DIFFERENTIATION OF ENTEROBACTERIACEAE BY BIOCHEMICAL REACTIONS

REVISED

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ABSTRACT. *Differentiation of ENTEROBACTERIACEAE by Biochemical Reactions,* Revised 1973. W. H. Ewing, CDC Publication, Center for Disease Control, Atlanta, Ga. 30333. Tabular data on the biochemical reactions of ENTEROBACTERIACEAE are presented, which, if used properly, should enable investigators to identify 98 percent or more of the members of this family seen in daily practice. The data, including percentages, are given in 52 tables. The majority of these contain the results of tests that are of particular usefulness for differentiation of members of the various genera and species within the family. An emended outline of the classification and nomenclature used is included.

Introduction

The author and co-workers have studied the biochemical reactions given by relatively large numbers of cultures of each of the genera and species of ENTEROBACTERIACEAE in an attempt to produce tabular data that might be of value to microbiologists in laboratories of all kinds and, particularly to those engaged in work with specimens of human and veterinary interest. Among other things, these investigations led to revisions of the classification of ENTEROBACTERIACEAE and of the definitions for the family, its tribes, and genera (*13,15,18,23,43*) and to recognition of additional tests and methods of value for identification of members of the species within the family. A number of bacterial types, which originally were thought to be atypical ENTEROBACTERIACEAE, later were found to be quite typical members of new tribes, genera, or species. Among these are *Edwardsiella, Enterobacter hafniae, Arizona, Citrobacter diversus,* and *Providencia.*

The procedures employed were the recommended or standard methods given in the 1958 Report of the International Subcommittee on Enterobacteriaceae, as revised and extended in subsequent publications (*26,68*). Unless otherwise indicated, the temperature of incubation was 35 to 37 C.

The numerical data given in this and other publications on biochemical reactions by the author and colleagues are based upon results obtained from examinations of comparatively large numbers of cultures (Table 1) from all States and Territories of the United States, and from many other parts of the world, over a period of about 30 years. Since most of these strains were submitted directly or indirectly from clinical laboratories, and since these cultures were, in most instances, quite typical, it is believed that the numerical data are representative and objective. Another publication is planned that will include summaries of all of the biochemical reactions of various members of the family as far as they have been studied by the author and co-workers.

It is the author's opinion that unless percentages are included in tabular data, the resulting tables are of little value. As in the past, 90 percent levels are employed in determining the signs to be applied. Some arbitrary level must be selected, and the author and colleagues believe that the 90 percent level is the most reasonable and practical. Thus a "+" sign means that 90 to 100 percent of strains tested on a particular substrate gave positive reactions within 1 or 2 days of incubation (usually within 24 hr). The addition of the actual percentage of such reactions (e.g., 91% or 98%) yields useful information; conversly, the same is true of negative results. If percentages are included, the sign "d" (for different reactions) assumes meaning in many instances. For example, if 60 to 90 percent of results obtained in a test are positive, then the "d" sign is meaningful, since most cultures yielded positive results. Similarly, the "d" sign is useful if most of the strains give negative results on a particular substrate (e.g., 11% to 25% or 30%). When the percentages are between 30 and 60 percent, the "d" sign usually indicates that the test is of little differential value in that particular segment of the family, but this cannot be determined if percentages of positive results are not listed. There are some instances, however, in which a "d" sign followed by about 40 percent positive, for example, is of value, as in the case of the differentiation of *Salmonella cholerae-suis,* inositol −: 0%+, and *Salmonella enteritidis* inositol d: 39%+, or in differentiating commonly occurring salmonellae from members of the genus *Arizona.*

Classification and Nomenclature

The system of classification and nomenclature employed herein (see below) is that proposed by the author in 1963 *(15)* as emended and extended in subsequent publications *(18,20,30,33,39,43,45)*. Because of changes made in the rules of nomenclature approved during the IX International Congress of Microbiology (see reference *53*), it became necessary to change the specific epithet *arizonae* in *Arizona arizonae*. For this, the specific epithet *hinshawii* was proposed (*20*) in recognition of Dr. William R. Hinshaw who did much of the pioneer work with members of the genus *Arizona*.

In 1966 (*17*) the author submitted a request for an opinion regarding a proposal for validation of the species name *Arizona arizonae* Kauffman and Edwards (*57*). As mentioned above, the specific epithet had to be changed. More recently, the author was informed (personal communication, 1969) by Dr. P. A. H. Sneath (Chairman, Judicial Commission of the International Committee on Nomenclature of Bacteria) that the Judicial Commission had ruled that the generic name *Arizona* was not validly published by Kauffman and Edwards (*57*) because the classification and names used by those investigators were suggested, not recommended (Rule 12c). However, the generic name *Arizona* was validly published by the author and co-workers (*13,15,36,41*). Since the genus *Arizona* was characterized and defined (*13,15,36,41*) and since the specific epithet was changed legitimately to *hinshawii* (*20*), the correct citations should be *Arizona hinshawii* Ewing and Fife) Ewing (see outline below). Of course, some European investigators regard *Arizona* as a subgenus of *Salmonella*, but there are good and sufficient reasons for maintaining *Arizona* as a separate genus. This subject is discussed elsewhere (*7,13,14,23*).

If *Arizona* Kauffmann and Edwards (*57*) was invalidly published, then *Providencia* Kauffman and Edwards also was. However, the genus *Providencia* was validly published, charaterized, and defined by the author (*15* and references therein).

Citrobacter diversus has been added as a second species within the genus *Citrobacter* for reasons given elsewhere (*27,30*). Some authors use other names, such as *Citrobacter koseri* (*48,52*) and *Levinia malonatica* (*76*), for these bacteria, but the species name *Citrobacter diversus* (Burkey) Werkman and Gillen was validly published and has priority (see references *27,30*).

The species *Enterobacter agglomerans* has been added to the genus *Enterobacter*. These bacteria apparently are saprophytic soil or water microorganisms that frequently are found on grasses and other plants, their leaves, seeds, and fruits, and sometimes in humans and other animals (see references *38-40,51*). In the past they usually have been classified in the genus *Erwinia* under a variety of specific epithets or in other genera such as *Pseudomonas, Xanthomonas, Flavobacterium, Escherichia (Escherichia adecarboxylata),* and *Bacterium (Bacterium typhiflavum)*. This synonomy is discussed elsewhere (*38-40,51*). Although several biogroups of *E. agglomerans* can be differentiated, they all are included in the single species, for the present at least. If good reasons are forthcoming, the species can be divided or subdivided as necessary or desirable, but to suggest this now would be premature (*39*).

The microorganisms formerly classified as *Enterobacter liquefaciens* (Grimes and Hennerty) Ewing have been transferred to the genus *Serratia* as *Serratia liquefaciens* (Grimes and Hennerty) Bascomb et al., and the addition of a third species, *Serratia rubidaea* (Stapp) Ewing et al., has been proposed (*32,33*). The bases for these changes are reviewed in references *32* and *33*; suffice it to add here that the deoxyribonucleic acids (DNAs) of *S. liquefaciens* and *S. rubidaea* are related to those of *Serratia marcescens* (*4*, and personal communication, 1973, Dr. D. J. Brenner, Division of Biochemistry, Walter Reed Army Institute of Research, Walter Reed Medical Center, Washington, D.C. 20012).

The work of Lessel (*59*) indicates that the citation for the authorship of the species name *Proteus morganii* should be changed from *Proteus morganii* (Winslow et al.) Rauss to *Proteus morganii* Winslow et al.) *Yale*.

Brenner et al. (*5*) reported that the DNAs of *Pectobacterium* and *Erwinia* are more closely related to each other than to other members of the family and that both of these genera belong in the family ENTEROBACTERIACEAE. These investigators (*5*) recommended maintenance of the tribe ERWINIEAE, which would include the genera *Erwinia*, exemplified by *Erwinia amylovora*, and *Peetobacterium*, exemplified by *Pectobacterium carotovorum*. The author has adopted these proposals; therefore, the genus *Pectobacterium* was removed from the tribe KLEBSIELLEAE, where the author placed it earlier (*18*) because of relationships of its biochemical reactions to those of KLEBSIELLEAE,

and returned to the tribe ERWINIEAE. The fact remains that the biochemical reactions of strains of the genera *Pectobacterium* and *Erwinia*, as constituted by Brenner et al. (*5*) and in the outline below, resemble those of members of the tribe KLEBSIELLEAE in many respects, and if a culture of either genus is isolated from a specimen of human origin or from a lower animal, differentiation from *Klebsiella, Enterobacter*, and *Serratia* still is required. This is illustrated by the fact that some bacteria described as species of *Pectobacterium* or *Erwinia* actually are *Klebsiella pneumoniae* and *Enterobacter cloacae* (*3,5* and unpublished data). Members of the tribe ERWINIEAE produce sporadic and epiphytic disease in plants and their fruits; they are of economic importance and are of primary interest to the plant microbiologist and the phytopathologist. The frequency with which members of the tribe ERWINIEAE occur in the normal human is not known, but in the experience of the author and colleagues, they are isolated very infrequently from specimens from patients in hospitals. Whether this might be a reflection of the temperature of incubation usually used, the choice of media for primary isolation, or other factors, is not known. *Erwinia* and *Pectobacterium* are not mentioned further. However, the biochemical reactions of pectobacteria are given in references *13,38*.

A definition and the classification and nomenclature of the family ENTEROBACTERIACEAE recommended by the author and colleagues follow:

The family ENTEROBACTERIACEAE consists of gram-negative, aerobic, facultatively anaerobic, asporogenous, rod-shaped bacteria that grow well on artificial media. Some species are atrichous, and nonmotile variants of motile species also may occur. Motile forms are peritrichously flagellated. Nitrates are reduced to nitrites, and glucose is utilized fermentatively with the formation of acid or of acid and gas. The indophenol oxidase test is negative and alginate is not liquefied. Pectate is liquefied by members of only one genus *(Pectobacterium)*.

THE NOMENCLATURE OF THE FAMILY ENTEROBACTERIACEAE IN OUTLINE

Family ENTEROBACTERIACEAE Rahn

TRIBE I ESCHERICHIEAE Bergey, Breed, and Murray

Genus I *Escherichia* Castellani and Chalmers
1. *Escherichia coli* (Migula) Castellani and Chalmers

Genus II *Shigella* Castellani and Chalmers
1. *Shigella dysenteriae* (Shiga) Castellani and Chalmers
2. *Shigella flexneri* Castellani and Chalmers
3. *Shigella boydii* Ewing
4. *Shigella sonnei* (Levin) Weldin

TRIBE II EDWARDSIELLEAE Ewing and McWhorter

Genus I *Edwardsiella tarda* Ewing and McWhorter

TRIBE III SALMONELLEAE Bergey, Breed, and Murray

Genus I *Salmonella* Lignieres
1. *Salmonella cholerae-suis* (Smith) Weldin
2. *Salmonella typhi* (Schroeter) Warren and Scott
3. *Salmonella enteritidis* (Gaertner) Castellani and Chalmers

Genus II *Arizona* Ewing and Fife
1. *Arizona hinshawii* (Ewing and Fife) Ewing

Genus III *Citrobacter* Werkman and Gillen
1. *Citrobacter freundii* (Braak) Werkman and Gillen
2. *Citrobacter diversus* (Burkey) Werkman and Gillen

TRIBE IV KLEBSIELLEAE Trevisan

Genus I *Klebsiella* Trevisan

1. *Klebsiella pneumoniae* (Schroeter) Trevisan
2. *Klebsiella ozaenae* (Abel) Bergey, Breed, and Murray
3. *Klebsiella rhinoschleromatis* Trevisan

Genus II *Enterobacter* Hormaeche and Edwards

1. *Enterobacter cloacae* (Jordan) Hormaeche and Edwards
2. *Enterobacter aerogenes* (Kruse) Hormaeche and Edwards
3. *Enterobacter hafniae* (Moeller) Ewing
4. *Enterobacter agglomerans* (Beijerinck) Ewing and Fife

Genus III *Serratia* Bizio

1. *Serratia marcescens* Bizio
2. *Serratia liquefaciens* (Grimes and Hennerty) Bascomb et al.
3. *Serratia rubidaea* (Stapp) Ewing et al.

TRIBE V PROTEEAE Castellani and Chalmers

Genus I *Proteus* Hauser

1. *Proteus vulgaris* Hauser
2. *Proteus mirabilis* Hauser
3. *Proteus morganii* (Winslow et al.) Yale
4. *Proteus rettgeri* (Hadley et al.) Rustigian and Stuart

Genus II *Providencia* Ewing

1. *Providencia alcalifaciens* (DeSalles Gomes) Ewing
2. *Providencia stuartii* (Buttiaux et al.) Ewing

TRIBE VI ERWINIEAE Winslow et al.

Genus I *Erwinia* Winslow et al.

*1. *Erwinia amylovora* (Burrill) Winslow et al.

