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## Review on microbiota in fruit piercing moths specifically from erebidea (Noctuoidea: Lepidoptera)

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### Abstract

Insects belong to order Lepidoptera include moths, comprising approximately 1,60,000 species acts as a major crop pest of the world. Many insects play as host to different microbes by forming close mutualistic relationships. The study on this topic reveals bacterial symbiont play important role on host's biology, it may impact on host's digestion, immunity, anti-pathogenic responses, nutrient absorption of host food etc.; Fruit piercing moths are common agricultural pest which damages the commercially important fruit crops such as moth of genus *Othreis* as a harmful pest on pomegranate, citrus and orange mostly found in Maharashtra state. Most of the moth vertically transmit the endosymbiotic microorganisms, which can affect the host in different parameters like fitness, reproduction, genetic diversity etc. according to study of genome sequencing of *Erebidae* moths focusing on bacterium *Wolbachia*. Different life stages like egg, larva, pupa, adult, varies in their gut microbiota. This review reveals the work done on the diversity and richness of gut microbiota, their role on host fruit piercing moth, in Maharashtra, India and different countries of world.

**Keywords:** Family- *Erebidae*, fruit piercing moth, microbiota, diversity

### Introduction

Fruit piercing moths are economically important, as they are serious pest on commercially important fruit crops through the sub-tropical and tropical countries (Waterhouse and Norris, 1987) (Banziger, 1982) [55], and microorganisms play key role in insects/hosts physiology as well as nutrition (Dillon and Charnley, 1995; Nardi *et al.*, 2002) [18, 56] like production of antifungal and antibacterial phenol in locust gut by *P. agglomerance* bacteria (Dillon and Charnley, 1995) [18], which is most abundantly found in gut of locust *S. gregaria* (J. Hunt and Charnley, 1981) [32]. Insects and their symbiotic bacteria's has evolved diversely and in their diverse interactions, which play role in insect nutrition (Wicker, 1983; Douglas, 1988; Douglas and prosser, 1992; Lal *et al.*, 1994; Bracho *et al.*, 1995; Bernays and Klein, 2002) [57, 21, 22, 34, 5, 4]. Bacteria's play role in the defense mechanism of host insect (Kellner and Dettner, 1996; Piel, 2002; Oliver *et al.*, 1999) [33, 58]. Insect gut has diversity of bacteria as gut of insects provide suitable habitat for bacterial association (Bignell, *et al.*, 1984) [59]. Symbiotic bacteria of host insect plays role in the reproductive fitness and development of host (Caspari and Watson, 1959; Gherna *et al.*, 1991; Hurst *et al.*, 1999) [11, 26, 31]. In many species of insects bacterial presence varies in species to species and even every life stages of insect shows different bacterial composition that is bacteria's are transient do not remain in gut in each life stage. However in some cases, nutrient supplying bacteria's to their host insect remain permanent in gut of host throughout the life and some possess obligate microbial endosymbionts that benefits the insects (Bridges, 1981) [7]. This bacteria's distinguished it in to two categories as intracellular and extracellular based on their location of habitat in the insect body as whether they live within cell or in gut lumen or body surface and lining of insect cavities (Dillon and Dillon, 2004; Engel and Moron, 2013; Hansen and Moron, 2014) [17, 23, 28]. Symbiotic bacteria's are considered primary or secondary depending on their role for the host insect whether they are benefited to host or provide non-essential benefit for hosts survival (Douglas, 2015) [20].

The bacterial species associated with a host can vary in their diversity across different life stages, that is bacterial communities can be stable or dynamic, symbiotic association can be temporarily persistence of microbes in host insect in different life stages and across generations (Zilber-Rosenberg, 2008; Engel P, Moron, 2013; Salem H. *et al.*, 2015) [54, 23, 48]. In agricultural and forest habitat of particular pest insects, variability in associated microbe community is found in relation to host diet, to enhance the dietary range of phytophagous insects (Dillon R, Dillon V, 2004; Hosokawa *et al.*, 2007; Douglas AE, 2009; Engel, Moon, 2013; Chu C, *et al.*, 2013) [17, 30, 19, 23, 15]. Depending on the diet bacterial associated communities varies in several insects (Broderic N.A. *et al.*, 2004; Belda E, *et al.*, 2011; Chandler J. A. *et al.*, 2011; Gayatri P.N. *et al.*, 2012) [8, 3, 12, 25]. But in some insects bacterial community may also be stable across diets (Sudakaran S *et al.*, 2012) [50]. Other factors that are responsible for bacterial variation are rearing history of host insect and geographic habitat of host insect population (Xiang H. *et al.* 2006; Adams A.S. *et al.*, 2010) [53, 1]. In Lepidoptera order wide variety of agricultural and forest pest which are most damaging worldwide, which have developed resistance against existing control measures like insecticides, genetically modified plants, genetic host plant resistance (Roush RT, 1987; Stoyenoff JL *et al.*, 1994) [47, 49]. The knowledge of gut microbiota composition and their roles they play in insect's life cycle and their different stages could lead to new approaches for pest management. The Gypsy moth (Erebidae) is most damaging, invasive defoliator of deciduous trees (Montgomery ME, W E Wallner, 1989) [39]. The Gypsy moth can causes economic losses to agriculture and forest industry, it can alters natural ecosystems, can be pathogenic for humans and enumerate 300 host species which shows highly polyphagous nature of it (Liebhold *et al.*, 1995) [35]. Gut microbiota associated with Lepidoptera are responsible for their host insects nutritional and physiological and other functions (Brand *et al.* 1975; Buchner P., 1965; Campbell B.C, 1989; Dasch *et al.*, 1984; Mittler T.E, 1988) [6, 9, 10, 16, 38].

