JOURNAL OF CLINICAL MICROBIOLOGY, Oct. 2005, p. 5202–5207 Vol. 43, No. 10
0095-1137/05/$08.00+0 doi:10.1128/JCM.43.10.5202–5207.2005

Molecular Characterization of Hand Flora and Environmental Isolates in a Community Setting

Preeti Pancholi,[1]* Mimi Healy,[2] Tracy Bittner,[2] Renee Webb,[2] Fan Wu,[1] Allison Aiello,[3] Elaine Larson,[4] and Phyllis Della Latta[1]

Clinical Microbiology Laboratory, Department of Pathology, New York Presbyterian Hospital, Columbia University Medical Center, New York, New York[1]; Spectral Genomics, Inc., Houston, Texas[2]; Center for Social Epidemiology & Population Health, Ann Arbor, Michigan[3]; and Department of Epidemiology, Joseph Mailman School of Public Health, Columbia University School of Nursing, and College of Physicians and Surgeons, Columbia University, New York, New York[4]

Received 5 April 2005/Returned for modification 16 June 2005/Accepted 7 July 2005

We analyzed 69 bacterial isolates, comprising seven species of gram-negative bacterial rods and three species of coagulase-negative staphylococci, recovered from both the hands of caretakers and their environment in households sampled in upper Manhattan. Repetitive sequence-based PCR and dendrogram analysis were used to determine strain similarity. Greater than 25% of individual species of *Acinetobacter*, *Enterobacter*, and coagulase-negative staphylococci recovered from the hands and immediate environment within each household shared the same genotype. This study is the first to demonstrate the frequency of bacteria shared within community households. These strains may serve as potential reservoirs for either community- or hospital-acquired infections.

Published studies that examine the microbial flora in community settings are sparse in comparison to those within hospital environments (1, 14). Nosocomial infections can emanate from endemic hospital strains or from microorganisms endogenous in the community. Recently, nosocomial transmission of a community-associated strain of methicillin-resistant *Staphylococcus aureus* (CA-MRSA) among postpartum women has been reported (28), underscoring the clinical importance of determining the frequency of shared or similar bacterial genotypes within households. Studies examining the microbial ecology of microorganisms within households are scarce but are clinically relevant, because they enhance our understanding of pathogen transmission within both community and hospital settings. The emergence of nosocomial infections due to gram-negative rods (GNR) is of paramount importance. Infections due to *Acinetobacter* species, particularly *Acinetobacter baumannii*, causing nosocomial bacteremia and pneumonia have a high mortality rate (9, 11). However, there is a lack of published reports concerning the epidemiology of clonal transmission of *A.baumannii* and other GNR within community settings.

The advent of molecular technology has facilitated the examination of strain similarities. Molecular methods available for genotyping isolates are separated into those using DNA amplification by PCR and non-nucleic acid amplification techniques (26, 35). Both pulsed-field gel electrophoresis and repetitive sequence-based PCR (rep-PCR) analyze large parts of the bacterial genome, and they offer greater discriminatory power than those that employ the use of small regions of single genes or operons (e.g., rRNA) (26, 33). PCR typing methods using specific primers designed on the basis of the repetitive and conserved sequences in bacteria and stringent annealing conditions generate complex fingerprint patterns for strain typing.

Some reports have shown that results obtained using rep-PCR are superior to those obtained using manual or automated ribotyping (6, 8, 19). The two types of repetitive elements most commonly used to differentiate a vast variety of strains are the enterobacterial repetitive intergenic consensus and repetitive extragenic palindromic sequences (7, 16, 31). The rep-PCR technology was chosen for strain typing in this study because of its rapidity, relatively low cost, ease of use, and applicability to typing a wide variety of strains, e.g., *S. aureus* (32), *Streptococcus* species (8, 23), *Clostridium difficile* (30), *Salmonella enterica* serovar Enteritidis (5), *Shigella* species (18), *Enterococcus faecalis* (20), *Enterobacter aerogenes* (21), *Burkholderia cepacia* (19), *Stenotrophomonas maltophilia* (22), *A. baumannii* (3), *Pasteurella multocida* (10), *Vibrio parahaemolyticus* (34), *Lactobacillus johnsonii* (33), *Helicobacter pylori* (4), and *Bartonella henselae* (29).

The goal of the present study was to determine if bacterial isolates recovered from the hands of caretakers were similar or different from those obtained from a designated environmental site within their households. To the best of our knowledge, this is the first study of its kind to determine the prevalence and frequency of bacterial strains, particularly GNR, shared within community settings.

MATERIALS AND METHODS

Specimen collection. A total of 238 primary caretakers living in inner-city multigenerational households in northern Manhattan, New York, with an immigrant population, predominantly from the Dominican Republic, were enrolled in this study. The participants were part of a larger clinical trial to determine the effect of hygiene practices on infectious disease symptoms (15).