Genus II *Pectobacterium* Waldee

*1. *Pectobacterium carotovorum* (Jones) Waldee

N.B. The first species listed in each genus is the type species.

*Only one species is listed in the genus *Erwinia* and in the genus *Pectobacterium*, but additional species (or subspecies, or biotypes) no doubt will be recognized, e.g., *Erwinia rubifaciens, Erwinia salicis, Erwinia tracheiphila; Pectobacterium chrysanthemi, Pectobacterium rhapontici, Pectobacterium carnegieanum.*

Readers who are interested in other classifications or definitions of this family should consult the references (*13,15,23,55-57*) and the several editions of *Bergey's Manual of Determinative Bacteriology*.

Isolation and Preliminary Identification

An outline of methods that may be used to isolate and identify ENTEROBACTERIACEAE from clinical specimens of all sorts is given in Figure 1. Proper selection of plating media and conditions of incubation for isolation of colonies obviously are very important. Most workers employ blood agar plates along with MacConkey's or eosin methylene blue agar or a similar combination for specimens of extraintestinal origin, and use a variety of differential and selective media for stool speciemens. Since the subject of media for isolation, enrichment, and preservation of ENTEROBACTERIACEAE is dealt with in detail in many other publications (e.g., reference *13*), it will not be discussed here.

The schema given in Figure 1 may be modified to meet the requirements of investigators in various kinds of laboratories and in accordance with the nature of a particular specimen. When colonies of ENTEROBACTERIACEAE have been isolated on solid media, methods and procedures for their further examination and identification are much the same, regardless of the source and nature of the specimen in which the bacteria originated.

Although there are many methods and devices (see references *50,61,69,71,73,75*) that may be employed for primary examination of colonies, the author and many colleagues (*16,54*) prefer to use lysine-iron agar (LIA) and motility-indol-ornithine (MIO) media (*11,54*) in conjunction with triple-sugar-iron (TSI) or Kligler's iron (KI) agar for this purpose (Figure 1). With a straight wire, inoculum is taken carefully from a selected colony and transferred to TSI or KI medium in the usual way. Then, *without* going back to the colony, a tube of LIA is inoculated directly by stabbing the butt of the medium twice and streaking the slant. Inoculation of a tube of semisolid MIO medium (by stab to the bottom of the tube), in turn, also is helpful. The use of paper strips impregnated with indol reagent (Kovacs' or that mentioned by Gillies, *50*, and by Johnson et al., *54**) and dried furnishes additional valuable information, particularly when tests are positive. The strips may be suspended in tubes of LIA or MIO medium, being held in place by the closure. Negative tests obtained by this method should be retested by conventional means, especially in instances where a negative result does not align itself with the results of other tests.

The aforementioned system may be extended to include Christensen's urea agar, Simmons' citrate, and a tube of peptone water (conventional medium for indol production). Material from a colony is inoculated into a tube of TSI agar, after which the other media are inoculated from the slant of the TSI medium. This six-tube system (TSI, LIA, MIO, and the three media just mentioned) has been used by Dr. W. J. Martin (Director, Microbiological Laboratories, UCLA Hospital and Clinics, The Center for the Health Sciences, Los Angeles, California 90024) and his colleagues. It has been found to be advantageous, since it permits identification of 90 percent or more of isolants from extraintestinal sources within 24 hours (Laboratory Procedures in Clinical Microbiology, Editor, J. A. Washington II, M.D., Little, Brown and Company, in press).

The reactions of ENTEROBACTERIACEAE in TSI, LIA, and MIO media are shown in Tables 2, 3, and 4. The combinations of results obtained by using the above-mentioned methods yield a great deal of useful information early in the examination of a specimen. In some instances specific identification can be made on the basis of the reactions in TSI, LIA, and MIO or in the six-tube procedure. Frequently, however, it is necessary to use the tests and substrates listed in Figure 1 under the heading of basic or minimal tests, together with those recommended for differentiation within the several tribes. The species of many strains can be determined by the tests given in the basic list. For other isolates, the basic tests are sufficient to determine the tribe and which of the additional substrates (Figure 1) are required for accurate differentiation of species that belong to a particular genus. Finally, occasional cultures may yield atypical reactions in one or more tests; with these all known biochemical tests may have to be performed in order to be certain of the species to which they belong, including additional carbohydrate broths, alginate medium, oxidation-fermentation tests, or incubation of cultures at 22 to 26 C etc. (*13,26*).

*The formula for this reagent is given in references *13,50,54*. Apparently, it was described by Braun, H., and Silberstein, W., 1940, (Tib. Fakültesi, Mecmuasi Yil: *3*, Sayi *12*), but this publication was unavailable to the author. The formula is given, however, by Braun and Özek (*2*).

Differentiation of Genera and Species

The data presented in Tables 5 to 52 largely are self-explanatory, but a few remarks and notes about some of them may be helpful.

The urease reactions given by members of the various genera and species in Christensen's medium should be mentioned. In addition to negative tests, there are three kinds of urease reactions, based upon their speed and strength. Strains of *Proteus* produce rapid, strong, positive reactions that are apparent in a few minutes to a few hours; almost all cultures are positive within 4 hours. When positive, the urease reactions given by members of the genera *Citrobacter, Enterobacter,* and *Serratia* are weak and do not become apparent until after 18 to 24 or more hours of incubation. Reactions yielded by *K. ozaenae* also are weak and delayed, whereas those given by cultures of *K. pneumoniae* are quite strong and usually are apparent after overnight incubation. They are not as rapid as those given by *Proteus,* however, and apparently a certain amount of growth must take place before positive reactions occur in Christensen's medium.

The biochemical reactions listed for *E. coli* (Tables 6, 11) were extracted from data derived from the examination of more than 2,000 cultures (*34*). None of these produced hydrogen sulfide in TSI or peptone-iron (PI) agar. However, the occurrence of occasional isolants of certain bioserotypes of *E. coli* (Alkalescens-Dispar) has been known for some time (*49,* and unpublished data 1949, 1950), but cultures of this sort have not been seen in recent years. Since about 1962, however, the author and co-workers have received strains of *E. coli* that produce abundant hydrogen sulfide in TSI and PI agar. Cultures of this sort had not been seen for a long time either by the author or by other investigators (e.g., personal communication, 1970, Dr. I Ørskov, State Serum Institute, Copenhagen). It now is known (*10,58,72*) that abundant hydrogen sulfide formation by isolants of *E. coli* is mediated by an episome or plasmid. Thus, the appearance of an atypical character in a culture that is otherwise typical may be explained, in many instances at least, by recombinations in which an episome or plasmid carrying genetic material for that character is involved. Darland and Davis (*10*) studied 204 hydrogen sulfide-positive strains identified as *E. coli*, received between 1966 and 1972. In the same period, 4,048 isolates of normal *E. coli* were received for serotyping; therefore, the incidence of hydrogen sulfide-positive cultures in this group was 4.8 percent. However, the author does not believe that the true incidence of such isolants among all *E. coli* is that high. An estimate of about 0.1 percent seems more reasonable. Most hydrogen sulfide-positive forms of *E. coli* are more resistant to ampicillin and tetracycline than are typical strains of that species (*10,72*). It usually is possible to isolate hydrogen sulfide-negative forms from the atypical variants by plating the cultures on a medium such as PI agar, i.e., the episome or plasmid can be lost, and under these circumstances the hydrogen sulfide-negative (normal) form remains stable (*10*).

The most commonly occurring form of *S. choleraesuis* in the United States is the hydrogen sulfide-positive (abundant production) Kunzendorf variant, which is monophasic (6,7:-:1,5), so the percentage of hydrogen sulfide-positive strains of this species probably is higher than indicated in Table 14.

S. enteritidis bioserotype Paratyphi-A is an exception to the rule that salmonellae decarboxylate lysine (final reading at 4 days), and it also has several other rather atypical characteristics (Table 15). This microorganism no longer occurs frequently in the United States, but some carriers may exist, and, it might be imported at any time (*19,25*); hence, bacteriologists should be familiar with it. *S. enteritidis* bioserotype Typhisuis also is lysine-negative and fails to ferment mannitol (*25*).

S. enteritidis bioserotypes Pullorum and Gallinarum (Table 16) are of particular interest to veterinary bacteriologists, but bioser Pullorum is known to occur occasionally in specimens of human origin. Edwards et al. (*12*) reported the identification of 37 cultures of bioser Pullorum from human sources among a total of 12,331 strains of salmonellae. Twenty of these were isolated from a single outbreak of gastroenteritis (see *12,19* for references). The remaining cultures were from 16 sporadic cases of diarrheal disease in babies and adults, and one was from a carrier (*12*). The author has seen one or two strains of bioser Pullorum that were isolated from the bloodstream of children ill with enteric fever. Bioser Gallinarum is host-adapted to chickens and turkeys and has been reported in humans only once or twice (*12,19*). When they occur, isolants of these two bioserotypes must be differentiated from each other and from *S. typhi* (Table 16). Only small numbers of cultures were tested for fermentation of melibiose (Table 16), hence the value of this substrate for this

particular differentiation requires confirmation.

Cultures of *C. diversus* must be differentiated from *C. freundii*, particularly from the hydrogen sulfide-negative variants of the latter species. Means for this differentiation are given in Tables 17 and 18.

Some strains of *C. freundii* do not grow on Simmons' citrate medium. Of 582 isolates studied by Davis and Ewing (*9*), 31 were citrate-negative and indol-negative, whereas 39 were indol-positive and citrate-positive. Two cultures that were indol-positive and citrate negative have since been recognized (*27*), however.

Tests and substrates that are of value for differentiation of the three species of *Klebsiella* are given in Table 19. The ENTEROBACTERIACEAE are not known to liquefy sodium alginate in a nutrient medium (*8* and unpublished data), but a few members of the family, notably *K. pneumoniae* (Table 19), are able to utilize that substrate as a carbon source in a medium similar to Simmons' citrate agar but with 0.25 percent sodium alginate instead of sodium citrate. This alginate medium is useful, therefore, for differentiation within the genus *Klebsiella* (Table 19) and for differentiating cultures of *K. pneumoniae* and *E. aerogenes* (Table 21).

Some information that is not discernible in Table 19 may be of interest. The indol and gelatin reactions given by 3,560 strains of *K. pneumoniae* were as follows:

Gelatin +, indol +	31	(0.9%)
Gelatin +, indol –	2	(<0.1%)
Gelatin –, indol +	189	(5.3%)
Gelatin –, indol –	3,338	(93.7%)
Total	3,560	(100%)

Similar analyses of the results of methyl red (MR) and Voges-Proskauer (VP) tests with 2,855 cultures of *K. pneumoniae* yielded the following information:

MR +, VP +	62	(2.2%)
MR +, VP –	43	(1.5%)
MR –, VP +	2,747	(96.2%)
MR –, VP –	3	(0.1%)

Among other things, the data given above indicate that the MR and VP reactions are not always inverse to each other. Some workers use only the MR test and assume that if a culture is MR-positive, it must be VP-negative and vice versa. This is not recommended. If only one of these tests is used, it should be the VP, which can be done with cultures incubated for 18 to 24 hours.

Of the 2,855 strains of *K. pneumoniae*, 6 (0.2%) were citrate-negative, 51 (1.8%) were anaerogenic, and 4 (0.1%) failed to reduce nitrate. Other biochemical aberrancies occur, of course, as mentioned elsewhere (*13,47*). The decarboxylase reactions of klebsiellae are given in Table 52.

Although it is inadvisable to attempt to identify bacteria on the basis of their incidence, data on frequency of occurrence often is helpful as ancillary information. In the author's experience, *K. pneumoniae* occurs much more frequently than either of the other species: among 5,575 cultures, 5,269 (94.5%) were *K. pneumoniae*, 256 (4.6%) were *K. ozaenae*, and 50 (0.9%) were *K. rhinoschleromatis*. These figures are given for comparison only; investigators should compile their own incidence data for various species of bacteria in their location and circumstances.

K. pneumoniae probably is the most reactive microorganism in the family of terms of the variety and extent of substrates utilized, and some strains, at least, of this species are capable of fixing nitrogen (*62*).

Most cultures labeled *E. aerogenes* submitted to the author and co-workers during the past 25 years actually were *K. pneumoniae*. However, judicious use of the tests and substrates listed in Table 21 should prevent such misidentifications. No strains of *E. aerogenes* examined (*47*) were both nonmotile and ornithine decarboxylase-negative. In addition, Brenner et al. (*4*) reported that the degree of relatedness of the DNAs will distinguish between the most atypical isolates of *Klebsiella* and *E. aerogenes*. The work of these investigators on DNA relatedness indicates that there is no justification for distinguishing taxonomically between motile and nonmotile variants of *E. aerogenes* as suggested by Bascomb et al. (*1*). Data from these studies (*4*) show that DNA relatedness among species of *Klebsiella* is substantially greater than relatedness between *E. aerogenes* and any of the species of *Klebsiella*. Therefore, there is no justification for recognition or use of the specific or subspecific names introduced by Cowan et al. (*6*).

