Epidemiology and Spatial Relationships of Bacteria Associated with *Periplaneta americana* (Blattodea: Blattidae) in Central Texas[1]

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J. Agric. Urban Entomol. 24(4): 205–216 (October 2007)

ABSTRACT Identifying cockroach (Order: Blattodea) populations is important to understanding the ability of surrogate species indirectly affecting humans by their ability to vector disease-causing organisms including bacteria. These interactions may have potentially deleterious health consequences on animal and/or human populations. In this study, American cockroaches, *Periplaneta americana* were sampled from 12 locations throughout College Station, Texas from January through May 2008. Cockroach distribution was examined as well as prevalence of *Escherichia coli* including the O157:H7 strain and *Campylobacter* spp. on their external surfaces.

Bacteria isolated from total populations collected indicated a high prevalence (92.3%) of microbes carried on the exoskeleton of *P. americana*. Gram-negative bacteria acquisition and dissemination of organisms such as *E. coli* was prevalent throughout the campus. Screening for *E. coli* O157:H7 and *Campylobacter* spp. resulted in no positive colony growth. The lack of *Campylobacter* spp. growth from cuticular surfaces may have resulted from undesirable conditions required to sustain colony growth. Data from this study corroborate the potential ability of cockroaches to mechanically transmit pathogens.

KEY WORDS *Periplaneta americana*, *Campylobacter* spp., *Escherichia coli*, central Texas

Cockroaches (Order: Blattodea) are important vectors of pathogens due in part to their unsanitary lifestyle. Cockroach cuticle can harbor several *Enterobacteriaceae* species including *Salmonella* spp., *Klebsiella* spp., and *Escherichia* spp. (Mpuchane et al. 2006). A few medically important pathogens that can be vectored by the American cockroach, *Periplaneta americana* (Linnæus) (Blattodea: Blattidae), include: *Campylobacter* spp., *E. coli*, *Salmonella* spp., *Shigella* spp., *Staphylococcus* spp., *Streptococcus* spp., and *Toxoplasma gondii* (Barcay 2004). Cockroaches are also able to transmit pathogens such as anthrax, cholera, diphtheria, pneumonia, tetanus, and tuberculosis (Baumholtz et al. 1997). Many of which could be used as bioterrorism agents targeting animal or human populations (Lane et al. 2001, Moran 2002).

Understanding the nature of pathogen transmission from urban insect pests to humans could clarify the epidemiology of many illnesses. The epidemiology of

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these pathogens needs to be thoroughly examined as they relate to cockroaches. Certain disease causing pathogens commonly associated with cockroaches result in gastro-intestinal related illnesses. Pathogens, such as *E. coli* and *Campylobacter* spp., commonly transmitted by cockroaches may be overlooked during diagnosis of sudden ailments with symptoms being similar to food-borne illnesses, including abdominal cramping, diarrhea, nausea, and fever.

Campylobacter spp. are not part of a normal bacterial fauna in humans but have been found in individuals displaying symptoms such as diarrhea and fever (Blaser et al. 1979). In human patients with symptoms of diarrhea, *C. jejuni* has been isolated and causes diarrhea-like symptoms more than *Shigella* spp., *Salmonella* spp., and *E. coli* O157:H7 (Blaser et al. 1979, Blaser 1997).

Diseases associated with *Campylobacter* spp. result from ingesting undercooked poultry, or mishandling raw poultry and cross-contaminating surfaces. *Campylobacter jejuni* is enteric in livestock such as cattle, swine, poultry, companion animals (i.e., dogs and cats), and wild animals such as rodents and raccoons (Blaser 1997, Sahin et al. 2002). *Campylobacter jejuni* is susceptible to atmospheric desiccation and oxygen can inhibit growth in locations such as livestock feed and water (Sahin et al. 2002). However, human interactions with livestock increase the potential risk of contamination.

In human cases, there are several strains of *E. coli* that produce varying effects, ranging from mild fevers to hospitalizations and even death. *Escherichia coli* titers in the environment corresponded with levels of fecal contamination (Le Guyader et al. 1989, Rivault et al. 1994). Transmission of these organisms can follow an unsuspected fecal-oral interaction, such as using a contaminated hand towel and then touching food or the mouth area.