Hand cultures were obtained during a home visit using a modification of the "glove juice" method (14). Briefly, a randomly selected hand was cultured immediately after washing by placing in a sterile polyethylene bag containing 50 ml

* Corresponding author. Mailing address: University Hospital East, 3N, 1492 East Broad Street, Columbus, OH 43205. Phone: (614) 257-3488. Fax: (614) 257-2405. E-mail: pd23@columbia.edu.

TABLE 1. Rep-PCR analysis of paired bacterial strains from hands and household environments

Bacteria	No. of pairs tested	Strain characterization of paired isolates	
		% Different (no./no. tested)	% Similar (no./no. tested)
GNR			
A. baumannii	15	60 (9/15)	40 (6/15)
A. lwoffii	14	85.7 (12/14)	14.3 (2/14)
E. agglomerans	12	91.7 (11/12)	8.3 (1/12)
E. cloacae	2	50 (1/2)	50 (1/2)
K. pneumoniae	9	55.6 (5/9)	44.4 (4/9)
P. fluorescens/P. putida	3	66.7 (2/3)	33.3 (1/3)
S. maltophilia	1	100 (1/1)	0
Total	56	73.2 (41/56)	26.8 (15/56)
GPC			
S. warneri	9	77.8 (7/9)	22.2 (2/9)
S. epidermidis	3	33.3 (1/3)	66.7 (2/3)
S. haemolyticus	1	0 (0/1)	100 (1/1)
Total	13	61.5 (8/13)	36.5 (5/13)
Total (GNR + GPC)	69	71 (49/69)	28.9 (20/69)

