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Phylogenetic and morphometric observations of the *Periplaneta americana* (L.) gut microbiome from collections of Gaya Ji region

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Abstract*— In Indian urban habitats, especially Gaya, the American cockroach, Periplaneta americana (L.), is a common synanthropic bug. Because of its scavenging behavior, P. americana frequently harbors a variety of intestinal nematodes that are significant from an epidemiological standpoint. In order to establish a thorough account of nematode parasites associated with the gut microbiome of P. americana from an Indian perspective, this study synthesizes available parasitological data and integrates morphological and phylogenetic insights derived from recent observations, with an emphasis on the applicability of these findings to Gaya.

***Keywords*— Nematodes, Parasites, American Cockroach, Gaya.**

I. INTRODUCTION

Study of insect-parasitic nematodes serves three purposes. First, it expands knowledge of biodiversity in poorly examined host groups. Second, it contributes to host–parasite ecology by revealing how parasites persist in insect populations. Third, it provides material for systematics and phylogeny, particularly where host association helps interpret nematode relationships. Although entomopathogenic genera such as Steinernema and Heterorhabditis attract attention for biological control, the intestinal oxyurids of cockroaches remain significant for taxonomy, ecology, and biodiversity documentation (Adams and Nguyen 2008).

In India, insect-parasitic nematodes have received less sustained attention than plant-parasitic or soil nematodes. Several classical records predate molecular tools, and many require reassessment through integrative methods. Regional surveys supported by molecular data are therefore needed to strengthen the taxonomy of insect-associated nematodes.

The phylogenetic diversity of insect-parasitic nematodes provides opportunities for comparative biology. By sequencing nematodes from different host groups and regions, researchers can reconstruct the evolutionary history of host–parasite associations. Such comparative frameworks have been constructed for some nematode groups (Blaxter et al. 1998; Holterman et al. 2006), but *thelastoma*tids remain under-represented relative to their ecological importance. The hindgut of *P. americana* is a biologically active compartment containing host tissues, dietary residues, and dense microbial communities. For intestinal nematodes, it offers protection from external conditions and continuous access to nutrients, but it also imposes constraints: limited space, competition, peristalsis, and dependence on host behaviour for transmission. The hindgut can support more than one nematode species simultaneously. Connor and Adamson (1998) demonstrated niche overlap among pinworms in the hindgut of *P. americana* and showed that multiple species coexist in this confined intestinal space.

Leidy (1849, 1850) was among the earliest workers to describe nematodes from invertebrate hosts, establishing the genus *Thelastoma*. Hammerschmidt (1838) contributed early descriptions of forms later placed in *Hammerschmidtiella*. Travassos (1929) formalised the classification of oxyuroid nematodes at the family level. Chitwood (1932) then produced a major synopsis of nematodes parasitic in insects of the family Blattidae, connecting nematode morphology with cockroach hosts and providing a framework for later workers.

Connor and Adamson (1998) showed that multiple pinworm species can coexist in the cockroach hindgut through partial spatial and temporal separation. The present findings support this interpretation: the three major genera differ in body size (*Thelastoma* largest, *Schwenkiella* smallest), anterior-end morphology, and tail form, suggesting possible microhabitat differentiation within the posterior gut.

II. MATERIALS AND METHODS

This investigation was designed as an integrative taxonomic study of nematode parasites from *Periplaneta americana* collected in Gaya, Bihar. The work combined field collection, laboratory dissection, microscopic examination, morphometric recording, DNA extraction, PCR amplification of ribosomal markers, sequencing, and phylogenetic analysis. The target genera were *Hammerschmidtiella, Thelastoma, Leidynema,* and *Schwenkiella,* all *thelastomatid* oxyurids.

Both morphological and molecular markers were used because *thelastomatid* nematodes may show overlapping diagnostic characters, and several older records were based only on morphology (Adamson and Van Waerebeke 1992; Jex et al. 2006). The methodological framework was intended to verify whether recovered nematodes could be assigned confidently to known genera and representative species and whether molecular placement agreed with morphological identity.

Adult *P. americana* were collected manually or by baited traps during evening and night hours. Specimens from each site were kept separately in labelled, ventilated containers. Damaged or dead cockroaches were excluded. A total of 200 adult cockroaches were examined. Identification followed the diagnostic characters of Bell, Roth, and Nalepa (2007): large body size, reddish-brown colour, flattened body form, long antennae, spiny legs, fully developed tegmina, and a pale yellowish pronotal margin. Nymphs and uncertain specimens were excluded.

