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Preliminary inventory of native natural enemy species of the new pest; the Fall Armyworm, *Spodoptera frugiperda* (J.E. Smith)

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Abstract

Fall Armyworm (*Spodoptera frugiperda*), which began to spread in Bhutan in recent years, is a highly destructive pest of maize that is native to the Americas. Due to the limited information available for developing sustainable management options for the new pest, current management practices are largely chemical-dependent. For use as a biological control option, we present a preliminary list of native natural enemies associated with Fall Armyworm that is collected from infested maize fields in west-central Bhutan. Visual observation, sweep net collection, and rearing of different Fall Armyworm stages were used for detection of the natural enemies. We identified 48 natural enemy species associated with the Fall Armyworm infested maize fields. Parasitism was noted for *Chelonus formosanus, Trichogramma* sp., *Winthemia trinitatis, Linnaemya* sp. and *Campoletis sonorensis* while predation was observed by *Polistes olivaceus, Stenodynerus smilibaronii, Tetraponera allaborans, Crematogaster rogennhoferi* and *Dolichoderus thoracicus* on the Fall Armyworm.

Keywords: Fall armyworm, native, parasitoids, predators, west-central Bhutan

Introduction

Fall Armyworm (FAW), *Spodoptera frugiperda* (G.E. Smith) is a highly destructive (Day *et al.*, 2017) lepidopteran pest. FAW is a highly mobile (Westbrook *et al.*, 2016)) pest that has spread to other parts of the world (Goergen *et al.*, 2016; CABI, 2020; Nagoshi *et al.*, 2020) from the Americas. It typically occurs in tropical regions with annual temperature ranging from 18 to 26 ºC and 500 to 700 mm annual precipitation (Early *et al.*, 2018). FAW migrates to warmer areas for overwintering (Sparks, 1979) becoming a sporadic pest (Hardke *et al.*, 2015). Polyphagous in nature (Hoy, 2019), it feeds on more than 350 plant species (CABI, 2020) including economically important crops such as maize, wheat, potato and rice (Navasero *et al.*, 2019; CABI, 2020).

In Bhutan, maize is widely grown as a staple crop closely associated with household food security (Katwal, Wangchuk, Dorji, 2013; DAMC, 2019). Maize has diversified uses such as animal feeds, poultry feeds, and human consumption (such as Kharang, Tengma, cornmeal and flour) besides other uses (DAMC, 2019). Maize plants are sensitive to insect pest infestations despite their high genetic yield potential (Khatri *et al.,* 2020).

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In 2020, Mahat *et al*. (2020) reported FAW in the country plausibly C-strain (Pashley, 1986) that infests maize. It can suddenly become a major pest (De Groote *et al.,* 2020) due to its short development period and high fecundity (Navasero and Navasero, 2020) posing a significant threat to the country's food security. Presence of ideal climatic conditions and maize -based cropping systems present in many parts of the country further aggravates the threat.

Current FAW management practices in Bhutan largely include chemical insecticides as practised in most of the recently invaded areas (Guo *et al.*, 2020). The injudicious use and reliance on agrochemicals results in reduced efficacy of natural pest control processes (Harrison *et al*., 2019; Mantzoukas and Eliopoulos, 2020), while also leading to the development of insecticide resistance (Gutirrez-Moreno *et al.*, 2019). Further, the usual practice of chemical spray is when the larvae are at a late stage where contact pesticides are not effective (Goergen *et al.,* 2016). The farmers' inclination towards chemical pesticides as the first choice of control indicates the need to identify and provide sustainable control options rooted in the country's organic policy/vision (DoA, 2019).

The integrated pest management (IPM) concept mainly focuses on the use of the natural enemy of the pest as a biological control method that is environmentally friendly and sustainable (Khatri *et al.,* 2020). Natural enemies such as predatory arthropods and parasitic wasps play important role in managing arthropod pest populations (Hajek and Eilenberg, 2018) in agro-ecosystems (El-Wakeil and Volkmar, 2013; Ali *et al*., 2018). For use in IPM and enhancing current FAW management strategies, we present a preliminary list of natural enemies in west-central Bhutan.