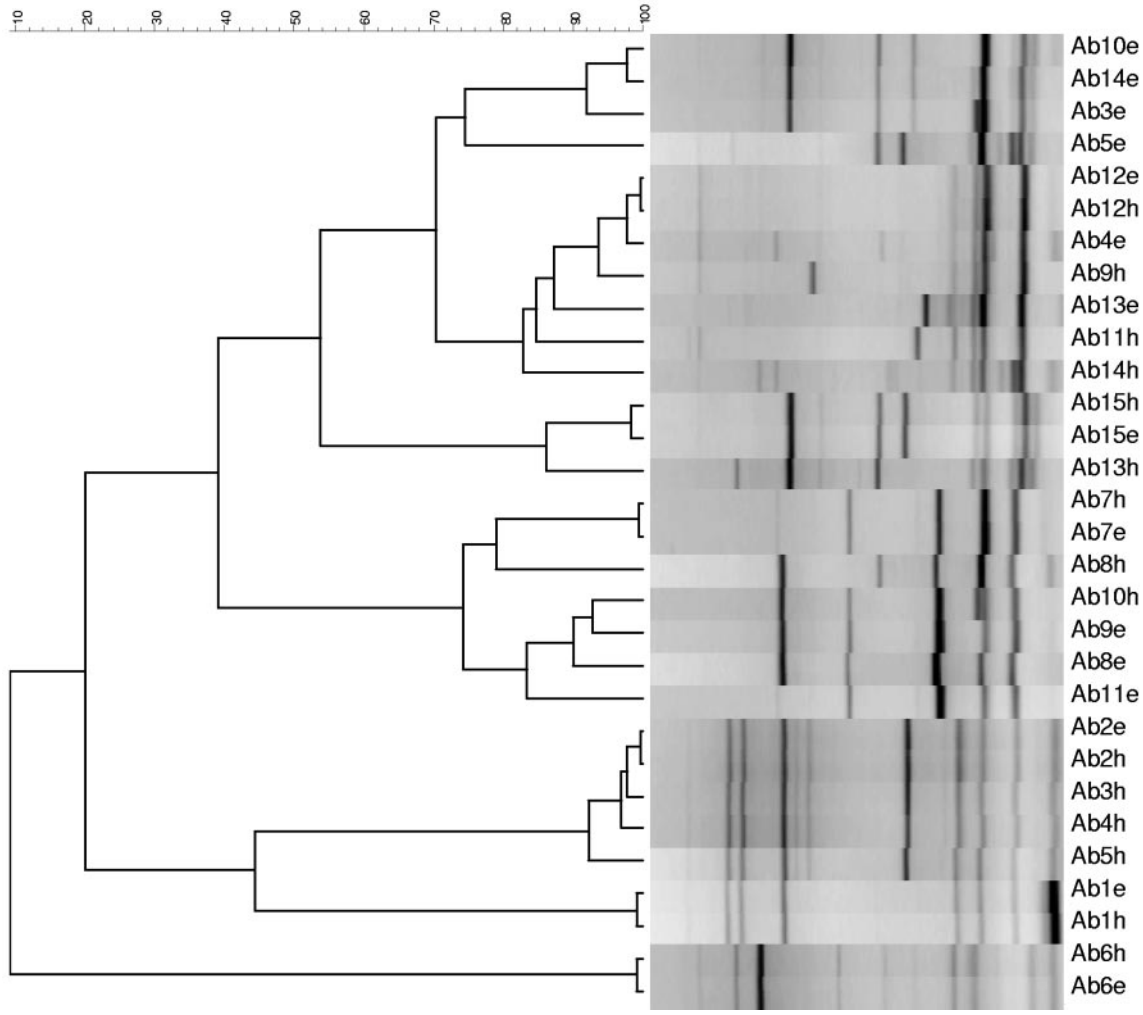

FIG. 1. Rep-PCR gel images and dendrogram of *A. baumannii* strains isolated from hands (h) and the environment (e), showing the degree of strain similarity.

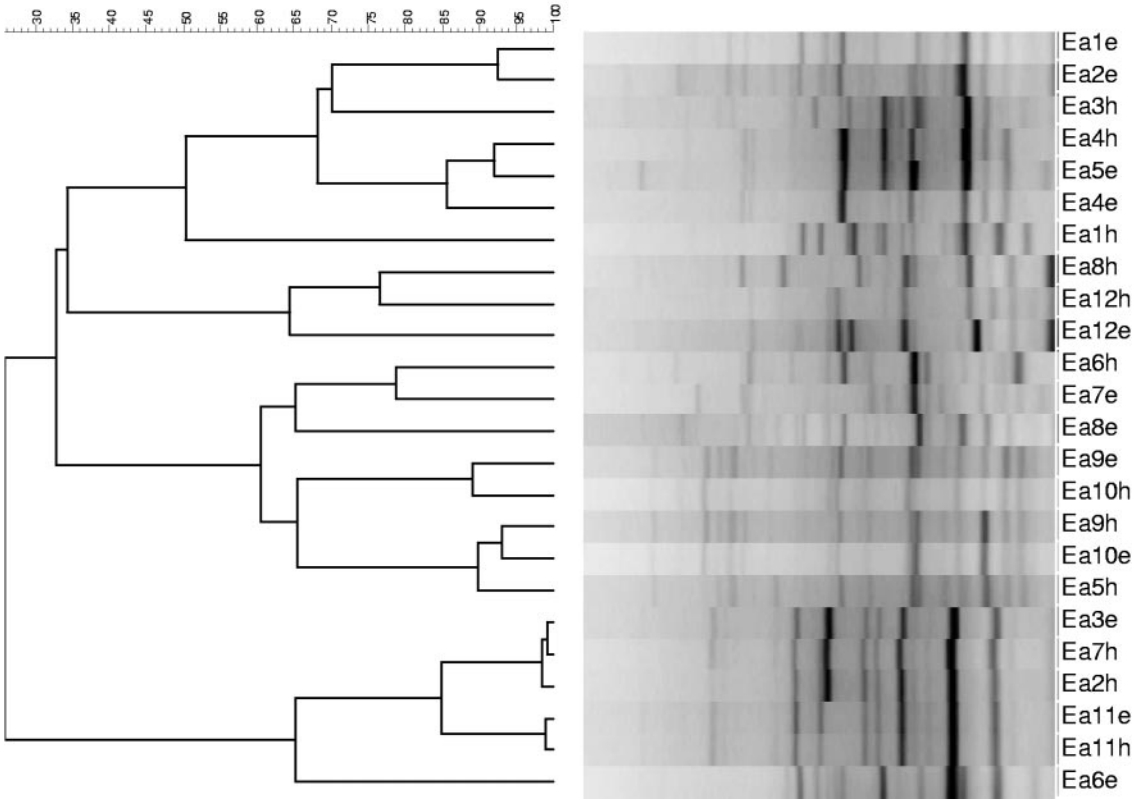

FIG. 2. Rep-PCR gel images and dendrogram of *E. agglomerans* strains isolated from hands (h) and the environment (e), showing the degree of strain similarity.

of sampling solution (0.075 M phosphate buffer, pH 7.9, containing 0.1% polysorbate 80 and 0.1% sodium thiosulfate). The hand was massaged through the wall of the bag for 60 seconds by a trained data collector, and samples were hand delivered to the clinical microbiology laboratory for processing.

To detect environmental reservoirs, an area about the size of a quarter was sampled from the table used for eating meals, using a premoistened cotton-tipped swab (Mini-Tip culturette collection and transport system; Becton Dickinson Microbiology Systems, Sparks, MD). This environment site was chosen since the subjects reported eating most of their meals within the home at the table and, therefore, it reflected a high-contact/high-traffic area.