The frequency of occurrence of members of the four species of *Enterobacter* submitted during the past 20 to 25 years was as follows: *E. cloacae*, 1,259 (50.7%); *E. aerogenes*, 121 (4.9%); *E. hafniae*, 565 (22.8%); and *E. agglomerans*, 536 (21.6%). *E. hafniae* was not recognized as a distinct entity until about 1955, and most of the cultures of *E. agglomerans* were received during the last 10 years. These figures are included for comparative purposes only.

All strains of *E. hafniae* studied by Moeller (*66,67*) and by the author (*37,47*, unpublished data) have been indol-negative. However, Sakazaki (*70*) reported that 16 (5.4%) of 294 isolants of this species formed indol. The author certainly does not doubt that indol-positive cultures of this species occur, but he cannot refrain from wondering whether the appearance of this atypical character might not be episomal in origin (see work on *E. coli* cited above and the discussion in reference *13*, p. 62).

Their biochemical reactions clearly show that strains of *E. hafniae* are quite different from other species of *Enterobacter* (Tables 24, 25, 35, and references *37,47*). Analogous differences are reflected in the results of investigations (*4*) of the relatedness of their DNAs to those of other ENTEROBACTERIACEAE. These aggregate facts may indicate that the taxonomic position of *E. hafniae* should be changed in the future. For the present they are left where they are (*37*).

Means by which cultures of *E. agglomerans* (*21,38-40*) may be distinguished from other ENTEROBACTERIACEAE that characteristically fail to produce hydrogen sulfide in TSI agar are given in Tables 26 to 40. The anaerogenic forms of this microorganism have been divided into seven biogroups on the basis of nitrate reduction, production of indol, and VP reactions (*21,38-40*). Four biogroups of aerogenic *E. agglomerans* are recognized. Determination of the biogroup of a culture of this species facilitates its identification, in the author's opinion, and knowledge of the biogroup should be of epidemiological value as well. Extensive tabular data that deals with differentiation of strains of *E. agglomerans* from other ENTEROBACTERIACEAE is given elsewhere (*21,38,40*) and need not be repeated here.

Tests and substrates that are of value for differentiation of the three species of *Serratia* from each other and for differentiating cultures of *S. marcescens* and certain species of *Enterobacter* are listed in Tables 36 and 38 and 41 to 47. Only 49 strains of *S. rubidaea* have been studied (*32,33*), and these produced pink to red pigment (Table 37). The percentage of such isolants may be reduced as more cultures are isolated and characterized.

The author and co-workers received 1,402 cultures of *S. marcescens* between 1955 and 1970. These constituted 89.9 percent of the total of serratiae examined. In about the same period (1957 to 1972), 109 strains of *S. liquefaciens* and 49 isolants of *S. rubidaea* were studied. These made up 7 percent and 3.1 percent of the total, respectively.

It is known that a rare culture of *P. vulgaris* may fail to form indol (*29*) and that an occasional strain of *P. mirabilis* may produce indol (*29,65*), as indicated in Table 49. Cultures of *P. morganii* and *P. rettgeri* examined through 1968 (*29*) all formed indol, but in the period 1969 to 1972 a few cultures were received that failed to produce this substance. Current data indicated that 0.5 percent of isolants of *P. morganii* and 2.9 percent of *P. rettgeri* were indol-negative. A few of the latter cultures also were mannitol-negative.

DECARBOXYLASE REACTIONS OF ENTEROBACTERIACEAE

Practical tests for detection of decarboxylation of lysine, arginine, and ornithine by various ENTERIOBACTERIACEAE were introduced by Moeller (*77,67*). These tests have proven to be of great value, not only throughout the family ENTEROBACTERIACEAE, but in other families as well, e.g., for differentiation of aeromonads and vibrios (*31*).

The results of investigations into the decarboxylase activities of the various species are scattered in the literature (e.g., publications cited in Table 1, wherein the work of others is reviewed); therefore, it was thought advisable to tabulate the results obtained by the author and co-workers in one table. This has been done in Table 52; in it results derived from the examination of 15,831 cultures are summarized.

Although numerous isolates of *S. typhi* were examined earlier, none were encountered that failed to decarboxylate lysine in the recommended 4-day period until 1971 (*44*). As noted above, cultures of *S. enteritidis* bioser Paratyphia-A characteristically fail to decarboxylate lysine (*24,25,31*). *S. enteritidis* bioser Typhisuis also is lysine-negative and mannitol-negative (*24,25,31*). Isolates of *S. typhi* and bioser Gallinarum do not decarboxylate ornithine (*24,74*).

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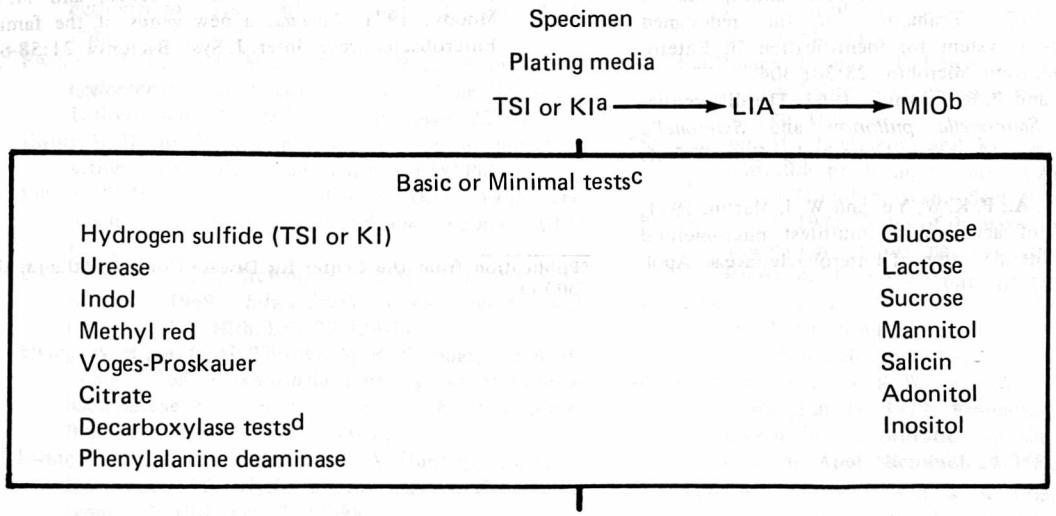
Figure 1

Isolation and Identification of ENTEROBACTERIACEAE



Additional tests for:

Escherichieae	Edwardsielleae	Salmonelleae	Klebsielleae	Proteeae
Esculin	See tables	Dulcitol	Sorbitol	Mannose
Mucate		Malonate	Arabinose	Xylose
Sodium acetate		KCN	Raffinose	Maltose
Christensen's citrate		Gelatin	Rhamnose	Erythritol
See tables		ONPG	Melibiose	Esculin
		Jordan's tartrate	Alginate	Alpha methyl glucoside
		See tables	Gelatin	Lipase
			Lipase or DNase	See tables
			See tables	

[a]Many workers use KI agar with specimens from extraintestinal sources.

[b]Indol test papers may be suspended over LIA or MIO agar.

[c]Inclusion of media for oxidase and nitrate reduction tests is recommended.

[d]Use of all three amino acids and a control is recommended.

[e]Durham insert tubes should be placed in all fluid carbohydrate media.

Table 1

Numbers of cultures studied and references

Species	No. of cultures	References
E. coli	2,505	13, 34
S. dysenteriae	656	13, 22
S. flexneri	3,166	13, 22
S. boydii	704	13, 22
S. sonnei	640	13, 22
E. tarda	494	42, 43, and unpublished data
S. cholerae-suis	20	24, 25
S. typhi	81	24, 25, 44
S. enteritidis	995	24, 25, 64, 74
A. hinshawii	315	36, 41, and unpublished data
C. freundii	616	9, 27, 30
C. diversus	226	27, 30, and unpublished data
K. pneumoniae	1,192	47 and unpublished data
K. ozaenae	256	47 and unpublished data
K. rhinoschleromatis	50	47 and unpublished data
E. cloacae	460	47 and unpublished data
E. aerogenes	121	47 and unpublished data
E. hafniae	286	47 and unpublished data
E. agglomerans	536	21, 38–40
S. marcescens	1,402	28
S. liquefaciens	109	32, 33
S. rubidaea	49	32, 33
P. vulgaris	70	29 and unpublished data
P. mirabilis	372	29 and unpublished data
P. morganii	208	29 and unpublished data
P. rettgeri	170	29 and unpublished data
P. alcalifaciens	674	35
P. stuartii	217	35
Total	16,590	

The work of other investigators is reviewed in the publications cited.

Table 2

Reactions of ENTEROBACTERIACEAE in TSI agar

Genera and species	Slant	Butt	Gas	H_2S
Escherichia	A (K)	A	+ (–)	–[a]
Shigella	K	A	–	–
S. typhi	K	A	–	+ (–)
Other *Salmonella*	K	A	+ (–)	+++ (–)
Arizona	K	A	+	+++
Citrobacter freundii	K (A)	A	+	+++ (–)
C. diversus	K (A)	A	+	–
Edwardsiella	K	A	+	+++
Klebsiella	A	A	++	–
Enterobacter	A	A	++	–
E. hafnia	K	A	+	–
Serratia	A (K)	A	–	–
Proteus vulgaris	A (K)	A	+	+++
P. mirabilis	K (A)	A	+	+++
P. morganii	K	A	– (+)	–
P. rettgeri	K	A	–	–
Providencia	K	A	+ (–)	–

[a]See text.
K, alkaline.
A, acid.
Symbol enclosed in parentheses indicate occasional reactions.

Table 3

Reactions of ENTEROBACTERIACEAE in LIA medium

Genera and species	Slant	Butt	Gas	H_2S
Escherichia	K	K or N	– or +	–[a]
Shigella	K	A	–	–
Salmonella	K	K or N	–	+ (–)
S. typhi	K	K	–	+ (–)
Paratyphi-A	K	A	+ or –	– or +
Arizona	K	K or N	–	+ (–)
Citrobacter freundii	K	A	– or +	+ or –
C. diversus	K	A	– or +	–
Edwardsiella	K	K	– or +	+
Klebsiella	K or N	K or N	+ or –	–
Enterobacter cloacae	K or N	A	+ or –	–
E. aerogenes	K	K or N	+ (–)	–
E. hafniae	K	K or N	– or +	–
E. agglomerans	K	A	– or +	–
Serratia	K or N	K or N	–	–
Proteus vulgaris	R	A	–	– (+)
P. mirabilis	R	A	–	– (+)
P. morganii	K (R)	A	–	–
P. rettgeri	R	A	–	–
Providencia	R	A	–	–

[a]See text.
K, alkaline.
A, acid.
R, red (oxidative deamination).

Symbols in parentheses indicate occasional reactions.

Table 4

Reactions of ENTEROBACTERIACEAE in MIO medium[a]

Genera	Motility	Indol	Ornithine
Escherichia	+ or –	+	– or +
Shigella	–	– or +	– or +[b]
Edwardsiella	+	+	+
Salmonella	+ (–)	–	+
S. typhi	+ (–)	–	–
Arizona	+	– (+)	–
Citrobacter freundii	+ (–)	–	– or +
C. diversus	+ (–)	+	+
Klebsiella	–	–	–
Enterobacter	+	–	+
E. agglomerans	+ or –	– (+)	–
Serratia	+	–	+
S. rubidaea	+	–	–
Proteus vulgaris	+ (–)	+ (–)	–
P. mirabilis	+ (–)	– (+)	+
P. morganii	+ or –	+	+
P. rettgeri	+	+ (–)	–
Providencia	+	+	–

[a]See text and other tables for details.

[b]*S. sonnei* is ornithine – positive.

Symbols enclosed in parentheses indicate occasional reactions.

Table 5

Differentiation of the tribes of ENTEROBACTERIACEAE by biochemical methods

Test or Substrate	Tribes				
	ESCHERICHIEAE	EDWARDSIELLEAE	SALMONELLEAE	KLEBSIELLEAE	PROTEEAE
Hydrogen sulfide (TSI)	–	+	+	–	+ or –
Urease	–	–	–	– or (+)	+ or –
Indol	+ or –	+	–	–	+ or –
Methyl red	+	+	+	–	+
Voges-Proskauer	–	–	–	+	–
Citrate (Simmons')	–	–	+	+	d
KCN	–	–	– or +	+	+
Phenylalanine deaminase	–	–	–	–	+
Mucate	d	–	d	+ or –	–
Mannitol	+ or –	–	+	+	– or +

Note: *S. typhi, S. enteritidis* bioserotype Paratyphi-A and some rare bioserotypes fail to utilize citrate. Cultures of *S. enteritidis* bioser Paratyphi-A and some rare bioserotypes may fail to produce hydrogen sulfide; an occasional strain of almost any serotype of salmonellae may be hydrogen sulfide-negative. Some cultures of *P. mirabilis* may yield positive Voges-Proskauer tests. Some strains of *E. agglomerans* deaminate phenylalanine.

