mites, and diseases of tea crop, and summarizes the challenges and perspectives on this issue. Discussions are also expanded on the compatibility of FBCAs with insecticides or fungicides, and their limitations which are not addressed in earlier reviews. Moreover, gaps and future research prospects for the innovative use of FBCAs as dual microbial/insect control agents are summarized.

Major pests and diseases of tea crop and their economic impact

Tea is considered as most consumed non-alcoholic beverage secondary to water. Globally, total tea production was estimated about six million tons (MT) in a growing area of around five million ha. Among the tea-growing countries, China (annually 2.5 MT) is the major tea producer, followed by India (1.3 MT), Kenya, Sri Lanka, Vietnam, Argentina, and Indonesia (FAO 2018). Tea with several medicinal values has been used as an antioxidant in human medication (Yang et al. 2014). Tea-containing flavanols (20–50%) and caffeine are the major phytochemical constituents, which benefit consumers' health and also contribute to the quality as well as stress responses (Punyasiri et al. 2005; Engelhardt 2010).

There are multiple insect pests, mites, and fungal pathogens that are threatening tea production by causing damages to tea bushes. All parts of the tea bushes, such as foliage, stem, and root, are

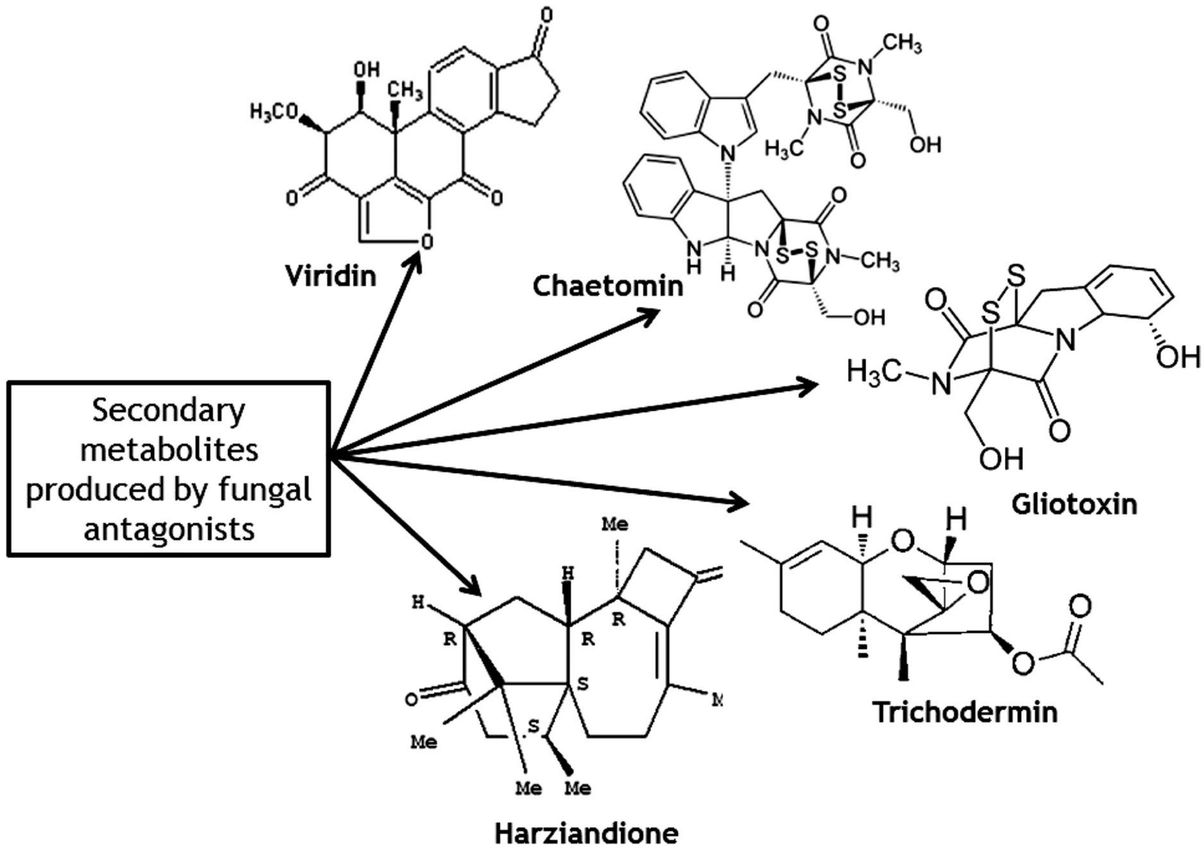

Fig. 3 Basic chemical structures of secondary metabolites of *Trichoderma* sp. (Trichodermin, gliotoxin), *T. viride* (viridin), *T. harzianum* (harziandione) and *Chaetomium* sp. (chaetomin)

susceptible to insect and mite pests as well as fungal diseases, which have co-evolved with tea plants for decades in tea-growing countries. The major pests include tea mosquito bug, thrips, jassids, aphids, tea looper complex, red spider mites, and termites (Table 1), and cause a maximum of up to 90% yield loss in the tea production (Zhang et al. 2017). Among foliar, stem, and root rot diseases, foliar diseases are more harmful due to damage to harvestable shoots (two leaves and a bud) and may result in a direct crop loss as high as 43% (Chen et al. 2018). A list of distribution of major tea diseases and their economic impact on harvestable shoots are summarized in Table 1. The major yield-limiting diseases are brown blight, grey blight, blister blight, canker, black rot, and root rots as well as *Fusarium* shoot dieback and bird's eye spot as emerging diseases.