After collection, swabs were placed into 2 ml of sampling solution, capped, and delivered to the clinical microbiology laboratory.

Specimen processing. All specimens were processed within 2 to 3 h of collection or refrigerated for up to 12 h before processing. Samples were diluted 10-fold in 0.85% normal saline, up to 10^{-3}, and plated on five different medium types to optimize the recovery of bacterial and fungal organisms. The enriched and selective agar media included Columbia agar with 5% sheep blood, MacConkey, colistin-nalidixic acid, Sabouraud's with chloramphenicol and gentamicin, and bile esculin (Becton Dickinson Microbiology Systems, Sparks, MD). The plates were incubated at 35°C and examined daily for microbial growth over 48 h.

Species identification. Gram-negative and gram-positive bacteria were identified to the species level using the MicroScan system (Dade Behring Inc., Deerfield, IL). A high percentage (≥95%) was utilized as the acceptance criterion for identification by MicroScan. Staphylococci were additionally identified by coagulase and Staphaurex (Remel Europe Ltd., Kent, United Kingdom.). All isolates were frozen at −70°C for further analyses.

Epidemiologic strain typing. From each study household, paired specimens containing the same bacterial species isolated from both the hand of the primary caretaker and the environmental area were evaluated. For each pair, isolates were genotyped using rep-PCR (DiversiLab DNA fingerprinting kits; Spectral Genomics, Houston, TX) to determine strain similarities.

Rep-PCR. Cultures were incubated overnight (37°C), and the DNA was extracted from 1 μl of culture with the UltraClean microbial DNA isolation kit (Mo Bio Laboratories, Inc., Solana Beach, CA). The DNA samples were standardized to 25 ng/μl, and genomic integrity was verified using conventional agarose gel electrophoresis. DNA (50 ng) from staphylococci was amplified using the DiversiLab *Staphylococcus* kit (Spectral Genomics) and from GNR using a DiversiLab DNA fingerprinting kit and UprimeE1 plus UprimeB2 primers.

The following thermal cycling parameters were used: initial denaturation at 94°C (2 min), 35 cycles of denaturation at 94°C (30 s), annealing at 55°C (or 45°C for staphylococci) (30 s), extension at 70°C (90 s), and a final extension at 70°C (3 min). The amplicons were separated by 1.5% agarose gel electrophoresis, and digital images of the gels were captured with the AlphaImager 2200 (Alpha Innotech Corp., San Leandro, CA). Samples were analyzed with BioNumerics software package using the Pearson's correlation coefficient and unweighted pair group method with arithmetic means to create dendrograms (Applied Maths, Inc., Austin, TX). The relatedness was determined by cluster analysis and guidelines provided by the manufacturer. Isolates were categorized as different, similar, or indistinguishable. Because homogeneous organisms (such as MRSA) have less variation between genomes while heterogeneous organisms (such as *Pseudomonas* and *Acinetobacter*) have a larger natural presence in the environment and have greater genomic variations, the following criteria were applied to determine relatedness. In general, "different" was defined as <95% similarity and two band differences for homogeneous organisms or three band differences for heterogeneous organisms. "Similar" was defined as <97% similarity and one band difference for homogeneous organisms or up to two band differences for heterogeneous organisms. "Indistinguishable" was defined as >95% similarity and no banding differences, including no variation in intensities of individual bands, although overall intensities may differ. DNA from a *Pantoea agglomerans* strain was included as a positive kit control, while water was included as a negative control for the performance of rep-PCR.

RESULTS

Bacterial strains. There were 69 instances in which the same bacterial species were recovered from both the hand and the environmental surface sampled (Table 1). Of these, 81% were