KEY:

+	90% or more positive within 1 or 2 days.
–	90% or more no reactions.
(+)	delayed positive (3 or more days).
d	different biochemical reactions, +, (+), –.
+ or –	most cultures positive, some negative.
– or +	most strains negative, some positive.
– or (+)	most cultures negative, some positive delayed.

Table 6

Differentiation within the tribe ESCHERICHIEAE

Test or Substrate	*Escherichia*			*Shigella*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	+	96.3		– or +	37.8	
Lysine decarboxylase	d	80.6	(1.5)	–	0	
Arginine dihydrolase	d	16.3	(39.1)	d	7.6	(5.6)
Ornithine decarboxylase	d	57.8	(8)	– or +	20[a]	
Mucate	+	91.6		–	0[a]	
Sodium acetate	+ or (+)	83.8	(9.7)	–	0[b]	
Christensen's citrate	d	18.1	(22.6)	–	0	
Gas from glucose	+	92		–	2.1[c]	
Lactose	+	91.6	(4.2)	d	0.3	(11.4)[a]
Sucrose	d	53.7	(5.5)	d	0.9	(31.1)[a]
Salicin	d	36	(12.3)	–	0	
Esculin	d	30.9	(19.7)	–	0	
Motility	+ or –	62.1		–	0	

*Figures in parentheses indicate percentages of delayed positive reactions (3 days or more).

[a]Cultures of *S. sonnei* usually ferment lactose and sucrose slowly and strains of this species decarboxylate ornithine. Some isolates of *S. sonnei* utilize mucate weakly and slowly.

[b]See Table 8.

[c]Certain biotypes of *S. flexneri* 6 form gas.

Note: Obviously there is no difficulty in the differentiation of typical cultures of *E. coli* and shigellae. However, the anaerogenic nonmotile forms of *E. coli, some* of which often are referred to as Alkalescens-Dispar bioserotypes, may require closer examination before they can be classified as *E. coli.* In attempting to classify a particular strain as *E. coli* or as a member of the genus *Shigella,* the biochemical reactivities of the culture should be considered as a whole. Shigellae are much less reactive than *E. coli* and a culture that produces acid promptly (i.e., within 24 hr.) from all, or most of a wide variety of carbohydrates, such as maltose, rhamnose, xylose, sorbitol, and dulcitol, undoubtedly is not a *Shigella.*

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 7

The decarboxylase reactions of shigellae and *E. coli* inclusing nonmotile anerogenic biotypes such as the "Alkalescens-Dispar" biotypes

Genera and Species	No. tested	Lysine			Arginine			Ornithine		
		Sign	%+[a]	(%+)[b]	Sign	%+	(%+)	Sign	%+	(%+)
S. dysenteriae	400	–	0	0	d	1.5	(11.3)	–	0	0
S. flexneri	872	–	0	0	d	7.9	(1.6)	–	0	0
S. boydii	400	–	0	0	d	18.1	(31.9)	–	2.5[c]	0
S. sonnei	633	–	0	0	d	0.5	(5)	+	99.8[d]	0
E. coli	1,887	d	80.6	(1.5)	d	16.3	(39.1)	d	57.8	(8)
"Alkalescens-Dispar" biotypes	618	d	73.6[e]	(8.1)	d	6.8	(42.9)	d	12	(3.4)

[a] %+, percentage of positive reactions that occurred within 1 or 2 days.
[b] (%+), percentage of positive reactions after 3 or 4 days.
[c] These few cultures all were *S. boydii* ser 13, a rare serotype.
[d] Decarboxylation of ornithine is characteristic of *S. sonnei.*
[e] 81.4% of A-D O group 1 (*E. coli* biotype, O group 1) gave positive reactions in lysine medium within 4 days.

KEY:

+	90% or more positive within 1 or 2 days.
–	no reaction (90% or more).
d	different reactions, +, (+), –.

Table 8a

The reactions of shigellae and *E. coli* in acetate, Christensen's citrate, and mucate media

Genera and Species	Sodium acetate				Christensen's citrate				Sodium mucate			
	No. tested	Sign	%+[a]	(%+)[b]	No. tested	Sign	%+	(%+)	No. tested	Sign	%+	(%+)
S. dysenteriae	100	–	0	0	291	–	0	0	400	–	0	0
S. flexneri[c]	425	–	0	0	1,375	–	0	0	823	–	0	0
S. boydii	100	–	0	0	442	–	0	0	123	–	0	0
S. sonnei	100	–	0	0	209	–	0	0	209	– or +[w]	16.4	
E. coli	186	+ or (+)	83.8	(9.7)	469	d	18.1	(22.6)	344	+	91.6	
"Alkalescens-Dispar" biotypes	249	+ or (+)	83	(7)	211	d	35	(35)	120	– or +	38	

[a]Percent positive within 1 or 2 days.

[b]Percent positive in 3 to 7 days.

[c]Includes all serotypes of *S. flexneri* (see also Table 8b).

w, weakly positive reaction.

Table 8b

Reactions of cultures of *S. flexneri* serotype 4 in sodium acetate medium

Subserotype and bioserotype of *S. flexneri* 4	No. tested	%+	(%+)[a]
4a	52	0	(8[w])
4a (mannitol negative)	50	0	(43[w])
4b	52	0	0

[a]Positive in 2 to 7 days.

w, weakly positive reaction.

Table 9

Reactions of 778 cultures of *E. coli* in lysine iron agar

	24 hours			48 hours		
	K/A	K/N	K/K	K/A	K/N	K/K
Number	155	457	166	26	88	664
Percent	19.9	58.7	21.4	3.3	11.3	85.4
		623	(80.1%)		752	(96.7%)

K, alkaline; N, neutral; A, acid.

In these tests the butt of the medium was stabbed only once. It since has been learned that results similar to those listed under 48 hours may be obtained earlier by stabbing the butt of the medium twice.

Table 10

Differentiation of species of *Shigella*

Test or Substrate	*S. dysenteriae*			*S. flexneri* 1 to 5			*S. flexneri* 6			*S. boydii*			*S. sonnei*		
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	– or +	43.7		+ or –	61.5		–	0		– or +	28.8		–	0	
Arginine dihydrolase	d	1.5	(11.3)	–	0[b]		d	48.9	(10.3)	d	18.1	(31.9)	–	0.5	(5)
Ornithine decarboxylase	–	0		–	0		–	0		–	2.5[d]		+	99.4	
Mucate	–	0		–	0		–	0		–	0		– or +	16.4	
Jordan's tartrate	+ or –	78		–	0		–	0		– or +	13		+	100	
Gas from glucose	–	0		–	0		– or +	18.1		–	0		–	0	
Lactose	–	0	(1.6)[a]	–	0	(< 0.1)	–	0		–	1		d	1.8	(88.1)
Sucrose	–	0	(4.2)	d	1.5	(41.9)	–	0		–	0		d	0.1	(85.4)
Mannitol	–	0		+	93.7		+ or –	82.5		+	97.6		+	98.9	
Dulcitol	–	4.5	(0.5)	–	0		d	9.4	(72.2)	d	6.7	(10.4)	–	0	(1)
Sorbitol	d	29.2	(29.5)	d	30.6	(1.5)	(+) or +	30.2	(59.8)	d	41.8	(36.3)	–	1	(1)
Arabinose	d	43.6	(7.2)	d	65	(8.7)	+ or (+)	54.6	(39.3)	+	94.1		+	94.2	(2.9)
Raffinose	–	0		d	52.8	(28.4)	–	0		–	0		d	2.5	(81.5)
Rhamnose	d	32.4	(5.5)	d	6	(6.2)	–	1.6	(3.7)	–	0.2	(1.6)	+ or (+)	77.1	(21)
Maltose	d	12	(77)	d	28.4	(45.3)	(+) or +	16	(74.4)	d	16.6	(66)	+ or (+)	86.4	(6.8)
Xylose	d	3.9	(7.6)	–	1.8	(0.4)[c]	d	0.5	(18.2)	d	11.2	(57.2)	–	1	
Trehalose	+ or (+)	89.8	(7.5)	+ or (+)	77.8	(12.2)	(+) or +	7.4	(92.6)	+ or (+)	85.2	(11.2)	+	100	
Celloboise	–	0		–	0		–	0		–	0		d	10.6	(1.8)
Glycerol	d	12.3	(72.5)	–	0		+ or (+)	60	(31.1)	+ or (+)	55.5	(34.8)	d	13	(32.7)
Beta galactosidase (ONPG)	– or +	49.9[a]		–	0.8		–	0		– or +	11.1		+	95	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Some strains of *S. dysenteriae 1* ferment lactose slowly; all are ONPG positive.

[b]A few doubtful reactions occurred, but these were regarded as negative.

[c]Xylose was fermented by some cultures of the mannitol negative bioserotype of *S. flexneri 4*, but not by other strains of serotypes 1 to 5.

[d]Only cultures of *S. boydii 13* are positive.

+	90% positive within one or two days' incubation.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	majority of strains positive, some cultures negative.
– or +	majority of cultures negative, some strains positive.
(+) or +	majority of reactions delayed, some occur within 1 or 2 days.
d	different reactions: +, (+), –.

Table 11

Differentiation of *Escherichia* and *Edwardsiella*

Test or Substrate	*Escherichia*			*Edwardsiella*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Hydrogen sulfide (TSI)	–	0[a]		+	99.6	(0.2)
Arginine dihydrolase	d	16.3	(39.1)	–	0	(0.2)
Ornithine decarboxylase	d	57.8	(8)	+	99	(0.2)
Lactose	+	91.6	(4.2)	–	0	
Sucrose	d	53.7	(5.5)	–	0.2	
Mannitol	+	97.5		–	0	
Dulcitol	d	49.3	(18)	–	0	
Salicin	d	36	(12.3)	–	0	(0.2)
Sorbitol	d	80.3	(1)	–	0.2	
Arabinose	+	99.3	(0.5)	d	10.7	(0.2)
Raffinose	d	49.4	(2.1)	–	0	
Rhamnose	d	83.5	(3.4)	–	0	
Xylose	d	82.8	(6.6)	–	0	
Trehalose	+	98.2	(1.8)	–	0.3	
Esculin	d	30.9	(19.7)	–	0	
Mucate	+	91.6		–	0	
Jordan's tartrate	+	97.6		–	0	
Sodium acetate	+ or (+)	83.8	(9.7)	–	0	

*Figures in parentheses indicate percentages of reactions delayed 3 or more days.

[a]Some strains produce hydrogen sulfide, but an exact percentage cannot be calcuated at present (see text).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 12
Differentiation of genera within the tribe SALMONELLEAE

Test or Substrate	*Salmonella*[a]			*Arizona*[b]			*Citrobacter*[c]		
	Sign	%+	(%+)*	Sign	%+	(z+)*	Sign	%+	(%+)*
Urease	–	0		–	0		d	70.8	(7.5)
Indol	–	1.1		–	5.1		– or +	21.9	
KCN	–	0.3		–	5.7		+ or –	80.6	
Gelatin (Kohn's)	–	0	(1.1)	+ or (+)	12.1	(84.8)	–	0	(1)
Lysine decarboxylase	+	94.6		+	99.4	(0.3)	–	0	
Ornithine decarboxylase	+	92.7		+	100		d	30.7	(0.1)
Lactose	–	0.8		d	69.8	(15.6)	d	38.3	(50.9)
Sucrose	–	0.5		–	2.9		d	15.5	(7.9)
Dulcitol	d	86.5	(2.7)	–	0		d	58.7	(0.6)
Adonitol	–	0		–	0		– or +	16.3	
Inositol	d	34.5	(0.8)	–	0		–	2.7	(1.6)
Cellobiose	d	6.5	(76.4)	d	0.5	(69.4)	+ or (+)	66.3	(32.2)
Malonate	–	0.5		+	94.6		– or +	32.7	
Jordan's tartrate	+ or –	84.6		–	6		+ or –	83.6	
Beta galactosidase	–	2.1		+	97.8[d]		+	<90	
Organic acid media									
citrate	+ or (+)	87	(4.6)	+ or (+)	75.6	(21.3)	(+) or +	49.2	(49.5)
D-tartrate	+ or (+)	84.3	(5.7)	(+) or –	0	(80.3)	(+)	0	(90.9)

*Figures in parentheses indicate percentages of positive reactions delayed 3 days or more.

[a]Based on 371 cultures; includes representatives of all three species.

[b]Based on 315 cultures.

[c]Based on 695 strains; includes representatives of both species.

[d]In another study (60), 92.8% of the isolates tested were ONPG-positive.

