to antimicrobials and the serotypes involved. The objectives of this study were to describe the
temporal development of quinolone and macrolide resistance in \textit{C. jejuni} and \textit{C. coli} isolated
from Thai children during 1991-2000 and to analyze possible associations between antimicrobial
resistance patterns, strain origin, and Lior serotypes. A total of 971 \textit{C. jejuni} and 201 \textit{C. coli}
isolates were obtained from Thai children ≤10 years-old with and without diarrhea during
1991-2000. The predominant serotypes among diarrhea cases were \textit{C. jejuni} Lior 36, 2, 11, 4,
7 and 9 representing about 40-50\% of all serotypes isolated in urban and rural areas. Similar
serotypes were isolated from controls. About 20\% of \textit{C. jejuni} strains from diarrhea cases were
untypable. For \textit{C. coli}, serotypes Lior 8, 29, 55, 20 and 44 were the most common serotypes
found in diarrhea cases. Between 30-45\% of \textit{C. coli} from cases and controls were untypable.
No association between origin of strains (urban versus rural) and particular serotypes were
detected. Resistance to nalidixic acid and/or ciprofloxacin in \textit{C. jejuni} from cases increased
dramatically from 3\% in 1992 to 80\% in 1994 and remained at this level. \textit{C. jejuni} from cases
and controls did not show difference on resistance to nalidixic acid and/or ciprofloxacin adjusted
for origin by Mantel-Haenzel chi square. Resistance to erythromycin and azithromycin in \textit{C. jejuni}
was found in 0-7\% from both cases and controls, in contrast to \textit{C. coli} of which approximately
20\% were resistant to erythromycin and azithromycin throughout. The results suggest that
ciprofloxacin will have little therapeutic effect where as erythromycin and azithromycin might
be expected to be effective in treatment of diarrhea caused by \textit{C. jejuni}. The relative frequent
occurrence of macrolide resistant \textit{C. coli} is of concern and should be monitored closely.

\textbf{American Society for Microbiology 105th General Meeting. Georgia World Congress
Center, Atlanta, GA. 5-9 June 2005. (Poster)}

\textbf{TYPHOID FEVER: A MASSIVE, SINGLE-POINT SOURCE, MULTIDRUG-
RESISTANT OUTBREAK IN NEPAL}

\textbf{Lewis MD, Serichantalergs O, Pitarangsi C, Chuanak N, Mason CJ, Regmi LR, Pandey P,
Laskar R, Shrestha CD, Malla S}

\textbf{Background:} In the summer of 2002, a total of 5963 cases of typhoid fever were recorded in
Bharatpur, Nepal (population, 92,214) during a 7-week period. A team from the Armed Forces
Research Institute of Medical Sciences in Bangkok, Thailand, and the CIWEC Travel medicine
Clinic (Kathmandu, Nepal) assisted the Nepal national Public Health Laboratory (Kathmandu,
Nepal) in the further investigation of this large, explosive febrile disease outbreak.

\textbf{Methods:} Investigators conducted a thorough epidemiologic and laboratory investigation to
assess the size and scope of the outbreak. In addition to subculturing of previously collected
samples, blood samples were obtained from 100 febrile patients, and culture and susceptibility
testing were done by standard laboratory methods. Pulsed field gel electrophoresis (PFGE) and
plasmid analysis were done.

\textbf{Results:} The majority of the isolates, including 1 from the municipal water supply, were multi-
drug resistant. The minimum inhibitory concentrations (MICs) of ciprofloxacin ranged from
0.19 µg/mL to 0.125 µg/mL. With use of PFGE, all isolates had a plasmid encoding for resistance,
and those with resistance to nalidixic acid had a single-point mutation.
Conclusion: To the best of our knowledge, this outbreak is the largest single-point sources outbreak of multidrug-resistant typhoid fever yet reported, and it was molecularly traced to the city’s single municipal water supply. Isolates were uniformly resistant to nalidixic acid, there was a decrease in their susceptibility as measured by MIC of fluoroquinolones and 90% of isolates obtained were resistant to >1 antibiotic.


DEVELOPMENT AND EVALUATION OF FLA- AND VIAB-BASE TAQMAN PCR ASSAYS FOR IDENTIFICATION OF SALMONELLA SPP. AND S. TYPHI

Phasuk R, Sethabutr O, Srijan A, Bodhidatta L, Mason CJ

Background: The Salmonella detection based on microbiological culture methods may take several days to get diagnostic result. For rapid, sensitive and specific diagnosis of Salmonella infection, we developed two sets of TaqMan PCR assay base on the nucleotide sequence encoding the flagella (fla-gene, FAM signal) and Vi antigen (viaB region, VIC signal).

Method: The fla assay, 29 bp of TaqMan probe bound to 87 bp specific amplicon, specifies for Salmonella spp. detection, including S. enterica, S. enteritidis, S. paratyphi, S. typhi, S. arizona, S. typhimurium and S. newport. The viaB assay, 38 bp of TaqMan probe bound to 101 bp specific amplicon, specifies for S. typhi. These assays were tested with 64 Salmonella isolates (27 serovars) and 29 other Enterobacteriaceae isolates (e.g. Campylobacter, E. coli, Plesiomonas, Shigella, and Vibrio).

Result: The fla assay could identify 56 of 64 Salmonella isolates by showing FAM signal in positive Salmonella culture. The rest (8/64) belong to 4 different rare serovars (Gr.I, Gr.L, Gr.M and some group of poly F). No FAM signal was shown in any of 29 non-Salmonella isolates. The viaB assay correctly identified S. typhi by showing VIC signal in positive S. typhi culture (n = 15). No VIC signal was shown in any of 49 other Salmonella spp. and 29 non-Salmonella isolates. The detection limit of fla and viaB assay was < 2 cell per test. These assays were evaluated with 148 Salmonella strains isolated from clinical samples from Nepal in 2002. The sensitivity and specificity of fla assay were 99.32% and 100.00% respectively, of viaB assay was 99.02% and 100.00% respectively.

Conclusion: The fla and viaB assay demonstrated high sensitivity and specificity in Salmonella isolates. However, these assays should be further evaluated in clinical samples before using as a rapid diagnostic tool for identification of Salmonella spp. and S. typhi.

American Society for Microbiology 105th General Meeting, Georgia World Congress Center, Atlanta, GA. 5-9 June 2005. (Poster)