Materials and Methods

Study site and survey period

Sampling of natural enemies was done from Bajo (27°29'21.4"N, 89°53'53.6"E), Dzomlingthang (27°36'10.3"N, 89°51'11.7"E) and Tashidhing (26°56'21.0"N, 90°00'06.0"E) in west-central Bhutan. In Bajo, sampling was done in research plots whereas in Dzomlingthang and Tashidhing sampling was done in farmers' fields. The survey was conducted during the vegetative to maturity stage of maize from February to May 2021.

Sampling methods

Maize plants were observed for eggs starting at the early vegetative stage, which coincided with a number of moth catches in surveillance traps. Predation activities by natural enemies were monitored during morning hours (9 to 11 am) in the fields. Different stages of the FAW were collected and reared in laboratory until nymphosis. Egg clusters were randomly picked from the sites and put in boxes to observe the hatching of egg parasitoids. The freshly hatched neonate larvae (10 random batches) and bigger larvae (mostly suspected to be infested by parasitoids) were collected and fed with fresh maize leaves until nymphosis in the laboratory. Pupae were collected from soil and put into boxes until the emergence of adults. Fabricated insect boxes were used for insect rearing. Adult moths from sex pheromone traps were collected and kept in boxes to detect the emergence of natural enemies. A sweep net was used for collecting the potential beneficial insects associated with the FAW infestation in maize fields, with five complete sweepings at random points in 'W' or 'X' patterns. Samples were preserved using 85% alcohol (Siregar *et al.,* 2017) or as dry (Upton and Chapman, 2010). For identification, the specimens were sorted, observed under a light microscope, and matched with key descriptors available in published documents. The species recorded were categorized into different orders, followed by the nature of roles as parasitoid and predator.

Results and Discussion

Diversity of natural enemies of maize found in the study is presented in Table 1. A total of

48 native natural enemies were associated with FAW infested maize fields. Natural enemies were from the orders of Coleoptera, Diptera, Hemiptera, Hymenoptera, Mantodea, and Neuroptera (Figure 1.) The highest diversity of species was observed at Bajo (40) followed by Punakha (13) and Dagana (6). Dagana and Punakha sites have a history of pesticide spray as per information provided by farmers during the survey. Pesticide affects species diversity (Ndakidemi *et al*., 2016).

Parasitoids

Hymenopterans were the largest group of parasites distributed in eight families. Two tachinid flies were also recorded. We observed *Trichogramma* sp. attacking *S. frugiperda* egg masses in Wangdue. Similarly, *Chelonus formosanus* was found laying eggs on the FAW eggs at Punakha and Wangdue. Using sweep nets, we collected adults of *Telenomus* spp., *Stauropoctonus* sp., *Amyosoma* sp., *Aleiodes, Glyptapanteles, Brachymeria* spp., *Eriborus argenteopilosus, Epitranus* sp., *Eurytoma* sp., *Vulgichneumon saturatorius, Ischnojoppa luteator* and *Alomya debellator* with the largest species count at Wangdue. In the laboratory, *Campoletis sonorensis* and *Winthemia trinitatis* emerged from larvae while *Linnaemya* sp. emerged from moth carcasses.

Releasing *Trichogramma pretiosum* was reported to increase maize yield by about 19.4% (de Lourdes Corrêa Figueiredo *et al*., 2015). *Telenomus* spp. (remus) reportedly has great potential for biological control (Liao *et al*., 2019) of FAW in the field (Cave, 2000) with parasitism reaching about 90% through inundative release (Ferrer *et al.,* 2001). *Chelonus formosanus* is an egg and larval parasitoid of *S. frugiperda* amenable to laboratory mass production and a potential biocontrol agent for use in the field (Gupta *et al*., 2020).

The *Linnaemya* spp. parasitizes lepidopteran larvae including Fall armyworm (Molina-

