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The Role of Bacterial Decarboxylases as a Possible Tool for Chemotaxonomic Characterization

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ABSTRACT. An attempt to differentiate 40 members of bacteria, belonging mostly to the family Enterobacteriaceae, on the basis of their capacity to decarboxylate 24 different amino acids has been carried out.

The results showed that the majority of the bacteria were able to decarboxylate threonine (29 members), ornithine (25 members), lysine (24 members), histidine (22 members) and arginine (21 members). Very few members were able to decarboxylate alanine, aspartic acid, or methionine (4 members for each), cystine, glycine or leucine (3 members for each), hydroxyproline, iso-leucine, nor-leucine, proline, tryptophan or valine (2 members for each). Only one bacterium decarboxylated tyrosine while none could decarboxylate dihydroxyphenyl alanine.

Within the limits of these results, a preliminary key is proposed for the identification of each or a small group of closely related species and/or strains of one genus.

The Enterobacteriaceae is a large family comprising many interrelated types displaying every conceivable combination of biochemical characteristics compatible with the definition of the family.

Identification of members of the Enterobacteriaceae is commonly based on diagnostic schemes (Kauffman 1954, and Breed *et al.* 1957, Ewing and Edwards 1960, Cowan and Steel 1965, and Ewing 1973) using a range of conventional tests.

Ernst (1982) suggested a two stage determination for the differentiation of the Enterobacteriaceae. In the first stage, 7 reactions are performed (formation of gas from dextrose, production of acid from lactose, formation of hydrogen sulphide, urease activity, indole formation, motility and lysine decarboxylation). The resulting pattern of reactions produces a code number. If more than one species

falls in one code number, then it is necessary to carry out further tests (stage two).

The lysine decarboxylase test for the Enterobacteriaceae is considered of great taxonomic value and is used for the identification and characterization of the Enterobacteriaceae (Möller 1955), since it can differentiate *Citrobacter* (lysine negative) from *Salmonella* and *Arizona* strains (lysine positive) (Davis *et al.* 1960, and Stroup 1974) as well as identify new members of the Enterobacteriaceae found in clinical specimens (Rohde *et al.* 1975, Sakazaki *et al.* 1976, Farmer *et al.* 1981, and Brenner *et al.* 1982).

The aim of this investigation is to find out whether the presence or absence of different types of amino acid decarboxylases in selected members of the Enterobacteriaceae as well as other pathogenic bacterial genera, could serve as a differential character for identifying such bacteria.

Material and Methods

The organisms used in this investigation were a selection of the most common clinical isolates from patients suffering from bilharzia and receiving medical treatment in the Institute of Research for Tropical Medicine, Cairo. These were chemically and serologically characterized using conventional methods (Edwards and Ewing 1972, Tatum *et al.* 1974, Buchanan and Gibbons 1974, Hickman and Farmer 1978 and Hickman *et al.* 1980).

These bacteria as well as authenticated specimens (kindly provided by the Hygiene Institute, National Salmonella Center; Hamburg) together with two plant pathogens (Saleh 1977) were used in the determination of the various amino acid decarboxylases.

Detection of Amino Acid Decarboxylases

Preliminary investigations, using one dimensional ascending paper chromatography to identify the formation of one or several decarboxylation products of the different amino acids (Smith 1969, and Goldschmidt and Lockhart 1971) showed that:

1. The triple sugar iron (T S I) medium was a better medium for efficient initiation of decarboxylases during the 24 hours incubation to produce the experimental biomass.

2. Impurities in the inoculum interfered with the decarboxylase activity and/or metabolism of the organisms leading to the appearance of several unwanted ninhydrin - positive spots. This might be the reason for the superiority of the basal

salt medium (inorganic nitrogen medium) for running the decarboxylase test over the Falkow (1958) medium or modified Möller medium (organic nitrogen medium), Saleh (1977).

3. Two hours incubation is not enough for the detection of the decarboxylation products especially for the slow growing organisms.

4. The presence or absence of ammonium sulphate does not significantly affect the results.

5. Increased size of the inoculum misleads the results especially with long term incubation periods, required for the slow organisms.