For each host, infection status, number of worms recovered, and preliminary genus-level identity were recorded. Mixed infection was noted when more than one genus occurred in the same host. Prevalence, mean intensity, and mean abundance were calculated as:

Prevalence (%) = (Number of infected hosts / Total hosts examined) × 100

Mean intensity = Total parasites recovered / Number of infected hosts

Mean abundance = Total parasites recovered / Total hosts examined

III. RESULT AND DISCUSSION

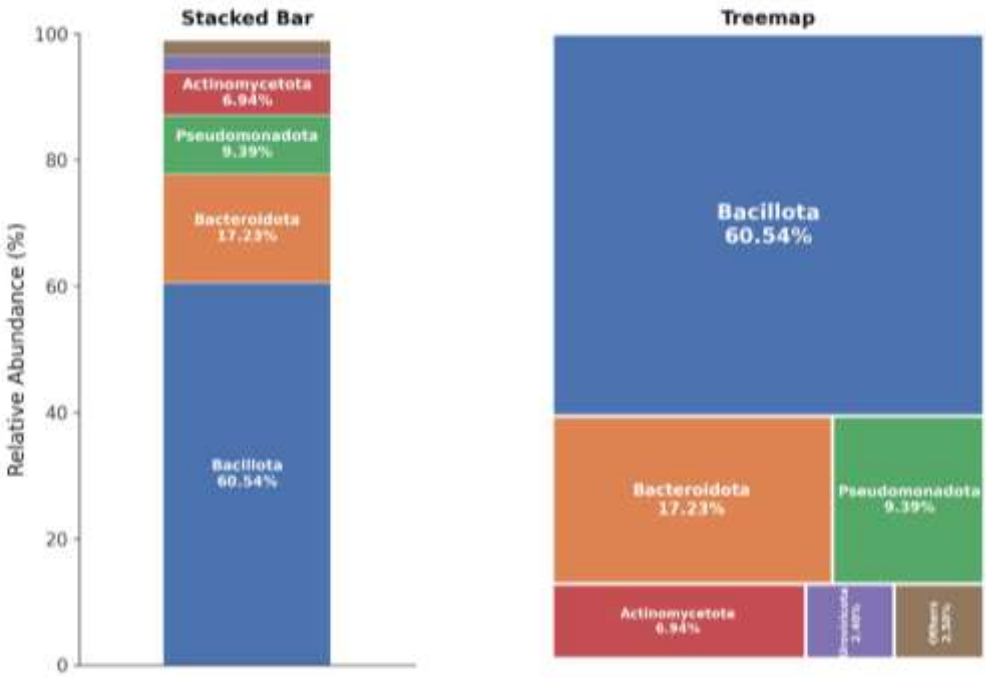
The associated cockroach gut microbiome provides the verified molecular publication component. It reported a complex hindgut community dominated by Bacillota (60.54%), followed by Bacteroidota (17.23%), Pseudomonadota (9.39%), and Actinomycetota (6.94%).

Table :1. Phylum-level composition of the P. americana gut microbiome.

Phylum	Relative Abundance (%)	Read Count	Observation
Bacillota	60.54	2,331,212	Dominant phylum in the hindgut community
Bacteroidota	17.23	663,352	Second most abundant phylum
Pseudomonadota	9.39	361,743	Fermentative and succinate-producing groups
Actinomycetota	6.94	267,402	Moderately abundant phylum
Uroviricota	2.40	Not specified	Viral/bacteriophage component
Thermodesulfobacteriota	1.00	Not specified	Minor phylum
Euryarchaeota	0.70	Not specified	Methanogenic archaeal component

Table: 2. Dominant taxa and diversity in the cockroach gut microbiome.

Taxonomic Level	Dominant/Important Taxa	Reported Value	Interpretation
Family	Oscillospiraceae	24.72%	Major fermentative family
Family	Lachnospiraceae	24.65%	Co-dominant fermentative family
Genus	*Bacteroides*	8.10%	Most abundant genus
Genus	*Vescimonas*	7.83%	Highly represented gut-associated genus
Genus	*Blautia*	6.77%	SCFA-associated fermentative genus
Species	*Bacteroides fragilis*	4.52%	Most abundant species
Species	*Vescimonas coprocola*	4.45%	Second most abundant species
Diversity	64 phyla; 584 families; 2,018 genera	–	High taxonomic richness



Histogram: 1. Multi-level taxonomic composition of the P. americana gut microbiome from the associated cockroach gut microbiome.

