

# **Epidemiology Bulletin**

- 167 A Study of Cockroaches as  
Vectors of *Entamoeba*  
*histolytica*  
173 Cases of Notifiable and  
Reportable Diseases,  
Taiwan-Fukien Area
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## **A Study of Cockroaches as Vectors of *Entamoeba histolytica***

### **1. Introduction**

Amebic dysentery is a notifiable disease in Taiwan. The primary mode of transmission is contamination of either food or water by the cysts in feces<sup>(1)</sup>. The disease occurs more often in crowded and unsanitary places such as jails and mental institutions, but also occurs among homosexuals.<sup>(2)</sup> The world-wide prevalence rate of amebic dysentery is about 10%<sup>(1)</sup>, and there have been outbreaks in Taiwan in recent years in schools and mental institutions<sup>(3,4)</sup>. Symptoms of the disease include diarrhea, occasionally abscess of the liver and brain<sup>(2,5)</sup>; death can even result from the disease because of dehydration from diarrhea. Amebic dysentery is a communicable disease it is important to prevent and to control. Survey of potential sources of infection in the environment is absolutely essential to make such efforts effective.

It has been reported that the cockroach is capable of transmitting communicable diseases through such pathogenic agents as bacteria, fungi and the eggs and cysts of parasites<sup>(6,7)</sup>. Cockroaches can be found in many varieties, and those common in Taiwan are *Blatella germanica*, *Periplaneta americana* and *Periplaneta brunnea*<sup>(8)</sup>. Cockroaches breed rampantly under the warm weather conditions on the Island, feeding on odds and ends from human food to feces. The present study was conducted because cockroaches are a likely vector for *Entamoeba histolytica*.

### **2. Materials and Methods**

#### **a) Study Method**

(1) in a laboratory situation:

The cockroaches *Periplaneta american* and *Blatella germanica*, both originally free cysts of *Entamoeba histolytica*, were bred in the laboratory to observe their transmission of ameba cysts.

They were then exposed to 10 gm of human feces containing 100/gm to 1,000/gm of cysts of *Entamoeba histolytica* for 24 hours; the control group was exposed to human feces without *E. histolytica*.

(2) through field survey:

In May 1995, from each of the 11 Kaohsiung districts where school lunches were offered, one primary school was selected at random for the collection of cockroaches. Each school was given six collecting boxes for placement along the walls of the dining room and the kitchen.

b) Examination of Cockroaches for Ameba Cysts:

The method developed by Fotedar et al. (1991)<sup>(9)</sup> was used.

(1) to examine for ameba cysts on the cockroach surfaces: Each insect collected was placed in a sterile test tube and immobilized by freezing at 0°C for five minutes. After 2 ml of sterile saline solution was added, and the tube was shaken for two minutes, it was centrifuged at 2,000 rpm for five minutes. Then, 2 ml of merthiolate-iodine-formaldehyde (MIF) solution was added for color before observation under the microscope.

(2) to examine in Cockroaches digestive tracts for Ameba Cysts:

After examination of the cockroach surface, the cockroaches were placed in 70% alcohol for five minutes, dried at room temperature then placed in saline solution for three minutes. Their digestive tracts were removed, ground with 2 ml of saline solution, centrifuged and color dyed per the procedure described next above, before microscopic observation.

(3) Counting of *E. histolytica* Cysts:

The method developed by Kao and Unger (1994)<sup>(10)</sup> was used to count *E. histolytica* cysts on both the surface and in the digestive tracts of the cockroaches in a bloodcell counting plate under microscopic observation. The total number of *E. histolytica* cysts on the surface or in the digestive tracts was calculated by:

$$\text{No. of cysts in each chamber} \times \frac{\text{total volume of each cockroach examined}}{\text{total volume of roaches placed in a given chamber}}$$

(4) Statistical Analysis:

The tests applied were Fisher's exact test, Wilcoxon rank sum test,  $\chi^2$ -test, proportional test.