6. Low concentrations of the amino acids results in very faint spots even at 0.1-0.3% and two hours incubation.

Accordingly, screening for amino acid decarboxylases was carried out using inocula prepared from several times washed biomass, grown, for 24 hours, on T S I medium. 0.5 ml of this saline suspension was mixed with 1 ml basal salt medium amended with 0.5% of the tested amino acid. The mixture was incubated for 4 hours followed by chromatography of the produced amines.

Results and Discussion

Forty bacteria including isolates from bilharzia patients (*Citrobacter* sp.; *Pseudomonas* sp.; *Salmonella montevideo; S. paratyphi* B; *S. typhimurium; Klebsiella* sp.; *Proteus morganii; P. vulgaris; Shigella boydii; S. dysenteriae* 8; *S. dysenteriae* 10; *S. flexneri* 1; *S. flexneri* 6; *S. sonnei*) as well as representative members of the culture collection donated by the National Salmonella Center, namely *Arizona* 30:32-25; *Ar.* (5), 29:33-21; *Ar.* 6:13:14; *Ar.* 20:29-25; *Citrobacter* 048_1, 48_3, 48_4; *Escherichia coli* DM 3219; *E. coli* K12; *E. coli* 018; *Salmonella anatum; S. cilbek; S. dar-es-salaam; S. enteritidis; S. farmsen; S. kralendyk; S. newport; S. offa; S. pomona; S. saint-paul; S. sofia; S. wassenaar; S. wayne* and *Yersinia* sp., together with two species of *Erwinia* and two species of *Vibrio,* were tested for their amino acid decarboxylation activities.

Table 1 shows that the majority of the organisms were able to decarboxylate threonine (29 members); ornithine (25 members); lysine (24 members); histidine (22 members) and arginine (21 members). Very few members were able to decarboxylate alanine, aspartic acid or methionine (4 members for each); cystine, glycine or leucine (3 members for each); dihydroxyproline, iso - leucine, nor - leucine, proline, tryptophan or valine (2 members for each). Only one member was able to decarboxylate tyrosine and none were able to decarboxylate hydroxyphenyl alanine.

The four strains of Arizona could be differentiated from each other by the following:

i. *Arizona* 30:32-25 was the only strain decarboxylating arginine.
ii. *Arizona* (5), 29:33-21 was the only strain decarboxylating cysteine.
iii. *Arizona* 6: 13, 14:- decarboxylated α-aminobutyric acid but not threonine.
iv. *Arizona* 20: 29-25 decarboxylated threonine but not α-aminobutyric acid.

Erwinia carotovora v. *citrullis* was the sole organism decarboxylating tyrosine. *E. coli* DM 3219 could be differentiated from the other two strains by the ability to decarboxylate lysine and ornithine. *E. coli* K12 and *E. coli* 018 had similar decarboxylating activity. *Proteus morganii* could be differentiated from *P. vulgaris* by the ability to decarboxylate ornithine.

The following decarboxylation criteria could primarily differentiate the tested seventeen *Salmonella* organisms from each other:

i. Cysteine decarboxylase appeared only in *S. wayne*.
ii. Phenylalanine decarboxylase appeared only in *S. kralendyk*.
iii. Both *S. saint-paul* and *S. wayne* decarboxylate proline.
iv. *S. anatum; S. enteritidis; S. farmsen; S. pomona; S. typhimurium;* and *S. wassenaar* can decarboxylate arginine, histidine, lysine, ornithine, serine and threonine.
v. *S. sofia* and *S. typhi* did not have arginine and histidine decarboxylases. The former did not have lysine decarboxylase while the latter did not have arginine and serine decarboxylases.
vi. *S. montevideo, S. newport, S. offa* and *S. paratyphi* B did not have serine decarboxylase but did have the other five decarboxylases except *S. offa* that lacked arginine decarboxylase.
vii. *S. cilbek* and *S. dar-es-salaam* did not have serine decarboxylase but did have histidine and lysine decarboxylase. *S. cilbek* lacked also arginine and ornithine decarboxylase whereas *S. dar-es-salaam* lacked threonine decarboxylase.