For the management of tea pests, cultural (Lehmann-Danzinger 2000; Zhang et al. 2017) and chemical (Ponmurugan and Baby 2007) practices are in regular use, and due to perennial nature, host-plant resistance is not much successful in the mitigation of tea pests (Akbar et al. 2017). Besides, the overuse of chemical fungicides/pesticides has resulted in an adverse effect on the atmosphere and residue levels in processed tea, which is harmful to human health (Fronza et al. 2017). Therefore, tea growers are eager for alternatives to reduce the load of chemical insecticides/fungicides in tea crop. In this regard, an interest is increasing in the control of insect pests, mites, and diseases in tea crop through the use of FBCAs due to their efficient and eco-friendly nature. In the present review, we focus on the efficiency of some potential fungal entomopathogens and antagonists against major insect pests, mites, and diseases of tea crop.

Table 1 Pests and diseases of tea crop, their geographical distribution and economic impacts

Biotic stresses*	Causal organism	Yield loss (%)	Geographical distribution	References
Insect and mite pests				
Aphids	*Toxoptera aurantii* Boyer de Fonscolombe, *Myzus persicae* Sulzer	30–45	India, Kenya, Bangladesh, China, and India	Fang and Pu (2005)
Jassid	*Empoasca flavescens* Fab	30–50	India, China, Taiwan, and Japan	Bordoloi et al. (2012); Ye et al. (2014)
Red spider mite	*Oligonychus coffeae* Nietner	45–60	India, Sri Lanka, Bangladesh, China, Indonesia, Taiwan, Kenya, Burundi, Malawi, Japan, Uganda, Vietnam, Mauritius, and Zimbabwe	Ahmed et al. (2011); Liao et al. (2010)
Tea looper complex	*Andraca bipunctata* Walker*, Buzura suppressaria* Guenée*, Hyposidra talaca* Walker, *H. infixaria* Walker	50–90	China, India, Thailand, Myanmar, Sri Lanka, and Nepal	Zolotuhin (2012)
Tea mosquito bug	*Helopeltis theivora* Waterhouse	40–80	Bangladesh, Sri Lanka, Java and Sumatra, Laos, Vietnam, West Malaysia, Taiwan, Papua New Guinea, Uganda, Kenya, and China	Ahmed et al. (2013); Roy and Muraleedharan (2014)
Termite	*Microcerotermes* sp., *Odontermes* sp.	30–50	India, Sri Lanka, Kenya, and Bangladesh	Ahmed et al. (2011)
Thrips	*Scirtothrips dorsalis* Hood, *S. bispinosus* Bianchi	30–50	Sri Lanka, India, China, Indonesia, Bangladesh, Taiwan, Hawaii, and Japan	MacLeod and Collins (2006)
Diseases				
Root rot				
Charcoal/stump rot	*Ustulina zonata* (Lév.) Sacc., *Kretzschmaria deusta* (Hoffm.) Martin (= *U. deusta* (Hoffm.) Lind)	–	India, Sri Lanka, Indonesia, and Malaysia	Balasuriya (2008); Mishra et al. (2014)
Red root rot	*Poria hypolateritia* Berk. ex Cooke	–	Sri Lanka, South India, and Indonesia	Chakraborty et al. (2016)
Foliar and stem				
Bird's eye spot	*Cercospora theae* Petch	–	India	Gnanamangai and Ponmurugan (2012)
Black rot	*Corticium theae* Bernard	–	India, Taiwan, and Bangladesh	Muraleedharan and Chen (1997)
Blister blight	*Exobasidium vexans* Massee	40–50	Sumatra and Java (Indonesia), Bangladesh, India, and Sri Lanka	Uddin et al. (2005); Barthakur and Dutta (2006)
Brown blight and anthracnose	*Colletotrichum* spp.	–	Sri Lanka, India, China, Indonesia, Bangladesh, Taiwan, Hawaii, and Japan	Yoshida et al. (2010); Wang et al. (2016)

Table 1 (continued)

Biotic stresses*	Causal organism	Yield loss (%)	Geographical distribution	References
Cankers, twig dieback	*Macrophoma theicola* Siemaszko, *Phomopsis theae* Petch., *Fusarium solani* (Mart.) Sacc	40–70	Sri Lanka, India, Taiwan, Bangladesh, China, Japan, Kenya, and Hawaii	Ahmed et al. (2013); Balasuriya (2008); Keith et al. (2006)
Fusarium dieback	*F. solani*	–	India	Muraleedharan and Chen (1997)
Grey blight	*Pestalotiopsis theae* (Sawada) Steyaert, *P. longiseta* (Desm.) Steyaert	20	Japan, India, Sri Lanka, Korea, Kenya, and China	Chen et al. (2018)

*The common name of pests and diseases are given in alphabetical order

FBCAs for potential control of insect and mite pests of tea crop

In the kingdom fungi, approximately 700 fungi are putative entomopathogens. The fungi to be exploited for IPM fall into the orders Entomophthorales, Neozygitales (Entomophthoromycota), Hypocreales, and Onygenales (Ascomycota) (Mora et al. 2017). Over the last few decades, entomopathogenic fungi have attracted the attention of researchers for control of insect and mite pests, and plant growth promotion as well as for several other benefits in crops. The agro-ecosystem with a RH of more than 80% is suitable for better performance of entomopathogens (Sain and Pandey 2016).