### Microbiota of Various moth pests

The initial studies on lepidopteron gut microorganisms, then little was known about gut bacterial association suggested the possibility that gut microbiota provided assistance to important biochemical function related to host food ingestion and digestion or provide essential nutrients. In Gypsy moth: *Lymantria disper* L. (Lepidoptera: Erebidae), the effect of diet on bacterial composition has been examined the found study stated that some species shown similar bacterial composition among insects fed on similar diet and variations were found in microbial composition in insect those who fed on different diets like sterilized artificial diet these bacterial analysis has been done by 16s rRNA sequencing as well as culture based analysis, study has provide influence of host plant on gut microbial composition of lepidopteron insects (Broderick *et al.*, 2004) [8]. Insects provide environmental reservoirs for bacteria's which are resistant to antibiotic. To know the antibiotic resistance of bacterial community in larval mid-gut of Gypsy moth (Lepidoptera: Erebidae) by using Metagenomics for identification of bacterial community and their genotypes which are responsible for resistance (Heather K. Allen *et al.*, 2009) [29]. The association of bacterial community of insects are mostly depend on the subsequent structuring by extreme conditions within hosts gut, environmental acquisition and maternal transmission, to study

this in gypsy moth: *Lymantria disper* L. (Lepidoptera: Erebidae), comparative analysis of interaction between egg mass and larval midgut microbiome and effect of diet and internal host gut environment on their bacterial community by using 16s rRNA gene pyro-sequencing, found midgut bacterial community which reflects its diet weather its foliar diet of wild or laboratory maintained diet, the environmental acquisition can lead to similar bacterial community assemblance (C.J. Mason, K.F. Raffa, 2014) [13]. Behavior of insect can affect the associated bacterial community composition by direct transmission between insects within colonies and gregarious species (Minkley *et al.* 2006; Moran *et al.*, 2012; Woodbury *et al.*, 2013) [37, 40, 52]. The various insect proportionately carries pathogenic bacteria's, like soft rot Enterobacteriaceae (SRE) which can infect vegetables like potato and fruit plants, insects like *Delia floralis*, *Plutella xylostella*, *Chrysoperla carnea* carries this pathogenic bacteria which are harmful to crops (S. Rossmann *et al.*, 2018) [46].

Comparative study of gut microbiome of two fruit piercing moths peach fruit moth and oriental fruit moth which have different habitat and host plant showed common bacterial genera but had difference in their richness despite having different fruit habits and common genera in both the fruit piercing moths, the abundance of bacteria is depend on the functional parameters like their role in metabolism, translation and membrane transport etc. (Q. Gong, J. Cao, *et al.* 2020) [60]. The fall webworm, *Hyphantria cunea* Drury (Erebidae: Hyphantria), is a phytophagous pest species of forest and agricultural ecosystems; where its larvae feed on Deciduous tree leaves (Schowalter T. *et al.*, 2017) [61], the responsible genes for aggregating behavior, chemosensory, nutrient-related and silk-web-related genes of *H.cunea* has been studied and found by metagenomics and this genes can be responsible for host *H. cunea* environmental adaptations and its cause of rapid invasion at genetic level (Chen *et al.*, 2020) [14].

The fruits piercings moth Othreis: *Eudocima* (Erebidae family) is polyphagous pests. This serious pest was firstly recorded in India by Lefroy and Hawletl, 1909. At Chennai, Isolation and the characterization of roles of bacteria from gut of different instars of *Bombyx mori*, these bacteria's help in degradation of polysaccharides by producing digestive enzymes (A. Anand *et al.*, 2009) [62]. In Maharashtra among the population density of fruit piercing moth in harvesting season of citrus fruits moths belong to *Eudocima*: Erebidae found the most (Mohite and Deshmukh, 2014) [63]. At Bangalore, the identification of gut bacteria has been done to compare gut microbial flora of Tomato fruit borer *Helicoverpa armigera* of the wild and laboratory habitat from fourth and fifth instars by 16s rRNA gene sequencing, concluded that habitat impacts on gut microbial flora, founded variation in the bacterial species (S. Madhusudan, S. K. Jalali, *et al.* 2011) [36]. At New Delhi the polyphagous Eri silk moth, *Samia ricini* has been subjected to isolation of gut bacteria's by different methods like 'Culture-dependent 16s rRNA' that reveals diversity in number of species of gut bacteria, the 'Metagenomics' analysis found diversity in culturable as well un-culturable bacterial community and their inhibition in three parts of gut, and by the 'Enzymatic assays' found role of aerobic and un-aerobic bacteria's in digestion and nutrition of host *S.ricini* (K. MsangoSoko *et al.*, 2020) [41]. The recent study at Delhi, on gut bacteria of cotton bollworm *Pectinophora gossypiella* by targeting Metagenomics of 16s

