## Unusual Enterobacteriaceae: a Salmonella cubana that is Urease Positive

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This is the first report of a naturally occurring *Salmonella* that is urea positive. The strain was identified as *Salmonella cubana* and it was typical in all biochemical, serological, and bacteriophage reactions, except that it produced urease strongly.

Genetic interchanges among different species of *Enterobacteriaceae* have produced some unusual recombinants in otherwise typical species. These unusual strains have been isolated from clinical specimens and some striking examples include lactose (Lac)- and/or sucrose-positive *Salmonella* (2),  $H_2S^+$  *Escherichia coli* (1) and Lac<sup>+</sup> *Proteus* (6). In this note we describe a naturally occurring strain of *Salmonella enteritidis* serotype Cubana which is urea positive on Christensen's urea agar (3) (designation: *S. cubana* urea +).

The strain was isolated from the urine of a 77-year-old male, and was submitted to the Center for Disease Control as number 3237-71. Except for urease production, the biochemical reactions were quite typical of those for *S. enteritidis*, whose composite reactions are included for comparison (3) in Table 1. Serological testing by the Tennessee Department of Public Health and Center for Disease Control indicated that the isolate was a typical serotype Cubana. In addition, the strain was lysed by the "Salmonella specific O1 phage" (4).

The population in the original tube was almost entirely urea<sup>+</sup>; however, after 55 colonies were picked, a urea-negative (urea<sup>-</sup>) clone was isolated. It also typed as serotype Cubana. After the original tube had been stored at room temperature for 30 months, the population was about 30% urea<sup>-</sup>. Single colonies which were urea<sup>+</sup> also produced urea<sup>-</sup> clones after storage, and the latter typed as serotype Cubana. If a urea<sup>-</sup> clone had been isolated first, there would have been no question about the identification of this strain as a typical *Salmonella* (Table 1).

The definition of the tribe Salmonelleae includes the clause "urease is not produced"; however, a strain which is urea<sup>+</sup> should not be excluded from the tribe only on the basis of this

Test	Reaction given by:		
	S. cubana urea+	Salmonella enteritidis	
		-	0%
Nitrate to nitrite	+	+	100
Indole	-	-	1.2
Methyl red	+	+	100
Voges-Proskauer	-		0
Simmons' Citrate	+	v	89
H₂S on TSI	+	+	94
KCN	_	-	0.3
Motility	+	+	94
Gelatin	-	-	<1
Lysine	+	+	95
Arginine	+	v	65
Ornithine	+	+	97
Phenylalanine	_	-	0
Mucate	Weak	V	82
Malonate	_	_	0.6
Glucose acid	+	+	100
Glucose gas	+	+	96
Lactose	-	-	0.9
Sucrose	-	-	0.6
Mannitol	+	+	100
Dulcitol	+	+	96
Salicin	-	-	<1
Adonitol	-	_	0
Inositol	+	V	38
Sorbitol	+	+	94
Arabinose	+	+	99
Raffinose	-	-	3
Rhamnose	+	+	94
Urea (Christensen's)	+	-	0

TABLE 1. Biochemical reactions of the unusual strain compared to percentage reactions of Salmonella enteritidis<sup>a</sup>

 $^{a}$  +, Positive reaction within 48 h. -, Negative reaction at 48 h. (V) Variable.

<sup>b</sup> Percent positive reactions in 1 to 2 days for S. enteritidis as given by Edwards and Ewing (3). property if it is otherwise typical. Lewis and Rosen (Abstr. Annu. Meet. Amer. Soc. Microbiol. 1973, G218, p. 62) showed that Proteus rettgeri can transfer to Salmonella typhi and S. typhimurium a plasmid which codes for urease production. The origin of the strong urease in our isolate is not known. Its genetic material which codes for urease production may have originated in a Proteus and been transferred by conjugation or transduction. This would be similar to lactose-positive Salmonella which probably acquire lactose genes from E. coli in nature (2). Or, this may be a mutant which has lost the ability to regulate the amount of urease it produces, since most Salmonella we have tested hydrolyze urea, but extremely slowly (30 to 90 days).

The identification of S. cubana urea<sup>+</sup> would pose a problem for clinical laboratories which rely on the combination of triple sugar iron agar and urea agar to rule out *Proteus* because the strain is a strong urease producer (the urea slant becomes red in 1 to 3 h and the entire tube is red at 18 h). However, a more complete set of biochemical tests (phenylalanine, indole, KCN, and lysine decarboxylase or lysine iron agar would be particularly useful) would clearly rule out *Proteus* and make *Salmonella* suspect. Two other simple tests can be particularly helpful. Proteus species are very strong catalase producers but *Salmonella* are much weaker and slower (5). Place a drop of commercial 3% hydrogen peroxide on a suspect colony (usually Lac<sup>-</sup>); instantaneous and vigorous bubbling would rule out Salmonella in almost all instances. Antibiograms are also useful in differentiating Proteus and Salmonella; Proteus is uniformly resistant to colistin but Salmonella is uniformly sensitive. Urea<sup>+</sup> Salmonella must be extremely rare in clinical specimens; however, we wonder if they have been overlooked in the past. Perhaps this report will stimulate others to isolate urea<sup>+</sup> Salmonella so that their incidence can be more accurately established.

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