JOURNAL OF CLINICAL MICROBIOLOGY, Oct. 1977, p. 373-378

Vol. 6, No. 4
Printed in U.S.A.

# Unusual *Enterobacteriaceae*: "*Proteus rettgeri*" That "Change" into *Providencia stuartii*

J. J. FARMER III,[1]* F. W. HICKMAN,[1] DON J. BRENNER,[1] MINNIE SCHREIBER,[2] AND DAVID G. RICKENBACH[3]

*Enteric Section, Center for Disease Control, Atlanta, Georgia 30333[1]; General Bacteriology Section, Office of Laboratory Services, Jacksonville Central Public Health Laboratory, Jacksonville, Florida 32201[2]; and Microbiology Section, St. Vincent's Medical Center, Jacksonville, Florida 32204[3]*

Received for publication 8 July 1977

A blood culture bottle from a patient with bacteremia contained both *Proteus rettgeri* biogroup 5 and *Providencia stuartii* (described in *Bergey's Manual of Determinative Bacteriology* [8th ed., 1974] as *Proteus inconstans*), which had the same unusual antibiotic resistance pattern. Single colonies of this *P. rettgeri* biogroup 5 isolate were shown to produce urea$^-$ clones. If current taxonomy is used, the strain changed from *P. rettgeri* to *P. stuartii* in the laboratory and probably also in the patient. Urea$^-$ clones were also found in three of six other strains of *P. rettgeri* biogroup 5. No urea-negative clones were found in two isolates each of *P. rettgeri* biogroups 1 and 3. Previous data from deoxyribonucleic acid-deoxyribonucleic acid hybridization, biochemical reactions, and serological cross-reactions all have indicated that the taxon now called *P. rettgeri* biogroup 5 should be reclassified as *P. stuartii* urea$^+$. We propose that this taxonomic change be made. Urease production is probably plasmid mediated in *P. stuartii* urea$^+$ and can easily be lost, as shown in our case report and in three stock cultures. Urea hydrolysis will no longer be the key test for differentiating *P. rettgeri* from *P. stuartii*. Rather, acid production from trehalose, D-arabitol, adonitol, and D-mannitol will be the key tests. Whereas *P. rettgeri* is usually trehalose$^-$, D-arabitol$^+$, adonitol$^+$, and D-mannitol$^+$, *P. stuartii* has the opposite reactions.

Because the biochemical reactions of some strains of *Proteus rettgeri* (especially Penner's biogroup 5 strains) are very close to those of *Providencia stuartii* (3, 5), the two species are sometimes difficult to differentiate (see Tables 1 and 2). *Bergey's Manual of Determinative Bacteriology* (2) lists *Providencia stuartii* and *Providencia alcalifaciens* together as *Proteus inconstans*. We prefer the two-species classification (3) and will use *P. stuartii* and *P. alcalifaciens* throughout this paper, a practice more consistent with the nomenclature used in many clinical microbiology laboratories. Although urea$^+$ strains of *Providencia* were reported over 20 years ago (7, 8), urease has become the single test criterion most often used to differentiate *P. rettgeri* from *P. stuartii*. Urea$^+$ strains are called *P. rettgeri*, and urea$^-$ strains are called *P. stuartii* (3).

Recently, several lines of evidence have indicated that some strains currently identified as *P. rettgeri* should be reclassified as *P. stuartii*. As early as 1973 (1), Brenner showed by deoxyribonucleic acid (DNA)-DNA hybridization that several strains identified as *P. rettgeri* by the Enteric Section, Center for Disease Control (CDC), Atlanta, Ga., were actually *P. stuartii*. In 1975, Penner defined five biogroups of *P. rettgeri* and showed that biogroups 1 to 4 were typical *P. rettgeri*, but that biogroup 5 was much closer biochemically to *P. stuartii* (Table 2). All of the strains that Brenner had called *P. stuartii* based on DNA-DNA hybridization (but CDC had called *P. rettgeri* because they were urea$^+$) were biochemically tested by one of us (F.W.H.), and all fell into Penner's biogroup 5. Thus, evidence from DNA-DNA hybridization (1), biochemical reactions (5, 6), computer analysis (J. J. Farmer III, unpublished data), and serological typing (6) indicate that strains now called *P. rettgeri* biogroup 5 really belong in the species *P. stuartii*.