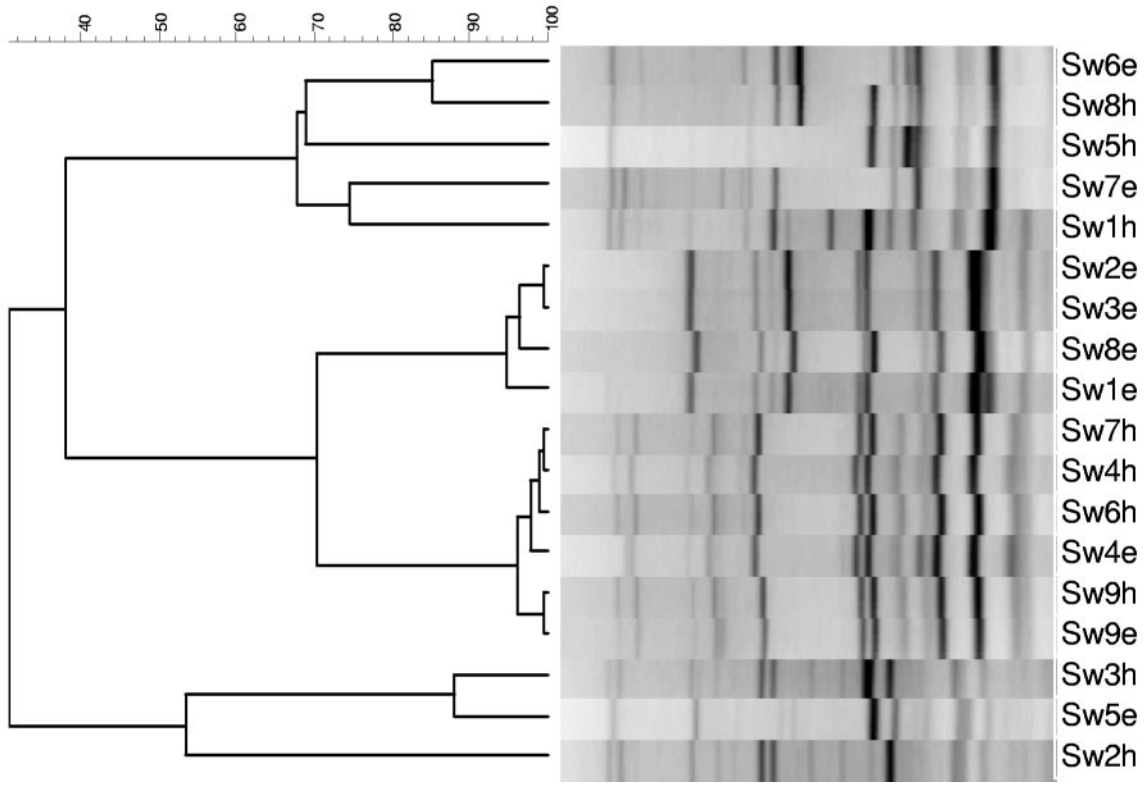

FIG. 3. Rep-PCR gel images and dendrogram of *S. warneri* strains isolated from hands (h) and the environment (e), showing the degree of strain similarity.

GNR and 19% were gram-positive cocci (GPC). Among the GNR matched pairs, 29 were *Acinetobacter* species (52%), of which 15 *A. baumannii* and 14 *Acinetobacter lwoffii* isolates were found. The *Acinetobacter* strains were susceptible to the majority of antimicrobial agents tested, and results have been published elsewhere (37). The remaining GNR included 12 *Enterobacter agglomerans*, 2 *Enterobacter cloacae*, 9 *Klebsiella pneumoniae*, 3 *Pseudomonas fluorescens*/*P. putida*, and 1 *S. maltophilia* isolate. All GPC matched pairs were coagulase-negative staphylococci and included nine *Staphylococcus warneri*, three *Staphylococcus epidermidis*, and one *Staphylococcus haemolyticus* isolate.

Strain differentiation by rep-PCR. DNA fragment patterns by rep-PCR were obtained for each matched bacterial species. The dendrogram analyses revealed significant correlations between hand and the environmental isolates. Similar strains were identified between the paired isolates from the hands and environment by using a Pearson's correlation coefficient of >95% similarity.

(i) Relationships between paired hand and environmental strains. Results indicated that nearly 29% of all paired hand and environmental isolates tested were similar (26.8% GNR and 36.5% GPC) (Table 1). For GNR, *A. baumannii*, 6/15 (40%) matched hand and environmental pairs were similar or indistinguishable (Ab1, Ab2, Ab6, Ab7, Ab12, and Ab15) and 9/15 (60%) pairs were different (Table 1; Fig. 1). For *A. lwoffii*, 2/14 (14%) matched pairs were similar and 12/14 (86%) pairs were different. For *E. agglomerans*, 1/12 (8%) paired isolates were similar (Ea11) and 11/12 (92%) pairs were different (Table 1; Fig. 2). Of two paired *E. cloacae* isolates, one was similar and one was different. For *K. pneumoniae*, 4/9 (44%) paired isolates were similar and 5/9 (56%) pairs were different (Table 1). For *P. fluorescens*/*P. putida*, 1/3 (33%) were similar and 2/3 (67%) pairs were different (Table 1).

One pair of *S. maltophilia* isolates was different. Among GPC, 2/9 (22%) *S. warneri* matched pairs were similar (Sw4 and Sw9) (Fig. 3) and 7/9 (78%) were different. For *S. epidermidis*, 2/3 (67%) matched pairs were similar and 1/3 (33%) pairs were different. The single matched pair of *S. haemolyticus* was similar.

