

Metabolic Variations of *Proteus* in the Memphis Area and Other Geographical Areas

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The number of strains of *Proteus* studied was 413, and these were obtained from all clinical materials with the exception of fecal specimens. Lactose was fermented by 37 strains (*P. inconstans*, 29%; *P. rettgeri*, 16%; *P. mirabilis*, 4.2%; *P. morganii*, 3.6%; and *P. vulgaris*, 0%) of which 33 were from the genitourinary system. These 33 strains constituted 12.7% of the 260 strains isolated from this source. Biochemically, *P. mirabilis* was the least variable, and *P. rettgeri* was the most variable of the five species of *Proteus* tested. *P. inconstans* and *P. rettgeri* resembled each other more closely than any of the other species of *Proteus*. Comparison of results obtained in the Memphis area with those found in other locations showed that biochemical characteristics varied most with the substances citrate, salicin, xylose, trehalose, and mannitol. In contrast to earlier reports from Israel and England, none of the strains of *P. inconstans* in the present study was able to attack urea. All five species of *Proteus* tested (by the disc method) were highly susceptible to methenamine mandelate. *P. mirabilis*, *P. morganii*, and *P. vulgaris* were also highly susceptible to nitrofurantoin. All strains of *P. mirabilis* were susceptible to ampicillin. *P. inconstans* was the most resistant species of *Proteus*. Of the other 356 urease-positive strains tested, 79% were susceptible to chloramphenicol, whereas only 3.8% of the 56 urease-negative strains (*P. inconstans*) were susceptible. When tested with streptomycin, 61% of urease-positive strains were susceptible and 1.8% of the urease-negative strains were susceptible. Of 36 lactose-positive strains, 33.8% were susceptible to chloramphenicol, whereas 72.8% of all lactose-negative strains were susceptible. Again, of the lactose-positive strains, 17% were susceptible to streptomycin, whereas 56.3% of all lactose-negative strains were susceptible.

The study of variations in biochemical characteristics within the genus *Proteus* has resulted in the recognition of five species of *Proteus*. In addition, it is been recognized that there is considerable variation within each species of *Proteus*. Lactose-fermenting variants of *Proteus* were practically unrecognized until 1962, when Sutter and Foecking (18) in California found over a 2-year period 286 lactose-fermenting strains of *Proteus*; 258 of these were *P. rettgeri* and 11 of them were *P. inconstans*. Until that time, there had been, to our knowledge, only seven lactose-fermenting strains of *Proteus* reported in the literature. These were as follows. Welch and Poole (20) in 1934 reported one lactose-fermenting variant of *Proteus* X19. Rustigian and Stuart (13) in 1945 observed one *P. vulgaris* and one *P. mirabilis* isolate that fermented lactose. Singer and Bar-Chay (15) in 1954 observed one lactose-fermenting *P. rettgeri* strain. Three lactose-fermenting strains of *P.*

inconstans have been seen, two by Shaw and Clarke (14) in 1955, and one by Proom (11) in 1955.

Because of the high incidence of lactose-fermenting strains of *Proteus* reported by Sutter and Foecking, it was decided to do a study on the *Proteus* genus with the following objects in mind: (i) to determine the incidence of lactose-fermenting *Proteus* strains in the Memphis area; (ii) to determine the range of biochemical variations in *Proteus* and compare these findings with those of other investigators; and (iii) to determine whether susceptibility to antibiotics is correlated to any of the above variations.

The number of strains of *Proteus* studied was 413. They were from all clinical sources except fecal specimens. The study was conducted over two different periods of time. During the first period, from 9 January 1963 to 14 March 1963, 133 cultures were studied of which 6.8% were lactose-fermenting strains. The second period,

