

## Intestinal Parasites in Cockroaches and Houseflies and Household Stool Positivity in Labuhan Deli Indonesia

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### ABSTRAK

**Latar belakang:** Deteksi parasit pada serangga tidak langsung membuktikan penularan kepada manusia. Penelitian ini menilai kontaminasi parasit pada kecoa dan lalat serta hubungannya dengan positivities tinja rumah tangga.

**Metode:** Penelitian potong lintang dilakukan pada 100 rumah tangga di Kecamatan Labuhan Deli, Sumatera Utara, pada 2022. Kriteria meliputi lama tinggal  $\geq 1$  tahun dan riwayat gangguan gastrointestinal keluarga dalam setahun terakhir. Tinja diperiksa dengan sediaan langsung dan sedimentasi formalin-eter; endapan pencucian serangga diperiksa secara mikroskopis. Rumah dinyatakan positif jika  $\geq 1$  penghuni yang diperiksa positif. Analisis menggunakan rasio prevalensi (PR), interval kepercayaan (IK) 95% logaritmik, dan uji Fisher dua sisi.

**Hasil:** Sebanyak 81 rumah tangga (81%) positif helminth dan 71 (71%) positif protozoa. Serangga teridentifikasi sebagai *Periplaneta americana* dan *Musca domestica*. Kecoa memiliki empat taksa helminth dan lima protozoa; lalat masing-masing tiga. Positivitas parasit kecoa berasosiasi dengan positivities protozoa rumah tangga (PR 1,57; IK 95% 1,18–2,09;  $p=0,001$ ) dan helminth (PR 1,53; IK 95% 1,22–1,92;  $p<0,001$ ). Estimasi lalat tidak presisi karena hanya dua rumah memiliki sampel lalat positif parasit.

**Simpulan:** Kontaminasi parasit kecoa berasosiasi dengan positivities tinja rumah tangga dalam sampel terseleksi ini. Desain potong lintang dan mikroskopi tidak menetapkan viabilitas atau arah penularan. Perbaikan sanitasi dan pengendalian serangga merupakan implikasi kesehatan lingkungan; efektivitasnya tidak diuji.

**Kata kunci:** Cacing; Kecoa; Lalat; Protozoa; Sanitasi

### ABSTRACT

**Background:** Parasite detection on insects does not establish transmission to humans. This study examined cockroach and housefly contamination and its association with household stool positivity.

**Methods:** A cross-sectional study included 100 households in Labuhan Deli District, North Sumatra, in 2022. Eligibility required residence  $\geq 1$  year and recent family gastrointestinal illness. Stool specimens underwent direct microscopy and formalin-ether sedimentation; insect-wash sediments were examined microscopically. A household was positive if  $\geq 1$  examined resident tested positive. Analyses used prevalence ratios (PRs), approximate log-based 95% confidence intervals (CIs), and two-sided Fisher exact tests.

**Results:** Helminths and protozoa were detected in 81 (81%) and 71 (71%) households, respectively. The insects were *Periplaneta americana* and *Musca domestica*. Cockroaches yielded four helminth and five protozoan taxa; houseflies yielded three of each. Cockroach parasite positivity was associated with household protozoan positivity (PR 1.57; 95% CI 1.18–2.09;  $p=0.001$ ) and helminth positivity (PR 1.53; 95% CI 1.22–1.92;  $p<0.001$ ). Housefly estimates were imprecise because only two households had parasite-positive fly samples.

**Conclusion:** Cockroach contamination was associated with household stool positivity in this selected sample. Cross-sectional microscopy does not establish viability or transmission direction. Sanitation and insect control are environmental-health implications; their effectiveness was not evaluated.

