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A review on entomopathogenic nematodes as potential bio-pesticide in North Eastern States of India

Ankita Dey[1], Sobita Singha[1], Piyush Pandey[1], Baby Singha[2*]
[1] Department of Microbiology, Assam University, Silchar, Assam, India
[2] Department of Zoology, Gurucharan College, Silchar, Assam, India
[*]Corresponding Author: babysingha@gmail.com

Abstract
Background: Over 90% of bioinsecticide market dominated by Bacillus thuringiensis (BT) but with time insects developed resistance against BT toxins. The menace of pesticides and demand for organically produced agricultural products has forced the Indian Government to promote organic agriculture in the North-eastern states through special schemes and programs such as 'Technology Mission for North East and through setting up of a special Agricultural Export Zone. Two bacteria belong to the genera *Photorhabdus* and *Xenorhabdus*, which are symbiotically associated with entomopathogenic nematodes of the families Heterorhabditidae and Steinernematidae respectively can parasitize insects, and are widely used worldwide for insect-pest management in different agro-ecosystems. Moreover, no resistance against *Xenorhabdus* or *Photorhabdus* has been reported to date. Hence, in the absence of insecticides, Entomopathogenic Nematodes (EPNs) and their symbiotic Entomopathogenic Bacteria (EPB) are the best and most useful agents for the management of insect pests ranging from Lepidoptera, Diptera to Coleoptera. Here, we present a brief review of the various species of EPN and EPB isolated from soils of North East India and the current trend of research on the efficacy of EPN as bio-pesticide in northeastern agro-climate.
Results: Reports on EPN research and its uses as bio-pesticide are available only from 4 out of 8 states of North East India. A total of 24 EPN species comprising14 from Assam, 5 from Meghalaya, 4 from Mizoram, and 1 from Manipur were identified under three genera- *Heterorhabditis* (7 spp.), *Steinernema* (15 spp.), and *Oscheius* (2 spp.). Studies on efficacy testing reported that EPN species *Heterorhabditis indica* showed great efficacy against tea mosquito bug, tea termite, white grubs, tobacco cutworm, mustard sawfly, and greater wax moth in Northeast India. This encouraging current status of EPN research in NE India will provide a footing ground for further exploration, isolation, characterization, and biosystematics of EPNs and their symbiotic bacteria (EPBs) from the soils of varied agro-climatic conditions of this region.
Conclusions: We conclude that isolation together with testing of the efficacy of EPN strains through massive field trials against pests of seasonal crops are essential to realizing the potential of EPNs formulations and EPBs bioinsecticide as potent biological pest management tools to promote organic agriculture in North East India.

Keywords: entomopathogenic nematode, *heterorhabditis*,*steinernema*, *oscheius*, organic agriculture, biopesticide

Introduction
Entomopathogenic nematodes (EPN) are recognized as the lethal parasites of insect larvae which are widely used as a biocontrol agent against many important insect pests of vegetable crops. Therefore EPNs are grabbing the attention of entomologists as well as nematologists in the field of bio-control research worldwide (Kalita *et al*., 2019; Devi *et al*., 2017; Ganguly, 2006) [18, 10, 16]. EPNs represent a group of soil-inhabiting nematodes that parasitize and kill a wide range of host insects within 24-48 hours of infection and are harmless to plant and non-target organisms. These nematodes belong to two families viz., Steinernematidae and Heterorhabditidae which have a mutualistic partnership with strain-specific anaerobic bacteria, and together they kill a wide range of insect species. *Xenorhabdus* bacteria associated with *Steinernema* and *Photorhabdus* bacteria associated with *Heterorhabditids*. Nematodes after entering into the host insect body release their symbiotic bacteria into the hemolymph which kills the insect by massive septicemia. The bacteria degrades insect-larval tissues making it as the food source for the nematodes. It facilitates them to mature and multiply. Usually, 1 or 2 generations of adult nematodes are produced within the insect cadaver. The progeny of the last adult generation reassociates with few bacterial cells and nurture them in their intestine. Then nematodes move out of the insect cadaver into the soil and wait for another insect to parasitize (Ganguly, 2006) [16].
Several studies have been conducted to isolate and identify EPN strains from across NE India, many of which are new to this part of India. The earliest report by Nematologists on EPNs was the isolation of *Steinernema* and *Heterorhabditis* from around the root zones of different plants across Assam (Deuri *et al*., 2000) [5]. Studies on

EPNs from the Northeast can be traced back to 2010 when Promodini and Mohilal reported for the first time on the occurrence of Heterorhabditis strain from Manipur. In the same year, the author of the present paper reported three strains of EPN viz., *S. carpocapsae* (GenBank Acc. No. MH204152), *S. surkhetense* (MG 976890), and *Steinernema sp*. (MG976891) from Cachar district of Southern Assam through molecular and microscopic assays. Similarly, a report on the isolation of another new species *Oscheius indicus* also came from the Cachar district of Assam (Kumar, *et al*., 2019) [21].