Table 1: Beneficial insects associated with FAW infested maize fields

District	Detection Method	Observed Species
Wangdue	Rearing	*Winthemia trinitatis, Linnaemya* sp., *Campoletis sonorensis*
	Visual	*Crematogaster rogennhoferi* (Mayr, 1979), *Dolichoderus thoracicus., Trypoxylon petiolatum, Chrysoperla rufilabris*
	Sweep net	*Megalocaria dilatata* Fabricius (1755), *Coccinella septumpunctata* Linnaeus (1758), *Harmonia octamaculata* Fabricius (1781), *Coelophora bissellata* Mulsant (1850), *Cryptogonous quadriguttatus* Weise (1895), *Micraspis univittata, Oenopia sauzeti* Mulsant (1866), *Propylea luteopustalata, Paederus* sp., *Geocoris* sp., *Ichthyurus* sp., *Nabis capsiformis* Germar (1838), *Scipinia horrida* Stal (1859), *Aleiodes* sp., *Glyptapanteles* sp., *Chelonus formosanus, Brachymeria perflavipes, Brachymeria* sp., *Epitranus* sp., *Tetraponera allaborans* Walker (1859), *Ichneumon* sp., *Vulgichneumon saturatorius, Alomya debellator, Amyosoma* sp., *Eriborus argenteopilosus, Stauropoctonus* sp., *Telenomus* sp., *Trichogramma* sp., *Polistes olivaceus* De Geer (1773), *Ropalidia aritfex, Ropalidia* sp., *Sceliphron destillatorium, Ischnojoppa luteator, Ammophila* sp., *Tetraponera allaborans, Stenodynerus smilibaronii* (Ma & Li, 2016)
Punakha	Visual	*Coccinella septumpunctata, Micraspis univittata, Tetraponera allaborans, Ichthyurus tridens, Dolichoderus thoracicus, Polistes olivaceus, Ammophila* sp., *Aleiodes* sp.
	Sweep net	*Chelonus formosanus, Geocoris* sp., *Glyptapanteles* sp., *Trichogramma* sp.
Dagana	Visual	*Coelophora bissellata, Coccinella septumpunctata,*
	Sweep net	*Glyptapanteles* sp., *Scolia* sp., *Polistes olivaceus, Ammophila* sp.

Ochoa *et al*., 2003; Bixby-Brosi and Potter, 2012; CABI, 2021e; Fite *et al*., 2021;) while *Winthemia trinitatis* parasitizes the fifth and sixth instar larvae of *S. frugiperda* (Prasanna *et al*., 2018) and contributes to the reduction of future pest generations.

Five species of *Campoletis* were previously reported as parasites of FAW (Hoballah *et al*., 2004; CABI, 2020) but we found only *C. sonorensis* in this study. Ophioninae such as *Ophion flavidus* is a natural enemy of *Spodoptera frugiperda*, *Autographa californica*, *Mythimna unipuncta*, *Peridroma saucia*, and *Spodoptera eridania* (CABI, 2021g) but there are no reports of *Stauropoctonus* spp. parasitizing FAW.

Species recorded under genera Aleiodes, Glyptapanteles, and Brachymeria are parasites of FAW (CABI, 2020). Actual parasitism for other species collected through sweeping is not known. But these species may also be potential parasites as these are natural enemies of other lepidopteran pests as indicated in different studies. *Amyosoma* sp. parasitizes cereal stem borers (Achterberg and Polaszek, 1996; Rahmani *et al*., 2020). *Eriborus argenteopilosus* was reported as the natural enemy of some species under the genus *Spodoptera* (CABI, 2021a). Chiranjeevi and Patange (2017) reported *Epitranus* sp. and *Eurytoma* sp. as parasites of *Melanagromyza obtusa* (Malloch) larvae and pupae. *Eurytoma* is a natural enemy of some lepidopterans like *Helicoverpa armigera* and *Uraba lugens* (CABI, 2021b). *Epitranus* sp. was also reported to parasitize some lepidopterans (pupal stage) such as crambid, pyralid and tineid moths (Binoy *et al.,* 2020). *Vulgichneumon saturatorius* is natural enemy of *Mamestra brassicae* (CABI, 2021i). *Ischnojoppa luteator* parasitizes *Cnaphalocrocis medinalis*, *Marasmia exigua*, *Marasmia patnalis*, *Pelopidas mathias*, *Scirpophaga incertulas*, and *Scirpophaga innotata* (CABI, 2021d). Scolid wasps parasitize scarab beetle larvae (Gupta and Jonathan, 2003) while *Alomya debellator* parasitizes hepialid moths (Cameron, 1950).

Parasitism was low which might have resulted from infection by entomopathogens and predation by generalist predators as reported by Firake and Behere (2020). Determination of the rate of parasitism by these parasites depends on the architecture of different stages of *S. frugiperda* and other factors require detailed investigation for adoption and use in the IPM for FAW in Bhutan.