Note: The majority of salmonellae ferment dulcitol promptly, but *S. typhi, S. cholerae-suis*, and *S. enteritidis* bioserotypes Paratyphi-A and Pullorum do not. Members of the genus *Arizona* are uniformly negative on this substrate. Bioser Paratyphi-A is lysine-negative. *S. typhi* is ornithine negative.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 13

Differentiation of *S. enteritidis*, *A. hinshawii*, *C. freundii*, and *C. diversus*

Test or Substrate	*S. enteritidis*[a]			*A. hinshawii*			*C. freundii*			*C. diversus*		
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Hydrogen sulfide	+	93.7		+	98.7		+ or –	81.6		–	0	
Urease	–	0		–	0		d	69.4[w]	(6.9)	+ or (+)	85.8[w]	(6.2)
Indol	–	1.2		–	5.1		–	6.7		+	100	
KCN	–	0.1		–	5.7		+	96.2		–	0	
Gelatin (22 C)	–	1.1		(+) or +	12.1	(84.8)	–	0	(0.9)	–	0	(1)
Lysine decarboxylase	+	94.9	(0.1)	+	99.4	(0.3)	–	0		–	0	
Ornithine decarboxylase	+	96.7		+	100		d	17.2	(0.2)	+	99.6	
Lactose	–	0.9		d	69.8	(15.6)	(+) or +	39.3	(50.8)	d	40.3	(47.8)
Sucrose	–	0.6		–	2.9		d	15.3	(9.4)	– or +	20.8	
Dulcitol	+	95.7	(0.3)	–	0		d	59.8	(0.7)	+ or –	52.2	
Adonitol	–	0		–	0		–	0		+	99.6	
Inositol	d	38.2	(0.9)	–	0		–	3.3	(1.9)	–	0	
Malonate	–	0.6		+	94.6		– or +	21.8		+	94.3	
Jordan's tartrate	+ or –	84.7		–	6		+	96.2		+ or –	71.1	
Beta galactosidase	–	2.1		+	97.8[c]		+	>90		+	100[d]	

*Figures in parentheses indicate percentages of delayed positive reactions.

[a]Summary of reactions given by 335 cultures (includes bioserotypes such as Paratyphi-A).

[c]See footnote, Table 12.

[d]Only lactose-negative strains were tested.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 14

Differentiation of species of *Salmonella*

Test or Substrate	*S. cholerae-suis*[a]			*S. typhi*[a]			*S. enteritidis*[b]					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Hydrogen sulfide (TSI)	d	60	10	$+^{w}$	93.8		+	93.7		+	98	
Citrate (Simmons')	(+)	0	(90)	–	0^{c}		+ or (+)	88.7	(2.1)	+	99.3	(0.7)
Ornithine decarboxylase	+	100		–	0		+	96.7		+	100	
Gas from glucose	+	95		–	0		+	96.1		+	97.7	
Dulcitol	d	5	(25)	d	1.2	(33.4)	+	95.7	(0.3)	+	98.3	
Inositol	–	0		–	0		d	38.2	(0.9)	d	42.8	(1)
Trehalose	–	0		+	100		+	98.8	(1.2)	+	100	
Arabinose	–	0		–	0	(6.3)	+	98.8	(0.6)	+	99.3	
Rhamnose	+	100		–	0		+	94	(1.2)	+	95	
Cellobiose	–	0		d	6.3^{w}	(31.3)	(+) or +	7	(83.5)	(+)	5	(92.8)
Erythritol	d	4^{w}	(68^{w})	–	0		–	0.6^{w}		–	0	
Sodium acetate	– or $(+^{w})$	0	(20)	–	0		d	86.9	(2.1)	+	97.3	(2)
Mucate	–	0		–	0		+ or –	81.5		+ or –	88.3	
Stern's glycerol fuchsin	–	0		–	0		+	98.2		+	98.2	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a] See Table 1.

[b] The percentages given in the fourth column of this table are based on 335 cultures (including such bioserotypes as Paratyphi-A, Pullorum, and Gallinarum. The percentages in the fifth column are based upon 299 strains of commonly occurring serotypes (Paratyphi-A, Pullorum, and Gallinarum excluded).

[c] Rarely, a culture of *S. typhi* may grow slowly and weakly on Simmons' citrate medium (*64*).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 15

Differentiation of *S. enteritidis* bioserotype Paratyphi-A

Test or Substrate	Bioser Paratyphi-A[a]			*S. enteritidis*[b]		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Hydrogen sulfide (TSI)	– or +[w]	12.5		+	93.7	
Citrate (Simmons')	– or (+)	0	(25)	+ or (+)	88.7	(2.1)
Lysine decarboxylase	–	0		+	94.9	
Inositol	–	0		d	38.2	(0.9)
Xylose	–	0		+	93.7	(0.3)
Cellobiose	d	12.5	(6.2)	(+) or +	7	(83.5)
Glycerol	(+)	0	(100)	d	5.1	(14.1)
Stern's glycerol fuchsin	–	0		+	98.2	
Jordan's tartrate	–	0		+ or –	84.7	
Sodium acetate	–	0	(6.2)	d	86.9	(2.1)
Mucate	–	0		+ or –	81.5	
Organic acid media						
citrate	–	0		+ or (+)	86.2	(4.8)
D-tartrate	–	0		d	83.5	(6)
i-tartrate	–	0		d	4.2	(54.6)
l-tartrate	–	0		d	11.8	(73.5)

*Figures in parentheses indicate percentages of reactions delayed 3 days or more.

[a]Based on 16 cultures (references *24,25*).

[b]Based on 335 cultures (see Table 13).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 16

Differentiation of *S. enteritidis* bioserotypes Pullorum and Gallinarum from each other and from *S. typhi*

Test or Substrate	*S. enteritidis*[a]						*S. typhi*		
	bioser Pullorum			bioser Gallinarum					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Motility	–	0		–	0		+	99	
Arginine dihydrolase	– or (+)	0	(30)	–	0		– or (+)	0	(22)
Ornithine decarboxylase	+	100		–	0		–	0	
Mucate	–	0		+	90.3		–	0	
Jordan's tartrate	–	0		+	100		+	91.4	
Cysteine-gelatin	–	0		+	98.1	(1.6)	–	0	
Dulcitol	–	0		+	100		d	1	(33)
Sorbitol	d	20	(40)	(+) or –	0	(70)	+	100	
Rhamnose	+	100		+ or (+)	50	(40)	–	0	
Maltose	–	0		+	98.1	(1.9)	+	100	
Melibiose[b]	–	0		–	0		+	100	
Cellobiose	–	0		d	60	(30)	d	6	(31)
Glycerol	–	0		(+)	0	(90)	(+)	0	(94)
Organic acid media									
citrate	–	0		– or (+)	0	(40)	+	100	
D-tartrate	–	0		+ or (+)	90	(10)	+ or (+)	25	(69)
i-tartrate	–	0		(+)	0	(100)	–	0	
l-tartrate	–	0		(+) or –	0	(80)	–	0	

*Figures in parentheses indicate percentages of reactions delayed 3 or more days.

[a]Based on 103 cultures of each bioserotype.

[b]Only small numbers of strains were tested on melibiose (see text).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 17

Differentiation of hydrogen sulfide-negative cultures of *C. freundii* and *C. diversus*

Test or Substrate	*C. freundii*[a] Sign	%+	(%+)*	*C. diversus*[b] Sign	%+	(%+)*
Indol	+ or –	50.4		+	100	
KCN	+	96.8		–	0	
Ornithine decarboxylase	+ or –	57.5		+	99.6	
Adonitol	–	0		+	100	
Raffinose	d	33.1	(0.8)	–	0	
Malonate	– or +	17.3		+	94.3	
Alpha methyl glucoside	d	11.9	(6.8)	(+) or +	35.8	(57.4)

*Figure in parentheses indicate percentages of reactions delayed 3 or more days.

[a]Based on 127 cultures that failed to produce hydrogen sulfide in TSI medium.

[b]Based on 226 strains.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 18

Differentiation of indol-positive, hydrogen sulfide-negative cultures of *C. freundii* from *C. diversus*

Test or Substrate	*C. freundii*[a] Sign	%+	(%+)*	*C. diversus*[b] Sign	%+	(%+)*
KCN	+	96.9		–	0	
Ornithine decarboxylase[c]	+	96.9		+	99.6	
Dulcitol	– or +	10.9		+ or –	52.2	
Adonitol	–	0		+	100	
Malonate	– or +	14.1		+	94.3	
Alpha methyl glucoside	–	3.4	(1.7)	(+) or +	35.8	(57.4)

*Figures in parentheses indicate percentages of reactions that were delayed 3 days or more.

[a]Based on 64 cultures that produced indol, but failed to form hydrogen sulfide in TSI medium (references *9,27*).

[b]Based on 226 cultures.

[c]Included to show that most cultures of *C. freundii* in this group decarboxylated ornithine.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 19

Differentiation within the genus *Klebsiella*

Test or Substrate	*K. pneumoniae*			*K. ozaenae*			*K. rhinoschleromatis*		
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	+	95.4	(0.1)	d	14.8[w]	14.8	–	0	
Methyl red	– or +	11.3		+	97.7		+	100	
Voges-Proskauer	+	93.7		–	0		–	0	
Citrate (Simmons')	+	96.8	(0.6)	d	28.1	(32.4)	–	0	
Lysine decarboxylase	+	97.2	(0.1)	– or +	35.8	(6.3)	–	0	
Malonate	+	92.5		–	6		+ or –	50	
Mucate	+	92.8		– or +	25		–	0	
Sodium alginate (utilization)	+ or (+)	88.5	(9.2)	– or (+)	0	(11)	–	0	
Gas from glucose	+	96		d	55.5	(9.4)	–	0	
Lactose	+	98.7	(1)	d	26.2	(61.3)	d	6	(70)
Dulcitol	– or +	33		–	0		–	0	
Organic acid media									
citrate	+ or –	64.4		– or +	18		–	0	
D-tartrate	+ or –	67.1		– or +	39		–	0	

*Figures in parentheses indicate percentages of positive reactions that were delayed 3 or more days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 20

Differentiation of *K. pneumoniae* and *E. cloacae*

Test or Substrate	*K. pneumoniae* Sign	%+	(%+)*	*E. cloacae* Sign	%+	(%+)*
Urease	+	95.4	(0.1)	+ or –	74.6[w]	
Motility	–	0		+	92.4	
Gelatin	–	1.9	(0.4)	(+)	0.6	(94.2)
Lysine decarboxylase	+	97.2	(0.1)	–	0	
Arginine dihydrolase	–	0.6		+	92.4	(2)
Ornithine decarboxylase	–	0		+	93.7	(1.3)
Adonitol acid	d	89	(0.2)	– or +	22.2	
gas	d	84.4	(0.3)	– or +	21.7	
Inositol acid	+	97.2	(0.9)	d	13	(8)
gas	+	92.5	(1.5)	–	4.1	(1.5)
Glycerol acid	+	97.6	(1.9)	d	43	(45)
gas	+	92.2	(2.9)	d	5.5	(16)
Esculin	+	98.9	(1.1)	– or +	29.5	
Jordan's tartrate	+	94.4		– or +	27.4	
Sodium alginate (utilization)	+ or (+)	88.5	(9.2)	–	0	

*Figures in parentheses indicate percentages of positive reactions that were delayed 3 or more days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 21

Differentiation of *K. pneumoniae* and *E. aerogenes*

Test or Substrate	*K. pneumoniae* Sign	%+	(%+)*	*E. aerogenes* Sign	%+	(%+)*
Urease	+	95.4	(0.1)	–	5[w]	
Motility	–	0		+	91.7	
Gelatin (22 C)	–	1.9	(0.4)	(+) or –	0	(77.3)
Ornithine decarboxylase	–	0		+	95.9	(0.8)
Sodium alginate (utilization)	+ or (+)	88.9	(8.9)	–	0	

*Figures in parentheses indicate percentages of positive reactions that were delayed 3 or more days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 22

Differentiation of *K. rhinoschleromatis* and *Shigella*

Test or Substrate	*K. rhinoschleromatis* Sign	%+	(%+)*	*Shigella* Sign	%+	(%+)*
Indol	–	0		– or +	37.8	
KCN	– or +	42		–	0	
Ornithine decarboxylase	–	0		d	20	(0.3)
Lactose	d	6	(70)	d	0.3	(11.4)
Sucrose	+ or (+)	52	(44)	d	0.9	(31.1)
Salicin	+	100		–	0	
Adonitol	+	98	(2)	–	0	
Inositol	+	90	(10)	–	0	
Malonate	+ or –	50		–	0	
Xylose	+	100		d	4.3	(11.1)
Cellobiose	+	100		–	2.9	(0.4)
Esculin	+	95	(5)	–	0	

*Figures in parentheses indicate percentages of positive reactions that were delayed 3 or more days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 23