Tropical and subtropical tea plantations feature a diversity of entomopathogens either in the rhizosphere or as endophytes (Ye et al. 2014). Most of the entomopathogenic fungi survive at higher humidity and planted tea bushes provide the suitable microclimatic conditions essential for their growth and multiplication. However, for the management of insect and mite pests, very few of them have been utilized as bio-formulations. In general, *Beauveria* spp., *Metarhizium* spp. (Cheramgoi et al. 2016; Canassa et al. 2020), *Purpureocillium* sp., *Cordyceps* spp. (syn. *Paecilomyces* sp.), *Akanthomyces* spp. (syn. *Cephalosporium* spp.) (Fronza et al. 2017; Lu et al. 2019), and *Akanthomyces lecanii* (Zimm) Spatafora (syn. *Lecanicillium lecanii* Zare & Gams) (Abdel-Raheem et al. 2020) have been used as bio-insecticides for effective control of pests.

In particular, in tea plantations, the population of red spider mite, *Oligonychus coffeae* Nietner (Acari: Tetranychidae), is the main target of entomopathogens through external cuticle infection. The in vitro studies from Sri Lanka revealed that some entomopathogenic fungi, including *Akanthomyces* spp. (syn. *Lecanicillium* spp.) and *Entomopthora* sp., are pathogenic to *O. coffeae* (Vitarana 2000). Under field conditions, the formulations of *M. anisopliae sensu lato (s.l.)* Sorokın (=*M. anisopliae*) and *B. bassiana s.l.* (Bals) Vuill (=*B. bassiana*) isolates have shown efficient activity against a wide range of pests (Cheramgoi et al. 2016).

Beauveria bassiana s.l.

Beauveria bassiana s.l. is an important fungal pathogen of insect and mite pests and is a base of fungal insecticides against the prevalent pests hampering tea crop. The efficacy of *B. bassiana s.l.* in terms of pathogenicity has been assessed against several pests of tea crop in both the laboratories and in the field (Pu et al. 2005; Babu and Kumhar 2014). During the 1990s, Guangyuan et al. (1995) from Fujian province of China found that *B. bassiana s.l.* controlled the tea weevil, *Myllocerinus aurolineatus* Voss (Coleoptera: Curculionidae), where 65% of the pupae and 80% of the adults were controlled by spraying of spore suspensions. In the same year, Wu et al. (1995) also reported that spraying of 15–30 kg of *B. bassiana* ($1–2 \times 10^8$ spores ml^{-1}) suspension controlled 80% of tea weevil adults in the field. Similarly, in Kenya, two indigenous isolates of *B. bassiana s.l.* sprayed as solid (in wheat bran) and liquid formulations significantly reduced tea weevils (*Sprigodes mixtus* Kolbe and

Entypotrachelus meyeri micans/Kolbe) populations in tea crop (Cheramgoi et al. 2016).

Beauveria bassiana s.l. and its formulated products have also been found pathogenic to the tea loopers (Ghatak and Reza 2007). The field application of mineral oil-based emulsified formulation of *B. bassiana s.l.* (1.7×10^{13} conidia ha^{-1}) with imidacloprid (50 g ha^{-1}) significantly reduced the population of false-eye leafhopper, *Empoasca vitis* Gothe (Hemiptera: Cicadellidae), in Yunnan Province of China (Feng et al. 2004). Similar results were reported by Feng and Pu (2005), who observed that a mixed formulation of *B. bassiana s.l.* (1×10^{10} to 2×10^{10} conidia ml^{-1}) and imidacloprid (3 mg ml^{-1}) were more effective against *E. vitis* than the standalone application in Hangzhou tea plantations, Zhejiang, China. In addition, spraying of formulated *B. bassiana s.l.* in 100-fold dilution in terms of conidial concentration (150 l ha^{-1}) by a knapsack air-blast sprayer with 1.6-horsepower gasoline engine was more effective in the control of tea leafhopper than that of hand-operated backpack hydraulic sprayer and micro-ulva hand-held sprayer methods used by Pu et al. (2005).

Some studies also reported *Beauveria* as an efficient entomopathogen for the control of termites in tea plantations (Gurusubramanian et al. 1999). In this regard, Singha et al. (2006) reported that *Microtermes obesi* Holmgren (Blattodea: Termitidae) was very susceptible to *B. bassiana s.l.* with the LD_{50} (lethal dose) values ranged from 35 to 140 conidia per termite. Conversely, *B. bassiana s.l.* (5.0 g l^{-1}) was reported as poor entomopathogen than that of *M. anisopliae s.l.*, *Cordyceps fumosorosea* Zare and Gams (= *Paecilomyces fumosoroseus* (Wize) Br. & Sm.), and *Akanthomyces lecanii* (Zimm.) Spatafora (= *Verticillium lecanii* (Zimmerman) Viegas) in control of red spider mite in Bangladesh (Mamun et al. 2014). In addition, interestingly, the application of the highest dose of *B. bassiana s.l.* (10^7 and 10^8 spores ml^{-1}) was more effective against the shot hole borer, *Euwallacea fornicatus* Eichhoff (Coleoptera: Curculionidae) infesting tea plants compared to the lower doses in the south Indian tea plantations (Muraleedharan 2006). Moreover, this entomopathogen (4 g l^{-1}) has also been used as an effective bio-insecticide against tea mosquito bug (TMB), *Helopeltis antonii* Signoret (Hemiptera: Miridae), with 91.7% nymphal mortality, and was more effective than the botanicals and chemical insecticides (Navik et al. 2015).