TABLE 1. Biochemical reactions of 413 cultures of *Proteus*^a

Test	<i>P. vulgaris</i> (18)			<i>P. mirabilis</i> (262)			<i>P. morgani</i> (28)			<i>P. rettgeri</i> (48)			<i>P. inconstans</i> (57)		
	+	(+)	-	+	(+)	-	+	(+)	-	+	(+)	-	+	(+)	-
Glucose.....	18			262			28			48			57		
Gas from glucose....	11	4	3	205	43	14	25	1	2	10	1	37			57
Xylose.....	18			252	10				28	8		40	5		52
Arabinose.....	2		16			262			28			48			57
Rhamnose.....			18			262			28	25	1	22			57
Lactose.....			18			251	1		27	8		40	17		40
Sucrose.....	18			52	210			3	25	12	36		25	32	
Maltose.....	16	2			2	260			28			48		2	55
Trehalose.....		14	4	250	11	1	3	1	24	9		39	41		16
Mannitol.....			18			262			28	41	5	2	2	1	54
Adonitol.....			18			262			28	39		9			57
Dulcitol.....			18			262			28			48			57
Inositol.....			18			262			28	21	26	1	35	22	
Sorbitol.....			18			262			28	7	41	2	53	2	
Glycerol.....	6	12		197	65		2	24	2	5	42	1	11	38	8
Salicin.....	4	8	6	1	6	255			28	18		30			57
Malonate.....			18			262			28			48			57
Gelatin.....	9	9		68	190	4			28			48		1	56
Citrate, Simmons....		2	16	198	64				28	45	3		57		
Urease.....	18			262			28			48					57
Hydrogen sulfide, TSI.....	16		2	251	1	9		26	VW 2			48			57
Indole.....	11	1	6			262	28			47	1		57		
Tryptophan deami- nase.....	18			257	5*		11	17*		39	9*		43	14*	
Phenylalanine de- aminase.....	18			262			28			48			57		
Lysine deaminase....	10	6	2	262				6	22	48			57		
Arginine decarboxyl- ase.....			18			262			28			48			57
Lysine decarboxylase.			18			262			28			48			57
Ornithine decarboxyl- ase.....			18	258		4	27		1			48	1		56

^a Symbols: +, positive reaction promptly produced; (+), positive reaction delayed (after 2 days); -, negative reaction (14 days); VW, very weak reaction; *, orange color.

which extended from 3 November 1963 to 16 March 1964, resulted in 280 isolations of *Proteus*, and during this time 10% of the cultures were lactose-fermenting.

All cultures were isolated from patients at Veterans Administration Hospital. The majority of the patients came directly from their homes in Tennessee, Mississippi, and Arkansas, located for the most part not more than 70 miles from Memphis.

MATERIALS AND METHODS

All organisms that appeared to be members of the *Enterobacteriaceae* were first characterized with the following tests: urease, phenylalanine and tryptophan deaminase, malonate, lactose, dextrose, sucrose, maltose, mannitol, indole, and citrate. Those which appeared to be *Proteus* were then given the full battery

of 28 tests as listed in Table 1. After completion of these 28 tests and after a consideration of all of the results, the 413 strains of organism were given a species designation. The standard used for this evaluation was that published by Ewing, Suassuna, and Suassuna (6). These investigators know the history of the genus *Proteus*, they understand the rules of nomenclature, and they have studied many strains of *Proteus* from many parts of the world. Their publication shows the results of 37 tests on 255 cultures. Following their standard, we have not found it difficult to designate species, as there are certain key biochemical characteristics that are practically without exception. These key characteristics, as listed in the publication by Ewing, Suassuna, and Suassuna, and in Table 1 of this paper, are as follows: urease production, gas from glucose, sucrose fermentation, mannitol fermentation, inositol fermentation, adonitol fermentation, indole production, citrate utilization, H₂S production in Triple Sugar Iron Agar (TSI), gelatin liquefaction,

and ornithine decarboxylation. When the rare exceptions do occur, there are still a sufficient number of other constant characteristics to identify these strains properly. In identifying *P. inconstans*, the guide used was the publication by Ewing, Tanner, and Dennard (7). In their study, 611 cultures were used.