Differentiation of *E. cloacae* and *E. aerogenes*

Test or Substrate	*E. cloacae*			*E. aerogenes*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	+ or –	74.6[w]		–	5[w]	
Lysine decarboxylase	–	0		+	97.5	
Arginine dihydrolase	+	92.4	(2)	–	0	
Jordan's tartrate	– or +	27.4		+ or –	78.3	
Adonitol acid	– or +	22.2		+	97.5	
gas	– or +	21.7		+	94.2	(0.8)
Inositol acid	d	13	(8)	+	96.7	
gas	–	4.1	(1.5)	+	93.4	(0.8)
Glycerol acid	d	43	(45)	+	99.1	
gas	d	5.5	(16)	+	94.4	(2.8)
Esculin	– or +	29.5		+	98	

*Figures in parentheses indicate percentages of positive reactions that were delayed 3 or more days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 24

Differentiation of *E. cloacae* and *E. hafniae*

Test or Substrate	*E. cloacae* Sign	*E. cloacae* %+	*E. cloacae* (%+)*	*E. hafniae* Sign	*E. hafniae* %+	*E. hafniae* (%+)*
Urease	+ or –	74.6[w]		–	6.6[w]	(2.5)
Methyl red	–	3.3		– or +	35	
Voges-Proskauer	+	100		+ or –	83.6	
Citrate (Simmons')	+	98.9	(0.4)	d	5.6	(63)
Gelatin (22 C)	(+)	0.6	(94.2)	–	0	
Lysine decarboxylase	–	0		+	99.6	
Arginine dihydrolase	–	92.4	(2)	d	4.6	(6.6)
Lactose	+ or (+)	76.3	(21.8)	d	2.8	(11.9)
Sucrose	+	94.1	(0.9)	d	7	(46.5)
Salicin	+ or (+)	69.1	(26.5)	d	11.2	(5.9)
Adonitol	– or +	22.2		–	0	
Inositol	d	13	(8)	–	0	
Sorbitol	+	90.4		–	0	
Raffinose	+	90.7		–	3.8	(1.1)
Mucate	+ or –	75.5		–	0	
Gas from glycerol	d	5.5	(16)	+	95	
Alpha methyl glucoside	+ or (+)	84.1	(12.9)	–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 25

Differentiation of *E. aerogenes* and *E. hafniae*

Test or Substrate	*E. aerogenes* Sign	%+	(%+)*	*E. hafniae* Sign	%+	(%+)*
Methyl red 37 C	–	1.6		+ or –	35	
22 C				–	0.4	
Voges-Proskauer 37 C	+	100		+ or –	83.6	
22 C				+	99.6	
Citrate (Simmons') 37 C	+	92.6	(2.4)	d	5.6	(63)
22 C				d	3	(79)
Gelatin (22 C)	(+) or –	0	(77.3)	–	0	
Mucate	E	94.7		–	0	
Adonitol	+	97.5		–	0	
Inositol	+	96.7		–	0	
Sorbitol	+	98.3		–	0	
Raffinose	+	96.7		–	3.8	(1.1)
Salicin	+	99.2	(0.8)	d	11.2	(5.9)
Alpha methyl glucoside	+	96	(2)	–	0	
Esculin	+	98		–	6	(2)

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 26

Differentiation of *E. agglomerans* and *E. coli*

Test or Substrate	*E. agglomerans*						*E. coli*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	0	
Indol	– or +	13.7		– or +	37.2		+	96.3	
Methyl red	– or +	45.4		– or +	42.5		+	99.9	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	–	0.2	(0.3)
KCN	– or +	28.4		+ or –	55.8		–	2.6	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	
Lysine decarboxylase	–	0		–	0		d	80.6	(1.5)
Arginine dihydrolase	–	0		–	0		d	16.3	(39.1)
Ornithine decarboxylase	–	0		–	0		d	57.8	(8)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Malonate	+ or –	60.9		+ or –	66.1		–	0	
Gas from glucose	–	0		+	100		+	92	
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	<1	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 27

Differentiation of *E. agglomerans* and *Shigella*

Test or Substrate	*E. agglomerans*						*Shigella*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	0	
Methyl red	– or +	45.4		– or +	42.5		+	99.9	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	–	0	
KCN	– or +	28.4		+ or –	55.8		–	0	
Motility	+ or –	89.6		+ or –	88.5		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Malonate	+ or –	60.9		+ or –	66.1		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	3.5[w] [a]	
Gas from glucose	–	0		+	100		–	2.1	
Salicin	d	54.8	(26.7)	+	96.4		–	0	
Adonitol	–	2.9	(0.2)	– or +	21.2		–	0	
Xylose	+	91.4	(3.9)	+	97.3		d	4.3	(11.1)
Esculin	d	53.4	(32.3)	+	93.7	(0.9)	–	0	
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Some cultures of *S. sonnei* attack mucate weakly and slowly.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 28

Differentiation of *E. agglomerans* and hydrogen sulfide-negative (TSI) strains of *C. freundii*

Test or Substrate	*E. agglomerans*						*C. freundii*[a]		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	– or +	13.7		– or +	37.2		+ or –	50.4	
Methyl red	– or +	45.4		– or +	42.5		+	100	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	(9.7)[b]
Arginine dihydrolase	–	0		–	0		d	52	(31.5)
Ornithine decarboxylase	–	0		–	0		+ or –	57.5	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Jordan's tartrate	–	0		–	6.3		+ or –	59	
Gas from glucose	–	0		+	100		+ or –	81.1	
Sucrose	d	77.5	(1.1)	d	75.2	(2.7)	d	36.2	(1.6)
Salicin	d	54.8	(26.7)	+	96.4	(1.8)	d	27.6	(41.9)
Adonitol	–	2.9	(0.2)	– or +	21.2		–	0	
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	95.3	(0.8)
Esculin	d	53.4	(32.3)	+	93.7		d	5.1	(24.5)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Based on 127 strains that failed to produce hydrogen sulfide in TSI medium (*9,27*).
[b]Delayed 22–30 days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 29

Differentiation of *E. agglomerans* and *C. diversus*

Test or Substrate	*E. agglomerans*						*C. diversus*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8	(9.7)	d	40	(3.5)	+ or (+)	85.8	(6.2)
Indol	– or +	13.7		– or +	37.2		+	100	
Methyl red	– or +	45.4		– or +	42.5		+	100	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	+	99.7	(0.4)
KCN	– or +	28.4		+ or –	55.8		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	(1)
Arginine dihydrolase	–	0		–	0		+ or (+)	62.4	(2.6)
Ornithine decarboxylase	–	0		–	0		+	99.6	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Sucrose	d	77.5	(1.1)	d	75.2	(2.7)	– or +	20.8	
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	+ or –	52.2	
Adonitol	–	2.9	(0.2)	– or +	21.2		+	100	
Sorbitol	d	16.1	(5.4)	+ or –	60.2		+	98.2	(0.9)
Raffinose	d	14.4	(3.3)	d	63.7	(1.8)	–	0	
Jordan's tartrate	–	0		–	6.3		+ or –	71.1	
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	(+) or +	35.8	(57.4)
Esculin	d	53.4	(32.3)	+	93.7	(0.9)	d	2.6	(61.6)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 30

Differentiation of *E. agglomerans* and *K. pneumoniae*

Test or Substrates	*E. agglomerans*						*K. pneumoniae*		
	Anerogenic			Aerogenic					
	Sign	%	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	+	95.4	(0.1)
Indol	– or +	13.7		– or +	37.2		–	6.8	
Methyl red	– or +	45.4		– or +	42.5		– or +	11.3	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		+	93.7	
Motility	+ or –	89.6		+ or –	88.5		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	1.9	(0.4)
Lysine decarboxylase	–	0		–	0		+	97.2	(0.1)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Sodium alginate utilization	–	0		–	0		+ or (+)	88.9	(8.9)
Jordan's tartrate	–	0		–	6.3		+	94.4	
Gas from glucose	–	0		+	100		+	96	
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	– or +	33	
Adonitol	–	2.9	(0.2)	– or +	21.2		d	89	(0.2)
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	+	97.2	(0.9)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	99.4	(0.3)
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	+ or (+)	86.3	(13.7)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 31

Differentiation of *E. agglomerans* and *K. ozaenae*

Test or Substrate	*E. agglomerans*						*K. ozaenae*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	– or +	13.7		– or +	37.2		–	0	
Methyl red	– or +	45.4		– or +	42.5		+	97.7	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Motility	+ or –	89.6		+ or –	88.5		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	
Lysine decarboxylase	–	0		0			d	35.8	(6.3)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Malonate	+ or –	60.9		+ or –	66.1		–	6	
Jordan's tartrate	–	0		–	6.3		– or +	40	
Gas from glucose	–	0		+	100		d	55.5	(9.4)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0	
Adonitol	–	2.9	(0.2)	– or +	21.2		+	91.8	(3.1)
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	d	54.3	(16.8)
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	d	62	(25)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 32

Differentiation of *E. agglomerans* and *K. rhinoschleromatis*

Test or Substrate	*E. agglomerans*						*K. rhinoschleromatis*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	0	
Indol	– or +	13.7		– or +	37.2		–	0	
Methyl red	– or +	45.4		– or +	42.5		+	100	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	–	0	
Motility	+ or –	89.6		+ or –	88.5		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	0	
Adonitol	–	2.9	(0.2)	– or +	21.2		+	98	(1)
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	+	90	(10)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	98	(1)
Raffinose	d	14.2	(3.3)	d	63.7	(1.8)	+ or (+)	86	(14)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 33

Differentiation of *E. agglomerans* and *E. cloacae*

Test or Substrate	*E. agglomerans*						*E. cloacae*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	– or +	13.7		– or +	37.2		–	0	
Methyl red	– or +	45.4		– or +	42.5		–	3.3	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		+	100	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	+	98.9	(0.4)
Arginine dihydrolase	–	0		–	0		+	92.4	(2)
Ornithine decarboxylase	–	0		–	0		+	93.7	(1.3)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Gas from glucose	–	0		+	100		+	99.3	
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	d	15.2	(0.4)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	90.4	
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	+ or (+)	84.1	(12.9)
Esculin	d	53.4	(32.3)	+	93.7	(0.9)	– or +	29.5	
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	<1	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 34

Differentiation of *E. agglomerans* and *E. aerogenes*

Test or Substrate	*E. agglomerans*						*E. aerogenes*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	5[w]	
Indol	– or +	13.7		– or +	37.2		–	0.8	
Methyl red	– or +	45.4		– or +	42.5		–	1.6	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		+	100	
KCN	– or +	28.4		+ or –	55.8		+	97.5	
Lysine decarboxylase	–	0		–	0		+	97.5	
Ornithine decarboxylase	–	0		–	0		+	95.9	(0.8)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Jordan's tartrate	–	0		–	6.3		+ or –	78.3	
Gas from glucose	–	0		+	100		+	95.9	(0.8)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	4.1	
Adonitol	–	2.9	(0.2)	– or +	21.2		+	97.5	
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	+	96.7	
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	+	96	(2)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days..
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 35

Differentiation of *E. agglomerans* and *E. hafniae*

Test or Substrate	*E. agglomerans*						*E. hafniae*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	6.6	(2.5)
Indol	– or +	13.7		– or +	37.2		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	d	5.6	(63)
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	
Lysine decarboxylase	–	0		–	0		+	99.6	
Ornithine decarboxylase	–	0		–	0		+	98.6	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	0	
Jordan's tartrate	–	0		–	6.3		+ or –	58.6	
Gas from glucose	–	0		+	100		+	98.9	
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	d	2.8	(11.9)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	2.4	
Salicin	d	54.8	(26.7)	+	96.4	(1.8)	d	11.2	(5.9)
Adonitol	–	2.9	(0.2)	– or +	21.2		–	0	
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	–	0	
Sorbitol	d	14.2	(0.5)	+ or –	60.2		–	0	
Raffinose	d	14.4	(3.3)	d	63.7	(1.8)	–	3.8	(1.1)
Alpha methyl glucoside	–	2.7	(1.5)	d	21.4	(8.1)	–	0	
Esculin	d	53.4	(32.3)	+	93.7	(0.9)	–	6	(2)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 36

Differentiation of *E. agglomerans* and *S. marcescens*

Test or Substrate	*E. agglomerans*						*S. marcescens*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Indol	– or +	13.7		– or +	37.2		–	0.1[w]	
Methyl red	– or +	45.4		– or +	42.5		– or +	18.5	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		+	98.7	
Lysine decarboxylase	–	0		–	0		+	99.6	
Ornithine decarboxylase	–	0		–	0		+	99.6	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Malonate	+ or –	60.9		+ or –	66.1		–	1.6	
Mucate	– or +	35.4		+ or –	76.8		–	0	
Jordan's tartrate	–	0		–	6.3		+	99.5	
Lipase (corn oil)	–	0		–	0		+	98.4	(0.7)
DNase	–	0		–	0		+	97.2	(2.8)
Gas from glucose	–	0		+	100		+ or –	52.6[a]	
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	–	1.3	(4.6)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0	
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	d	77.3	(6.4)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	99.1	
Arabinose	+	97.9	(0.4)	+	98.2		–	0	
Raffinose	d	14.4	(3.3)	d	63.7	(1.8)	–	1.2	(0.8)
Rhamnose	d	82.8	(5.3)	+	98.2	(0.9)	–	0	
Xylose	+	91.4	(3.9)	+	97.3		d	7.1	(17.2)
Pigment	+ or –	77.1 yellow		+ or –	51.3 yellow		– or +	16.8 pink to red	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 5 or 10%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 37