The overall prevalence of 84.0% (168/200) confirms that *P. americana* in Gaya sustains active nematode transmission. This value is comparable to high-prevalence records from other Indian and global studies. Basir (1956) noted regular occurrence of *thelastomatids* in Indian cockroaches, and Shah (2007) reported frequent infection in cockroaches from Manipur. Internationally, Jex et al. (2006) found *thelastomatids* in a large proportion of Australian cockroaches examined for molecular work, and Blanco et al. (2012) recorded *H. diesingi* consistently from *P. americana* in Argentina.

Infection was highest in restrooms and toilets (91.4%), followed by garbage and waste areas (88.6%), kitchens (87.5%), and storehouses (85.7%). Lower prevalence occurred in damp soil and outdoor crevices (72.0%) and miscellaneous localities (73.3%). This gradient reflects the relationship between habitat moisture, organic matter, host density, and egg survival.

Morphological Interpretation

Morphological characters successfully separated the four genera. *Hammerschmidtiella* (2.35–3.12 mm body length) was identified by its triradiate stoma, filiform tail (280–350 µm), and slightly posterior vulva (52–56%). These measurements fall within the range reported for *H. diesingi* by Blanco et al. (2012), who measured body lengths of 2.2–3.5 mm in Argentinian material, and by Basir (1956) for Indian specimens.

Histogram: 2. Heatmap of key metabolic pathway completeness across eight MAGs from the associated cockroach gut microbiome.

Thelastoma (4.20–5.85 mm) was distinguished by its prominent buccal cavity (20–25 µm depth), muscular bulb, and tapering tail (350–450 µm). The larger body and deeper buccal cavity separate it clearly from *Hammerschmidtiella*. These characters are consistent with *T. periplaneticola* descriptions from Shah (2007) and Spiridonov and Guzeeva (2009).

Leidynema (3.85–4.92 mm) was characterised by its elongated stoma (18–22 µm), deeper cervical annulations, and conical tail (180–230 µm). The conical tail, in contrast to the filiform tail of *Hammerschmidtiella* and the tapering tail of *Thelastoma*, provides a reliable field character. These morphometrics agree with *L. appendiculata* descriptions from Ozawa et al. (2014).

The morphological results demonstrate both the value and the limits of classical taxonomy. While genus-level separation was achievable, measurements overlap between genera (e.g., body width ranges overlap for *Hammerschmidtiella* and *Schwenkiella*), and damaged or immature specimens could be difficult to assign. This reinforces the need for molecular confirmation, a conclusion shared by recent integrative studies in nematode taxonomy (Cantalapiedra-Navarrete et al. 2024; Munawar et al. 2024; Salazar-García et al. 2025).

IV. CONCLUSION

The high prevalence is explained by the ecology of both host and parasite. *P. americana* is nocturnal, omnivorous, aggregation-prone, and tolerant of unsanitary environments. Nematode eggs pass with host faeces and are ingested by other cockroaches during feeding or substrate contact. This direct life cycle, combined with high host density in indoor habitats, creates favourable conditions for sustained transmission.

The finding that uninfected hosts constituted only 16.0% of the sample suggests either efficient re-infection or that the hindgut environment is so consistently suitable that clearance is rare. In either case, *P. americana* in Gaya functions as a reliable reservoir for *thelastoma*tid nematodes.

Prevalence values for *thelastoma*tids in *P. americana* tend to be high (>50%), supporting the general conclusion that this host–parasite association is biologically robust. The consistency of high prevalence across different localities within Gaya (range: 72.0–91.4%) further supports this interpretation.

P. americana from Gaya, Bihar, harboured a diverse assemblage of *thelastoma*tid nematodes. The overall infection prevalence was 84.0% (168/200). *Hammerschmidtiella* was the dominant genus (68.5%), followed by *Thelastoma* (51.5%), *Leidynema* (40.5%), and *Schwenkiella*-like forms (16.0%). Infection was highest during the monsoon season.

From an epidemiological perspective, the 84.0% prevalence should be interpreted alongside other parasitological parameters. Mean intensity (average number of parasites per infected host) and mean abundance (average number per all examined hosts) would further characterise the infection level but require accurate worm counts that may be affected by the difficulty of recovering all small worms from gut contents. Nevertheless, the prevalence alone is sufficient to establish that *P. americana* populations in Gaya maintain a high baseline level of *thelastomatid* infection.

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