Incubation of cultures was performed at 37 C unless otherwise stated. Fermentation studies were done by adding 1% fermentable substances to Nutrient Broth containing bromocresol purple as an indicator; tubes were observed for at least 2 weeks. For gelatin liquefaction, Difco Nutrient Gelatin was used, and incubation was at room temperature for 30 days. Difco Simmons Citrate Agar was used to determine utilization of citrate. For urease production, Difco Urea Agar Base was employed. The Ehrlich-Boehme technique was employed in testing for indole. For phenylalanine deaminase production, the agar slant method of Ewing, Davis, and Reavis (5) was used. For malonate utilization, Ewing's modification of Leifson's method was used (4). For the tryptophan deaminase test, the method of Singer and Volcani (16) was used, and for the lysine deaminase test the method of Edwards and Fife (2) was employed. For the carboxylation studies, the Falkow method (8) was used. Oil seals were used with this medium.

In performing the tests for susceptibility of the organisms to antibacterial agents by the two-disc method, seeded pour plates were used; these were

incubated overnight and read the following day. An organism was considered susceptible to the agent if there was a clear zone around both discs of the two potencies stated below. An organism was considered moderately susceptible if there was growth around the lower concentration disc but a clear zone around the higher concentration disc. An organism was considered resistant if there was no clear zone around either disc. For susceptibility testing, concentrations of 5 and 30 µg of the following agents were used: tetracycline, chloramphenicol, demethylchlortetracycline, novobiocin, kanamycin, polymyxin B, oxytetracycline, and nalidixic acid. For streptomycin, colistin, and ampicillin, concentrations of 2 and 10 µg were used, for nitrofurantoin a 100 µg-disc was used, and for methenamine-mandelate a 3 µg-disc was used.

A group of 48 additional cultures were tested for kanamycin susceptibility by the tube dilution method and by the disc method. These 48 cultures are not included in the original group of 413.

RESULTS AND DISCUSSION

The percentages of different species of *Proteus* found were as follows: *P. mirabilis*, 62.9%; *P. inconstans*, 13.8%; *P. rettgeri*, 11.6%; *P. morgani*, 6.7%; and *P. vulgaris*, 4.3%.

The biochemical reactions of the 413 cultures of *Proteus* are shown in Table 1. With *P. mirabilis*, variability was very slight. *P. rettgeri*, however, showed a high degree of variability. This species was slow in the fermentation of mannitol in five instances and negative in two instances. This finding is of considerable interest because of the rather generally held belief that the fermentation of mannitol by *P. rettgeri* sets it apart from the other species of *Proteus*. It is also of interest to note the similarity of *P. rettgeri* and *P. inconstans*, particularly when mannitol fermentation and urease production are not taken into consideration.

The data in Table 2 show that *P. inconstans* was the leading species in the fermentation of lactose, and it was followed by *P. rettgeri*. That these two species of *Proteus* showed this relationship is of interest in view of the fact that these two species also resemble each other more than

TABLE 2. Fermentation of lactose by different species of *Proteus*^a

Species	No. of cultures		Lactose-positive		Percentage lactose-positive	
	Study 1	Study 2	Study 1	Study 2	Study 1	Study 2
<i>P. vulgaris</i>	81	18	7	0	9.0	0.0
<i>P. mirabilis</i>	931	262	4	11	0.4	4.2
<i>P. morgani</i>	66	28	6	1	9.0	3.6
<i>P. rettgeri</i>	619	48	258	8	40.0	16.0
<i>P. inconstans</i>	116	57	11	17	9.0	29.0
Total	1813	413	286	37	16.0	8.9

^a Study 1: Sutter and Foecking, California, 1962 (18). Study 2, Suter et al., Memphis, 1965 (present study).

TABLE 3. Clinical source of lactose-fermenting *Proteus*^a

Source	No. of cultures		Lactose-positive		Percentage lactose-positive	
	Study 1	Study 2	Study 1	Study 2	Study 1	Study 2
Genitourinary system	1,357	260	263	33	19.4	12.7
Respiratory system	61	76	1	2	1.6	2.6
Wounds	344	53	13	1	3.8	1.9
Other	51 (blood)	24	9	1	17.6	4.2

^a Study 1, Sutter and Foecking, California, 1962 (18). Study 2, Suter et al., Memphis, 1965 (present study).