A recent study in India revealed that formulated *B. bassiana s.l.* (1.68×10^6 spores ml^{-1}) at 21.87 ml l^{-1} concentration caused a 50% reduction in *Helopeltis theivora* Waterhouse adults after 96 h of application in tea crop (Ekka et al. 2019). Besides, more recently, some *B. bassiana s.l.* formulations made from Indian isolates and applied at the rate of ~ 1×10^{13} conidia ha^{-1} (Manimaran et al. 2019) or 1×10^9 spores ml^{-1} (Kumhar et al. 2020) were effective against *H. theivora.* The variation in the dose and efficacy of the same entomopathogenic species may be due to their isolates from different regions or variability in the biotypes of insect pests (Cheramgoi et al. 2016). In a study, spraying of conidial suspensions of *B. bassiana s.l.* in potato crop below the canopy level efficiently killed larvae of potato beetle (Wraight and Ramos 2002). However, as far as tea crop is concerned, it would be difficult to spray the microbial formulations from below the canopy levels using a commercial backpack manual hydraulic sprayer. This advocates a requirement for a spray method to augment the attachment of active fungal conidia/spore to the external cuticle of tea leafhoppers, beetles and other insect pests to improve the management practices.

Beauveria has also been found to be compatible with chemical insecticides/acaricides, but not fungicides. For example, some acaricides, such as pyridaben, hexythiazox, and propargite were highly compatible with *B. bassiana s.l.* during application against eggs of a spider mite, *Tetranychus cinnabarinus* Boisduval (Trombidiformes: Tetranychidae) (Shi et al. 2005). Conversely, Liu et al. (2012) studied the efficacy of six pesticides on the growth and spore development of *B. bassiana s.l.* and found that all the six pesticides, including bifenthrin, imidacloprid (insecticides), mancozeb, carbendazim (fungicides), and veratrine, qingyuanbao (botanical insecticides) had an inhibitory effect. However, bifenthrin had no significant negative impact on the growth of *B. bassiana s.l.* at lower doses than the commonly used doses. Therefore, to enhance the bio-efficacy of FBCAs, more investigations are required in this domain to search the more compatible insecticides so that they can be applied as an integrated approach when *Beauveria* alone failed to mitigate insect and mite pests.

Metarhizium anisopliae s.l.

The genus *Metarhizium* has demonstrated potential bio-efficacy against many soil-borne and foliar insect pests in different crop conditions, including tea. In India, termite pests (Singh et al. 2011; Debnath et al. 2012) and tea thrips (Shanmugapriyan et al. 2011) were significantly controlled through the application of *M. anisopliae s.l.* Another formulation of *M. anisopliae s.l.*, named Bioterminator, showed efficient activity against termites (86% mortality) damaging the tea bushes in Bangladesh (Hoque et al. 2016). The developed Bioterminator maintained its efficacy at the nine months of shelf life. In the same series, Kalichakra (1%WP) formulated with *M. anisopliae s.l.* controlled tea thrips with 100% mortality achieved at spray rate 0.375 ml l^{-1} after 72 h application (Hoque et al. 2016). In addition, the wettable powder formulation of *M. anisopliae s.l.* effectively controlled populations of red spider mite (Kumhar et al. 2020), tea termite, *M. obesi* (Singha et al. 2011), as well as *H. theivora* and *O. coffeae* (Anitha et al. 2019) at field levels. *Metarhizium anisopliae s.l.* at 10^8 spores ml^{-1} concentration was pathogenic to red spider mite, which showed a maximum of 90% mortality at all developmental stages after 2–3 days of spraying under laboratory conditions (Baruah and Deka 2017).

As far as compatibility is concerned, insecticides such as imidacloprid and abamectin have been found highly compatible, and thiamethoxam compatible with *M. anisopliae s.l.* (Niassy et al. 2012). The lambda-cyhalothrin has been found toxic with *Metarhizium* (Niassy et al. 2012). However, the spiromesifen (acaricide) has been found moderately toxic, and the fungicides such as copper-hydroxide, carbendazim, and probineb have been found extremely toxic with *Metarhizium* (Niassy et al. 2012). In another study, gamma- and lambda-cyhalothrin have also been reported compatible with *M. anisopliae s.l.* and *M. robertsii* Bisch (Pelizza et al. 2018). It has been found that these insecticides did not affect the vegetative growth, conidial viability, and conidial production of entomopathogen at higher doses. However, luphenuron and methoxyfenozide significantly affected the growth parameters of *Metarhizium* at the higher doses 200 ppm and 288 ppm, respectively (Pelizza et al. 2018). Likewise, lambda-cyhalothrin, novaluron, emamectin benzoate, and indoxacarb insecticides have been found compatible with both *M. anisopliae s.l.* and *B. bassiana s.l.* (Joshi et al. 2018). Among the fungicides tested by Joshi et al. (2018), only mancozeb was compatible with both entomopathogens to some extent at lower concentrations (0.5 and 0.25%). However, carbendazim, hexaconazole, and propiconazole had complete inhibitory effect on entomopathogens (Joshi et al. 2018). These results suggest that the application of *M. anisopliae s.l.* and *B. bassiana s.l.* with agrochemicals has to be evaluated carefully prior to the field intervention. The use of luphenuron, methoxyfenozide, carbendazim, hexaconazole, and propiconazole with entomopathogens based bio-pesticides should be avoided during application in tea crop.

Other insect mycopathogens

Several species of the genus *Cordyceps* spp. (= *Paecilomyces* spp.) such as *C. fumosorosea* and *C. tenuipes* (Peck) Kepler have been utilized for control of insect and mite pests of crop, but few species have been evaluated against insect pests of tea crop (Ye et al. 2014). For example, *C. fumosorosea* (10^8 spores ml^{-1}) has been found highly pathogenic to leaf roller and aphid infesting tea crop (Ramesh et al. 2002). A formulation of *P. cinnamomeus* (Petch) Samson & Gams (10^7 conidia ml^{-1}) has been used for control of whitefly, *Aleurocanthus camelliae* Kanmiya & Kasai (Hemiptera: Aleyrodidae) in the tea plantations of Japan, resulting in 90% mortality of nymphal whiteflies (Saito et al. 2012). Recently, Lu et al. (2019) reported that *P. variotii* Bainier showed dual potential as activities of entomopathogenicity and plant growth promotion.