Even more encouraging was the report of successful isolation of a new species of EPN, *Steinernema sangi* and its symbiotic bacteria *Xenorhabdus vietnamensis* from Mizoram for the first time in North East India (Lalramnghaki *et al*., 2017) [22]. Following that success, *Heterorhabditis baujardi* was also reported for the first time from Mizoram, which was originally described in Vietnam (Vanlalhlimpuia *et al*., 2018) [31]. Nematologists from Mizoram contributed greatly to EPN research by identifying three EPN associated bacteria (EPBs), *viz*., *Xenorhabdus vietnamensis*, *Xenorhabdus stockiae*, and *Photorhabdus luminescens* which could produce a great potency against pathogenic fungi, *Fusarium oxysporum* (Lalramchuani *et al*., 2020) [23].

Many studies, especially from four states of the North East confirmed the potency of the isolated EPNs against one or the other pests. Although *Heterorhabditis indica* is the first indigenous isolate of Meghalaya which showed potency against Greater Wax Moth, *Galleria mellonella* (Lalramliana *et al*., 2005) [14], *Steinernema meghalayensis* is the first new species of this genus isolated in Meghalaya whose efficacy was tested also against *Galleria mellonella*. (Ganguly *et al*., 2011) [15]. From Assam Borgohain (2015) [4] isolated and identified as *Heterorhabditis sonorensis*. *Heterorhabditis bacteriophora* isolated from Assam proved to be potential against agricultural pest White grubs, *Lepidiota albistigma* (Devi *et al*., 2016 and 2020) [6]. *H bacteriophora* and *H. indica* from Assam also exhibited great potential in killing pests like tea mosquito bug and tea termite respectively (Amuri *et al*., 2020; Singha *et al*.,2014) [1, 2, 19].

The aim of this paper is to review the studies on EPN undertaken in North East India in the last 20 years in order to assess the trend of study of EPNs to assess the diversity and potential in insect pest management in this part of India. Accordingly vigorous Journal and literature searches were made utilizing all available resources including Search Engines For Academic Research' like Research Gate, Google Scholar, Science Direct, Shodhganga, etc. Sincere attempts have been made to merge the investigations that were carried out to see the diversity, distribution, and efficacy of EPNs in Northeast India with a view to understanding the potential of EPNs as bio-pesticide in this agriculturally diverse land of India. Through this piece of work, the authors also tried to throw light on the prevailing knowledge gap in EPN research towards its successful implementation as a potential tool in biological pest management in North Eastern agro-ecosystem and suggesting some possible strategies to reach the goal.

EPN Systematics (Ref: Ganguly, 2006; Sharma *et al* 2016, Devi *et al*.; 2017)
Phylum: Nematoda
Class: Secernentea
Order: Rhabditida
Suborder: Rhabditina
Superfamily: Rhabditoidea
Family: i) Steinernematidae ii) Heterorhabditidae iii) Rhabditidae
Genus: i) *Steinernema, Neosteinernema;* ii) *Heterorhabditis;* iii) *Oscheius*

Global reports on EPN Strains

A total of 100 species were recorded to date under Genus *Steinernema* while 21 species were recorded under *Heterorhabditis* from different countries of the world (Bhat *et al*., 2020). Genus Oscheius has been divided into two groups, viz., Insectivora and Dolichura. Under the Insectivora group, 31 species were reported while 14 species have been identified under Dolichura. Thus, a total of 166 EPN species have been reported across the World to date.