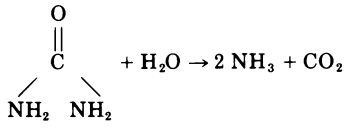
Recently, we investigated a case and some additional cultures that helped us clarify this taxonomic dilemma. Analysis of all the facts in the case led to only two possible conclusions: (i) *P. rettgeri* biogroup 5 can "change" into *P. stuartii* both in vivo and in vitro; or (ii) *P. rettgeri* biogroup 5 should be reclassified as *P. stuartii* urea$^+$.

## MATERIALS AND METHODS

**Bacterial strains.** All cultures were sent to the Enteric Section, CDC, for identification. The culture numbers are listed in Tables 1, 3, and 5. The standard nomenclature used in the Enteric Section recognized *P. stuartii* and *P. alcalifaciens,* but not *P. inconstans,* the name used in *Bergey's Manual of Determinative Bacteriology* (2).

**Media and biochemical tests.** The standard biochemical tests used in the Enteric Section have been described in detail by Edwards and Ewing (3). The standard test for urea hydrolysis was done on Christensen urea agar (3). All plating media were from commercial sources and were prepared according to the instructions of the companies.

**Urea segregation agar.** Urea segregation agar (USA) was designed and formulated to reduce the amount of ammonia produced around colonies of *Proteus* that hydrolyze urea:



$$\begin{array}{l} O \\ \| \\ C \\ / \quad \backslash \\ NH_2 \quad NH_2 \end{array} + H_2O \rightarrow 2\,NH_3 + CO_2$$

Colonies of urea$^+$ *Proteus* produce large amounts of ammonia when plated on Christensen urea agar and change the light orange (pH 7.0) medium to red for several centimeters around the colony. Thus, Christensen urea agar cannot be used to search for urea$^-$ colonies in a population of *P. rettgeri* that is predominantly urea$^+$. USA was made with two components. Component 1 contained: 4 g of yeast extract, 4 g of peptone (Difco Laboratories, Detroit, Mich.), 0.34 g of $NaH_2PO_4$, 1.03 g of $Na_2HPO_4 \cdot 7H_2O$, 15 g of agar-agar, and 900 ml of distilled water. Component 1 was autoclaved at 121°C for 15 min and then cooled to 50°C in a water bath. Component 2 contained: 29 g of Christensen urea agar base (0283-02-6, Difco), 8 g of D-glucose, and 100 ml of distilled water. It was filtered through a 0.22-nm nitrocellulose filter (Millipore Corp., Bedford, Mass.). USA was prepared by adding 100 ml of component 1 to 900 ml of component 2. The medium was poured into petri dishes (100 by 15 mm, 20 ml each; or 150 by 20 mm, 60 ml each), placed in sealed bags, and stored in a refrigerator at 4°C with no deleterious effect until used. Urea$^+$ colonies of *P. rettgeri* changed USA from light orange to red, but the size of the zone was much smaller and developed more slowly than reactions on Christensen urea agar. This probably resulted from two factors. Acid produced during glucose fermentation (8 g/liter in USA, compared to 1 g/liter in Christensen urea agar) neutralizes some of the ammonia produced during the hydrolysis of urea. USA also contains additional peptone, yeast extract, and phosphate buffer, which increases the buffering capacity of the medium. Because the red areas around urea$^+$ colonies were smaller and developed more slowly on USA, the medium was suitable for detecting the urease reaction of from 50 to 200 colonies on a single, 100-mm plate.