In addition, *A. lecanii* (1×10^7 spores ml^{-1}) has also been reported pathogenic to tea thrips, and the addition of jaggery to *A. lecanii* formulation enhanced insecticidal activity against thrips (Subramaniam et al. 2021). *Akanthomyces lecanii* also killed 70–80% of tea aphid, *Toxoptera aurantii* Boyer de Fonscolombe (Hemiptera: Aphididae) adults, and 70% population of a scale insect, *Coccus viridis* Green (Hemiptera: Coccidae), which resulted in the protection of 83% of the infested seedlings in the nursery (Hazarika and Puzari 2001). Despite several fungi pathogenic to red spider mite (Vitarana 2000), only *C. fumosorosea* (Muraleedharan 2006) has been formulated for application. The moderate temperature and high RH induced the infectivity of entomopathogens, and the rainfall helps the entomopathogen to grow abundantly on the insects (Hazarika and Puzari 2001).

However, some ascomycetes such as *Aspergillus flavus* Link, *Aspergillus niger* van Teighem, and *Cladosporium* sp. have shown pathogenicity against *H. theivora* under laboratory conditions (Bordoloi et al. 2012). In Bangladesh, *A. flavus* and *A. niger* have also shown biocontrol potential against red spider mite at respective concentrations 1×10^7 and 1×10^8 conidia ml^{-1} by exhibiting 91% mortality after 96 h application (Mazid et al. 2015). However, these FBCAs could not be validated and commercialized, and hence, they need to evaluate at multi-locations to ascertain their efficacy at the field levels.

FBCAs for potential control of diseases of tea crop

Many fungal pathogens are threatening the health of tea crop by causing several yield-limiting root rot, stem, and foliar diseases. Efforts to mitigate these diseases have been carried out worldwide, and several fungal antagonists have been evaluated against fungal pathogens of tea plants causing prevalent diseases. Here, we summarize the progress in efforts to control the important root rot, stem, and foliar diseases of tea plants through the use of antagonistic fungi under the following sections.

Direct suppression of the fungal pathogens of tea crop

The beneficial fungi which act as FBCAs against plant pathogens can directly suppress the pathogens through several mechanisms, such as competition for ecological niches for foods, mycoparasitism, and by the production of various secondary metabolites (Steyaert et al. 2003; Ownley et al. 2008). In tea ecosystem, for disease management, FBCAs have been capturing the attention of researchers to be used as alternatives to chemical fungicides because of their eco-friendly nature, easy biodegradability, and negligible effect on the host plants, environment, and humans. Several fungal antagonists have been identified as aggressive against major fungal pathogens of tea crop (Table 2). Unfortunately, most of these agents were screened in vitro, and few were evaluated at the field level (Table 2). Table 2 shows that the frequently used fungal antagonists evaluated against fungal pathogens of tea plants were *Trichoderma* spp. (Kumhar et al. 2020). The other report from Thailand (Phong et al. 2014) revealed that another antagonist, *Chaetomium lucknowense* Rai & Tewari (1000 µg ml^{-1}) was capable of reducing grey blight incidence (57%) and was more virulent than the strain *C. globosum* Kunze.

Also listed in Table 2, the efficacy of fungal antagonists was not consistent in different trials. For example, for blister blight, 20 to 50% disease control efficacy was observed (Rayati 2011). The use of FBCAs or consortium has also been found to be more effective for control of grey blight of tea crop than the use of individual antagonists alone (Nepolean et al. 2015). Researchers also found that fungal isolates of the same species showed variable activity against the same pathogen from different environments or different pathogens in the same environment. The variations in efficacy may be due to various isolates/strains of FBCAs from diverse origins, types of formulations in various carriers, and application methods (Murolo et al. 2019).

In recent years, the use of genomic tools has offered opportunities to understand the action modes of FBCAs and genes associated with them (Piombo et al. 2018). However, more research is required in this domain, especially for tea pathogens. In addition, the potential FBCAs from various experiments need field evaluation prior to recommendation of application in tea crop (Table 2). It will be advantageous that FBCAs are applied together with organic matter (cattle manure, plant residues, vermicompost), which facilitates fungal colonization and proliferation in soil against underground pests.

Induction of systemic plant resistance

Induced Systemic Resistance (ISR) in plants occurs when the plants are inoculated with weak pathogens or non-pathogenic microbial agents. Among these, the fungal antagonists are found to play a major role in ISR by stimulating the mechanical and physical strength of the host plant cell wall. These fungal antagonists are also found to change the biochemical and physiological reactions of hosts during which the synthesis of several defense-related metabolites/chemicals against target pathogens may take place. Several pathogens related (PR) proteins (β-1, 3 glucanase, chitinase), phenylalanine ammonia lyase