(ii) Relationships among strains from different households. The majority of gram-negative and gram-positive bacteria recovered from different households were unique strains that were readily distinguishable by rep-PCR. Despite their isolation from different households, some strains of *A. baumannii* (Ab10e, Ab14e and Ab2h, Ab2e, Ab3h, Ab4h, and Ab5h), *E. agglomerans* (Ea3e, Ea7h, and Ea2h), and *S. warneri* (Sw2e, Sw3e, Sw8e, Sw1e and Sw7h, Sw4h, Sw6h, Sw4e, Sw9h, and Sw9e) displayed indistinguishable patterns (Fig. 1, 2, and 3, respectively). This may reflect the presence of clonal strains within the community.

DISCUSSION

While the majority of paired isolates occurred as unique, source-specific strains, greater than 25% of *Acinetobacter* species (Fig. 1), *E. agglomerans* (8%) (Fig. 2), and *S. warneri* (22%) (Fig. 3) organisms isolated from the hands and environ-

ments of individual households shared common genotypes. Although *Acinetobacter* colonization on human skin and fingertips and prolonged survival in the environment (27 days) has been reported (2, 12, 17), this is the first study to document same-strain sharing between hands and environmental surfaces within a community setting. Over the past two decades, *Acinetobacter* species have become virulent pathogens, responsible for nosocomial infections and outbreaks, particularly in intensive care units (36). Therefore, the majority of published studies have concentrated on the hospital epidemiology of these organisms (27) and not on those within the community. Interestingly, MRSA, previously limited to causing nosocomial infections, has emerged as a virulent pathogen in the community among individuals without established risk factors for MRSA infection (24, 25). Saiman et al. recently reported that a virulent CA-MRSA strain had caused a nosocomial outbreak among postpartum women in our medical center (28), underscoring the importance of transmission of community strains in hospital settings.

Nosocomial *A. baumannii* isolates, compared to those isolated from the community, have been shown to be multidrug resistant and clonally related (12, 13, 37), suggesting that the hospital environment serves as the reservoir for epidemic *A. baumannii* strains. Although *A. baumannii* isolates in the community are more drug susceptible, they also are clonal and could be encountered as potential pathogens within the hospital environment (37).

To the best of our knowledge, no data have been reported demonstrating shared genotypes of *A. baumannii*, *E. agglomerans*, and *S. warneri* between the hands of caretakers and their household environments. The sampled environmental surface represented only one high-contact/high-traffic source area within the home; we cannot comment on whether there are other reservoirs harboring genetically similar shared species among other areas within the home.

Because transmission of community strains (e.g., recent example of CA-MRSA) into the hospital can occur, we speculate that these strains may comprise a natural reservoir of short- or long-term duration within the community that could serve as a source for nosocomial infections.

ACKNOWLEDGMENT

This study was funded in part by the National Institutes of Health, National Institute of Nursing Research, RO1 NR05251 (Home Hygiene Intervention to E.L.L.).