Escherichia coli 0157:H7 is a medically important strain initially reported in 1982 (McGee et al. 2004). It can cause bloody diarrhea, hemolytic uremic syndrome (HUS), and death (McGee et al. 2004). *Escherichia coli* O157:H7 had reported outbreaks in the United States, Great Britain, and Canada, with 20,000 infections and 100 deaths in the United States (Michino et al. 1999).

The objective of this study was to analyze spatial distributions of *E. coli* and *Campylobacter* spp. in relationship to different cockroach populations. This information may determine the spatial distribution of bacterial fauna and identify locations with high bacterial titers.

Materials and Methods

Sampling technique for cockroaches. *Periplaneta americana* (L.) were collected from February 2007–May 2008 within 50 m of neighboring urban structures in the Texas A&M University campus, College Station, Texas. Collecting sites on campus were selected from locations with the highest cockroach populations during preliminary trapping conducted the previous year. Once locations were established, three collecting containers were placed within a 1.83 km^2 square at each trapping location. The north quadrant was approximately 0.29 km^2. The central quadrant was approximately 0.40 km^2. The south quadrant was approximately 0.32 km^2, and the west quadrant had an area of approximately 0.58 km^2. Coordinates of each site were determined with a Garmin, Blaser 1997 eTrex® Vista Cx GPS unit (Garmin Ltd., Olathe, KS, USA) and data points uploaded to Google Earth™.

Containers used for collecting roaches were glass mason jars (430 mL) coated with Elmer's Acid Free Craft Bond© (Elmer's Products, Inc., Columbus, Oh, USA) and rolled in Quickrete® Playsand (Quickrete® International, Inc., Atlanta, GA, USA), according to Granovsky (1983). The top 2 cm of the jar opening was lined with H-E-B brand petroleum jelly (H-E-B, San Antonio, TX, USA) and baited with approximately 51.76 mL beer (Miller Brewing Co., Milwaukee, WI, USA), and 7.04 g of H-E-B brand white bread (H-E-B, San Antonio, TX, USA) for attracting and collecting cockroaches (Barcay 2004). Baited containers were placed in the field at dusk immediately after adding the beer/bread mixture and collected from the field after 8–12 h. Cockroaches collected were stored in a freezer at −20°C until analysis.

Adult cockroaches were collected from each jar and stored in individual plastic bags (16.5 × 14.9 cm), with up to three plastic bags containing roaches from each site. This method should not negatively influence bacterial colony growth (Szalanski et al. 2004). Voucher specimens were placed in the Texas A&M University insect collection.

Screening for *Escherichia coli* activity. Media used for screening *Escherichia coli* followed the CHROMagar™ ECC media manufacture's recipe (CHROMagar, Paris, France). *Escherichia coli* O157:H7 specific media was made using CHROMagar™ 0157 (CHROMagar, Paris, France) ratio.

Agar was poured into sterile petri dishes (100 × 15 mm, VWR International, West Chester, PA, USA). Petri dishes were divided into thirds and appropriately labeled for the specimen. Working under sterile conditions, forceps were flame sterilized using 95% ethanol and cooled prior to touching the cockroach to be plated. Dorsal and ventral sides of each cockroach were plated within their designated areas. Once the cockroach was plated it was moved to an isolated area, the forceps were sterilized using the aforementioned flaming technique. The process was repeated for all *P. americana* collected.

Escherichia coli samples plated on CHROMagar ECC and CHROMagar 0157 were incubated in a Percival Environmental Chamber Model I36LLVL (Percival Scientific, Inc., Perry, IA, USA) at 37°C for 24–48 h. Blue colored colonies were identified as *E. coli*, red colonies were coliform forming bacteria, and colorless colony forming units were non-coliform forming gram-negative bacteria and counted. Screening for *E. coli* O157:H7 followed the same technique, but with positives indicated by a mauve coloration

Colonies that were positive for *E. coli* were stored in sterile 1.5 mL microtubes with snap caps (VWR International, West Chester, PA, USA) in a 60% Tryptic soy agar (Fisher Scientific, Pittsburg, PA, USA)/40% glycerin (Fisher Scientific, Fair Lawn, NJ, USA), and frozen at −80°C, according to Hanahan et al. (1995).