**Keywords:** Cockroaches; Flies; Helminth; Protozoa; Sanitation

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## INTRODUCTION

Cockroaches and houseflies inhabit environments where human activity, organic waste, and food preparation overlap. Their contact with fecal material provides opportunities for mechanical carriage of intestinal parasites, making insect contamination relevant to environmental health.<sup>1</sup> Vector surveillance and environmental sanitation are complementary components of prevention.<sup>2</sup>

Environmental sanitation has been investigated in relation to cockroach occurrence on passenger ships in Indonesia.<sup>3</sup> Protozoan forms have also been identified in German cockroach fecal pellets, demonstrating the importance of distinguishing insect-associated organisms from potential human pathogens.<sup>4</sup> Both the collection setting and the identity of the detected organism are therefore relevant to interpreting parasite carriage.

Primary surveys have detected helminth forms on cockroaches in food premises and parasitic organisms on flies in market environments.<sup>5,6</sup> Household cockroach studies and reviews of their medical importance further support investigation of carriage in human environments.<sup>7,8</sup> However, the identification of an organism on an insect does not alone establish that it remains viable or reaches a susceptible person.

Household studies in Nigeria and Egypt have examined parasites associated with cockroaches and houseflies.<sup>9,10</sup> Studies of houseflies in Iraq and Sudan have also investigated parasite carriage.<sup>11,12</sup> Findings must distinguish helminth eggs or larvae from protozoan cysts, oocysts, or other morphological forms; these stages cannot be treated as interchangeable evidence of infectivity.

An important epidemiological question is whether parasite-positive insects and stool-positive residents occur in the same households. Joint household measurements can describe this association, but shared exposure to contaminated water, waste, or sanitation conditions can produce concordant findings without insect-to-human transmission.

Household-level evidence from Labuhan Deli can therefore complement studies of insect carriage by linking insect findings with resident stool results from the same residential setting. This approach provides local information on the co-occurrence of insect contamination and resident stool positivity within households.

This study aimed to describe intestinal parasite findings in cockroaches and houseflies, quantify the proportion of sampled households with stool-positive residents, and evaluate associations between insect parasite positivity and household helminth or protozoan positivity.

## MATERIALS AND METHODS

### Study area

The study was conducted in 2022 in a settlement with poor sanitation in Labuhan Deli District, Deli Serdang Regency, North Sumatra Province, Indonesia. The study population comprised households in this setting with a family history of gastrointestinal illness. The eligibility criteria selected a particular residential population rather than a representative sample of all district residents.

### Study design and data collection

This cross-sectional study included 100 households. Eligibility required residence for at least one year and a family member with a history of gastrointestinal illness during the preceding year. Data collection involved interviews, household observations, resident stool examinations, and collection of cockroaches and houseflies around the participating homes.

Stool sampling targeted household heads and family members with a history of gastrointestinal illness. Collection equipment was provided for stool specimens. Microscopic examination used direct preparations and formalin–ether sedimentation. Cockroaches and houseflies were collected using sticky traps, killed using chloroform applied to tissue, and placed in containers labeled by collection location. Examinations were performed at the Medical Parasitology Laboratory, Universitas Sumatera Utara.

The insects were identified under a dissecting microscope using taxonomic keys. For parasite examination, insects were placed in tubes containing 12 mL of NaCl solution and shaken vigorously for 30 minutes to release attached material. The wash fluid was centrifuged at 2,000 rpm for 10 minutes. Sediment was transferred to a glass

slide, mixed with Lugol solution, and examined by light microscopy for parasite forms. This procedure evaluated material recovered from insect-body washes.

### Household definitions and statistical analysis

The household was the unit of association analysis. Resident outcomes were defined by detection of helminths or protozoa in at least one examined household member. Insect exposure was classified by the household parasite-positive or parasite-negative status of cockroach or housefly samples, rather than by insect presence alone. Each table contrasted positive and negative insect-sample status within the same insect type.

Unadjusted prevalence ratios compared household outcome positivity between insect-parasite-positive and insect-parasite-negative groups. Approximate 95% confidence intervals were calculated on the natural-log scale:  $\ln(PR) \pm 1.96 \times \sqrt{[1/a - 1/(a+b) + 1/c - 1/(c+d)]}$ , where a and b were outcome-positive and outcome-negative households in the insect-positive group, and c and d were the corresponding counts in the insect-negative group. Two-sided Fisher exact tests were used for all four 2×2 comparisons;  $p < 0.05$  was the nominal significance threshold. These exploratory comparisons were unadjusted for confounding. Log-based intervals for houseflies require particular caution because the exposed group comprised only two households.