Studies on EPN in Northeast India

There are many reports on the isolation and characterization of EPN from Northeastern states. EPN strains identified across North East India till date tabulated in table 1

EPN reports from Assam

The soil of Assam rich in entomopathogenic agents. Ample of reports on isolation and molecular characterization of EPN available from this state. In a survey conducted in Sorbhog, Barpeta, Assam, EPNs were isolated cadavers of white grubs, *Lepidiota albistigma*. 10 samples tested EPN positive out of 100 cadavers. Based on morphological and morphometrical studies, the isolated nematode was identified as *Heterorhabditis bacteriophora* (Devi *et al*., 2020) [1, 2]. Similarly, there are reports of isolation of EPNs from 1 out of 200 soil samples from tea plantation areas of Jorhat through baiting technique using Galleria larvae. A new strain of EPN *Heterorhabditis bacteriophora* was identified through comparative Morphological and Morphometrical analysis (Amuri *et al*., 2020) [1, 2]. In another study by the same author, *Oscheius chongmingensis* was isolated from the tea garden soil sample. Identification was based on morphological similarities like body length, body width, anterior end to excretory pore, anterior end to nerve ring, anterior end to esophagus base, tail length, anal body

width, and distance from anterior end to the vulva as a percentage of length, etc. as described by Zhang *et al*., 2008 from Chongming Island in eastern China (Amuri *et al*., 2020) [1, 2]. Report of new species *Oscheius indicus* under Insectivora group obtained also from Cachar district of southern Assam based on morphometrical, morphological observations and molecular phylogenetic analysis. (Kumar, *et al*., 2019) [21]. Using baiting technique with Galleria larvae, Devi *et al*., (2019) [8] successfully isolated, characterized, and identified another strain *S. kushidai* from 1 out of 100 soil samples from the Golaghat district. Kalita *et al*. (2019) [18] identified two strains *viz*., *Heterorhabditis bacteriophora* and *Oscheius chongmingensis* based on the morphological and morphometric character from 8 positive soil samples out of 200 collected from Assam Agricultural University Campus. The first report of EPN from Barak Valley of southern Assam was the isolation and identification of *Steinernema sp.* from rhizospheric soil of Bamboo and Crop field during rabi and monsoon season (Sharma *et al*., 2016, 2018) [27, 28]. In another soil EPN survey conducted by Singha *et al*. (2019) [30], 2 out of 100 garden soil samples were found EPN positive. Based on molecular characterization the EPN Genus was identified as *Heterorhabditis* (Singha *et al*., 2019) [30]. In 2010 the author reported the occurrence of a strain of Steinernema from the soil of Cachar, Barak Valley which was subsequently characterized through molecular and microscopic assays as *S. carpocapsae* (Fig.1) (Acc. No. MH204152). Singha *et al*. also successfully isolated two more isolates of Steinernema from their subsequent investigations which are identified and submitted to the Gen Bank as *S. surkhetense* (MG 976890) and as *Steinernema sp.* (MG976891). These are the known records of occurrences of EPN of the genus *Steinernema* from the soils of Cachar district, Barak Valley, Southern Assam (Sharma R. 2018) [27, 28]. Very recently, another local strain *H.indica* has been characterized from the Cachar district of Assam through microscopic (Fig.2) and molecular approaches, and subsequently its symbiotic bacterium *Photorhabdus sp*. has also been confirmed through culture (Accession no. MF417383, Nath *et al*., 2021) [25]. Devi *et al*., (2017) [10] conducted a survey across Assam and collected 100 soil samples. Out of these only 5 samples were found to be EPN positive, which comprises of 2 from Barak Valley, 2 from hill areas, and 1 from Upper Brahmaputra Valley. Based on morphological and morphometrical analysis three strains were identified as *Steinernema abbasi, S. ceratophorum*, and *S. tami*. The author also successfully identified EPNs Heterorhabditis and Steinernema in Soil and dead white grub *Lepidiota mansueta* from the white grub endemic field of Majuli, the world's largest river island. Moreover, morphometric and cross-breeding studies *Heterorhabditis* isolates were identified as *H. bacteriophora* (Devi *et al*., 2016) [6]. With the objective to identify EPNs across Assam Borgohain (2015) [4] collected 305 soil samples from various regions of Assam. He identified four EPN species *viz*., *Steinernema abbasi, S. karii, Heterorhabditis bacteriophora*, and *H. sonorensis* from 45 EPN positive soil samples. Interestingly, this was the first report of *H. sonorensis* from India. According to the author, the sample positivity rate was found to be highest in Karbi Anglong while lowest in the Dhubri district of Assam. With the aim to isolate indigenous EPN Deuri *et al*. (2000) [5] reported a large number of *Steinernema* and *Heterorhabditis* species around the root zones of different plants across Assam.