**Quantitation of urea$^+$ and urea$^-$ colonies in strains of *P. rettgeri.*** All of the *P. rettgeri* strains studied at CDC were urea$^+$ when submitted. This urea reaction was confirmed on the whole culture and on the single colony used to make the stock culture. The purpose of this experiment was to determine whether urea$^+$ cultures of *P. rettgeri* contained urea$^-$ clones. After the strains had been stored for 2 to 15 months (at room temperature, 25 ± 5°C), they were inoculated into Trypticase soy broth and grown for 24 h. Dilutions of each strain were made so that 0.1 ml of the last dilution contained 50 to 200 colony-forming units. After 0.1 ml of this last dilution was placed on a dry plate of USA and spread with a glass rod, the plate was incubated for 16 to 24 h. A few plates were incubated for 8 h, refrigerated overnight, and incubated for 8 h the next day. This interrupted incubation was necessary for strains that hydrolyzed urea rapidly and strongly. The plates were removed from the incubator each hour and examined for urea$^+$ and urea$^-$ colonies. All urea$^+$ colonies were then completely removed (after the colony was marked as being positive) from the surface of the agar with a Pasteur pipette attached to a vacuum line. After the urea$^+$ colony was removed, the area of urea hydrolysis did not increase in size or interfere with the urea reactions of nearby colonies. Newly developed urea$^+$ colonies were removed in this manner at 1 h intervals throughout the incubation period. At the end of the incubation period (16 to 24 h), the presumptive urea$^-$ colonies were picked and individually tested for urea hydrolysis on Christensen urea agar, and tabulations were then made of the number of urea$^+$ and urea$^-$ colonies in the population.

## RESULTS AND DISCUSSION

**Case report: a *P. rettgeri* that appeared to "change" into *P. stuartii.*** A 79-year-old white male was admitted to a Florida hospital in late August 1975 for gram-negative septicemia, hypovolemic shock, diffuse arteriosclerosis, and possible abdominal aneurysm. After hospitalization for 1 month, he had partially recovered and was discharged to a nursing home. In late September, the patient's urinary catheter apparently became clogged, and he was eventually readmitted to the hospital with fever, chills, and bloody urine. During this later 2-week hospitalization, three blood samples for culture were taken (two were positive; one was negative). The strains isolated by one of us (D.G.R.) identified as *P. rettgeri* and *P. stuartii* were from the same blood-culture bottle. The blood-culture bottle was later sent to the Jacksonville Central Public Health Laboratory where another of us (M.S.) isolated two cultures identified as *P. rettgeri* and two cultures identified as *P. stuartii.* In fact, isolates with four different biochemical patterns (Table 1) were obtained from this bottle.

These four isolates were sent to CDC and underwent the usual series (3) of biochemical tests (Table 1). The taxonomic schema currently used at CDC allowed no choice but to report

TABLE 1. *Biochemical reactions of P. rettgeri and P. stuartii as they are currently defined (3) and biochemical reactions of the four isolates from a single blood-culture bottle*