TABLE 4. Number of cultures giving positive reactions in various tests by different investigators

Determination	Investigator ^a							
	1	2	3	4	5	6	7	8
<i>P. vulgaris</i>								
No. of cultures	18 (100%)	39 (87.2%)		69		14	7	
Xylose	18 (77.8%)	34 (94.9%)						
Trehalose	14 (66.7%)	37 (97.4%)						
Indol	12 (57.1%)	38 (97.4%)						
Citrate	2 (9.1%)	10 (25.6%)						
Salicin	12 (57.1%)	29 (74.4%)						
Gelatin	18 (100%)							
<i>P. mirabilis</i>								
No. of cultures	262 (94.7%)	116 (98.3%)		205 (100%)		86 (45.3%)	23 (26.1%)	
Gas from glucose	248 (100%)	114 (98.3%)		196 (95.6%)		39 (98.8%)	6 (82.6%)	
Citrate	262 (100%)					85 (98.8%)	19 (26.1%)	
Sucrose	7 (2.7%)	72 (62.1%)		19 (9.3%)		13 (15.1%)	4 (17.4%)	
<i>P.morganii</i>								
No. of cultures	28			102			24 (20.8%)	
Xylose	0						5 (20.8%)	
<i>P. rettgeri</i>								
No. of cultures	48 (25.0%)	58 (5.2%)	76	78	30	11	9	
Sucrose prompt	48 (100%)	35 (60.3%)						
Sucrose	39 (81.3%)	54 (93.1%)	76 (100%)	75 (96.2%)	29 (96.7%)		9	
Adonitol	26 (54.2%)	43 (74.1%)						
Rhamnose	11 (22.9%)	8 (13.8%)						
Gas from glucose	8 (16.7%)		66 (86.8%)	7 (9.0%)	6 (20.0%)		0	
Xylose	18 (37.5%)			26 (33.3%)		8		
Salicin	0							
H ₂ S	0							9 ^b
Arabinose	0							3
Trehalose	9 (18.8%)							0

Sorbitol	7 (14.6%)				0	611 ^d
<i>P. inconstans</i>						
No. of cultures	57			86	153	
H ₂ S	0				153 (100%)	
Urea	0			12 (14.0%)	11 (7.2%)	
Citrate	57 (100%)				96 (62.7%)	
Sucrose	57 (100%)				146 (95.4%)	530 (86.7%)
Maltose	2 (3.5%)				9 (5.9%)	
Trehalose	41 (71.9%)			84 (97.7%)	32 (20.9%)	
Glycerol	49 (86.0%)			86 (100%)	80 (52.3%)	
Mannitol	3 (5.3%)			59 (68.6%)	17 (11.1%)	
Sorbitol	55 (96.5%)			2 (2.3%)	12 (7.8%)	24 (3.9%)
Inositol	57 (100%)				32 (20.9%)	87 (14.2%)
Adonitol	0				110 (71.9%)	499 (81.7%)
Salicin	0				9 (5.9%)	4 (0.7%)
Gas from glucose	0					486 (79.5%)

^a (1) Suter et al., 1965 (present study); (2) Ewing, Suassuna, and Suassuna, 1960 (6); (3) Sutter and Foecking, 1962 (18); (4) Rustigian and Stuart, 1945 (13); (5) Singer and Bar-Chay, 1954 (15); (6) Cook, 1948 (1); (7) Shaw and Clarke, 1955 (14); (8) Ewing, Tanner, and Dennard, 1954 (7).

^b Paper strip test.

^c Weak reaction.

^d Providence group.

they do any of the other species of *Proteus*. Lactose was fermented by 37 cultures of *Proteus*, and this amounted to 8.9% of all of the cultures. Sutter and Foecking in California found, in their study reported in 1962 (18), that 16% of all of their cultures of *Proteus* were able to ferment lactose. The study by Sutter and Foecking showed fermentation of lactose by 40% of their cultures of *P. rettgeri* as compared to 16% of the cultures of *P. rettgeri* in the present study. Whereas, in the study by Sutter and Foecking, lactose was fermented in only 9% of the cultures of *P. inconstans*, it was found in the present study that 29% of the cultures of this species were able to ferment lactose.