Studies on testing of the efficacy of EPN conducted on live wood-eating termites found abundantly across North-eastern tea estates, which cause nuisance by eating young and mature tea leaves. Field trials conducted in Assam tea gardens to check the potency of bio-pesticide against the termites in tea plantations indicated that EPN strain *Heterorhabditis indica* is effective both alone and IPM method in controlling the pest in comparison to the commonly used chemical thiamethoxam (Roy *et al*., 2020) [26]. Singha *et al*., (2014) [29] conducted a study of laboratory efficacy and potency of two EPN spp. *Heterorhabditis indica* and *Steinernema thermophilum* (obtained from Nematology Division, IARI, New Delhi) against tea termites and found that although both were efficient against the pest, *Heterorhabditis indica* exhibited higher potency.

EPN Reports from Mizoram

The first report of new species of EPN *Heterorhabditis baujardi* which was identified through morphological and multigene (ITS rRNA, 28S rRNA, and COI) sequence characterization also came from Mizoram. This species was originally described from Vietnam (Vanlalhlimpuia *et al*., 2018) [31]. Phylogenetic analysis revealed that *H. baujardi* belongs to *H. indica* clad. Following this report, Lalramnghaki *et al*., (2020) [24] isolated indigenous EPNs from fertile lands of Mizoram which include *Heterorhabditis indica, Heterorhabditis baujardi*, and *Steinernema sangi*. Although all species are highly effective against 3rd, 4th, and 5th instar larvae of tobacco cutworm, *Spodoptera litura*, *S. sangi* showed the highest pathogenicity against the pest in terms of LC50 at 48 hr post-inoculation. The pioneer and sole report of EPN associated bacteria (EPB) also came from Mizoram. Three identified EPB strain are *Xenorhabdus vietnamensis* from *Steinernema sangi*, *Xenorhabdus stockiae* from *S. surkhetense* and *Photorhabdus luminescens akhurstii* from both *Heterorhabditis indica* and *H. baujardi*. Out of three *X. stockiae* and *P. luminescens akhurstii* were reported for the first time from Mizoram. All three also showed great potency against pathogenic fungi, *Fusarium oxysporum* (Lalramchuani *et al*., 2020) [23]. This study reveals the possibility of future use of EPNs as potential biocontrol agents against many fungal diseases of agricultural crops. Morphological and molecular analysis (ITS rDNA for *Steinernema* and 16S rRNA for *Xenorhabdus*) revealed that *Steinernema sangi* is associated with the bacteria *Xenorhabdus vietnamensis* and also it is the first report from India (Lalramnghaki, 2017) [22].

EPN Reports from Meghalaya

Steinernema meghalayensis is the first new species of this genus isolated from the Eastern Himalayan region of India. Findings revealed that the new species can infect the larvae of *Galleria mellonella* and can induce

mortality, multiply (reproduce), and emerged from the cadavers within 6–8 days at a temperature ranging from 20–30°C (Ganguly *et al*., 2011) [15]. Three more indigenous EPNs *Heterorhabditis indica*, *Steinernema thermophilum* and *Steinernema glaseri* were also isolated from Meghalaya. However, out of three EPNs, *H. indica* and *S. thermophilum* showed great potency against mustard sawfly, *Athalia lugens* (Yadav, 2012) [36]. In another study of the efficacy of EPN, Yadav *et al*. (2012) [36] found that *S. glaseri* produced 100 % mortality in 48 hr against the last instar larvae of taro leaf beetle, *Aplosonyx chalybaeus* under laboratory condition. Devi *et al*. (2011) [11, 12, 13] also reported indigenous species *Steinernema carpocapsae* and Heterorhabditis indica from Jawai and Mawsynram towns of Meghalaya. Further, these species provide inconsistent suppression to Root-Knot Nematode Meloidogyne incognita on Tomato (Devi, 2011) [11, 12, 13]