| Test | *P. stuartii* | | *P. rettgeri* | | 480-76[c] | 481-76[c] | 479-76[c] | 482-76[c] |
|---|---|---|---|---|---|---|---|---|
| | Reaction[a] | % Positive[b] | Reaction[a] | % Positive[b] | | | | |
| Indole | + | 99 | + | 100 | + | + | + | + |
| Methyl red | + | 100 | + | 93 | + | + | + | + |
| Voges-Proskauer | − | 0 | − | 0 | − | − | − | − |
| Citrate (Simmons) | + | 96 | + | 96 | + | + | + | + |
| $H_2S$-triple sugar iron agar | − | 0 | − | 0 | − | − | − | − |
| Urea | − | 0 | + | 100 | + | + | − | − |
| Phenylalanine | + | 94 | + | 98 | + | + | + | + |
| Lysine | − | 0 | − | 0 | − | − | − | − |
| Arginine | − | 0 | − | 0 | − | − | − | − |
| Ornithine | − | 0 | − | 0 | − | − | − | − |
| Motility-36°C | V | 86 | + | 94 | − | + | + | + |
| Gelatin-22°C | − | 0 | − | 0 | − | − | − | − |
| KCN | + | 99 | + | 97 | + | + | + | + |
| Malonate | − | 1 | − | 1 | − | − | − | − |
| D-Glucose, acid | + | 100 | + | 100 | + | + | + | + |
| D-Glucose, gas | − | 0 | V | 12 | − | − | − | − |
| Acid from: | | | | | | | | |
| Lactose | − | 4 | − | 9 | + | + | − | − |
| Sucrose | V | 26 | V | 13 | + | + | (+[3]) | (+[5]) |
| D-Mannitol | V | 13 | V | 89 | − | − | − | − |
| Dulcitol | − | 0 | − | 0 | − | − | − | − |
| Salicin | − | 2 | V | 30 | − | − | − | − |
| Adonitol | − | 4 | V | 81 | − | − | − | − |
| *i*-Inositol | + | 98 | + | 93 | + | + | + | + |
| D-Sorbitol | − | 4 | − | 1 | − | − | − | − |
| L-Arabinose | − | 5 | − | 0 | − | − | − | − |
| Raffinose | − | 6 | − | 10 | + | + | − | − |
| L-Rhamnose | − | 0 | V | 68 | − | − | − | − |
| Maltose | − | 3 | − | 2 | − | − | − | − |
| D-Xylose | − | 10 | V | 15 | − | − | − | − |
| Trehalose | V | 87 | V | 16 | + | + | + | + |
| Cellobiose | V | 13 | − | 4 | − | − | − | − |
| $\alpha$-$CH_3$-glucoside | − | 0 | − | 2 | − | (+[4]) | − | (+[7]) |
| *meso*-Erythritol | − | 0 | V | 78 | − | − | − | − |
| Esculin | − | 0 | V | 30 | − | − | − | − |
| Glycerol | V | 34 | V | 66 | + | + | + | + |
| Mucate | − | 0 | − | 0 | − | − | − | − |
| Tartrate (Jordon) | + | 96 | + | 96 | + | + | + | + |
| Acetate | V | 84 | V | 60 | (+[5]) | (+[5]) | (+[2]) | (+[5]) |
| Lipase (corn oil) | − | 0 | − | 0 | − | − | − | − |
| Deoxyribonuclease-25°C | − | 0 | − | 0 | − | − | − | − |
| $NO_3^- \rightarrow NO_2^-$ | + | 100 | + | 99 | + | + | + | + |
| Oxidase | − | 0 | − | 0 | − | − | − | − |
| *o*-Nitrophenyl-$\beta$-D-galactopyranoside | − | | − | | + | + | − | − |

[a] Symbols: +, 90% or more positive at 48 h; V, 10.1 to 89.9% positive at 48 h; −, 0 to 10% positive at 48 h.

[b] At 48 h.

[c] Symbols: +, positive at 24 h (48 h for indole, methyl red, and Voges-Proskauer); −, negative at 7 days (1 day for oxidase and phenylalanine, 2 days for methyl red, Voges-Proskauer, KCN, malonate, and mucate); number in parentheses, day a delayed reaction became positive.

the two urea$^+$ cultures as *P. rettgeri* biogroup 5 and the two urea$^-$ cultures as *P. stuartii* (3). Because of reports that urease production is plasmid mediated in *P. rettgeri* (A. D. Lewis and I. G. Rosen, Abstr. Annu. Meet. Am. Soc. Microbiol. 1973, G218, p. 62), we postulated that *P. rettgeri* biogroup 5 contained a urease plasmid and could "change" into *P. stuartii* when it lost the plasmid. This hypothesis was tested by the following experiment.

The lac$^+$ urea$^+$ strain of *P. rettgeri* (481-76) was streaked on MacConkey agar, and after incubating for 24 h at 36°C (all incubations were at 36°C), an isolated colony was picked; it was lac$^+$ urea$^+$. This process was repeated four times so that the final lac$^+$ urea$^+$ single colony came from the fifth in a series of five separate single-colony picks from five consecutive streak plates. To dispel any criticisms that our final results were due to contamination or a careless mix-up, two of us (J.J.F. and F.W.H.) did each step independently, comparing results only after the experiment was finished. The lac$^+$ urea$^+$ clone after five purifications had the same biochemical