The six species of *Shigella* could be differentiated from each other by the following criteria:

i. *S. flexneri* 6 was the only species possessing alanine decarboxylase activity.
ii. *S. sonnei* was the only species decarboxylating aspartic acid and not serine.
iii. *S. boydii* was the only species sharing *S. flexneri* 6 in decarboxylating α-aminobutyric acid.

iv. *S. flexneri* 1 differed from *S. dysenteriae* 8 and *S. dysenteriae* 10 by the absence of arginine decarboxylase and the presence of histidine decarboxylase.

v. *S. dysenteriae* 8 lacked ornithine decarboxylase, but this enzyme was detected in *S. dysenteriae* 10 and *S. flexneri* 1.

Vibrio inaba could be differentiated from *V. ogawa* by the presence of α-aminobutyric acid and ornithine decarboxylases as well as by the absence of glutamic acid decarboxylase.

Apart from these observations, all organisms produced one ninhydrin - positive compound from each decarboxylated amino acid except arginine, cysteine, histidine and lysine where two components were observed. A few *Salmonella* and *Shigella* species gave two compounds from threonine, namely *Salmonella typhimurium* and *S. wassenaar* (at R_f 0.17 and 0.44) and *Shigella boydii*, *S. dysenteriae* 8 and *S. dysenteriae* 10 (at R_f 0.17 and 0.65).

Similarly, all bacteria decarboxylating α-aminobutyric acid gave one spot at R_f 0.68 except *Arizona* 6: 13, 14:- where a further ninhydrin - positive spot was observed at R_f 0.50.

In contrast, all decarboxylation of cysteine produced two spots except *Arizona* 20: 29-25, and *Klebsiella* that gave only one spot at the higher R_f (0.65). Similarly, all arginine decarboxylation showed two spots except the strains of *E. coli* and *Yersinia* that showed one spot at the higher R_f (0.26).

Shigella flexneri 6 produced an amine from alanine at a lower R_f (0.46) than by the three strains of *E. coli* (0.58). Similarly, the two *Proteus* species differed from the other four organisms decarboxylating phenylalanine in giving a product at a lower R_f (0.29 compared to 0.39). The decarboxylation product of methionine, by *Yersinia*, was at a lower R_f (0.21) than that produced by the *E. coli* strains (R_f 0.36). The product of ornithine decarboxylation, by *Shigella boydii* and *S. dysenteriae* 10, *S. flexneri* 1, was located at a higher R_f (0.53) than that of the rest of the positive organisms (R_f 0.15).

It is possible that the degradation products might help in the identification of the organisms. The strains of *E. coli* differed from *Shigella flexneri* 6 in utilizing the ethylamine produced as the first decarboxylation product of α-alanine. They produced a compound at a higher R_f (0.58 compared with 0.46). Similarly, the R_f of the proline decarboxylation product of *Salmonella saint paul* was lower than produced by *Salmonella wayne* (0.18 compared with 0.58). All organisms decarboxylating arginine produced two component amines except the three strains of *E. coli* as well as *Yersinia*. These produced only one compound at the higher R_f (0.26). This is further repeated with cysteine decarboxylase where *Arizona* (5) 29:

33-21 and *Klebsiella* produced one amine at R_f 0.48 whereas the rest of the decarboxylation had another compound (at R_f 0.65).

Furthermore, putrescine (the decarboxylation product of ornithine) seemed to be further metabolized by *Shigella boydii, S. dysenteriae* 10 and *S. flexneri* 1 (R_f 0.53). The isopropanol amine (formed by ornithine decarboxylase) seemed to be further metabolised by *Salmonella typhimurium* and *S. wassenaar* to another metabolite (R_f 0.43) that differed from that produced by *Shigella boydii, S. dysenteriae* 8 and *S. dysenteriae* 10. Again, *Yersinia* differed from the three strains of *E. coli* in decarboxylating methionine to a product of a lower R_f (0.21 compared to 0.36).