Apart from efficacy studies against pests, a few studies were also conducted to assess the effect of temperature and moisture on the viability and virulence of local EPN strains. In one such study co-relation between storage temperature on viability and virulence was tested. Result showed 25 °C is the optimum temperature for storage. However, *S. glaseri* survived well in all temperature (5 ± 2°C and 25 ± 2 °C) at different storage durations (Yadav, 2016) [35]. Similarly, optimum soil moisture for different nematode species was reported as *H. indica* (8–18%), *S. thermophilum* (6–20%), and *S. glaseri* (8–25%). Further, this study also revealed that a minimum of 6% soil moisture is essential for all the three nematode species for achieving 100% host mortality (Yadav *et al*., 2012) [36]. The viability and effectiveness of two indigenous EPNs *Steinernema carpocapsae* and *Heterorhabditis indica* were tested using various spray adjuvants like Sunflower oil, Glycerol, Tween 20, Triton X-100, and Paraffin liquid at 1% concentration. The result showed the toxicity of Triton X-100 and Tween 20 were less compared to Paraffin liquid and Sunflower oil (Devi, 2011) [11, 12, 13]. Effect of agrochemicals (Monocrotophos, Topcin M, Glyphosate, Dicofol 18.5 E and Oxamyl (0.1%) and botanicals (pine seed, pine needle, plant parts of *Tithonia spp*., plant parts of marigold (1%)) on these indigenous species indicated that both the genera can withstand agrochemicals and botanicals for 24 h. Mortality rate directly proportional to exposure duration (Devi, 2011) [11, 12, 13]. The potency of EPN *Heterorhabditis indica* isolated from the forest soil of Meghalaya was tested at various temperatures and relative humidities (RH). The result indicated that approximately 100 IJs of *H. indica* used to infect *Galleria mellonella* larvae successfully at 25 °C and 100% RH. IJs could emerge only at 15, 20, 25, and 30 °C. The earliest emergence (8 days) of IJs occurred at 25 °C and at 85, 96 (Lalramliana *et al*., 2005) [14].

EPN Report from Manipur

There is only one report on EPN from this state. From a preliminary survey conducted in Manipur one EPN spp. *Heterorhabditis sp*. isolated from soil by using grasshopper following insect trap method (Maibam *et al*., 2010).

Table 1: List of identified EPN strains across Northeast India state wise till date

State	No. of species under Genus *Heterorhabditis*	No. of species under *Steinernema*	No. of species under Genus *Oscheius*	Total EPN sp. state wise
Assam	*Heterorhabditis bacteriophora* *H. sonorensis* *H. indica* (MF417383)	*Steinernemakushidai* *S. aciari* *S. abbasi* *S. ceratophorum* *S. tami* *S. karii* *S.carpocapsae* (MH204152) *S. surkhetense* (MG 976890) *Steinernema* sp. (MG976891)	*Oscheius chongmingensis* *O. indicus*	14
Mizoram	*Heterorhabditis indica* *H. baujardi*	*Steinernema sangi* *S. surkhetense*	0	4
Meghalaya	*Heterorhabditis indica*	*Steinernema thermophilum* *S. glaseri* *S. meghalayensis* *S. carpocapsae*	0	5
Manipur	*Heterorhabditis sp.*	0	0	1
Total	7	15	2	24

Table 2: List of indigenous EPN strains and their efficacy against local insect pests in NE India

Indigenous EPN species	Efficacy study against	Method of Isolation and Identification	Reported from	Reference
Heterorhabditis bacteriophora	White grubs, *Lepidiota albistigma.*(Sugarcane)	Isolated from dead White Grub; Morphological and Morphometrical	Sorbhog, Barpeta, Assam	Devi *et al*., 2020
Heterorhabditis	Tea mosquito bug,	Baiting with Galleria	Jorhat, Assam	Amuri and Devi, 2020;