reactions and antibiogram as those of the original culture (481-76). The clone was inoculated into tubes containing Trypticase soy agar, triple sugar iron agar, and Christensen urea agar; incubated for 7 days; and then streaked onto MacConkey agar. About 10% of the colonies from the urea slant were lac$^-$, and, of the seven colonies tested, four were lac$^-$ urea$^+$ and three were lac$^-$ urea$^-$. Thus, if the classification shown in Tables 1 and 2 is used, we have shown that *P. rettgeri* biogroup 5 changed into *P. stuartii* in vitro, as it probably had done in the patient. The most probable genetic explanation for the observed results is summarized in Fig. 1, which is based on the previously cited work of Lewis and Rosen. In this case, the genes for lactose fermentation and urea hydrolysis may or may not be on the same plasmid.

**Stability of urease production in other strains of *P. rettgeri*.** The previous experiment showed that a lac$^+$ urea$^+$ strain of *P. rettgeri* can become lac$^-$ urea$^+$ and lac$^-$ urea$^-$. We used USA to test other strains of *P. rettgeri* for stability of urease production. Each of these had been stored as a stock culture (from a single urea$^+$ colony) for 2 to 15 months. Urea$^-$ variants were easily detected on USA for three of the *P. rettgeri* biogroup 5 strains (Table 3). The percentage of urea$^-$ colonies varied from 28 to 45. Urease production was stable in the three other

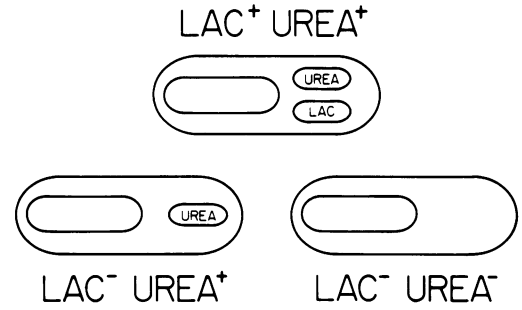



FIG. 1. *Most likely explanation for the observed variation lac$^+$ urea$^+$→urea$^-$ lac$^-$ in strain 481-76 (this strain probably also carries a resistance transfer factor). The small ovals with "urea" and "lac" inside represent plasmids; the larger, empty oval represents the bacterial chromosome.*

TABLE 2. *Biochemical reactions of P. stuartii, P. rettgeri biogroup 5, and P. rettgeri biogroups 1 to 4 as they are currently defined*

| Test | *P. stuartii* | | *P. rettgeri* biogroup 5 | | *P. rettgeri* biogroups 1 to 4 | |
|---|---|---|---|---|---|---|
| | Reaction[a] | % Positive[b] | Reaction[a] | % Positive[b] | Reaction[a] | % Positive[b] |
| Urea | − | 0 | + | 100 | + | 100 |
| Salicin | − | 2 | − | 0 | V | 52 |
| L-Rhamnose | − | 0 | − | 0 | V | 72 |
| D-Mannitol | V | 13 | − | 3 | + | 100 |
| Adonitol | − | 4 | − | 0 | + | 100 |
| D-Arabitol | − | 0 | − | 0 | + | 100 |
| *meso*-Erythritol | − | 0 | − | 0 | V | 73 |
| Trehalose | + | 100 | + | 100 | − | 0 |

[a] Symbols: +, ≥90% positive; −, ≤10% positive; V,10.1 to 89.9% positive (all at 48 h).
[b] At 48 h.

TABLE 3. *Determination of urea$^+$ and urea$^-$ colonies in strains of P. rettgeri that were originally urea$^+$*