As shown in Table 3, most of the lactose-fermenting strains of *Proteus* were isolated from the genitourinary system. In the present study, 12.7% of the strains that were isolated from this source were able to ferment lactose. In the study by Sutter and Foecking, 19.4% of the strains from this source were lactose-fermenting. In the present study, 89.2% of all of the lactose-fermenting *Proteus* were from the genitourinary system, and in the study by Sutter and Foecking 91.9% of the lactose-fermenting strains were isolated from the genitourinary system. Sutter and Foecking, in discussing their results stated, "The high incidence reported in this paper raises the question of whether these strains are more prevalent than heretofore reported. Our findings may very well be the result of an intensive screening and identification of lactose-fermenting enteric bacteria. They may also be an indication of the existence of a resident or endemic strain within the rather limited environment of this hospital, since most of them were from urine specimens of patients suspected of urinary tract infection, subsequent to instrumentation of the urinary tract."

Table 4 shows the results of certain selected tests on the different species of *Proteus*. These show selected results of different investigators located in different geographical areas. It must be pointed out, however, that some of the investigators did not limit themselves to cultures that had been isolated in their own respective countries. It is believed, however, from a study of the reports, that most of the cultures of Sutter and Foecking were from southern California that the majority of cultures of Rustigian and Stuart were from Providence, R.I., or neighboring locations, that most of the cultures of Singer and Bar-Chay were from Israel, and that most of the cultures of Cook were from near Oxford, England. It must be pointed out also that these studies were performed at different times, ranging over a 20-year period from 1945 to 1965.

As can be seen from Table 4, different investigators working with *Proteus* have obtained quite dissimilar results. The results shown here were selected to show differences between our findings and those of other investigators. Results that are in practical agreement have been omitted from Table 4. The ability of *P. mirabilis* to ferment salicin has shown divergent results, ranging from 2.7 to 62.1% in five different studies. The study of *P. morganii* by different investigators reveals that there is less variation in results with this species than with the other four species of *Proteus*. It is of interest, however, to note that in the present study none of the cultures of *P. morganii* was able to ferment xylose, whereas Shaw and Clarke found that 20.8% of their cultures of this species were able to ferment xylose. There is a wide range of results on the fermentation of sucrose and xylose by *P. rettgeri*. In the present study, xylose was fermented by only 16.7% of the cultures of *P. rettgeri* as compared to the 86.8% reported by Sutter and Foecking.

Table 4 also shows divergent data concerning the ability of *P. inconstans* to hydrolyze urea. Although in the present study none of these organisms hydrolyzed urea, a relatively high percentage of those studied by Singer and Bar-Chay and by Shaw and Clarke were able to produce urease.

One interesting observation from Table 4 is that none of the cultures of *P. inconstans* in the present study was observed to produce gas from glucose, whereas Ewing, Tanner, and Dennard reported that 79.5% of their *Providencia* cultures produced gas from glucose. The latter investigators grouped their 611 *Providencia* cultures into Biotype 1 (528) and Biotype 2 (83). Most of the cultures of Biotype 1 produced gas from glucose and quite often produced gas in the fermentation of other sugars, whereas the cultures of Biotype 2 were anaerogenic. It may be noted that in the present study all of the cultures were of the anaerogenic Biotype 2.

There can be no doubt that some of the differences in results by different investigators are the result of variations in methods, and these discrepancies can be overcome only by a standardization of procedures. This, of course, can never be completely accomplished, and it may not be desired, although it is reasonable to believe that for any particular test there must be a "best" and most reliable method. It is our opinion, however, that the majority of the procedures used in this type of study are fairly well stabilized, and this should be true for procedures used in carbohydrate fermentation studies. Why most *P. rettgeri* in California ferment xylose and why most of them in the Memphis area are unable

TABLE 5. Susceptibility of 412 strains of *Proteus* to antibacterial agents (disc method)