It is interesting to note that histidine and lysine formed similar decarboxylation products whereas threonine shared in only one component. This leads to the suggestion that the three amino acids shared the isopropanol amine (the first decarboxylation product of threonine). This could be derived from histamine (the first decarboxylation product of histidine) through the opening of the imidazole ring followed by cleavage to ethylamine and isopropanol amine. Cleavage of cadaverine (the first decarboxylation product of lysine) followed by hydroxylation may also lead to the formation of these two components.

Within the limits of our results, it seems possible to deduce a preliminary key for the identification of each or a small group of closely related species and/or strains of one genus. The latter could be further differentiated by other complementary reactions whether biochemical or serological.

It is interesting to note that arginine metabolism is not only useful as a tool to differentiate *Pseudomonas* from the other Gram-negative bacteria (King and Philips 1978) but also valuable in differentiating *Shigella dysenteriae* from *Shigella flexneri* (present results). Similarly, ornithine decarboxylase is not only a criterion to differentiate between *E. coli* 01: K1: H7: F11 and 01: K1: H−: F11 that were positive decarboxylators (Nimmish and Zingler 1984) and the serotypes 01: K1: H−: F9 and 01: K1: H− F− that were negative decarboxylators, but also can be used to differentiate between *Shigella dysenteriae* 8 and *Shigella dysenteriae* 10 (negative for the former and positive for the latter).

The present results further lend support to the observations of Finichiu (1984) regarding lysine decarboxylase activity since we observed that *Salmonella sofia* was the only bacterium able to decarboxylate lysine among the seventeen tested species.

It is worth noting that hydroxy− (highly polar electro-negative radical) substitution of alanine [serine] or the methyl substitution (highly nonpolar

electro-positive radical) of alanine [α-amino butyric acid] initiated the induction of decarboxylases of these amino acids in larger numbers of organisms (17 and 13 members compared to 4, in case of alanine). Coupling both substitutions [threonine] had an additive effect. On the other hand, substitution by sulphydryl (moderately polar electro-negative radical) or a benzene ring (aromatic non polar radical) had a slight stimulatory effect [7 members for cysteine and 8 members for phenylalanine]. Introduction of highly polar groups to the benzene ring arrested the potential of the amino acid for decarboxylase formation [one member for tyrosine and none for dihydroxyphenyl alanine]. These observations might be attributed to the inductive effect, hyper conjugation and mesomeric effects; the basic parameters of the effect of substitutions on conjugation (Williams and Fleming 1973).

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Table 1. The optical density of the decarboxylation products formed during the detection of decarboxylases of selected genera and species of Enterobacteriaceae and other pathogenic bacteria isolated from patients or plants.

(O.D. per 10^7 cells of each organism in 1.5 ml reaction mixture)

Alanine Decarboxylase

Organism	At R_f 0.46	0.58
E.coli DM 3219	–	2.5
E.coli K12	–	1.5
E.coli 018	–	2.0
Shigella flexneri 6	1.5	–

Aspartic decarboxylase

Organism	At R_f 0.23
E.coli DM 3219	3.1
E.coli K 12	1.3
E.coli 018	2.9
Shigella sonnei	2.1

Glutamic decarboxylase

Organism	At R_f 0.37
E.coli DM 3219	10.3
E.coli K 12	17.8
E.coli 018	15.0
Klebsiella	20.2
Proteus morganii	15.1
Proteus vulgaris	15.0
Shigella boydii	11.3
Shigella dysenteriae 8	10.0
Shigella dysenteriae 10	10.2
Shigella flexneri 1	11.4
Shigella flexneri 6	14.9
Shigella sonnei	11.3
Vibrio ogawa	0.5
Dihydroxyphenylalanine	None

α-amino butyric decarboxylase

Organism	At R_f 0.50	0.68
Arizona 30:32-25	–	1.5
Arizona 6:13,14:-	1.5	1.0
Erwinia carotovora v.citrullis	–	2.6
Erwinia toxica	–	2.1
E.coli DM 3219	–	3.0
E.coli K12	–	1.9
E.coli 018	–	2.8
Proteus morganii	–	2.1
Proteus vulgaris	–	1.8
Shigella boydii	–	2.0
Shigella flexneri 6	–	17.4
Vibro inaba	–	1.1
Yersinia	–	1.6

Cysteine decarboxylase

Organism	At R_f 0.48	0.65
Arizona (5),29:33-21	2.5	–
E.coli DM 3219	4.6	2.4
E.coli K12	3.0	1.5
E.coli 018	2.1	0.9
Klebsiella	1.3	–
Salmonella wayne	3.0	2.3
Yersinia	3.1	1.5

Cystine decarboxylase

Organism	At R_f 0.12
E.coli DM 3219	0.5
E.coli K 12	1.0
E.coli 018	0.6

Contd.