REFERENCES

1. **Aiello, A. E., J. Cimiotti, P. Della-Latta, and E. L. Larson.** 2003. A comparison of the bacteria found on the hands of "homemakers" and neonatal intensive care unit nurses. J. Hosp. Infect. **54:**310–315.
2. **Berlau, J., H. Aucken, H. Malnick, and T. Pitt.** 1999. Distribution of *Acinetobacter* species on skin of healthy humans. Eur. J. Clin. Microbiol. Infect. Dis. **18:**179–183.
3. **Bou, G., G. Cervero, M. A. Dominguez, C. Quereda, and J. Martinez-Beltran.** 2000. PCR-based DNA fingerprinting (REP-PCR, AP-PCR) and pulsed-field gel electrophoresis characterization of a nosocomial outbreak caused by imipenem- and meropenem-resistant *Acinetobacter baumannii*. Clin. Microbiol. Infect. **6:**635–643.
4. **Burucoa, C., V. Lhomme, and J. L. Fauchere.** 1999. Performance criteria of DNA fingerprinting methods for typing of *Helicobacter pylori* isolates: experimental results and meta-analysis. J. Clin. Microbiol. **37:**4071–4080.
5. **Chmielewski, R., A. Wieliczko, M. Kuczkowski, M. Mazurkiewicz, and M. Ugorski.** 2002. Comparison of ITS profiling, REP- and ERIC-PCR of *Salmonella enteritidis* isolates from Poland. J. Vet. Med. B **49:**163–168.
6. **Dawson, S. L., J. C. Fry, and B. N. Dancer.** 2002. A comparative evaluation of five typing techniques for determining the diversity of fluorescent pseudomonads. J. Microbiol. Methods **50:**9–22.
7. **Dombek, P. E., L. K. Johnson, S. T. Zimmerley, and M. J. Sadowsky.** 2000. Use of repetitive DNA sequences and the PCR to differentiate *Escherichia coli* isolates from human and animal sources. Appl. Environ. Microbiol. **66:**2572–2577.
8. **Dunne, W. M., Jr., K. S. Kehl, C. A. Holland-Staley, A. B. Brueggemann, M. A. Pfaller, and G. V. Doern.** 2001. Comparison of results generated by serotyping, pulsed-field restriction analysis, ribotyping, and repetitive-sequence PCR used to characterize penicillin-resistant pneumococci from the United States. J. Clin. Microbiol. **39:**1791–1795.
9. **Garcia-Garmendia, J. L., C. Ortiz-Leyba, J. Garnacho-Montero, F. J. Jimenez-Jimenez, C. Perez-Paredes, A. E. Barrero-Almodovar, and M. Gili-Miner.** 2001. Risk factors for *Acinetobacter baumannii* nosocomial bacteremia in critically ill patients: a cohort study. Clin. Infect. Dis. **33:**939–946.
10. **Gunawardana, G. A., K. M. Townsend, and A. J. Frost.** 2000. Molecular characterisation of avian *Pasteurella multocida* isolates from Australia and Vietnam by REP-PCR and PFGE. Vet. Microbiol. **72:**97–109.
11. **Husni, R. N., L. S. Goldstein, A. C. Arroliga, G. S. Hall, C. Fatica, J. K. Stoller, and S. M. Gordon.** 1999. Risk factors for an outbreak of multi-drug-resistant *Acinetobacter* nosocomial pneumonia among intubated patients. Chest **115:**1378–1382.
12. **Jawad, A., H. Seifert, A. M. Snelling, J. Heritage, and P. M. Hawkey.** 1998. Survival of *Acinetobacter baumannii* on dry surfaces: comparison of outbreak and sporadic isolates. J. Clin. Microbiol. **36:**1938–1941.
13. **Jones, R. N., L. Deshpande, T. R. Fritsche, and H. S. Sader.** 2004. Determination of epidemic clonality among multidrug-resistant strains of *Acinetobacter* spp. and *Pseudomonas aeruginosa* in the MYSTIC Programme (USA, 1999–2003). Diagn. Microbiol. Infect. Dis. **49:**211–216.
14. **Larson, E. L., C. Gomez-Duarte, L. V. Lee, P. Della-Latta, D. J. Kain, and B. H. Keswick.** 2003. Microbial flora of hands of homemakers. Am. J. Infect. Control **31:**72–79.
15. **Larson, E. L., S. X. Lin, C. Gomez-Pichardo, and P. Della-Latta.** 2004. Effect of antibacterial home cleaning and handwashing products on infectious disease symptoms: a randomized, double-blind trial. Ann. Intern. Med. **140:**321–329.
16. **Lipman, L. J., A. de Nijs, T. J. Lam, and W. Gaastra.** 1995. Identification of *Escherichia coli* strains from cows with clinical mastitis by serotyping and DNA polymorphism patterns with REP and ERIC primers. Vet. Microbiol. **43:**13–19.
17. **Liu, P. Y., and W. L. Wu.** 1997. Use of different PCR-based DNA fingerprinting techniques and pulsed-field gel electrophoresis to investigate the epidemiology of *Acinetobacter calcoaceticus-Acinetobacter baumannii* complex. Diagn. Microbiol. Infect. Dis. **29:**19–28.
18. **Liu, P. Y., Y. J. Lau, B. S. Hu, J. M. Shyr, Z. Y. Shi, W. S. Tsai, Y. H. Lin, and C. Y. Tseng.** 1995. Analysis of clonal relationships among isolates of *Shigella sonnei* by different molecular typing methods. J. Clin. Microbiol. **33:**1779–1783.
19. **Liu, P. Y., Y. J. Lau, B. S. Hu, J. M. Shyr, Z. Y. Shi, W. S. Tsai, Y. H. Lin, and C. Y. Tseng.** 1995. Comparison of different PCR approaches for characterization of *Burkholderia* (*Pseudomonas) cepacia* isolates. J. Clin. Microbiol. **33:**3304–3307.
20. **Malathum K., K. V. Singh, G. M. Weinstock, and B. E. Murray.** 1998. Repetitive sequence-based PCR versus pulsed-field gel electrophoresis for typing of *Enterococcus faecalis* at the subspecies level. J. Clin. Microbiol. **36:**211–215.
21. **Mammeri, H., G. Laurans, M. Eveillard, S. Castelain, and F. Eb.** 2001. Coexistence of SHV-4- and TEM-24-producing *Enterobacter aerogenes* strains before a large outbreak of TEM-24-producing strains in a French hospital. J. Clin. Microbiol. **39:**2184–2190.
22. **Marty, N.** 1997. Epidemiological typing of *Stenotrophomonas maltophilia*. J. Hosp. Infect. **36:**261–266.
23. **Matsumoto, M., Y. Suzuki, Y. Miyazaki, D. Tanaka, T. Yasuoka, K. Mashiko, R. Ishikita, and J. Baba.** 2001. Enterobacterial repetitive intergenic consensus sequence-based PCR (ERIC-PCR); its ability to differentiate *Streptococcus pyogenes* strains and applicability to the study of outbreaks of streptococcal infection. J. Exp. Med. **194:**205–212.
24. **Naimi, T. S., K. H. LeDell, D. J. Boxrud, A. V. Groom, C. D. Steward, S. K. Johnson, J. M. Besser, C. O'Boyle, R. N. Danila, J. E. Cheek, M. T. Osterholm, K. A. Moore, and K. E. Smith.** 2001. Epidemiology and clonality of community-acquired methicillin-resistant *Staphylococcus aureus* in Minnesota, 1996–1998. Clin. Infect. Dis. **33:**990–996.
25. **Naimi, T. S., K. H. LeDell, K. Como-Sabetti, S. M. Borchardt, D. J. Boxrud, J. Etienne, S. K. Johnson, F. Vandenesch, S. Fridkin, C. O'Boyle, R. N. Danila, and R. Lynfield.** 2003. Comparison of community- and health care-associated methicillin-resistant *Staphylococcus aureus* infection. JAMA **290:**2976–2984.
26. **Olive, D. M., and P. Bean.** 1999. Principles and applications of methods for DNA-based typing of microbial organisms. J. Clin. Microbiol. **37:**1661–1669.
27. **Presterl, E., R. Nadrchal, S. Winkler, A. Makristathis, W. Koller, M. L. Rotter, and A. M. Hirschl.** 1997. Molecular typing of *Acinetobacter bauman-*