Drug	<i>P. mirabilis</i> (262)			<i>P. morgani</i> (28)			<i>P. vulgaris</i> (18)			<i>P. rettgeri</i> (48)			<i>P. inconspans</i> (56)		
	S	MS	R	S	MS	R	S	MS	R	S	MS	R	S	MS	R
Tetracycline	1 0.4%	217 82.8%	44 16.8%	12 42.9%	6 21.4%	10 35.7%	9 50.0%	8 44.4%	1 5.6%	2 4.2%	8 16.7%	38 79.2%	0 0%	6 10.8%	50 89.3%
Chloramphenicol	248 94.7%	12 4.6%	2 0.8%	19 67.9%	5 17.9%	4 14.3%	12 66.7%	6 33.3%	0 0%	4 8.3%	12 25.0%	32 66.7%	2 3.6%	15 26.8%	39 69.7%
Streptomycin	183 69.8%	38 15.0%	41 15.6%	16 57.1%	5 17.9%	7 25.0%	12 66.7%	1 5.6%	5 27.8%	6 13.0%	7 14.6%	35 73.0%	1 1.8%	5 8.9%	50 89.2%
Novobiocin	255 97.3%	7 2.7%	0 0%	18 64.3%	7 25.0%	3 10.8%	15 83.3%	1 5.6%	2 11.1%	3 6.2%	20 41.7%	25 52.1%	11 19.6%	31 55.4%	14 25.0%
Kanamycin	261 99.6%	1 0.4%	0 0%	28 100%	0 0%	0 0%	18 100%	0 0%	0 0%	48 100%	0 0%	0 0%	56 100%	0 0%	0 0%
Polymyxin B	6 2.3%	17 6.5%	239 91.2%	2 7.1%	2 7.1%	24 85.7%	0 0%	1 5.6%	17 99.4%	3 6.2%	7 14.6%	38 79.2%	1 1.8%	2 3.6%	53 94.7%
Oxytetracycline	2 0.8%	1 0.4%	259 98.9%	12 42.9%	4 14.3%	12 42.9%	8 44.4%	5 27.8%	5 27.8%	2 4.2%	0 0%	46 95.8%	0 0%	0 0%	56 100%
Colistin	5 1.9%	6 2.3%	251 95.8%	1 3.6%	1 3.6%	26 92.9%	0 0%	2 11.1%	16 88.9%	3 6.3%	0 0%	45 93.8%	0 0%	1 1.8%	55 98.2%
Nitrofurantoin	257 98.1%	0 0%	5 1.9%	28 100%	0 0%	0 0%	17 94.4%	0 0%	1 5.6%	22 45.8%	0 0%	26 54.2%	17 30.4%	0 0%	39 69.7%
Methenamine mandelate	262 100%	0 0%	0 0%	28 100%	0 0%	0 0%	18 100%	0 0%	0 0%	48 100%	0 0%	0 0%	55 98.2%	0 0%	1 1.8%
Demethylchlor-tetracycline	4 1.5%	187 71.4%	71 27.1%	15 53.6%	4 14.3%	9 32.1%	11 61.1%	7 38.9%	0 0%	2 4.2%	9 18.8%	37 77.1%	1 1.8%	5 8.9%	50 89.3%
Ampicillin	62 100%	0 0%	0 0%	0 0%	1 14.3%	4 32.1%	0 0%	0 0%	4 38.9%	1 4.2%	3 18.8%	7 77.1%	2 1.8%	6 8.9%	7 89.3%
Nalidixic acid	56 90.3%	6 9.7%	0 0%	5 100%	0 0%	0 0%	5 100%	0 0%	0 0%	11 100%	0 0%	0 0%	15 100%	0 0%	0 0%

^a S, susceptible; MS, moderately susceptible; R, resistant.

TABLE 6. Susceptibility of *Proteus* to kanamycin

Species	No. of strains	Disc method		Tube dilution method, minimal inhibitory concn ($\mu\text{g/ml}$)										
		Susceptible to 5 μg	Not susceptible to 5 μg	0.39	0.78	1.56	3.12	6.25	12.5	25	50	100	>100	
<i>P. vulgaris</i>	10	9	1	7		1					1			1
<i>P. rettgeri</i>	10	10	0				3	3	2	2				
<i>P. mirabilis</i>	10	10	0							5	5			
<i>P. morgani</i>	10	10	0				2	2	6					
<i>P. inconstans</i>	8	5	3					1	2	2				3

to do so poses an interesting question to which we do not have the answer, assuming that methods are accurate.