Screening for *Campylobacter* species activity. *Campylobacter* specific media was made using the following procedure: 25 mL defibrinated sheep blood (Colorado Serum Co, Denver, CO, USA), one tube of antibiotic premix, 21.5 g BBL™ Brucella agar (BD, Becton, Dickinson and Co., Sparks, MD, USA), and 500 ml distilled water. Antibiotic premix was made by suspending 159.0 mg Cephalothin (MP Biomedicals, LLC., Solon, OH, USA), 50.0 mg Trimethoprim Lactate (Research Products International Corp., Prospect, IL, USA), 100.0 mg Vancomycin hydrochloride (Acros Organics, Morris Plains, NJ, USA), 3.22 mg Polymyxin B (InvivoGen, San Diego, CA, USA), and 20.0 mg Amphotericin B (Acros Organics, Morris Plains, NJ, USA) into 100 mL distilled, sterile water.

The total antibiotic premixture was divided into 20 tubes each containing 5 mL aliquots, covered with parafilm (American National Can™, Greenwich, CT, USA), and stored in a −20°C freezer. Plating methods previously described for screening for the presence of *E. coli* were also used when screening for *Campylobacter* spp.

Campylobacter spp. specific media was grown in an anaerobic environment for 96 h prior to checking for growth. An anaerobic environment was achieved by placing a BD BBL™ CampyPak™ Plus Microaerophilic system envelope with Palladium catalyst (Becton, Dickinson and Company, Sparks, MD, USA) in an acrylic canister (17.8 × 12.7 cm, Oggi Co., Anaheim, CA, USA) with a chrome locking clamp with a silicone gasket that sealed air tight. *Campylobacter* spp. selective media were removed from the anaerobic environment after 96 h followed by identification and prevalence of colonies.

Statistical analysis. JMP Statistical Discovery software version 5.1 (SAS Institute Inc., Cary, North Carolina) was used for the analysis of all results. Oneway ANOVA ($\alpha = 0.05$) was performed to analyze the mean total population numbers collected and quadrant counts. A Tukey-Kramer HSD was used to separate means. A linear regression was performed mean daily temperature and cockroach collected. Oneway ANOVA ($\alpha = 0.05$) was performed to analyze the mean bacteria colony forming units for *E. coli*, coliform forming gram-negative, and non-coliform forming gram-negative, and quadrant counts. Oneway ANOVA ($\alpha = 0.05$) was performed to analyze the mean number of bacteria colony forming units for *E. coli*, coliform forming gram-negative, and non-coliform forming gram-negative; cockroach stage of development; and quadrant counts.

Results

Periplaneta americana (L.) (n = 687), were collected from four designated areas, north, central, south and west, from the Texas A&M University campus College Station, Texas (Fig. 1). The mean number of cockroaches collected from January–May 2008 was 3.67 ± 4.23 (3.10 ± 3.31 nymphs and 0.56 ± 1.73 adults) per day. The north quadrant had the lowest mean of cockroaches collected with 1.86 ± 1.60 total (1.86 ± 1.25 nymphs and 0.00 ± 0.65 adults). The central quadrant had a mean of 2.21 ± 1.13 cockroaches with 2.14 ± 0.88 nymphs and 0.07 ± 0.46 adults. The south quadrant had a mean of 4.05 ± 0.94 total (3.25 ± 0.74 nymphs and 0.80 ± 0.39 adults). The mean number of cockroaches collected in the west quadrant was 7.29 ± 1.60 total (5.86 ± 1.25 nymphs and 1.43 ± 0.65 adults). There was no significant difference ($F = 2.746$; df = 4, 160; $P = 0.0542$) between population means within quadrants (north, central, south, and west).

There were five categories (Table 1) for building and/or structures from which cockroach populations were collected adjacent to: administration (primarily offices, some classrooms, and vending machines); lecture buildings (primarily lecture or research areas, some offices, and vending machines); dining halls (food establishments on campus with the primary purpose of food and beverage distribution); water tower; and garage. The prevalence of bacteria on cockroaches for each building type indicated that administration buildings had the highest positive rate of cockroaches (Table 1), while the dining hall maintained the lowest rate of prevalence on *P. americana* populations.