### Ethical approval

Ethical approval was granted by the Health Research Ethics Commission of Universitas Sumatera Utara (No. 723/KEPK/USU/2022).

## RESULTS AND DISCUSSION

### Household stool positivity

The sample comprised 100 eligible households. A household was classified as helminth-positive or protozoa-positive when at least one examined resident had the corresponding positive stool result. Accordingly, 81 households (81%) were helminth-positive and 71 (71%) were protozoa-positive (Table 1). These percentages describe household positivity, not individual prevalence, and the two outcomes are not mutually exclusive.

Table 1. Households with stool-positive residents among 100 sampled households

| Parasites                   | n  | %  |
|-----------------------------|----|----|
| <b>Helminth positivity</b>  |    |    |
| Positive                    | 81 | 81 |
| Negative                    | 19 | 19 |
| <b>Protozoan positivity</b> |    |    |
| Positive                    | 71 | 71 |
| Negative                    | 29 | 29 |

Note: The denominator is 100 households. Positivity indicates at least one examined resident with the corresponding stool finding. Helminth and protozoan outcomes may overlap; individual-level prevalence cannot be inferred from this table.

The high proportions of positive households should be interpreted within the eligibility criterion of a family history of gastrointestinal illness. Studies in other settings have evaluated intestinal parasites in symptomatic and asymptomatic populations and reported associations with environmental or behavioral factors.<sup>13–15</sup> Their individual-level estimates are not directly comparable with the household-level proportions in this study. The present results do not quantify current symptomatic infection or establish an impending outbreak.

### Parasite findings in cockroaches and houseflies

Cockroaches and houseflies were collected from all participating homes. The cockroaches were identified as *Periplaneta americana* and the houseflies as *Musca domestica*. Parasite findings were tabulated for the collected insects. Detection on insect-body washes indicates contamination; it does not establish infection of the insects or transmission to residents.

The taxon-specific counts in cockroaches summed to 46 helminth findings and 40 protozoan findings; the corresponding sums for houseflies were three and three (Table 2). *Ascaris lumbricoides* accounted for 26/46 (56.5%) of the tabulated cockroach helminth counts, and *Cryptosporidium sp.* accounted for 10/40 (25.0%) of the

protozoan counts. These are distributions of tabulated findings, not prevalence estimates among all insects. *Enterobius vermicularis*, *Blastocystis* sp., and *Iodamoeba buetschlii* were listed for cockroaches but not houseflies.

Table 2. Taxon-specific parasite counts and their distribution in cockroaches and houseflies

| Parasites                      | Cockroaches |      | Flies |      |
|--------------------------------|-------------|------|-------|------|
|                                | n           | %    | n     | %    |
| <b>Helminths</b>               |             |      |       |      |
| <i>Ascaris lumbricoides</i>    | 26          | 56.5 | 1     | 33.3 |
| <i>Trichuris trichiura</i>     | 6           | 13   | 1     | 33.3 |
| Hookworms                      | 5           | 10.9 | 1     | 33.3 |
| <i>Enterobius vermicularis</i> | 9           | 19.6 | 0     | 0    |
| <b>Protozoa</b>                |             |      |       |      |
| <i>Entamoeba coli</i>          | 9           | 22.5 | 1     | 33.3 |
| <i>Cryptosporidium</i> sp.     | 10          | 25.0 | 1     | 33.3 |
| <i>Blastocystis</i> sp.        | 9           | 22.5 | 0     | 0    |
| <i>Giardia lamblia</i>         | 7           | 17.5 | 1     | 33.3 |
| <i>Iodamoeba buetschlii</i>    | 5           | 12.5 | 0     | 0    |

Note: Percentages are calculated from the sums of the listed taxon-specific counts within each insect–parasite category: 46 helminth and 40 protozoan counts for cockroaches, and three counts in each category for houseflies. These compositional percentages are not prevalence among all examined insects. Zeros indicate no tabulated detection of the relevant taxon.