Table 2 Efficacy of FBCAs against fungal pathogens causing diseases in tea crop

Fungal pathogens#	Fungal antagonists	Methods/Dose	Efficacy impact* and yield
Athelia rolfsii	*Trichoderma viride* Pers	Dual culture	61% PGI (Shaigan et al. 2008)
Cercospora theae	*T. harzianum* Rifai (Th)	300 (2:3) l ha^{-1}	Th:44.1% DP, yield:3884 kg ha^{-1} (Gnanamangai and Ponmurugan 2012)
Corticum theae	*T. atroviride* Karsten	Dual culture	72.05 PGI (Thoudam and Dutta 2014)
Corticum invisum	*T. viride* (Tv), *T. harzianum* (Th)	Dual culture	Th:77% PGI, Tv:55–66% PGI (Kabir et al. 2016)
Macrophoma sp.	*Trichoderma* spp.	Dual Culture	All showed 2 cm zone of inhibition (Mareeswaran et al. 2015)
Fusarium solani	*T. virens*	Dual culture	77.6% PGI (Kumhar et al. 2015)
F. solani	*T. viride*	Foliar spray 400 l ha^{-1}	10% of *T. viride* caused 86.9% DP (Sarmah et al. 2017)
	Trichoderma strains	Dual culture	86.4% PGI (Kumhar and Babu 2019)
F. solani	21 isolates of *T. viride*	Dual culture	Isolate 15:76% PGI (Naglot et al. 2015)
Pestalotiopsis theae	*Chaetomium globosum* Kunze (CG), *C. lucknowense* Rai & Tewari (CL)	1000 µg ml^{-1}	CG:39.42% DP, CL:57% DP (Phong et al. 2014)
	T. viride	Dual culture	74.3% PGI (Barman et al. 2015)
	Trichoderma spp.	Dual culture	84.45% PGI (Sarkar et al. 2015)
	T. viride (TV), *T. harzianum* (TH)	Dual culture	TH: (75% PGI) TV: (55–63% PGI) (Kabir et al. 2016)
	21 isolates of *T. viride*	Dual culture	71% PGI (Naglot et al. 2015)
	Trichoderma strains	Dual culture	86.4% PGI (Kumar and Babu 2019)
Poria sp.	*Cunninghamella* sp.	Dual culture	100% PGI (Chong and Fazila 2012)

*DP: Disease protection, PGI: Percent Growth Inhibition

#Pathogens are given in alphabetical order

(PAL), chalcone synthase, phenolics, peroxidase (PO), callose, phytoalexins, and lignin are found to accumulate during the defense reaction in the host plants including tea (Saravanakumar et al. 2007). In tea crop, some FBCAs have been used against fungal diseases as bio-stimulants, such as Avascular Mycorrhizal Fungi (AMF) as a formulation for foliar or soil applications have shown a potential role in the management of *Athelia rolfsii* (Cruzi) Tu & Kimbr. (=*Sclerotium rolfsii* Sacc.), *Phellinus noxius* (Corner) Cunn., and *Poria hypobrunnea* Petch. by increasing activities of PAL, PO, chitinase, and α-1, 3 glucanase in tea plants (Chakraborty et al. 2016) through growth promotion, induced systemic resistance, and other action modes. Even though, while *Trichoderma* has been used to induce systemic resistance against other fungal pathogens (Rivera-Méndez et al. 2020), efforts are needed to evaluate their activities against prevalent diseases of tea crop.

Role of FBCAs in plant growth promotion

FBCAs also produce several growth hormones, such as auxins, cytokinin, and gibberellins. These hormones have a major role in the growth suppression of harmful pathogens as well as in the growth promotion of host plants (Barelli et al. 2020). A combination of bacterial and fungal antagonists (*Streptomyces griseus* + *T. harzianum*) increased green leaf yield and improved tea quality (Elango et al. 2015), while application of *T. asperellum* alone promoted the growth of tea seedlings (Shang et al. 2020). Sprays of tea bushes with the formulations of *B. bassiana s.l.*, *M. anisopliae s.l.*, and *Cordyceps* spp., as well as several *Trichoderma* resulted in increased yields of harvestable shoots (Ramesh et al. 2002; Saito et al. 2012; Cheramgoi et al. 2016; Ekka et al. 2019; Kumhar et al. 2020). In addition, rhizospheric application of vesicular-arbuscular mycorrhizal fungi in the tea plantations in Kenya increased nutrient uptake which can assist bushes to resistant against diseases (Sitienei et al. 2017). Thus, these FBCAs may serve as alternatives for the growth promotion and higher yield of harvestable tea shoots.

The advantage of using FBCAs over chemical pesticides

Worldwide, tea crop suffers many seasonal as well as perennial pests and diseases during its lifespan. In arable crops, such as rice, wheat, maize, etc., the pests and diseases are managed by tillage, rotation, and delayed seeding/sowing that disrupts the life cycle of pest/pathogen. However, such practices are not feasible for the perennial tea system, which is often controlled with chemical fungicides and insecticides (Mareeswaran et al. 2018). So far, only seven fungicides (active ingredients), and fourteen acaricide and insecticide are registered against the pests and pathogens of tea crop in India (Plant Protection code 2020). On the other hand, only five fungicides and insecticides are registered in Sri Lanka for tea pests' management (Advisory circular PU 1, 2020, TRISL). Although, the use of selective fungicides and insecticides in IPM programs of tea crop is a common practice. In addition, most of the chemical pesticides used in tea crop have tolerable effects on parasitoids and predators, beneficial microbes, and maximum residue level or phytotoxic effect (Devine and Furlong 2007; Ye et al. 2014). However, due to indiscriminate use, chemical pesticides may disturb the ecological balance between insect pests and natural enemies (parasitoids/predators).