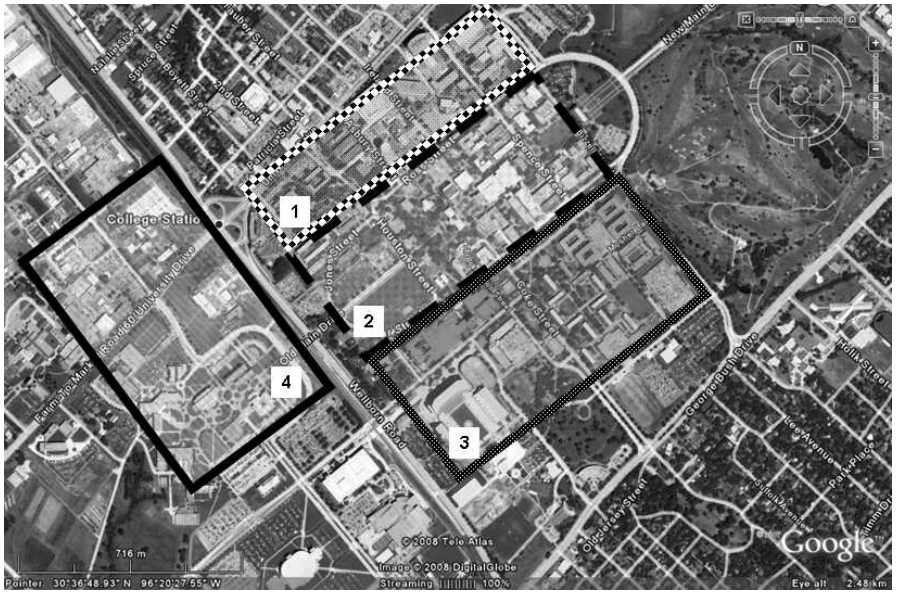

Fig. 1. The Texas A&M University campus, College Station, Texas, divided into four areas, north (checkered) 1, central (dashed) 2, south (dots) 3, and west (solid) 4, used for sampling cockroach populations. Images taken from Google™ Earth Plus v. 4.3.

Microbes isolated from total populations collected indicated a high prevalence (92.3%) of bacteria on the exoskeleton of *P. americana* (Table 2). Bacterial screening for *E. coli* resulted in a significant difference ($F = 2.468$; df = 4, 694; $P = 0.0437$) between quadrants (Table 3). There were also cockroaches that after plating had too many bacterial colony forming units to count. The north quadrant had one *E. coli*, seven coliform forming colonies, and zero non-coliform forming

Table 1. Positive rates of bacterial (*E. coli*, coliform forming gram-negative, and non-coliform forming gram-negative) prevalence for *P. americana* populations collected on the Texas A&M campus, College Station, Texas, as categorized by building function.

Building type	Cockroach population[a]
Administration	427/687 (62.2%)
Lecture building	103/687 (15.0%)
Dining hall	2/687 (0.3%)
Water tower	75/687 (10.9%)
Garage	80/687 (11.6%)

[a]Percentages based on the number of cockroaches collected at each building type compared to the total number of cockroaches collected from February 2007–May 2008.

Table 2. Prevalence of bacteria (*E. coli*, coliform forming gram-negative, and non-coliform forming gram-negative) from the total cockroach population collected on the Texas A&M University campus, College Station, Texas.

Location[a]	*E. coli*[b]	Coliform (G-)	Non-coliform (G-)	Total
North	46/104 (44.2%)	102/104 (98.1%)	87/104 (83.7%)	103/104 (99.0%)
Central	105/155 (67.7%)	145/155 (93.5%)	126/155 (81.3%)	154/155 (99.4%)
South	169/354 (47.7%)	271/354 (76.6%)	225/354 (63.6%)	310/354 (87.6%)
West	31/74 (41.9%)	54/74 (73.0%)	50/74 (67.6%)	64/74 (86.5%)
College Station	23/37 (62.2%)	36/37 (97.3%)	37/37 (100.0%)	37/37 (100.0%)
Total	374/724 (51.7%)	608/724 (84.0%)	525/724 (72.5%)	668/724 (92.3%)

[a]Collections from north, central, south, and west were quadrants on the Texas A&M University campus, College Station, Texas, and College Station specimens were from undisclosed locations in College Station, Texas.

[b]Percentages based on number of cockroaches with colony forming units compared to total number of cockroaches collected in each quadrant.