bacteriophora and *Oscheius chongmingensis*	*Helopeltis theivora* and bunch caterpillar, *Andraca bipunctata*	larvae; Morphometric characterization		Kalita *et al*., 2019
Heterorhabditis bacteriophora and *Steinernema*	White grubs, *Lepidiota mansueta*	From Soil and White Grub; Morphometric and Cross-breeding studies	Majuli, Assam	Devi *et al*., 2016
Heterorhabditis indica	Tea termite	Baiting technique with Galleria larvae; Morphological characterization	Cachar, Assam	Singha *et al*., 2014
S. khushidai		Baiting technique with Galleria larvae; Morphometric characterization	Golaghat, Assam	Devi *et al*., 2019
Heterorhabditis indica, H. baujardi and *Steinernema sangi*	LC_{50} at 48 hr against Tobacco cutworm, *Spodoptera litura*.		Mizoram	Lalramnghaki *et al*., 2020
Steinernema sangi, S. surkhetense,	Pathogenic fungi, *Fusarium oxysporum*		Mizoram	Lalramchuani *et al*., 2020
Heterorhabditis indica and *H. baujardi*	-	Morphological and multigene (ITS rRNA, 28S rRNA, and COI) sequence characterization; Phylogenetic analysis	Mizoram Assam	Vanlalhlimpuia *et al*., 2018 Nath *et al*., 2021
Heterorhabditis indica and *S. thermophilum*	Mustard sawfly, *Athalia lugens* Taro leaf beetle, *Aplosonyx chalybaeus*		Meghalaya	Yadav, 2012
Steinernema meghalayensis	Greater Wax Moth, *Galleria mellonella.*		Meghalaya	Ganguly *et al*., 2011
Steinernema carpocapsae and *Heterorhabditis indica*	Viability and effectiveness tested using various spray adjuvants like Sunflower oil, Glycerol, Tween 20, Triton X-100 and Paraffin liquid at 1% concentration		Jawai and Mawsynram, Meghalaya	Devi, 2011
Heterorhabditis indica	*Galleria mellonella;* varying temperature and Relative Humidity			Lalramliana *et al*., 2005
Heterorhabditis sp.	Grasshopper	From Soil; Insect Trap Method	Manipur	Pramodini *et al*., 2010

Conclusion

Organic agriculture is on the rise in the Northeastern states of India. In the absence of insecticides for the management of insect pests in bio-farming, entomopathogenic nematodes hold a promise to be a useful and most effective tool for pest control. With more and more documentation and realization of their efficacy, EPN research is certainly going to gain momentum with the attention of entomologists, nematologists, agricultural scientists, and environmentalists alike. The number of studies on EPNs in north-eastern states is very few. Earlier work was confined to the isolation of EPN, therefore, a large number of strains isolated from the NE states are yet to be identified. To date, only 4 out of 8 states of North East India viz., Assam, Mizoram, Meghalaya, and Manipur could produce some reports on EPN research (Fig.4). The highest number of EPN species isolated from Assam (14 sp) followed by Meghalaya (5 sp.), Mizoram (4 sp.), and Manipur (1 sp.) Reports are also rare from Arunachal Pradesh, Tripura, Nagaland, and Sikkim. In India, previously 35 strains of EPNs were isolated from different geographical regions of India representing different agro-climate zones (Kumar et. al., 2015) [19]. However, a total of 24 EPN species already identified across the Northeast under three genera viz., 15 species under genus *Steinernema*, 7 under genus *Heterorhabditis*, and 2 species under genus *Oscheius* (Table-1; Fig.3). Significantly, no other state of NE India reported EPN of Genus *Oscheius*, and both the species identified and reported from Assam indicate that the genus may be endemic to this state.

In addition to the isolation and identification of 4 indigenous species of EPNs, the Mizoram is the only state to report regarding isolation and identification of EPN associated bacteria (EPB). Not less than three EPB strain were described viz., *Xenorhabdus vietnamensis*, *Xenorhabdus stockiae* from *Steinernema sangi*, *S. surkhetense* while *Photorhabdus luminescens akhurstii* from *Heterorhabditis indica* and *H. baujardi*. Adding to the credit is the finding that these EPBs can effectively be applied against pathogenic fungi, *Fusarium oxysporum*. This report will undoubtedly open a new research avenue towards utilizing the EPNs against many fungal diseases of crops and vegetables.

Significantly, the trend of EPN research in NE India in the last 20 years indicates that in 2019 alone a total of 8 species have been reported from Assam (Fig.5). However, despite this encouraging trend it is surprising to observe that apart from isolation and identification of EPNs and laboratory efficacy testing, there was no such trial made in the agricultural field to test the efficacy of particular EPN or their combinations against pests of seasonal vegetables and crops. Therefore the present paper identifies the most important gap in the field of EPN research in NE India. It can now be realized that EPN research should go hand in hand with current Integrated Pest Management strategies in order to obtain greater success in Organic farming in NE India. In spite of the knowledge that local EPN strains are better adapted to local agro-climatic conditions than exotic EPN strains and will have better biofarming potential against seasonal vegetable pests, exploitation of such soil treasure towards biological pest management is still in its infancy.