In tea ecosystem, natural enemies serve as biocontrol agents against insect and mite pests for maintenance of ecological balance, hence called biological indicators of the tea agro-ecosystem (Ye et al. 2014). Unfortunately, few reports showed that chemical fungicides and insecticides have harmful effects on beneficial microbes, and on parasitoids and predators, such as *Geocoris ochropterus* Fieber (Hemiptera: Geocoridae), *Chrysoperla carnea* Stephens (Neuroptera: Chrysopidae), and *Stethorus gilvifrons* Mulsant (Coleoptera: Coccinellidae) in the tea ecosystem (Messelink et al. 2014). Besides, due to climate change, insect pests and plant pathogens have been developing resistance to insecticides and fungicides. For example, the pathogens of grey blight and canker diseases of tea crop have shown resistance against benzimidazole, strobilurin, and hexaconazole (Omatsu et al. 2012; Yamada et al. 2016). Likewise, in Japan, tea tortrix, *Adoxophyes honmai* Yasuda (Lepidoptera: Tortricidae), developed resistance to diamide insecticide (Uchiyama and Ozawa 2014), and tea green leafhopper was resistant to bifenthrin and acetamiprid (Wei et al. 2017). Nonetheless, indiscriminate use of chemical pesticides has also resulted in increased production costs for crops and potential risks to both humans and the environment, and the demand for contaminant-free, high-quality tea has led to a movement towards non-chemical pest management and sustainable tea cultivation. To restore the productivity and sustainability of soil as well as plants, efforts have been made to implement alternative with lower toxicity to beneficial organisms, eco-friendly and cost-effective pest and disease management strategies, and FBCAs are desirable in this regard.

Tea crop are considered highly suitable for biological control programs with FBCAs. The entomopathogens used as commercial bio-insecticides are non-pathogenic to some parasitoids and predators. For instance, *B. bassiana s.l.*, *Hirsutella* spp., *M. anisopliae s.l.*, and *Cordyceps* spp. were not pathogenic to *C. carnea*, *Coccinella septempunctata* L. (Coleoptera: Coccinellidae), *Dicyphus tamaninii* Wagner (Heteroptera: Miridae), *Oxyopes* sp. (Araneae: Oxyopidae), and *S. gilviforns* as well as *Heteromurus nitidus* Templeton (Entognatha: Entomobryidae), a beneficial soil insect (Mamun et al. 2014; Portilla et al. 2017), because these natural enemies were not affected at population levels after field application. More recently, wide-spectrum entomopathogen, *B. bassiana s.l.* has proved safe to apiculture (Peng et al. 2020). This shows that the entomopathogenic fungi could be quite specific and might infect only a certain type of host. Conversely, *M. anisopliae s.l.* was pathogenic to *C. carnea* and *D. tamaninii*, caused 4 and 10% mortalities, respectively (Thungrabeab and Tongma 2007). Therefore, before recommendation of the entomopathogens as bio-insecticides, it must be evaluated against the natural enemies.

Moreover, at present, the presence of maximum residue limit of chemicals in processed tea is a global concern because the European Union

(EU) has imposed strict rules on the use of chemical insecticides and fungicides in tea crop. EU has fixed the maximum residue limit (MRL) values (<0.1 mg kg^{-1}) for the commonly used insecticides/fungicides in manufactured tea that will not be met out in factual practice. Thus, the compliance with a MRL set for the pesticides as food safety standard is a major challenge in tea exporting countries like China, India, Sri Lanka, Kenya, and others. FBCAs are compatible with certain insecticides and fungicides and have a potential role in plant growth promotion (Liu et al. 2012; Feng and Pu 2005). Therefore, in the circumstances where FBCAs or fungicides/insecticides alone are not effective, the combined application of FBCAs and fungicides/insecticides could be a choice to reduce the pesticide residue level in tea crop. Insecticide residue levels above the EU recommendations have often appeared in harvestable tea shoots (Devine and Furlong 2007). Therefore, limited use of chemical pesticides can be recommended and emphasis should be made on the use of IPM so that indigenous natural enemies and beneficial microbes existing in tea agro-ecosystem could be conserved for sustainable crop protection (Messelink et al. 2014).

Challenges and perspectives

The FBCAs are recognized as agents that will continue to have a significant role in the management of insect pests and pathogens of tea crop for years to come. However, the microbes-based mitigation of pest infestation by entomopathogenic fungi were regarded untrustworthy during the early years of investigation because of their inconsistency in the field performance. For example, *B. bassiana s.l.* acts very slowly against termites. An ideal entomopathogen against termites is efficacious when it causes mortality after a realistic lag period to permit the contacts in maximum number with the infected carrier before death (Carruthers and Soper 1987). In other words, the workers in a termite colony that has elite up the conidia of the entomopathogen must be able to travel through the whole colony, and before dying encounter the other individuals. In this regard, isolates of *B. bassiana s.l.* can act as the leading candidates against termites. However, the main dispute which restricts the market share of this fungus is related to the bio-insecticidal property, that must be directly proportional to insect-killing speed, and the isolation mechanism that prevails in the termite colony is a matter of concern. Hence, there is a need to endorse research activities in the selection of potential isolates of entomopathogens since genetic changes have increased virulence to target pests (Tong and Feng 2020).

During the progressive era, entomopathogenic fungi were found to be efficient in reducing the population of a wide range of insect and mite pests of crops. This has encouraged the commercialization of many entomopathogens-based bio-insecticides in the markets. Consequently, it was examined that the pathogenicity of FBCAs can be enhanced in the laboratory to infect a given host. The chemical profile of the insect cuticle, temperature, moisture, and humidity are the major factors that significantly affect the infectious process (Tong and Feng 2020). The improvement of formulation technology of FBCAs based products by exploring nutrition of entomopathogens and environmental tolerance will enhance the survival of bio-products. In addition, both fungal entomopathogens and antagonists used for biological control of insect pests, mites, and diseases of tea crop are affected by changes in the climatic patterns and conditions. A better understanding of FBCAs-insects/pathogens and environment interaction is required to anticipate the likely patterns of variation that will be encountered under climate change and to adapt release strategies accordingly.