Table 3. Bacterial counts from cockroach populations screened from each quadrant on the Texas A&M University campus, College Station, Texas.

Bacteria	Location[a]	n	Bacterial mean ± SE[b]	95% Mean Upper	95% Mean Lower
E. coli	North	118	24.14 ± 3.86 ab	16.56	31.72
	Central	150	24.45 ± 3.42 ab	17.73	31.18
	South	318	25.75 ± 2.35 a	21.14	30.37
	West	72	19.51 ± 4.92 ab	9.81	29.22
	College Station	37	3.76 ± 6.89 b	−9.78	17.29
Coliform	North	106	93.92 ± 6.24 c	81.67	106.16
	Central	152	26.91 ± 5.21 e	16.68	37.14
	South	305	69.36 ± 3.67 d	62.14	76.58
	West	70	85.44 ± 7.67 cd	70.37	100.52
	College Station	37	22.35 ± 10.56 e	1.62	43.08
Gram-	North	120	17.73 ± 3.08 f	11.68	23.77
	Central	149	21.87 ± 2.76 f	16.44	27.29
	South	308	24.90 ± 1.92 f	21.13	28.67
	West	71	15.93 ± 4.00 f	8.08	23.78
	College Station	37	14.41 ± 5.54 f	3.53	25.29

[a]Collections from north, central, south, and west were quadrants on the Texas A&M University campus, College Station, Texas, and College Station specimens were from undisclosed locations in College Station, Texas.

[b]Same letters following means within a column were not significantly different ($P < 0.05$, Tukey-Kramer HSD).

colonies. Central quadrant had four *E. coli*, two coliform forming colonies, and five non-coliform forming colonies. The south quadrant had the most with 28 *E. coli*, 14 coliform forming colonies, and 11 non-coliform forming colonies. The west quadrant had no *E. coli*, two coliform forming colonies, and one non-coliform forming colony. Various locations on the Texas A&M University campus resulted in zero plates with too many to count (Table 4). Coliform forming bacteria were significantly different ($F = 24.728$; df = 4, 665; $P < 0.001$) between quadrants, while non-coliform forming gram-negative bacteria had no significant difference ($F = 2.0573$; df = 4, 680; $P = 0.0848$) (Table 3).

There was no significant difference ($F = 0.0420$; df = 2, 205; $P = 0.8379$) between mean number of adult and nymph cockroaches collected and *E. coli* forming units (Table 5). There was no significant difference ($F = 3.0748$; df =2, 216; $P = 0.0809$) between adult and nymph stages of cockroaches collected and coliform-forming bacteria units (Table 5). There was no significant difference ($F = 0.0003$; df = 2, 216; $P = 0.987$) between adult and nymph cockroaches collected and non-coliform forming bacteria units of (Table 5).

Screening for *E. coli* O157:H7 and *Campylobacter* spp. yielded no positive colony forming units for all of the samples screened (n = 724).

Discussion

The purpose of this study was to determine the amount and viability of bacteria harbored by *P. americana* in an outdoor, urban environment by observing commonly occurring and ubiquitous bacteria such as *E. coli* and *Campylobacter*. Outdoor collecting sites on campus provided insight into American cockroach population within an artificial environment. There were no significant differences between collecting sites in each quadrant and

Table 4. Prevalence of cockroach specimens plated for *E. coli*, coliform forming gram-negative, and non-coliform forming gram-negative that resulted in too many bacteria colony forming units to count for cockroaches collected on the Texas A&M University campus, College Station, Texas and various undisclosed locations in College Station, Texas.

Location[a]	*E. coli*[b]	Coliform (G-)	Non-coliform (G-)	Total
North	1/104 (0.009%)	7/104 (0.067%)	0/104 (0%)	8/104 (0.077%)
Central	4/155 (0.026%)	2/155 (0.013%)	5/155 (0.032%)	11/155 (0.071%)
South	28/354 (0.079%)	14/354 (0.040%)	11/354 (0.031%)	53/354 (0.150%)
West	0/74 (0%)	2/74 (0.027%)	1/74 (0.014%)	3/74 (0.041%)
College Station	0/37 (0%)	0/37 (0%)	0/37 (0%)	0/37 (0%)
Total	33/724 (0.046%)	25/724 (0.035%)	17/724 (0.023%)	75/724 (0.102%)

[a]Collections from north, central, south, and west were quadrants on the Texas A&M University campus, College Station, Texas, and College Station specimens were from undisclosed locations in College Station, Texas.