Predators

Predators were largely Coleopterans distributed in families of Coccinellidae (8), Cantharidae (2) and Staphylinidae (1). Hymenopterans were the second largest group with four Vespidae, three Formicidae and two Sphecidae. *Polistes olivaceus* was observed feeding on larger instars of Fall Armyworm at Wangdue. Adult *Stenodynerus smilibaronii* (Ma and Li, 2016), *Tetraponera allaborans*, *Crematogaster rogennhoferi* (Mayr, 1979), and *Dolichoderus thoracicus* were found feeding on neonate larvae at Punakha. The other listed predators were collected in sweep nets.

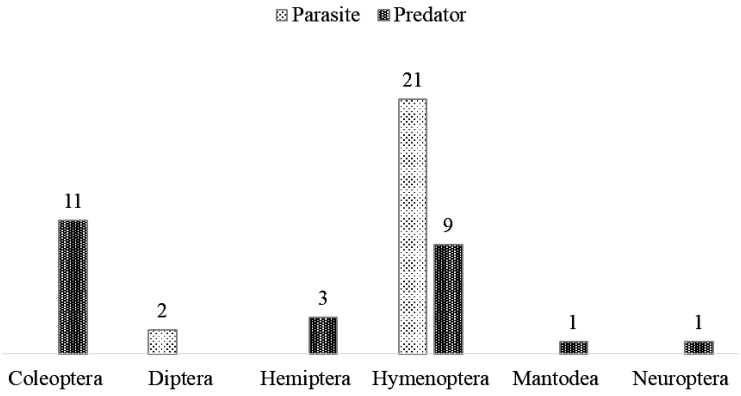

Figure 1: Natural enemy species counts in different orders

Geocoris punctipes preys on FAW (CABI, 2021c) but we netted a different species at Punakha. *Geocoris erythrocephalus* is reported to feed on cabbage aphids (Rajan *et al*., 2018) and there was no information on FAW predation. *Nabis capsiformis* also preys on this pest (CABI, 2021f). Gint-

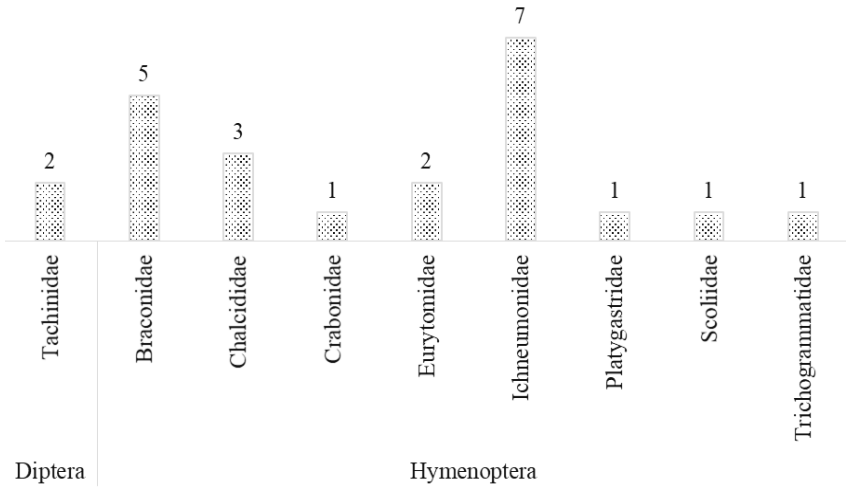

Figure 2: Parasite species distributed under different families

ing *et al*. (2021) reported *Coleomegilla maculata* as FAW natural enemy while Kalleshwaraswamy *et al.* (2019) observed *Harmonia octamaculata* and *Coccinella transversalis* abundantly in FAW infested maize fields. In this study besides these two species, we recorded six other coccinellid beetles associated with FAW infested maize fields.

Some *Tetraponera* sp. are reported to prey on Mosquito bugs (Beevi and Mahapatro, 2008) and *Hypsipyla robusta* (CABI, 2021h). Besides these, there are trophic associations with aphids (Gull-E-Fareen *et al*., 2020) and lac insects (Rajagopal and Manimozhi, 2005). Other predatory species collected in sweep net from different maize fields were generalist predators. Cantharid beetles prey on different kinds of insects (Pérez-Hernández, 2018) and we recorded two cantharid beetles in this study. Although actual predation by these entomophagous coleopterans was not sighted during our study, the immature stages of FAW might be preyed upon (Evans, 2009). *Paederus* sp. is known to predate on Diptera and other arthropods (Frank and Kanamitsu, 1987).