Differentiation of *E. agglomerans* and *S. liquefaciens*

Test or Substrate	*E. agglomerans*						*S. liquefaciens*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	d	3.7[w]	(11)
Indol	– or +	13.7		– or +	37.2		–	1.8[w]	
Lysine decarboxylase	–	0		–	0		+ or (+)	64.2	(31.2)
Ornithine decarboxylase	–	0		–	0		+	100	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0.9	
Malonate	+ or –	60.9		+ or –	66.1		–	0.9	
Mucate	– or +	35.4		+ or –	76.8		–	0.9	
Jordan's tartrate	–	0		–	6.3		+ or –	70.3	
Lipase	–	0		–	0		+ or (+)	85.9	(4.7)
DNase	–	0		–	0		+ or –	88.3	
Gas from glucose	–	0		+	100		d	72.5	(0.9)
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	d	15.6	(21)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0	
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	d	64.2	(25.7)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		+	97.3	
Rhamnose	d	82.8	(5.3)	+	98.2	(0.9)	d	16.5	(0.9)
Glycerol	d	15.3	(22.4)	d	31.3	(36.6)	+	92.2	(6.8)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 38

Differentiation of *E. agglomerans* and *S. rubidaea*

Test or Substrate	*E. agglomerans*						*S. rubidaea*		
	Anaerogenic			Aerogenic					
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	d	4[w]	(16)
Indol	– or +	13.7		– or +	37.2		–	2[w]	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		+	92	
Lysine decarboxylase	–	0		–	0		+ or (+)	61	(31)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	0	
Jordan's tartrate	–	0		–	6.3		+ or –	78	
Lipase (corn oil)	–	0		–	0		+	98	
DNase	–	0		–	0		+	100	
Gas from glucose	–	0		+	100		d	35[a]	(4)
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	+	100	
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0	
Adonitol	–	2.9	(0.2)	– or +	21.2		+ or (+)	88	(2)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		–	8	
Raffinose	d	14.4	(3.3)	d	63.7	(1.8)	+	96	
Rhamnose	d	82.8	(5.3)	+	98.2	(0.9)	–	4	(2)
Pigment	+ or –	77.1 yellow		+ or –	51.3 yellow		+ or –	61 pink to red	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 5 or 10%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 39

Differentiation of *E. agglomerans* from *P. morganii* and *P. rettgeri*

Test or Substrate	*E. agglomerans*						*P. morganii*			*P. rettgeri*		
	Anaerogenic			Aerogenic								
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	+	97.1	(0.5)	+	100	
Indol	– or +	13.7		– or +	37.2		+	99.5		+	95.9	(1.2)
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0		–	0	
Citrate (Simmons')	+ or (+)	67.8	(22.7)	d	61.9	(6.2)	–	0		+	96	(3.3)
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	(2)[a]	–	0	(2)[a]
Ornithine decarboxylase	–	0		–	0		+	96		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	0		–	0	
Phenylalanine deaminase	– or +	31.1		– or +	15.9		+	95		+	98	
Gas from glucose	–	0		+	100		+ or –	86[b]		– or +	12[b]	
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	–	0		d	10	(2)
Mannitol	+	100		+	100		–	0		+ or –	89	
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0		–	0	
Adonitol	–	2.9	(0.2)	– or +	21.2		–	0		d	81	(6)
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	–	0		+	93	(5)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		–	0		d	1	(10)
Arabinose	+	97.9	(0.4)	+	98.2		–	0		–	0	
Raffinose	d	14.4	(3.3)	d	63.7	(1.8)	–	0		–	0	
Rhamnose	d	82.8	(5.3)	+	98.2	(0.9)	–	0		+ or –	68	
Maltose	+ or (+)	86.7	(4.5)	+	97.3	(1.8)	–	0		–	2	(2)
Xylose	+	91.4	(3.9)	+	97.3		–	0		– or +	15	
Erythritol	–	0		–	0		–	0		d	78	(7)
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Gelatin liquefaction was weak and was not apparent until 30–35 days.

[b]Volumes of gas produced are small (bubble to 10 or 15%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 40

Differentiation of *E. agglomerans* from *P. alcalifaciens* and *P. stuartii*

Test or Substrate	*E. agglomerans*						*P. alcalifaciens*			*P. stuartii*		
	Anaerogenic			Aerogenic								
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	24.8[w]	(9.7)	d	40[w]	(3.5)	–	0		–	0	
Indol	– or +	13.7		– or +	37.2		+	99.4		+	98.6	
Voges-Proskauer	+ or –	70.7		+ or –	57.5		–	0		–	0	
Gelatin (22 C)	d	2.9	(84.2)	d	3.5	(64.6)	–	0	(1.4)	–	0	(4.3)
Phenylalanine deaminase	– or +	31.1		– or +	15.9		+	97.4		+	94.5	
Malonate	+ or –	60.9		+ or –	66.1		–	0		–	0	
Mucate	– or +	35.4		+ or –	76.8		–	0		–	0	
Gas from glucose	–	0		+	100		d	85.2	(0.6)[a]	–	0	
Lactose	d	28.6	(10.9)	+ or (+)	85	(11.5)	–	0.3		–	3.6	
Mannitol	+	100		+	100		–	1.9	(0.1)	d	11.8	(9.1)
Dulcitol	–	1.4	(0.5)	d	52.2	(0.9)	–	0		–	0	
Salicin	d	54.8	(26.7)	+	96.4	(1.8)	–	0.6	(0.3)	–	1.8	
Adonitol	–	2.9	(0.2)	– or +	21.2		+	94.3	(0.3)	– or +	12.4	
Inositol	d	16.1	(5.4)	–	7.1	(2.7)	–	0.6		+	97.2	(2.8)
Sorbitol	d	14.2	(0.5)	+ or –	60.2		–	0.6	(0.6)	d	3.4	(34.7)
Arabinose	+	97.9	(0.4)	+	98.2		–	0.7	(0.7)	–	4	(2.7)
Rhamnose	d	82.8	(5.3)	+	98.2		–	0	(1.1)	–	0	
Maltose	+ or (+)	86.7	(4.5)	+	97.3	(1.8)	–	0.6	(0.6)	–	3.3	
Xylose	+	91.4	(3.9)	+	97.3		–	0.7		–	5.5	(0.9)
Esculin	d	53.4	(32.3)	+	93.7	(0.9)	–	0		–	0	
Pigment (yellow)	+ or –	77.1		+ or –	51.3		–	0		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 10 or 15%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 41

Differentiation of species of *Serratia*

Test or substrate	*S. marcescens*			*S. liquefaciens*			*S. rubidaea*		
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Voges-Proskauer	+	98.7		– or +	49.5		+	92	
Lysine decarboxylase	+	99.6		+ or +	64.2	(31.2)	+ or (+)	61	(31)
Ornithine decarboxylase	+	99.6		+	100		–	0	
Malonate	–	1.6		–	0.9		+ or –	86	
KCN	+	98.9		+	91.7		– or +	22	
Lactose	–	1.3	(4.6)	d	15.6	(21)	+	100	
Adonitol	d	46.5	(13.8)	d	8.3	(5.5)	+ or (+)	88	(2)
Sorbitol, acid	+	99.1		+	97.3		–	8	
gas	–	0		d	57.8	(19.3)	–	0	
Arabinose	–	0		+	97.3		+	100	
Raffinose	–	1.2	(0.8)	+	90.8	(4.6)	+	96	
Xylose	d	7.1	(17.2)	+	99.1	(0.9)	+	98	
Glycerol, acid	+	97.2	(1.8)	+	92.2	(6.8)	d	29	(18)
gas	–	0		d	39.8	(30.1)	–	0	
Melibiose	–	0		d	73.8	(7.5)	+	96	

*Figures in parenthenses indicate percentages of delayed reactions (3 or more days).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 42

Differentiation of *S. liquefaciens* and *S. marcescens*

Test or Substrate	*S. liquefaciens* Sign	*S. liquefaciens* %+	*S. liquefaciens* (%+)*	*S. marcescens* Sign	*S. marcescens* %+	*S. marcescens* (%+)*
Urease	d	3.7[w]	(11)[w]	d	39.7[w]	(22.3[w])
Methyl red (37 C)	+ or –	64.2		– or +	18.5	
(22 C)	– or +	36.7		–	8.8	
Voges-Proskauer	– or +	49.5		+	98.7	
Lysine decarboxylase	+ or (+)	64.2	(31.2)	+	99.6	
Lactose	d	15.6	(21)	–	1.3	(4.6)
Adonitol	d	8.3	(5.5)	d	46.5	(13.8)
Sorbitol gas	d	57.8	(19.3)	–	0	
Arabinose	+	97.3		–	0	
Raffinose	+	90.8	(4.6)	–	1.2	(0.8)
Rhamnose	d	16.5	(0.9)	–	0	
Xylose	+	99.1	(0.9)	d	7.1	(17.2)
Cellobiose gas	d	7.1	(32.3)	–	0	
Glycerol gas	d	39.8	(30.1)	–	0	
Erythritol	–	0		d	1.6	(25.1)
Melibiose	d	73.8	(7.5)	–	0[a]	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]150 cultures tested.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 43

Differentiation of *S. liquefaciens* and *S. rubidaea*

Test or Substrate	*S. liquefaciens* Sign	%+	(%+)*	*S. rubidaea* Sign	%+	(%+)*
Voges-Proskauer	– or +	49.5		+	92	
KCN	+	91.7		– or +	22	
Ornithine decarboxylase	+	100		–	0	
Lactose	d	15.6	(21)	+	100	
Adonitol	d	8.3	(5.5)	+ or (+)	88	(2)
Sorbitol acid	+	97.3		–	8	
gas	d	57.8	(19.3)	–	0	
Rhamnose	d	16.5	(0.9)	–	4	(2)
Malonate	–	0.9		+ or –	86	
Cellobiose acid	d	27.3	(36.3)	+	90	(6)
gas	d	7.1	(32.3)	–	0	
Glycerol acid	+	92.2	(6.8)	d	29	(18)
gas	d	39.8	(30.1)	–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

Note: Delayed reactions given by *S. rubidaea* occurred within 3 to 7 days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 44

Differentiation of *S. rubidaea* and *S. marcescens*

Test or Substrate	*S. rubidaea* Sign	*S. rubidaea* %+	*S. rubidaea* (%+)*	*S. marcescens* Sign	*S. marcescens* %+	*S. marcescens* (%+)*
KCN	– or +	22		+	98.9	
Lysine decarboxylase	+ or (+)	61	(31)	+	99.6	
Ornithine decarboxylase	–	0		+	99.6	
Lactose	+	100		–	1.3	(4.6)
Adonitol	+ or (+)	88	(2)	d	46.5	(13.8)
Inositol	d	35	(16)	d	77.3	(6.4)
Sorbitol	–	8		+	99.1	
Arabinose	+	100		–	0	
Raffinose	+	96		–	1.2	(0.8)
Malonate	+ or –	86		–	1.6	
Xylose	+	98		d	7.1	(17.2)
Cellobiose	+	90	(6)	d	14.2	(25.2)
Glycerol	d	29	(18)	+	97.2	(1.8)
Erythritol	–	2	(4)	d	1.6	(25.1)
Melibiose	+	96		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

Note: Delayed reactions given by *S. rubidaea* occurred within 3 to 7 days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.
F	fermentative reaction

Table 45

Differentiation of *S. marcescens* and *E. cloacae*

Test or Substrate	*S. marcescens*			*E. cloacae*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	39.7[w]	(22.3)[w]	+ or –	74.6[w]	
Gelatin (22 C)	+ or (+)	84.4	(11.3)	(+)	0.6	(94.2)
Lysine decarboxylase	+	99.6		–	0	
Arginine dihydrolase	–	0.9[w]		+	92.4	(2)
Malonate	–	1.6		+ or –	80.5	
Mucate	–	0		+ or –	75.5	
Gas from glucose	+ or –	52.6[a]		+	99.3	
Lactose	–	1.3	(4.6)	d	76.3	(21.8)
Inositol	d	77.3	(6.4)	d	13	(8)
Arabinose	–	0		+	99.4	
Raffinose	–	1.2	(0.8)	+	90.7	
Rhamnose	–	0		+ or (+)	89.8	(1.2)
Lipase	+	98.4	(0.7)	–	0	
Xylose	d	7.1	(17.2)	+	98	(0.5)
Alpha methyl glucoside	–	0.8	(0.6)	+ or (+)	84.1	(12.9)
DNase	+	97.2		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 5 or 10%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 46
Differentiation of *S. marcescens* and *E. aerogenes*