[b]Percentages based on the number of specimens with too many bacteria colony forming units to count compared to the total number of specimens collected from each location.

Table 5. Comparison of bacteria counts for adults and nymphs in all quadrants collected on the Texas A&M University campus, College Station, Texas.

Bacteria	Stage	n	Bacteria Mean ± SE[a]	95% Mean Upper	95% Mean Lower
E. coli	Adult	77	18.64 ± 5.31 a	8.16	29.11
	Nymph	131	20.01 ± 4.07 a	11.98	28.04
Coliform	Adult	83	88.02 ± 21.34 a	45.97	130.08
	Nymph	136	135.51 ± 16.67 a	102.65	168.36
Non-coliform	Adult	83	85.98 ± 32.68 a	21.56	150.39
	Nymph	136	86.65 ± 25.53 a	36.33	136.97

[a]Same letters following means within a column were not significantly different ($P < 0.05$, Tukey-Kramer HSD).

populations of *P. americana* collected. Specific areas of campus did appear to yield higher populations based on observations.

Haines & Palmer (1955) determined that *P. americana* was a predominant species in sewer systems with low population densities indoors and around the home; although the restrooms of indoor facilities maintained the highest population numbers. Overall, building type does not play a significant role in the population densities of cockroaches. The assumption can be made that the same applies for an area such as a university where cockroaches were ubiquitous in the environment.

Pai et al. (2003) determined that adult populations of *P. americana* and *Blatella germanica* L. were significantly higher than nymph populations collected in hospitals, which fails to correspond with data found in this study. There were no significant differences between adult and nymph populations collected around campus. The difference between studies may result from a difference in collection techniques or that the Pai et al. (2003) study was conducted indoors, from a single structure (hospitals) type. Our study exploited various collecting locations and their outdoor structures.

Spatial distribution of natural population is typically patchy. Resource levels fluctuate over time in individual locations, thus population numbers will also change over time indicating a patchy distribution (Roughgarden, 1977). Population fluxes are normal because collecting cockroaches from outside coincided with weather. Population surges may result from rainfall, food availability, an overabundance of water in sewer systems, and/or external weather conditions.

Campylobacter spp. are not part of a normal bacterial fauna in humans but has been found in individuals displaying symptoms such as diarrhea and fever (Blaser et al. 1979). In human patients with symptoms of diarrhea, *C. jejuni* has been isolated to cause diarrhea-like symptoms more than *Shigella* spp., *Salmonella* spp., and *E. coli* O157:H7 (Blaser et al. 1979, Blaser 1997).

Cockroaches could be competent carriers of nosocomial infection agents, especially to patients in neonatal units, intensive care, and immunocompromised patients (Elgderi et al. 2006, Fotedar et al. 1991, Gliniewick et al. 2003,

Salehzadeh et al. 2007). Nosocomial infections may result from pathogens on food; a contaminated water supply; and/or unsanitary facilities, like bathrooms (Lemos et al. 2006). Salehzadeh et al. (2007) described cockroaches collected in hospitals to have greater bacterial counts than cockroaches found in residential areas. Hospital environments may be more conducive to bacterial acquisition from numerous contaminated sources such as water, food, and/or harborage thus resulting in higher rates of bacteria prevalence. Multiple drug-resistant bacterial strains of medical importance have also been isolated from cockroaches in several hospitals (Elgderi et al. 2006, Fotedar et al. 1991, Gliniewick et al. 2003, Salehzadeh et al. 2007).

Escherichia coli can be found on both internal and external surfaces of cockroaches (Rivault et al. 1994). The current study concurred with the Le Guyader al. (1989) study of gram-negative bacteria amounts not having a significant difference between adults and nymphs. Despite the stigma of cockroaches being filth laden, Bell et al. (2007) indicated cockroaches spending at least half of their time grooming and removing foreign objects from their body. The amount of time spent cleaning is inadequate because of contamination of the habitat and the capability to become re-inoculated with pathogens present in the environment. The ability to harbor bacteria on internal and external surfaces provides multiple means of pathogen transmission. In addition to direct contact with surfaces, cockroaches can disseminate internal organisms via defecation and/or regurgitation.