The different totals of parasite findings cannot establish a higher contamination probability in one insect species without comparable denominators and standardized trapping effort. Weather, the number of insects examined, pooling, and recovery efficiency can influence detection. Rain during collection is a possible sampling consideration, but no quantitative weather comparison establishes its effect on capture or parasite detection in this study. Interpretation is restricted to the body-wash procedure described in the methods; the results do not distinguish internal carriage or excretion.

Not all parasites associated with cockroaches are human intestinal pathogens. Studies of gregarines concern parasites of the insects themselves and should not be interpreted as evidence of human transmission.<sup>16</sup> In contrast, field and experimental research has examined cockroaches as potential mechanical disseminators of *Entamoeba histolytica*.<sup>17</sup> Such experimental exposure differs from the concurrent household observations made here.

Cockroach size, surface structures, and foraging behavior provide plausible explanations for retention of parasite forms.<sup>18</sup> These mechanisms were not measured in the present study. Differences in trapping effort, specimen numbers, or processing could also contribute to the observed counts and must be distinguished from biological differences in carriage.

Housefly surveys and reviews describe opportunities for contact with contaminated substrates and food.<sup>19,20</sup> These observations support the biological plausibility of mechanical carriage, but they do not determine the contribution of houseflies to infection within the sampled households.

#### Associations between insect contamination and household stool positivity

Households classified as cockroach-parasite-positive had protozoan positivity in 46/54 (85.2%) compared with 25/46 (54.3%) cockroach-parasite-negative households (PR 1.57; 95% CI 1.18–2.09;  $p=0.001$ ). Corresponding helminth positivity was 52/54 (96.3%) versus 29/46 (63.0%) (PR 1.53; 95% CI 1.22–1.92;  $p<0.001$ ). Only two households were classified as fly-parasite-positive. Their estimates were imprecise: PR 0.70 (95% CI 0.17–2.81;  $p=0.498$ ) for protozoa and PR 0.61 (95% CI 0.15–2.46;  $p=0.345$ ) for helminths (Table 3).

Table 3. Unadjusted associations between insect parasite positivity and household stool positivity

| Insect and outcome       | Insect-positive<br>n+/n- (%) | Insect-negative<br>n+/n- (%) | PR   | 95% CI    | p      |
|--------------------------|------------------------------|------------------------------|------|-----------|--------|
| Cockroaches<br>Protozoa  | 46/8 (85.2)                  | 25/21 (54.3)                 | 1.57 | 1.18–2.09 | 0.001  |
| Cockroaches<br>Helminths | 52/2 (96.3)                  | 29/17 (63.0)                 | 1.53 | 1.22–1.92 | <0.001 |
| Houseflies<br>Protozoa   | 1/1 (50.0)                   | 70/28 (71.4)                 | 0.70 | 0.17–2.81 | 0.498  |
| Houseflies<br>Helminths  | 1/1 (50.0)                   | 80/18 (81.6)                 | 0.61 | 0.15–2.46 | 0.345  |

Note: n+/n- = numbers of households with positive/negative resident stool outcomes. Percentages are row-specific household outcome positivity within each insect-sample status group. PR = prevalence ratio, with the insect-negative group as reference. CIs are approximate log-based intervals; p values are from two-sided Fisher exact tests. Fly estimates are especially uncertain because only two households were insect-positive. All four comparisons are unadjusted.

Studies of non-biting synanthropic flies demonstrate that carriage estimates depend on insect species and collection setting.<sup>21</sup> Indonesian work around a waste-disposal site also places fly findings within a specific sanitation context.<sup>22</sup> Cockroach surveys from other regions describe additional parasite findings, but comparability depends on sampling and diagnostic procedures.<sup>23</sup> Species-specific ecological context also matters: blow fly assemblages have been investigated across residential, agricultural, mountainous and forested habitats.<sup>24</sup> Such ecological findings do not quantify parasite carriage in the houseflies sampled here.