Casiraghi *et al.* (2001) reported that *Ammophila* (sp.: sabulosa) preys on caterpillars of Noctuidae, Geometridae, and Notodontidaes. Preys of *Polistes* spp. largely constitute lepidopteran larvae (Jeon *et al*., 2019; Parent *et al*., 2020). *Mantis religiosa* preys on diverse preys (Mook and Davies, 1966; Merlin Kamala and Kennedy, 2017). *Chrysoperla rufilabris* feeds on different arthropods (Hoballah *et al*., 2004; Alghamdi and Sayed, 2017; Samal and Sahu, 2021).

Biological control methods that have the advantage of not affecting non-target organisms like pollinators and natural enemies (Harrison *et al*., 2019) should be opted for Biological control using entomopathogenic fungi (Mantzoukas and Eliopoulos, 2020) which is one of the most effective non-chemical pest control options. The parasitism rate of different parasitoids and predation potential of the predatory species may be studied to assess their potential for use in biological control options for Fall Armyworm management. Research on the natural entomopathogens is recommended which the current study did not cover. Also, agricultural simplification at the landscape scale reduces parasitism rates of crop pests (Grab *et al.,* 2018) while non-crop (Gagic *et al*., 2018) vegetation and field borders provide refuge to natural enemies (Clem and Harmon-Threatt, 2021).

Conclusion

This study indicates that there is a good diversity of natural enemies and other beneficial insects for Fall Armyworm in west-central Bhutan. The first-hand information on the FAW's native natural enemies will be valuable information for the development of sustainable control/management strategies. However, parasitism rate and predation potential of different species were not studied. Limited by facilities of the centre and the capacity of the authors, no effort was made to report entomopathogens although a few incidents of lar-

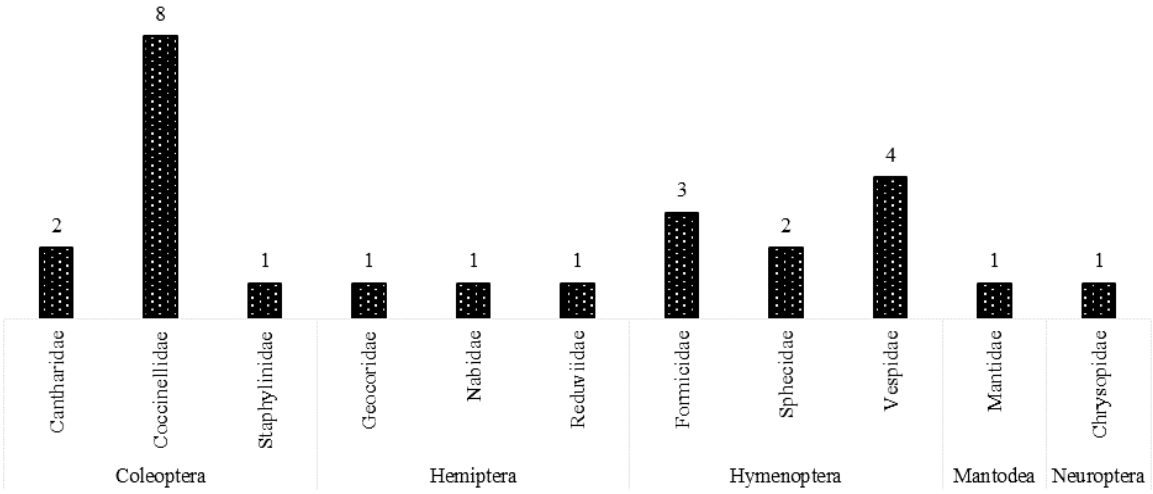

Figure 3: Predator species under different families associated with FAW infested Maize fields

vae were suspected of infection by entomopathogens. Green muscardine fungus and nuclear polyhedrosis virus were the suspected entomopathogens based on the symptoms of the affected carcases.

Although Bhutan currently has limited resources to develop biocontrol programmes, efforts to increase the population of these native natural enemies could prove useful. Further, agro-ecological options for Fall Armyworm management such as intercropping with appropriate companion plants and diversifying farm environment through the management of seminatural habitats at multiple spatial scales could be considered.

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