Therefore, we suggest that acceleration of collaborative research efforts among Agricultural Universities, other state and central Universities, and the Research Institutes of NE India is the only alternative to pave the way for establishing and promoting EPNs as the most useful bio-control agent and fill the prevailing knowledge gap. Industry-Academia exchange Programmes together with mass awareness of farmers is the key to substantially encourage both mass culture and production of local EPNs and their associated bacteria as well as their applications in the seasonal and vegetable crop fields in order to realize the potential of EPNs formulations as potent biological pest management tools to promote organic agriculture in North East India.

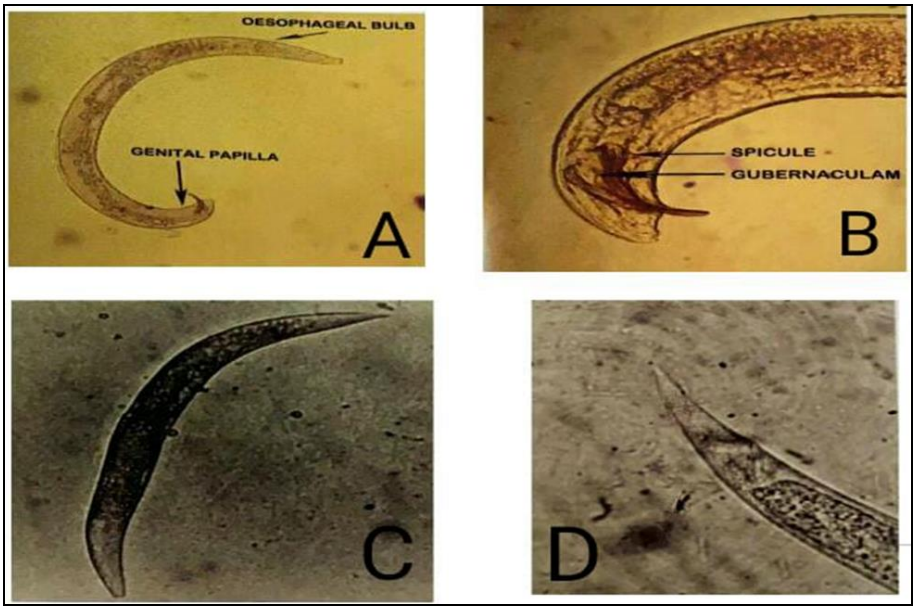

Fig 1: Light microscopic images of - A) Adult Male 1st generation (B) Spicule and Gubernacula of male of 1st generation (C) Infective juvenile (D) Tail region of infective juvenile of *Steinernema carpocapsae* (Adopted from Sharma *et al*., 2018)

Fig 2: Light microscopic images of- (A) Infective juveniles (10X) (B) 1st generation hermaphrodite (40X) of *Heterorhabditis indica* (Adopted from Nath *et al*., 2021)

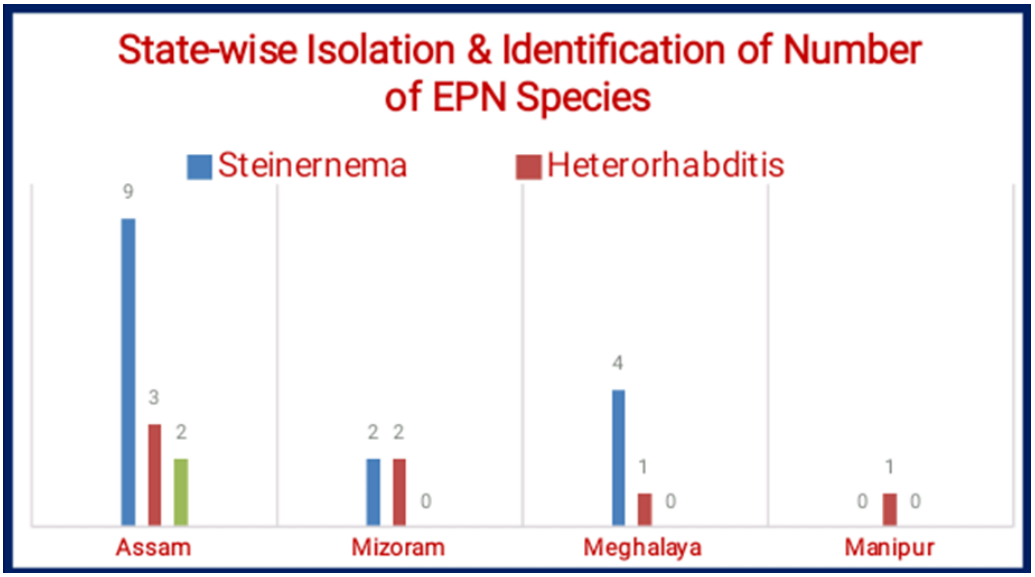

Fig 3: State-wise presentation of number of EPN species identified under three predominant genera - *Heterorhabditis*, *Steinernema* and *Oscheius* across Northeast India.