The cockroach associations concern household prevalence rather than the incidence of new infections. They do not show that cockroaches are independent causal risk factors. Sanitation, water supply, waste handling, socioeconomic conditions, and household size may influence both resident stool positivity and insect contamination. More people examined within a household also increase the opportunity to classify that household as positive. The small number of fly-positive households precludes a firm conclusion about the presence or absence of a housefly association.

The helminth taxa identified in this study overlap descriptively with reports from household cockroach research.<sup>10,25</sup> No formal comparison between studies was performed. Differences in setting, insect species, examination units, and microscopy methods limit comparisons of their frequencies.

*Ascaris lumbricoides* accounted for the largest share of the tabulated cockroach helminth findings. Its eggs must develop in the environment before becoming infective, and their detection does not itself establish an infective exposure.<sup>26</sup> Persistence and environmental contamination are relevant to its epidemiology,<sup>27</sup> but egg viability and developmental status were not measured by the procedures used here.

Environmental conditions can influence the occurrence of *Trichuris trichiura* and exposure to its eggs.<sup>28</sup> However, parasite production or environmental tolerance cannot be identified as the cause of the distribution in Table 2 from these cross-sectional observations. The study did not quantify environmental egg loads or species-specific survival.

*Enterobius vermicularis* findings on cockroaches are consistent with descriptions of mechanical carriage in household studies.<sup>10,29</sup> Nevertheless, their detection does not identify a source person or direction of spread. Similarly, hookworm eggs on an insect indicate contamination and do not establish the presence of the infective larval stage or a transmission route to residents.

Protozoan findings in insect surveys vary with location, insect species, and laboratory procedures.<sup>30</sup> In the present study, broad helminth and protozoan household outcomes do not establish same-species concordance between residents and insects. Microscopic identification also requires caution, particularly for small forms attributed to *Cryptosporidium*; body-wash findings do not by themselves establish viability or clinical disease.

The clinical meaning of parasite detection differs among taxa. *Entamoeba coli* and *Iodamoeba buetschlii* are generally regarded as nonpathogenic indicators of fecal exposure, whereas other detected taxa can be associated with disease. Detection alone does not establish that an organism caused a resident's symptoms. The broader literature supports environmental hygiene and insect-control measures as plausible prevention approaches,<sup>1,18</sup> but the present study measured neither their effectiveness nor clinical outcomes such as nutritional or cognitive impairment.

### Study limitations

The cross-sectional design cannot establish temporal sequence or the direction of contamination between residents, insects, and shared environmental sources. Selection based on a family history of gastrointestinal illness limits generalizability. Household positivity depends on the number and selection of residents examined and cannot substitute for individual prevalence. Microscopic body-wash findings do not establish internal insect carriage, parasite viability, infectivity, or same-species transmission. Limited comparability of insect sampling and the small fly-positive group constrain between-insect comparisons. The unadjusted associations may reflect sanitation, water, waste management, socioeconomic conditions, household size, or examination intensity; they do not demonstrate an independent cockroach effect. No intervention effectiveness was assessed.

### CONCLUSION

In this selected sample of 100 households, helminths and protozoa were detected in examined residents of 81% and 71% of households, respectively. Parasite-positive cockroach samples were associated with higher household stool positivity, whereas the housefly estimates were imprecise because only two households were fly-parasite-positive. These observations support further investigation of household environmental contamination but do not establish viable parasite carriage, insect-to-human transmission, or an independent causal effect. Sanitation, household hygiene, and insect management are relevant public-health considerations; their effectiveness was not evaluated in this study.

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### AUTHORS' CONTRIBUTION

Yoan C Panggabean and Lambok Siahaan contributed equally as first authors and were responsible for quality control, coordination, methodology, data analysis, manuscript editing, and integration. Adelina HS, Nurfida KA, and Dewi Saputri contributed to conceptualization and literature review. Janry Sinaga and M Ary Irawan were responsible for field data collection and reference and manuscript preparation. All authors have read and approved the final work.

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