Nevertheless, even though control of diseases and insect pests through the FBCAs is a less expensive tool and less harmful to the environment, the biological control practices in tea plantations are currently very limited compared to chemical pesticides. This fact mainly occurs because the mitigation strategies based on FBCAs are more specific and takes a longer time to achieve the desired results. Hence, more than one FBCA are often introduced to tackle an insect pest or pathogen. Also the potential collaboration between plant pathologists, entomologists, and advisory officers from both research organizations and universities are critical to take on this challenge. A decision to focus on the particular FBCAs, for either the classical or bio-pesticide approaches discussed in this review, should be grounded in their ability to exert the required negative effect on the target pests/pathogens.

Notably, FBCAs based bio-pesticides have a checked history and have not yet become a key component in IPM, despite increasing interest in sustainable and environmentally friendly pest and pathogen control methods by regulators, tea growers, and the general public. For example, in India, 30 products based on *M. anisopliae s.l.,* over 87 products of *B. bassiana s.l.*, over 60 products of *A. lecanii,* and only three of *C. fumosorosea* products, are registered for use against insect pests of different crops (Kumar and Babu 2019). However, at present, few are in regular use due to awareness among the farmers/tea growers. The commercialization of bio-pesticides based on FBCAs has been the measure of success in affluent countries, although few products that reached that stage did not necessarily remain on the market for long. Their vulnerability to slight fluctuations in ambient temperature and moisture conditions in the field has made them less reliable than bio-pesticides based on compounds produced by microorganisms or obtained from natural sources. Transformational advances in large-scale production and formulation to cost-effectively develop highly efficient bio-pesticides, combined with a more strategic selection of target pest/pathogen and legislative and social incentives to adopt such technology over chemical-based products will open opportunities for the future.

Gaps and future remarks

The search for FBCAs, their formulation methods, mass production, and their applications in tea crop have greatly advanced worldwide, reviewed date back by many researchers (Ye et al. 2014; Kumhar et al. 2020). Therefore, these commercially formulated entomopathogens may be utilized potentially in the tea ecosystem against pests and diseases as eco-friendly and safer bio-pesticides as well as one of the major components of an IPM strategy to minimize the pesticide load in the tea ecosystem. However, some gaps should be addressed in the future.

The FBCAs illustrated in this review have been identified through the classical and molecular methods based on sequencing of ITS regions of the rDNA, β-tubulin, and tef 1-alpha, as well as through several markers (Hassan et al. 2019). However, the frequent use of omics technologies, such as proteomics, genomics, and metabolomics will modernize our understanding of FBCAs-insect/pathogen interactions, which will impact decisions, particularly during the selection of FBCAs, and potential practices used in biological control. Such understanding will also be important in regulatory genetic modifications to improve bio-efficacy and tolerance of FBCAs, provided acceptance by a wide range of tea growers.

The mode of pathogenicity of FBCAs and related secondary metabolites has been studied against pests and pathogens of other crops (Xiao et al. 2012; Vinale and Sivasithamparam 2020), but not studied in tea crop and need future attention from a fundamental point of view. Identification of more potential isolates of FBCAs having multi-pathogenic activity from the tea ecosystem and subsequently their mass multiplication in the laboratory followed by their use in tea gardens may be practiced. Fungal entomopathogens, such as *M. rileyi* (Farlow) Kepler, Rehner and Humber (formerly in the genus *Botrytis, Spicaria* or *Nomuraea*) and *Hirsutella* species have been used for control of lepidopteran insect pests of several economically important crops (Fronza et al. 2017). Nonetheless, both entomopathogens have not been evaluated against insect and mite pests of tea crop and need to be addressed in future research projects.

The biological control of biotic stresses in tea crop has been also documented as an alternative to commercial insecticides/fungicides. Many entomopathogenic fungi and fungal antagonists either in combination with synthetics or alone have been evaluated with positive results in China, India, Kenya, and Sri Lanka (Agnihothrudu 1999; Cheramgoi et al. 2016). There are many reports demonstrating that FBCAs introduced for classical biocontrol brought down target insect pests and pathogens populations in tea ecosystems to manageable levels, but more post-introduction quantitative assessments are needed.

In addition, collecting data to show durability over the time of host range of FBCAs used in classical pest and pathogen control of tea crop is required to address the concerns that, after their introduction in tea plantations, they can readily jump to new hosts on ecological timescales. The practicality, feasibility, and implications under the guidance of non-profit organizations or smallholdings producing and distributing FBCAs based bio-pesticides in developing countries should be explored further. Likewise, exploitation of *B. bassiana, M. anisoplae, A. lacani,* for management of pests and *Trichoderma* for management of major diseases in the tropical and subtropical tea-growing countries is a potential area of research. Besides, research should also be focused on identifying bioactive compounds from the potential FBCAs and their subsequent deployment for tea pests' management.

In conclusion, it is evident from the reviewed literature that FBCAs, such as *Beauveria, Metarhizium, Cordyceps, Trichoderma,* etc., holds a great possibility for the formulation of bio-pesticides and their subsequent use in tea ecosystem against pests and diseases as well as for growth promotion. The use of FBCAs is safe, natural, and economical compared to other management strategies. The early registering and patenting of the fungicidal/insecticidal properties of these FBCAs would ensure future deployments of these potential bio-pesticides that form a part of the rich microbial diversity of the tea ecosystem.

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