| Culture no. | Previous identification | Identification based on recommendations in this paper | No. of colonies | |
|---|---|---|---|---|
| | | | Urea$^+$ | Urea$^-$ |
| 2450-75 | *P. rettgeri* biogroup 1 | *P. rettgeri* biogroup 1 | 85 | 0 (0)[a] |
| 2353-75 | *P. rettgeri* biogroup 1 | *P. rettgeri* biogroup 1 | 62 | 0 (0) |
| 1234-74 | *P. rettgeri* biogroup 3 | *P. rettgeri* biogroup 3 | 61 | 0 (0) |
| 1786-75 | *P. rettgeri* biogroup 3 | *P. rettgeri* biogroup 3 | 56 | 0 (0) |
| 505-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | 107 | 0 (0) |
| 9140-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | 153 | 0 (0) |
| 2346-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | 108 | 48 (31) |
| 1165-75 | *P. rettgeri* biogroup 5 (LAC$^+$) | *P. stuartii* urea$^+$ | 72 | 0 (0) |
| 697-75 | *P. rettgeri* biogroup 5 (LAC$^+$) | *P. stuartii* urea$^+$ | 115 | 44 (28) |
| 285-75 | *P. rettgeri* biogroup 5 (LAC$^+$) | *P. stuartii* urea$^+$ | 17 | 14 (45) |

[a] Number in parentheses represents percentage.

strains (505-76, 9140-76, and 1165-75) of *P. rettgeri* biogroup 5. Urease production was also stable in the two isolates each from *P. rettgeri* biogroups 1 and 3. It is tempting to postulate that urease production in *P. rettgeri* biogroups 1 to 4 is stable because the genes are located on the chromosome.

We propose moving Penner's biogroup 5 from *P. rettgeri* to *P. stuartii* and that it be designated as "*P. stuartii* urea$^+$." Independently, Penner and Hennessy have made a similar proposal (4). This proposal is based on the following considerations: (i) as early as 1973, DNA-DNA hybridization indicated that some strains of *P. rettgeri* were really *P. stuartii* (1); (ii) urease production is probably plasmid mediated and is readily lost from some urea$^+$ strains of *P. stuartii;* (iii) biochemically (Table 2) and serologically (5, 6), *P. rettgeri* biogroup 5 is much closer to *P. stuartii* than to *P. rettgeri* biogroups 1 to 4. Thus the interpretation of the case we have described is that the patient was infected with *P. stuartii* urea$^+$. When this strain multiplied, some of the progeny did not receive the urease plasmid. Thus, the original blood-culture bottle contained both urea$^+$ and urea$^-$ clones of *P. stuartii* that were derived from the urea$^+$ parent strain.

TABLE 4. *Schema for identifying P. rettgeri and P. stuartii based on the new definitions given in this paper*

| Test | *P. rettgeri* | | *P. stuartii* | |
|---|---|---|---|---|
| | Reaction[a] | % Positive[b] | Reaction[a] | % Positive[b] |
| D-Mannitol | + | 99 | − | 10 |
| D-Arabitol | + | 99 | − | 1 |
| Adonitol | + | 99 | − | 2 |
| Trehalose | − | 1 | + | 99 |
| Urea | + | 99 | V | 15[c] |

[a] Symbols: See footnote *a* to Table 2.

[b] The differences in these values compared to those in Table 2 reflect the addition of strains previously called "*P. rettgeri* biogroup 5." These values may need adjustment as more data become available. We have arbitrarily set the upper and lower values for the percentage positive at 99 and 1, rather than at 100 and 0.

[c] This value was estimated from Table 1 in the paper of Penner et al. (5). It will be corrected as additional data become available.

Urea hydrolysis will no longer be the key reaction for differentiating *P. rettgeri* and *P. stuartii* in the revised taxonomy, but acid production from D-arabitol, trehalose, adonitol, and D-mannitol will be used (Table 4). In the old classification, all strains of *P. stuartii* are urea$^-$, but with the new classification up to 15% will be urea$^+$. If urease production is plasmid mediated in all urea$^+$ strains of *P. stuartii,* it will be possible to see a urea$^+$ *P. stuartii* become urea$^-$ in the same patient or in the laboratory. We have documented this change in vitro for four different strains of *P. stuartii* urea$^+$. This mechanism also explains the report of Washington et al. (9), who showed that urine specimens from 22 different patients yielded both *P. rettgeri* biogroup 5 and *P. stuartii* (both gentamicin resistant). Urease production was stable in three strains of *P. stuartii* urea$^+$, but unstable in three others (Table 3).