From Table 1, it may be seen that some of the tests are of no value in differentiating the species of *Proteus*, since they are either positive for all of the species or negative for all of the species. These tests are: dulcitol and malonate fermentation, tryptophan and phenylalanine deamination, and arginine and lysine decarboxylation.

Table 5 shows the results of the susceptibility tests with the disc method.

It is of interest to note that all except 1 of the 412 strains of *Proteus* were susceptible to kanamycin, and the 1 nonsusceptible strain was moderately susceptible. This finding is in practical agreement with that of Ehrenkranz (3), who also used the disc method. Because some workers have found that there is a fairly significant incidence of kanamycin resistance among *Proteus* strains, it was decided to do some tube dilution tests. Therefore, 48 new cultures were done, both by the tube dilution method and by the disc method. The results indicated by the tube dilution method, which is considered by most workers to be more accurate, that indeed there was considerable resistance to kanamycin. These results are shown in Table 6. Studies by Turck and associates (19) indicated that, except for *Proteus* species, there was good agreement between results on antibacterial susceptibility obtained by measurements of the inhibitory zones around the disc and by tube dilution tests.

It is of interest, also, to note that all strains of *P. mirabilis* were susceptible to ampicillin, whereas all other species of *Proteus* did not show this striking susceptibility pattern. It may be seen, also, that a high percentage of strains of *P. mirabilis*, *P. morgani*, and *P. vulgaris* were susceptible to nitrofurantoin. Ehrenkranz (3) found a high degree of susceptibility of *Proteus* to this agent. Practically all strains of all species were also highly susceptible to methenamine mandelate. It should be pointed out that the

similarities of *P. rettgeri* and *P. inconstans* are extended into their susceptibility patterns.

P. inconstans is the most resistant species of *Proteus*, and that this is a quite striking difference from the other species can be seen from a study of Table 5. In 1959, the observation was made by two of the present authors (17) that paracolon 29911 (*P. inconstans*) was the most resistant organism found at that time in a study that included 12 different gram-negative species and 2 gram-positive species.

In the present study, when one compares the 56 urease-negative strains (*P. inconstans*) with all of the other 356 urease-positive strains (all other species), there is a very striking difference in the susceptibility to chloramphenicol and streptomycin. With chloramphenicol, 79% of urease-positive strains were susceptible as compared to 3.8% of the urease-negative organisms. With streptomycin, 61% of urease-positive strains were susceptible as compared to 1.8% of urease-negative strains. Potee and associates (10) reported that strains of *P. mirabilis* appeared to be somewhat more susceptible to streptomycin and chloramphenicol than those of the other three species, which included a greater proportion of strains that were highly resistant to these two antibiotics. *P. inconstans* was not included in their study. They used the twofold agar-plate dilution method.

In the present study, when one compares the 36 lactose-positive strains of *Proteus* with the 376 lactose-negative strains it is seen that again there is a difference in the susceptibility pattern with the two agents chloramphenicol and streptomycin. With chloramphenicol, 72.8% of the lactose-negative strains were susceptible as compared with 33.8% of the lactose-positive strains. With streptomycin, 56.3% of the lactose-negative strains were susceptible as compared to 17% of the lactose-positive ones. With the other antibacterial agents, there were no significant differences in the susceptibility patterns when comparing lactose-negative and lactose-positive strains.

The findings in this study in regard to the susceptibility of the lactose-negative urea-positive strains of *Proteus* to chloramphenicol and streptomycin agree favorably with those of Jeljaszewicz and Hawiger (9), Ehrenkranz (3), and Rantz and Rantz (12). None of these investigators included in their studies any strains that were lactose-positive, any that were urease-negative, or any that were both lactose-positive and urease-negative.

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