Arginine decarboxylase

Organism	At R_f	
	0.08	**0.26**
Arizona 32:32-25	7.5	3.6
Citrobacter $048_1,48_3,48_4$	2.4	3.5
E.coli DM 3219	–	4.7
E.coli K12	–	2.5
E.coli 018	–	5.1
Salmonella anatum	4.9	3.4
Salmonella dar-es-salaam	13.8	3.7
Salmonella enteritidis	10.0	3.1
Salmonella farmsen	12.7	3.4
Salmonella montevideo	9.9	3.0
Salmonella newport	13.8	4.0
Salmonella paratyphi B	11.3	2.6
Salmonella pomona	10.1	2.9
Salmonella typhimurium	11.3	4.7
Salmonella wassenaar	12.5	1.9
Shigella dysenteriae 8	12.7	3.6
Shigella dysenteriae 10	10.1	2.8
Shigella sonnei	12.6	1.9
Vibrio inaba	15.0	2.6
Vibrio ogawa	15.1	4.9
Yersinia	–	2.7

Phenylalanine decarboxylase

Organism	At R_f	
	0.29	**0.39**
E.coli K12	–	1.3
E.coli 018	–	1.8
Proteus morganii	1.5	–
Proteus vulgaris	1.0	–
Salmonella kralendyk	–	1.0
Shigella boydii	–	1.0

Glycine decarboxylase

Organism	At R_f
	0.30
E.coli DM 3129	1.5
E.coli K 12	1.0
E.coli 018	0.5

Histidine decarboxylase

Organism	At R_f	
	0.05	**0.17**
Arizona 30:32-25	7.5	3.5
Arizona 6:13,14:-	7.5	3.0
Arizona 20:29-25	5.0	4.0
Citrobacter $048_1,48_3,48_4$	15.0	3.5
Salmonella anatum	10.1	2.9
Salmonella cilbek	15.0	2.0
Salmonella dar-es-salaam	13.8	4.5
Salmonella enteritidis	15.1	3.6
Salmonella farmsen	12.5	3.0
Salmonella kralendyk	12.5	3.0
Salmonella montevideo	10.0	3.5
Salmonella newport	12.4	4.1
Salmonella offa	8.8	4.0
Salmonella paratyphi B	12.6	3.1
Salmonella pomona	13.8	4.0
Salmonella st. paul	10.1	2.3
Salmonella typhimurium	11.3	3.1
Salmonella wassenaar	11.3	3.6
Shigella flexneri 1	13.8	3.3
Shigella flexneri 6	12.5	4.0
Vibrio inaba	12.6	2.5
Vibrio ogawa	17.5	5.1

Contd.

Hydroxy-proline decarboxylase

Organism	At R_f
	0.52
E.coli K 12	2.0
E.coli 018	1.8

Ornithine decarboxylase

Organism	At R_f	
	0.15	**0.53**
Arizona 30:32-25	5.0	–
Arizona 6:13,14:-	5.1	–
Arizona (5),29:33,21	4.9	–
Arizona 20:29-25	4.0	–
E. coli DM 3219	3.8	–
Proteus morganii	5.0	–
Salmonella anatum	4.9	–
Salmonella dar-es-salaam	3.8	–
Salmonella enteritidis	5.0	–
Salmonella farmsen	3.8	–
Salmonella kralendyk	6.3	–
Salmonella montevideo	3.7	–
Salmonella newport	5.0	–
Salmonella offa	3.8	–
Salmonella paratyphi B	0.8	–
Salmonella pomona	3.7	–
Salmonella sofia	6.3	–
Salmonella typhimurium	5.0	–
Salmonella wassenaar	4.9	–
Salmonella wayne	5.1	–
Shigella boydii	–	1.0
Shigella dysenteriae 10	–	1.0
Shigella flexneri 1	–	1.5
Shigella sonnei	7.5	–
Vibrio inaba	7.5	–