nii from ten different intensive care units of a university hospital. Eur. J. Clin. Microbiol. Infect. Dis. **16:**740–743.
28. **Saiman, L., M. O'Keefe, P. L. Graham III, F. Wu, B. Said-Salim, B. Kreiswirth, A. LaSala, P. M. Schlievert, and P. Della-Latta.** 2003. Hospital transmission of community-acquired methicillin-resistant *Staphylococcus aureus* among postpartum women. Clin. Infect. Dis. **37:**1313–1319.
29. **Sander, A., M. Ruess, S. Bereswill, M. Schuppler, and B. Steinbrueckner.** 1998. Comparison of different DNA fingerprinting techniques for molecular typing of *Bartonella henselae* isolates. J. Clin. Microbiol. **36:**2973–2981.
30. **Spigaglia, P., and P. Mastrantonio.** 2003. Evaluation of repetitive element sequence-based PCR as a molecular typing method for *Clostridium difficile*. J. Clin. Microbiol. **41:**2454–2457.
31. **Stern, M. J., G. F. Ames, N. H. Smith, E. C. Robinson, and C. F. Higgins.** 1984. Repetitive extragenic palindromic sequences: a major component of the bacterial genome. Cell **37:**1015–1026.
32. **van der Zee, A., H. Verbakel, J. C. van Zon, I. Frenay, A. van Belkum, M. Peeters, A. Buiting, and A. Bergmans.** 1999. Molecular genotyping of *Staphylococcus aureus* strains: comparison of repetitive element sequence-based PCR with various typing methods and isolation of a novel epidemicity marker. J. Clin. Microbiol. **37:**342–349.
33. **Ventura, M., and R. Zink.** 2002. Specific identification and molecular typing analysis of *Lactobacillus johnsonii* by using PCR-based methods and pulsed-field gel electrophoresis. FEMS Microbiol. Lett. **217:**141–154.
34. **Wong, H. C., and C. H. Lin.** 2001. Evaluation of typing of *Vibrio parahaemolyticus* by three PCR methods using specific primers. J. Clin. Microbiol. **39:**4233–4240.
35. **Wu, F., and P. Della-Latta.** 2002. Molecular typing strategies. Semin. Perinatol. **26:**357–366.
36. **Zarrilli, R., M. Crispino, M. Bagattini, E. Barretta, A. Di Popolo, M. Triassi, and P. Villari.** 2004. Molecular epidemiology of sequential outbreaks of *Acinetobacter baumannii* in an intensive care unit shows the emergence of carbapenem resistance. J. Clin. Microbiol. **42:**946–953.
37. **Zeana, C., E. Larson, J. Sahni, S. J. Bayuga, F. Wu, and P. Della-Latta.** 2003. The epidemiology of multidrug-resistant *Acinetobacter baumannii*: does the community represent a reservoir? Infect. Control Hosp. Epidemiol. **24:**275–279.