rRNA of V3-V4 region, to find out the influence of sex of pink bollworm if exists in diversity of gut microbiota and their functions. According to alpha and beta diversity male and female gut bacterial community varies as well as unique in their bacterial diversity in both larval group, these variations in microbiota may help in pesticide resistance and even Bt-cotton gene resistance (H.S. Chaitra *et al.*, 2022) [27].

### Conclusion

The all above study reveals that the wide role of bacteria in host life cycle at all life stages, from hosts nutrition, physiology, digestion, resistance against any unfavorable conditions to even reproduction. The pest moth are important economic factor in agriculture as well as forest, the phytophagous nature of these pest moth is because of the symbiotic gut bacteria and different studies has done to find

out diversity and role of gut microbiota of different pest moths all over world. Most of the study of microbiota of pest moth belong to order Lepidoptera has done internationally to find out diversity of bacteria on basis of habitat weather wild or cultured, the effect of diet on composition gut bacteria of host, even some researchers reveals the genetic composition of bacteria is responsible for particular behavior of host moth. Most of the analysis has been done by using Metagenomics of 16s rRNA sequencing. The moth belong to Erebidae family are most common agricultural pest of vegetable as well as fruit crops. Microbial analysis of moths belong to family Erebidae can be helpful economically and agriculturally the most of the study has been done internationally, in India very few researches has been done on microbiota of pest moth, so there is huge scope for the new approaches in this field.

**Table 1:** Some International, National Researches on Microbiota of insects/moths

Sr. No.	Researcher's Name	Gut Microbe	Insect/Moth species	Purpose of Research
1.	J. Robert Bridges (1981) [7] (USA)	<i>Entobacter agglomerans</i> and <i>Entobacter</i> spp.	Bark beetles- <i>D. terbrans</i> <i>D. frontalis</i> <i>IPS avulsus</i>	Atmospheric nitrogen fixation in host plant.
2.	Hunt and Charnley (1981) [32]; (United Kingdom)	<i>Pantoea agglomerase</i> (domi.), <i>H. alveii</i> , <i>S. marcescens</i> , <i>S. liquefacies</i>	<i>S. gregaria</i> Desert locust	To find out gut bacterial flora.
3.	Dillon and Charnley (1995) [18]; (UK)	<i>Pantoea (Entobacter)</i> (domi.)	<i>S. gregaria</i> Desert locust	To prove Synthesis of fungitoxic Phenol by gut bacteria
4.	A. A. Prem Anand <i>et al.</i> (2009); [62] (Chennai, India)	G+: <i>B. circulans</i> G-: <i>P. vulgaris</i> , <i>K. Pneumonie</i> , <i>E. coli</i> , <i>C. freundii</i> and 7 more	<i>Bombex mori</i> L.	Degradation of cellulose, xylan, pectin and starch and their impact on digestion.
5.	C. J. Mason, <i>et al.</i> (2014) [13]	Similar bacterial community in diet associated bacteria, Different in egg mass.	<i>Lymantria dispar</i> L. Gypsy moth (Lepidoptera: Eribidae)	To investigate egg mass bacteria and environmental source (diet) of bacteria on larval mid-gut bacteria.
6.	H. Staudcher <i>et al.</i> (2016) [64]; (Germany)	Laboratory Larvae- <i>Enterococcus (domi)</i> Field larvae- <i>Methylobacterium</i> Eggs- <i>Entobacter</i> Adult- <i>Entobacter</i>	<i>Heliothis virescens</i>	Bacterial community varies in relation to stages, diet and population.
7.	Luis R. Paniagua Voirol, <i>et al.</i> (2018) [65] (Netherland)	Gut dominant bacteria families- <i>Enterobacteriaceae</i> , <i>Bacillaceae</i> , <i>Pseudomonadaceae</i>	Lepidoptera species (Almost 30 diff species)	1. To review composition of microbial flora across order Lepidoptera 2. Overview on drivers of the variability 3. Routes f transfer 4. Role of bacteria.
8.	Y. Zheng <i>et al.</i> (2020) [66] (China)	<i>Escherchia Shigella</i> and <i>Entobacter</i> (domi. Genera)	<i>Phthorimaea operculella</i> (Zellar) Potato tuber Moth	Richnes of bacterial communities was different depending on nutrient level of tissues consumed by moth.

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