Test or Substrate	*S. marcescens*			*E. aerogenes*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	39.7[w]	(22.3)[w]	–	5[w]	
Gelatin (22 C)	+ or (+)	84.4	(11.3)	(+) or –	0	(61.2)
Gas from glucose	+ or –	52.6[a]		+	95.9	(0.8)
Lactose	–	1.3	(4.6)	+	92.5	(5)
Gas from adonitol	–	0		+	94.2	(0.8)
Gas from inositol	–	0		+	93.4	(0.8)
Arabinose	–	0		+	100	
Raffinose	–	1.2	(0.8)	+	96.7	
Rhamnose	–	0		+	99.2	
Malonate	–	1.6		+ or –	74.7	
Mucate	–	0		+	94.7	
Lipase	+	98.4	(0.7)	–	0	
Xylose	d	7.1	(17.2)	+	100	
Gas from cellobiose	–	0		+	100	
Gas from glycerol	–	0		+	94.4	(0.8)
Alpha methyl glucoside	–	0.8	(0.6)	+	96	(2)
DNase	+	97.2		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).
[a]Volumes of gas produced are small (bubble to 5 to 10%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 47
Differentiation of *S. marcescens* and *E. hafniae*

Test or Substrate	*S. marcescens*			*E. hafniae*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	d	39.7[w]	(22.3)[w]	–	6.6[w]	(2.5)[w]
Citrate (Simmons')	+	97.6	(1.3)	d	5.6	(63)
Gelatin (22 C)	+ or (+)	84.4	(11.3)	–	0	
Gas from glucose	+ or –	52.6[a]		+	98.9	
Sucrose	+	99.4		d	7	(46.5)
Salicin	+	95.5	(1.1)	d	11.2	(5.9)
Adonitol	d	46.5	(13.8)	–	0	
Inositol	d	77.3	(6.4)	–	0	
Sorbitol	+	99.1	(0.9)	–	0	
Arabinose	–	0		+	99.3	
Rhamnose	–	0		+	95.4	(1.1)
Malonate	–	1.6		+ or –	67.2	
Lipase	+	98.4	(0.7)	–	0	
Xylose	d	7.1	(17.2)	+	97.6	
Gas from cellobiose	–	0		d	67	(22)
Gas from glycerol	–	0		+	95	
Esculin	d	72.7	(0.2)	–	6	(2)
DNase	+	97.2		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).
[a]Volumes of gas produced are small (bubble to 5 or 10%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 48

Differentiation of *P. vulgaris* and *P. mirabilis* from *P. morganii* and *P. rettgeri*

Test or Substrate	*P. vulgaris* and *P. mirabilis*			*P. morganii* and *P. rettgeri*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Hydrogen sulfide (TSI)	+	94.5		–	0	
Gelatin (22 C)	+	91.6	(6.4)	–	0	(2)[a]
Lipase (corn oil)	+ or (+)	89.6	(5.2)	–	0	
Mannose	–	0		+	100	
Swarm (2% agar)	+	94	(1)	–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Weakly positive after 30 to 35 days.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 49

Differentiation of *P. vulgaris* and *P. mirabilis*

Test or Substrate	*P. vulgaris* Sign	%+	(%+)*	*P. mirabilis* Sign	%+	(%+)*
Indol	+	91.4		–	3.2	
Voges-Proskauer (37 C)	–	0		– or +	15.6	
(22 C)	– or +	11.3		+ or –	51.6	
Citrate (Simmons')	d	10.5	(14.1)	+ or (+)	58.7	(37.1)
Ornithine decarboxylase	–	0		+	98.4	
Sucrose	+	94.7		d	18.9	(63.3)
Maltose	+	96.2	(1.9)	–	0.9	(0.4)
Salicin	d	58.2	(10.9)	d	0.8	(29.8)
Alpha methyl glucoside	d	79.5	(5.1)	–	0	
Esculin	d	59	(2.6)	–	0	(0.9)
DNase[a]	+ or –	50		–	0	

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Small number of strains tested (63).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 50

Differentiation of *P. morganii* and *P. rettgeri*

Test or Substrate	*P. morganii*			*P. rettgeri*		
	Sign	%+	(%+)*	Sign	%+	(%+)*
Citrate (Simmons')	–	0		+	95.6	(3.3)
Ornithine decarboxylase	+	95.7		–	0	
Gas from glucose	d	84.9	(0.9)[a]	– or +	12.2[a]	
Sucrose	–	1	(2.9)	d	13.3	(56.7)
Mannitol	–	0		+ or –	88.5	
Adonitol	–	0		d	80.9	(5.6)
Inositol	–	0		+	93.3	(4.5)
Salicin	–	0		d	30	(6.6)
Erythritol	–	0		d	78.3	(6.5)
Esculin	–	0		d	30.4	(8.7)
Xylose	–	0		– or +	15.1	
Cellibiose	–	0	(1.9)	d	3.7	(30.4)

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 10 or 15%).

Note: An occasional strain of either species may fail to form indol.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 51

Differentiation of *P. morganii* and *P. rettgeri* from *P. alcalifaciens* and *P. stuartii*

Test or Substrate	*P. morganii*			*P. rettgeri*			*P. alcalifaciens*			*P. stuartii*		
	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*	Sign	%+	(%+)*
Urease	+	97.1	(0.5)	+	100		–	0		–	0	
Ornithine decarboxylase	+	95.7		–	0		–	1.2		–	0	
Gas from glucose	d	84.9	(0.9)[a]	– or +	12.2[a]		d	85.2	(0.6)[a]	–	0	
Mannitol	–	0		+ or –	88.5		–	1.9	(0.1)	d	11.8	(9.1)
Adonitol	–	0		d	80.9	(5.6)	+	94.3	(0.3)	– or +	12.4	
Inositol	–	0		+	93.3	(4.5)	–	0.6		+	97.2	(2.8)
Erythritol	–	0		d	78.3	(6.5)	–	0		–	2.3	
Esculin	–	0		d	30.4	(8.7)	–	0		–	0	
Cellobiose	–	0	(0.9)	d	3.7	(30.4)	–	1.4	(1.8)	d	10.4	(45.9)

*Figures in parentheses indicate percentages of delayed reactions (3 or more days).

[a]Volumes of gas produced are small (bubble to 10 or 15%).

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 52

The decarboxylase reactions of ENTEROBACTERIACEAE

Species	Number	Lysine			Arginine			Ornithine		
		Sign	%+[a]	(%+)[b]	Sign	%+[a]	(%+)[b]	Sign	%+[a]	(%+)[b]
E. coli	872	d	80.6	(1.5)	d	16.3	(39.1)	d	57.8	(8)
A-D[c]	618	d	73.6	(8.1)	d	6.8	(42.9)	d	12	(3.4)
S. dysenteriae	400	–	0		d	1.5	(11.3)	–	0	
S. flexneri	1817	–	0		–	7.9	(1.6)	–	0	
S. boydii	400	–	0		d	18.1	(31.9)	–	2.5[d]	
S. sonnei	633	–	0		–	0.5	(5)	+	99.4	
E. tarda	494	+	100		–	0	(0.2)	+	99.7	(0.2)
S. cholerae-suis	20	+	95		(+)	0	(95)	+	100	
S. typhi	288	+	99.7		– or (+)	0	(22.2)	–	0	
S. enteritidis	995	+	94.9		+ or (+)	65.4	(27.4)	+	96.7	
A. hinshawii	315	+	99.4	(0.3)	(+) or +	25.1	(73.6)	+	100	
C. freundii	582	–	0		d	44.8	(45)	d	12.5	(0.2)
C. diversus	226	–	0		+ or (+)	62.4	(35)	+	99.6	
K. pneumoniae	3560	+	98.2	(0.1)	–	0.2[w]		–	0	
K. ozaenae	256	d	35.8	(6.3)	–	4.2	(4.2)	–	1	
K. rhinoschleromatis	50	–	0		–	0		–	0	
E. cloacae	460	–	0		+	92.4	(2)	+	93.7	(1.3)
E. aerogenes	121	+	97.5		–	0		+	95.9	(0.8)
E. hafniae	286	+	99.6		d	4.6	(6.6)	+	98.6	
E. agglomerans	536	–	0		–	0		–	0	
S. marcescens	1402	+	99.6		–	(0.9)		+	99.6	
S. liquefaciens	109	+ or (+)	64.2	(31.2)	–	0		+	100	
S. rubidaea	49	+ or (+)	61	(31)	–	0		–	0	
P. vulgaris	70	–	0		–	0		–	0	
P. mirabilis	372	–	0		–	0		+	98.4	
P. morganii	208	–	0	(1.5[w])	–	0		+	95.7	
P. rettgeri	170	–	0		–	0		–	0	
P. alcalifaciens	249	–	0	(0.8[w])	–	0	(0.4[w])	–	1.2[w]	
P. stuartii	162	–	0	(0.6[w])	–	0		–	0	
Erwinia spp.	16	–	0		–	0		–	0	
Pectobacterium spp.	95	–	0		–	0	(9.5)	–	0	

[a]Percent positive in 1 or 2 days.

[b]Percent positive in 3 or 4 days.

[c]A-D = Alkalescens-Dispar (bioserotypes of *E. coli*).

[d]These few positives all were *S. boydii* 13.

KEY:

+	90% or more positive within 1 or 2 days.
(+)	positive reaction after 3 or more days (decarboxylase tests: 3 or 4 days).
–	no reaction (90% or more).
+ or –	most cultures positive, some strains negative.
– or +	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed.
d	different reactions, +, (+), –.
w	weakly positive reaction.

Table 5part1

The identification of ENTEROBACTERIACEAE

Species	Number	Lysine			Arginine			Ornithine		
		Sign	%[a]	(24h)[b]	Sign	%[a]	(24h)[b]	Sign	%[a]	(24h)[b]
Aerobacter	852	+	50.0	(1.5)	d	16.7	(29.1)	+	72.8	(8)
A.liq.	815	d	72.0	(8.1)	d	5.9	(42.9)	d	12.5	(24.4)
Chromobacterium	460	+	0		+	1.4	(11.3)	-	0	
Cit.divernex	812	-	0		-	2.9	(1.0)	-	0	
Hafnia	480	+	0		d	15.1	(31.3)	-	1.5	
Kleb.oxytoca	473	+	0		+	0.5	(5)	+	99.3	
Kleb.pneum.	494	+	100		-	0.0	(0.2)	+	99.7	(0.2)
Providencia	20	+	95		(+)	0	(95)	+	100	
Proteus mir.	288	+	96.7		-	0	(22.2)	-	0	
Proteus vulg.	295	+	0.3		+	95.4	(22.4)	+	99.7	
Ps.aeruginosa	315	+	99.4	(0.3)	(+) or +	25.1	(73.6)	+	100	
Serratia	582	-	0		d	44.8	(43.9)	d	12.5	(0.2)
Shig.	220	-	0		+ or (+)	0.4	(35)	+	99.6	
Sp.paucimobilis	450	-	0.2	(0.1)		0.2		-	0	
Yersinia	250	d	25.4	(6.8)	-	2	(4.2)	-	1	
Citrobacter diver.	50	-	0		-	0		-	0	
Enterob.cloac.	440	-	0		+	92.4	(2)	+	93.9	(1.3)
Escherichia	171	+	97.5		-	0		+	95.9	(0.5)
Hafnia alvei	260	+	99.6		d	4.6	(8.6)	+	99.6	
Klebsiella	536	-	0		-	0		-	0	
Proteus rettgeri	1402	+	99.6		-	(0.9)		+	99.6	
Shigella dysen.	109	+ or (+)	94.2	(31.2)	-	0		-	0	
Shigella flexn.	40	+ or (-)	51	(6.1)	-	0		-	0	
Shigella sonn.	70	-	0		-	0		-	0	
Yersinia ent.	272	-	0		-	0		+	98.4	
Yersinia pseud.	208	-	0	(1.5[w])	-	0		+	95.7	
Yersinia sp.	170	-	0		-	0		-	0	
Salmonella	249	-	0	(0.8[w])	-	0	(0.4[w])	-	1.2[w]	
Serratia	162	-	0	(0.5[w])	-	0		-	0	
Providencia sp.	16	-	0		-	0		-	0	
Aeromonas - Plesiomonas	95	-	0		-	0	(0.5)	-	0	

Percent positive in 1 or 2 days

Percent positive in 3 or 4 days

A.D. = Alkalescens-Dispar (*Bordetella* spp. or *B. coli*).

[a] These new positives will serve to lower [illegible].

KEY:

	90% or more positive within 1 or 2 days
(+)	positive reaction after 3 or more days (decarboxylase tests 3 or 4 days)
	no reaction (90% or more)
(-) or -	most cultures positive, some strains negative.
+ or (+)	most strains negative, some cultures positive.
+ or (+)	most reactions occur within 1 or 2 days, some are delayed
d	different reactions, (+), -, ...
v	weakly positive reaction.