Compared to previous studies made indoors, the presence of bacteria on cockroaches appears to correlate with other studies with positives rates of bacteria in Ghana, France, and Taiwan (Agbodaze & Owusu 1989, Pai et al. 2004, Rivault et al. 1994). Overall, 92.3% of cockroaches collected from outdoor locations on campus carried gram-negative bacteria on their cuticular surfaces. Pai et al. (2005) determined there was no significant difference between *P. americana* and *B. germanica* incident rates of positive growth of bacterial colonies on the integument and the gut. Although, *P. americana* had significantly higher rate of gram-negative colonies than *B. germanica* (Pai et al. 2005). A previous study indicated cockroaches harbored bacteria present in the surrounding environment, as opposed to introducing new pathogens into the environmental fauna (Rivault et al. 1993).

During this study, it was assumed cockroaches were mechanically transmitting pathogens obtained in the environment and were capable of traveling while harboring these bacteria. This creates a public health concern if cockroaches inoculated with bacteria from outside migrated indoors and transmitted pathogens to sterile surfaces, such as areas in the kitchen. Chaichanawongsaroj et al. (2004) indicated *E. coli* levels on cockroaches coincided to *E. coli* levels in the environment. Rivault et al. (1993) discussed that not all bacteria would be able to survive on surfaces that a cockroach contacted.

Contamination rates of cockroaches compounded with their gregarious behavior could provide a mode for pathogens to spread to surfaces having direct contact with food. During this study, 51.7% of all cockroaches trapped were contaminated with *E. coli*. This was the lowest percentage of positive bacteria out of all the cockroaches screened for colony forming units. Despite having the lowest percentage of prevalence, one out of every two cockroaches on campus was carrying *E. coli*. A comparison was made to determine if the life stage (adult or

nymph) made an impact on bacteria associations with the cockroaches and found there to be no significant difference.

Data indicated collection locations as related to *E. coli*, coliform forming gram-negative bacteria were significantly different while there was no significant difference between non-coliform forming gram-negative bacterial species. It was interesting to note differences among collected populations and prevalence of bacteria, despite collecting sites being up to 1.44 km apart. A significant difference may indicate the environment of various collecting locations having differing compositions of bacteria. It is possible that the values for *E. coli* were not significantly difference for each quadrant even though the *P*-value indicated a significant difference. There were 75 specimens that resulted with too many bacteria colony forming units to count. These numbers should not have affected the overall significant difference between populations, quadrants, and bacteria species because the number of too many to count bacteria colony forming units were proportional to initial rates of prevalence among populations collected on campus. No differences for non-coliform forming gram-negative bacteria among collected populations implies that cockroaches may have obtained bacteria from common means throughout campus, such as soil in the flowerbeds or a common water source any of which may have been contaminated with bacteria.

A common water source that may have been easily accessible to all specimens is through the sewer systems. *P. americana* may have traveled from one area of campus to another through various methods of transportation. Cockroaches are capable of migration by ground movement, climbing vertical surfaces, swimming, and some limited flight capabilities (Bell et al. 2007). Jackson & Maier (1955) determined through capture and release experiments that cockroaches could travel through the sewer up to 107 m. It is possible cockroaches remained in locations until resources were depleted and then dispersed in search of food.

All specimens collected were negative for *E. coli* O157:H7. Presence of this pathogen usually occurs in livestock area because cattle and sheep act as reservoirs for the pathogen (McGee et al. 2004). There were no locations on campus that housed livestock which were regularly sampled for cockroach populations. This may have contributed to why there were no positives for *E. coli* O157:H7.

Overall, this study displayed the wide distribution of cockroach populations on campus and their ability to indiscriminately inhabit areas within an urban environment. Pathogen acquisition and dissemination of gram-negative bacteria, such as *E. coli*, was prevalent on campus but without detection of the highly pathogenic strain of *E. coli* O157:H7. Also, there was a lack of *Campylobacter* spp. growth from cuticular plating which may have resulted from undesirable conditions required to sustain colony growth.

Acknowledgment

The authors would like to thank Alan Szalanski for his input and review which improved an early version of the manuscript.

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