### 3. Results

#### a) Cockroach Transmittal of Cysts in the Laboratory:

Table 1 shows how the cockroaches transmitted *E. histolytica* cysts after having been exposed to cyst-containing feces. The transmission rate of *Periplaneta americana* exposed to feces containing 100/gm of *E. histolytica* cysts was 36.84% (7/19), transmitting  $27.05 \pm 9.46$  (mean  $\pm$  standard error) cysts. No statistically significant difference was noted between the transmission rate and the number of cysts transmitted *in vitro* (from the surface) (21.05%,  $11.32 \pm 5.92$ ) and the transmission rate and the number of cysts transmitted *in vivo* (through the digestive tract) (16.79%,  $15.74 \pm 8.60$ ). The transmission rate of *Periplaneta americana* exposed to feces containing 1,000/gm of *E. histolytica* cysts was 90.00%, transmitting  $123.35 \pm 29.89$  cysts. The *in vivo* transmission rate (85%) was significantly higher than the *in vitro* rate (55%), though the number of cysts transmitted *in vivo* ( $68.50 \pm 13.68$ ) was not significantly higher than the number transmitted *in vitro* ( $54.85 \pm 18.21$ ).

**Table 1. Cockroaches Transmitting *E. histolytica* Cysts**

Group	Transmission Rate (%)	No. Transmitted (Mean $\pm$ S.E.)
<i>Periplaneta americana</i>		
Control	0.00 ( 0/20)	0 $\pm$ 0
100/gm	36.84 ( 7/19) <sup>c</sup>	27.05 $\pm$ 9.46 <sup>b</sup>
<i>in vitro</i>	21.05 ( 4/19) <sup>EF</sup>	11.32 $\pm$ 5.92 <sup>ce</sup>
<i>in vivo</i>	16.79 ( 3/19) <sup>G</sup>	15.74 $\pm$ 8.60 <sup>fg</sup>
1,000/gm	90.00 (18/20) <sup>CD</sup>	123.35 $\pm$ 29.89 <sup>b</sup>
<i>in vitro</i>	55.00 (11/20) <sup>AE</sup>	54.85 $\pm$ 18.21 <sup>c</sup>
<i>in vivo</i>	85.00 (17/20) <sup>AGH</sup>	68.50 $\pm$ 13.68 <sup>f</sup>
<i>Blatella germanica</i>		
Control	0.00 ( 0/20)	0 $\pm$ 0
100/gm	29.17 ( 7/24)	13.08 $\pm$ 4.26
<i>in vitro</i>	0.00 ( 0/24) <sup>BF</sup>	0 $\pm$ 0 <sup>ade</sup>
<i>in vivo</i>	29.17 ( 7/24) <sup>B</sup>	13.08 $\pm$ 4.26 <sup>ag</sup>
1,000/gm	46.15 ( 6/13) <sup>D</sup>	90.31 $\pm$ 56.71
<i>in vitro</i>	23.08 ( 3/13)	47.92 $\pm$ 29.91 <sup>d</sup>
<i>in vivo</i>	30.77 ( 4/13) <sup>H</sup>	42.38 $\pm$ 29.21

- Notes: I. 100/gm: cockroaches exposed to 10 gm of feces containing 100/gm of *E. histolytica* cysts for 24 hours;  
 II. 1,000/gm: cockroaches exposed to 10 gm of feces containing 1,000/gm of *E. histolytica* cysts for 24 hours;  
 III. A:A, B:B, C:C, D:D, E:E, F:F, G:G, H:H Fisher's exact test,  $p > 0.05$ ;  
 VI. a:a, b:b, c:c, d:d, e:e, f:f, g:g, h:h  
 VII. Wilcoxon rank sum test,  $p > 0.05$ .

The transmission rate of *Blatella germanica* exposed to feces containing 100/gm of *E. histolytica* cysts was 29.17%, transmitting  $13.08 \pm 4.26$  cysts. The *in vivo* transmission rate (29.17%) was significantly higher than the *in vitro* rate (0%), though no significant difference was noted in the numbers of cysts transmitted ( $13.08 \pm 4.26$ , 0). The transmission rate for *Blatella germanica* when exposed to feces containing 1,000/gm of *E. histolytica* cysts was 46.15%, transmitting  $90.32 \pm 56.71$  cysts. No significant difference, however, was noted between the transmission rate and the number of cysts transmitted *in vitro* (23.08%,  $47.92 \pm 29.91$ ) and the transmission rate and number transmitted *in vivo* (30.77%,  $42.38 \pm 29.91$ ).

