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Invasive Strains of *Escherichia coli* Belonging to Serotype O121:NM

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Ten strains of *Escherichia coli* isolated from 10 travellers with sporadic diarrhea who were returning to Tokyo, Japan, from abroad were found to be of serotype O121:NM and were positive in the Serény test for invasiveness; this suggests that this serotype can cause a shigellosis-like illness in humans. The *E. coli* O121:NM strains were positive in the cell invasion test in HeLa cells. Analysis of the plasmid content of these strains showed that they contained a high-molecular-mass plasmid of 120 to 140 MDa which has been associated with invasiveness and were positive in an enzyme-linked immunosorbent assay for detection of the virulence plasmid-encoded proteins of *Shigella* spp. and enteroinvasive *E. coli*.

Some *Escherichia coli* strains are positive in the Serény test (9) for invasiveness and cause a shigellosis-like illness in humans (both children and adults) (14). Such *E. coli* strains have been described as belonging to at least the following 10 antigenic O groups: $O28_{ac}$, O29, $O112_{ac}$, O124, O136, O143, O144, O152, O164, and O167 (2, 11, 12, 14).

In this report, we describe 10 strains of *E. coli* serotype O121:NM which were positive in the Serény test. The strains were isolated in 10 sporadic cases of diarrheal disease among travellers returning to Tokyo, Japan, from abroad and are listed in Table 1.

Table 2 shows the biochemical reactions of the 10 strains. The reactions were carried out at 37°C with the media, reagents, and techniques recommended by Ewing (1).

In preliminary slide agglutination tests with *E. coli* O antisera O1 to O170, heated suspensions of strains were significantly agglutinated only by the O121 antiserum. When various serial dilutions of antiserum for *E. coli* O121 (standard strain W39) were used, the heated suspensions (100°C, 1 h) of 10 strains were agglutinated at the titer of the antiserum (1:2,560). Similarly, the heated suspension (100°C, 1 h) of *E. coli* O121 (W39) was agglutinated at the titer of the SL11 (representative strain) O antiserum (1:1,280). A cross-absorption test with heated suspensions (100°C, 1 h), showed that strains SL11 and W39 removed agglutinins from heterologous antiserum at a titer of less than 1:10. The present

TABLE 1. Sources of strains tested in this study

Yr of isolation	Country or countries visited	Age of patient (yr)	Strain designation
1984	Singapore	33	SL11
1986	Burma	31	86-256
1987	Indonesia and Malaysia	39	TEC475
1987	Taiwan	11	TEC496
1987	Thailand and Nepal	24	TEC509
1988	Thailand and Hong Kong	24	88-188
1989	India	21	89-237
1989	Thailand	22	89-410
1991	Thailand	23	91-367
1992	Bhutan and Laos	27	92-214

TABLE 2. Biochemical reactions of 10 strains of invasive *E. coli* O121:NM

Test	% Positive[a]
Oxidase	0
Methyl red	100
Voges-Proskauer	0
Christensen citrate	0
Simmons citrate	0
Sodium acetate	100
Mucate	10 (20)[b]
Malonate	0
Indole production	100
H_2S-triple sugar iron agar	0
Christensen urea	0
Phenylalanine deaminase	0
Lysine decarboxylase	0
Ornithine decarboxylase	70
Arginine dihydrolase	70
ONPG[c]	100
Motility	0
Glucose fermentation	100
Gas from glucose	100
Lactose fermentation	50 (100)[b]
Mannitol fermentation	100
Sucrose fermentation	0
Salicin fermentation	0
Dulcitol fermentation	0
Inositol fermentation	0
Adonitol fermentation	0
Raffinose fermentation	0
Sorbitol fermentation	100
Arabinose fermentation	100
Rhamnose fermentation	100
Xylose fermentation	100
Trehalose fermentation	100
Glycerol fermentation	100
Cellobiose fermentation	0
Maltose fermentation	100

[a] Test results at 1 or 2 days unless otherwise designated.
[b] Values in parentheses indicate percent positive at 14 days.
[c] ONPG, *o*-nitrophenyl-β-D-galactopyranoside.

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findings showed that the strains are antigenically identical to the standard strain (W39) of *E. coli* O group O121.

The 10 tested strains had the typical biochemical reactions of enteroinvasive *E. coli* in that they were negative for lysine decarboxylase and nonmotile (10, 13). They were positive not only in the Serény test with guinea pig eyes but also in the cell invasion test with HeLa cells (7). Analysis of the plasmid content of these strains (5) showed that they contained a high-molecular-mass plasmid of 120 to 140 MDa which has been associated with invasiveness (4) and that they were positive in an enzyme-linked immunosorbent assay (6, 8) for detection of the virulence plasmid-encoded proteins of *Shigella* spp. and enteroinvasive *E. coli* (3).

Our findings show that *E. coli* strains belonging to serotype O121:NM are invasive.

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