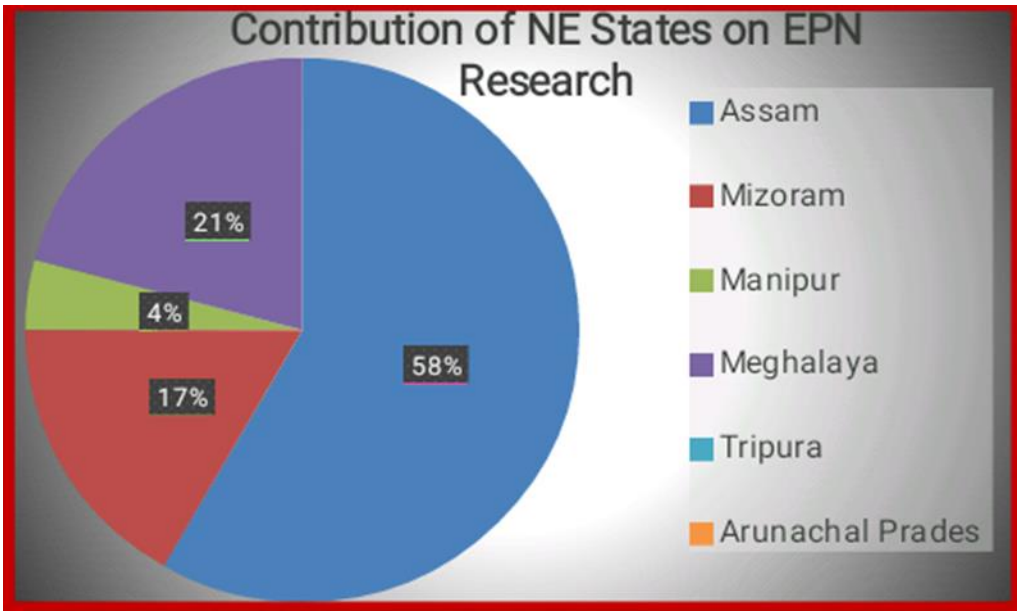

Fig 4: Represents the contribution of various states of NE India in EPN research

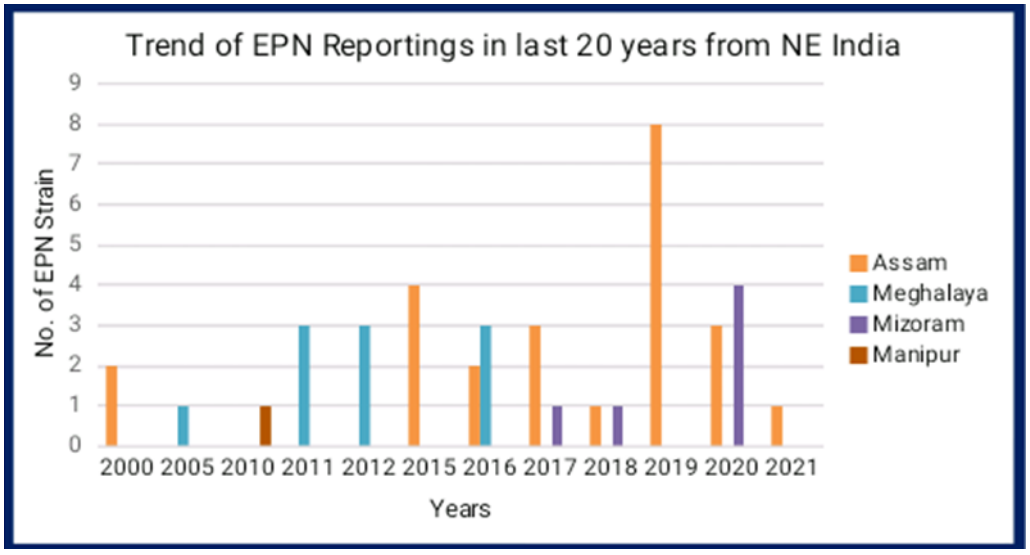

Fig 5: Trend of EPN Research in NE India in past 20 years

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