The difference in the transmission rates of *Periplaneta americana* (90%) and *Blatella germanica* (46.15%) when each was exposed to feces containing 1,000/gm of *E. histolytica* cysts was statistically significant. The difference in the transmission rates of the two cockroaches both exposed to feces containing 100/gm of cysts was not statistically significant, however.

#### b) Field Survey Cockroach Transmittal of Cysts

Four hundred and ninety-five *Periplaneta americana* and 53 *Blatella germanica* were collected from dining rooms and kitchens of the randomly-selected primary schools in Kaohsiung City. Table 2 shows how the roaches transmitted cysts. The rate of *Periplaneta americana* transmitting *E. histolytica* cysts was 25.42% (76/299); that of transmitting *ameba coli* cysts was 13.38% (40/299). The rate of *Blatella germanica* transmitting *E. histolytica* cysts was 10.34% (3/29); that of transmitting *ameba coli* cysts was 6.90% (2/29). No difference was noted between the rates of either cockroach's transmitting *E. histolytica* cysts. In both types, the *in vivo* transmission rates were significantly higher than the *in vitro*.

#### 4. Discussion

Several reports have indicated that cockroaches are vectors of some protozoa<sup>(11-13)</sup>. Not only do the roaches mechanically transfer pathogenic agents, but they also carry pathogenic agents to become a potential source of infection. Findings of the present study show that transmission rate of *Periplaneta americana* exposed to feces containing 1,000/gm of *E. histolytica* cysts was as high as 90%, significantly higher than the 46.15% of *Blatella germanica*. In feces containing 100/gm of cysts, the transmission rates of both cockroaches differed only slightly in this study which used only adult cockroaches. *Periplaneta americana* is the largest of the household cockroaches, some 35-40 mm in length; *Blatella germanica* is the smallest, from 10-15 mm in length<sup>(8)</sup>. It seems, therefore, that the concentration of cysts in feces, the variety of cockroaches and their sizes might affect the rate of transmission. No significant difference was noted in either the *in vitro* or *in vivo* transmission rates with the exception that *Periplaneta americana*, under 1,000/gm concentration, showed a higher *in vivo* than *in vitro* transmission rate.

Table 2. School-Collected Cockroaches Transmitting Ameba Protozoa

Cockroaches	Transmitting <i>E. histolytica</i>		Transmitting Ameba Coli			
	Rate (%)	No.		Rate (%)	No.	
		Min	Max		Min	Max
<i>Periplaneta americana</i>	25.42 (76/299) <sup>a</sup>	22	3,422	13.38 (40/299) <sup>c</sup>	16	1,000
<i>in vitro</i> *	8.36 (25/299)	22	150	4.68 (14/299)	16	1,000
<i>in vivo</i> *	18.49 (54/299)	44	3,422	8.70 (26/299)	50	817
<i>Blatella germanica</i>	10.34 ( 3/29) <sup>b</sup>	44	444	6.90 ( 2/29) <sup>d</sup>	50	89
<i>in vitro</i> <sup>#</sup>	0.00 ( 0/29)	0	0	0.00 ( 0/29)	0	0
<i>in vivo</i> <sup>#</sup>	10.34 ( 3/29)	44	444	6.90 ( 2/29)	50	89

a:b,  $\chi^2 = 3.28$ ,  $p > 0.05$

c:d,  $\chi^2 = 1.16$ ,  $p > 0.05$

\*:\*, proportional test,  $p < 0.05$

#:#, Fisher's exact test,  $p < 0.05$

The transmission rate of 25.42% of *Periplaneta americana* in the primary school survey did not significantly differ from that of *Blatella germanica*'s 10.34%. The *in vivo* transmission rate for both cockroaches was higher than the *in vitro* rate (that is, fewer cysts of *E. histolytica* were found on the surface). This finding does not agree with the results in the laboratory experiment noted above in that no difference was noted between the *in vitro* and *in vivo* rates. A likely reason is that the *E. histolytica* cysts on the surface of the school cockroaches could have already contaminated the environment before the cockroaches were caught. It can, however, be concluded that avoiding the eating of any food contaminated by cockroaches is important as a preventive medicine measure.

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