This taxonomic change should cause few difficulties in clinical microbiology laboratories because correct identifications can still be made with simple biochemical tests. We will be interested to know whether our proposed taxonomic change alters the physician's approach to infections caused by *P. rettgeri* and *P. stuartii.* Because many antibiotic-resistant strains of *Proteus* are *P. rettgeri* biogroup 5, which will now be called *P. stuartii* urea$^+$, the clinical significance, ecology, and epidemiology of *P. rettgeri* and *P. stuartii* should be reexamined. Beginning 1 July 1977, the Enteric Section, CDC, will report strains as "*P. stuartii* urea$^+$" instead of "*P. rettgeri* biogroup 5." The overwhelming evidence for this change has been given in this and other papers (1, 5, 6). Table 5 gives the key

TABLE 5. *Results in key biochemical reactions for 10 clinical isolates (all those received by CDC in 1975 and 1976) that were formerly called P. rettgeri biogroup 5 but will now be called P. stuartii urea$^+$*

| Culture no. | Previous identification | New identification | Biochemical tests[a] | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Tre | Arl | Adt | Man | Ery | Rha | Sal | Urea |
| 285-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 286-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 697-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 1165-75 | *P. rettgeri* biobroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 1631-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 2346-75 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 505-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 1557-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | +$^w$ | − | − | − | − | − | − | + |
| 1586-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |
| 1806-76 | *P. rettgeri* biogroup 5 | *P. stuartii* urea$^+$ | + | − | − | − | − | − | − | + |

[a] Abbreviations: Tre, trehalose; Arl, D-arabitol; Adt, adonitol; Man, D-mannitol; Ery, erythritol; Rha, L-rhamnose; Sal, salicin (each test is defined as acid production). Symbols: +, positive at 24 h; −, negative at 7 days; +$^w$, weak positive reaction at 24 h.

biochemical reactions for all the strains received at CDC in 1975 and 1976 that would be affected by this change.

## ACKNOWLEDGMENTS

We thank John Penner for confirming some biotypes of *P. rettgeri* and for sharing with us some unpublished work on the serology and biochemical reaction of *P. rettgeri* and thank A. G. Steigerwalt and G. R. Fanning for doing many of the DNA-DNA hybridization experiments referred to in reference 1.

## LITERATURE CITED

1. **Brenner, D. J.** 1974. DNA reassociation for the clinical differentiation of enteric bacteria. Public Health Laboratory **32**:118–130.
2. **Buchanan, R. E., and N. E. Gibbons (ed.).** 1974. Bergey's manual of determinative bacteriology, 8th ed. The Williams & Wilkins Co., Baltimore.
3. **Edwards, P. R., and W. H. Ewing.** 1972. Identification of *Enterobacteriaceae*, 3rd ed. Burgess Publishing Co., Minneapolis, Minn.
4. **Penner, J. L., and J. N. Hennessy.** 1977. Reassignment of the intermediate strains of *Proteus rettgeri* biovar 5 to *Providencia stuartii* on the basis of the somatic (O) antigens. Int. J. Syst. Bacteriol. **27**:71–74.
5. **Penner, J. L., N. A. Hinton, and J. Hennessy.** 1975. Biotypes of *Proteus rettgeri*. J. Clin. Microbiol. **1**:136–142.
6. **Penner, J. L., N. A. Hinton, G. R. Whiteley, and J. N. Hennessy.** 1976. Variation in urease activity of endemic hospital strain of *Proteus rettgeri* and *Providencia stuartii*. J. Infect. Dis. **134**:370–376.
7. **Shaw, C., and P. H. Clarke.** 1975. Biochemical classification of *Proteus* and *Providencia* cultures. J. Gen. Microbiol. **13**:155–161.
8. **Singer, J., and J. Bar-Chay.** 1954. Biochemical investigation of *Providence* strains and their relationship to the *Proteus* group. J. Hyg. **52**:1–8.
9. **Washington, J. A., II, D. H. Senjem, A. Haldorson, A. H. Schutt, and W. J. Martin.** 1973. Nosocomially acquired bacteriuria due to *Proteus rettgeri* and *Providencia stuartii*. Am. J. Clin. Pathol. **60**:836–838.