Proline decarboxylase

Organism	At R_f	
	0.17	**0.58**
Salmonella st.paul	2.0	–
Salmonella wayne	–	1.5

Leucine decarboxylase

Organism	At R_f
	0.45
E.coli DM3219	0.8
E.coli K12	0.8
E.coli 018	0.8

Lysine decarboxylase

Organism	At R_f	
	0.05	**0.17**
Arizona 30:32-25	11.3	3.0
Arizona 6:13,14:-	11.0	2.9
Arizona (5),29:33,21	10.1	2.5
Arizona 20:29-25	7.6	3.1
E.coli DM 3219	7.5	2.6
Klebsiella	10.0	4.0
Salmonella anatum	11.3	3.9
Salmonella cilbek	15.0	3.5
Salmonella dar-es-salaam	14.9	4.1
Salmonella enteritidis	1.5	3.5
Salmonella farmsen	12.5	3.0
Salmonella kralendyk	12.5	3.0
Salmonella montevideo	11.3	2.1
Salmonella newport	12.5	3.3
Salmonella offa	15.1	3.5
Salmonella paratyphi B	10.0	3.1
Salmonella pomona	15.0	4.1
Salmonella st.paul	11.3	3.5
Salmonella typhi	10.1	2.9
Salmonella typhimurium	15.1	4.6
Salmonella wassenaar	10.0	3.0
Salmonella wayne	10.1	3.0
Vibrio inaba	12.5	3.8
Vibrio ogawa	15.0	2.5

Tryptophan decarboxylase

Organism	At R_f
	0.29
Proteus morganii	2.5
Proteus vulgaris	1.0

Contd.

Iso-leucine decarboxylase

Organism	At R_f 0.36
E.coli K12	0.5
E.coli 018	0.8

Tyrosine decarboxylase

Organism	At R_f 0.3
Erwinia carotovora-V.citrullis	3.0

Serine decarboxylase

Organism	At R_f 0.34
E.coli DM3219	1.5
E.coli K12	0.8
E.coli 018	2.0
Salmonella anatum	0.8
Salmonella enteritidis	1.3
Salmonella farmsen	1.5
Salmonella kralendyk	1.0
Salmonella pomona	1.5
Salmonella st.paul	1.3
Salmonella sofia	1.5
Salmonella typhimurium	1.6
Salmonella wassenaar	1.0
Shigella boydii	1.8
Shigella dysenteriae 8	1.5
Shigella dysenteriae 10	1.5
Shigella flexneri 1	1.3
Shigella flexneri 6	1.0

Methionine decarboxylase

Organism	At R_f 0.21	At R_f 0.36
E.coli DM 3219	–	1.5
E.coli K12	–	1.6
E.coli K018	–	1.8
Yersinia	1.25	–

n-Leucine decarboxylase

Organism	At R_f 0.44
E.coli K12	0.8
E.coli 018	1.0

Contd.

Threonine decarboxylase

Organism	At R_f			
	0.17	**0.43**	**0.45**	**0.65**
Arizona 30:32-25	7.5	–	–	–
Arizona 20:29-25	11.3	–	–	–
Citrobacter $048_1,48_3,48_4$	7.6	–	–	–
Erwinia carotovora V.citrullis	8.8	–	–	–
Erwinia toxica	8.8	–	–	–
E. coli DM 3219	8.7	–	–	–
E. coli K12	5.0	–	–	–
E. coli 018	4.9	–	–	–
Proteus morganii	5.2	–	–	–
Proteus vulgaris	7.5	–	–	–
Salmonella anatum	6.3	–	–	–
Salmonella cilbek	5.0	–	–	–
Salmonella enteritidis	7.6	–	–	–
Salmonella farmsen	6.3	–	–	–
Salmonella kralendyk	5.1	–	–	–
Salmonella montevideo	6.3	–	–	–
Salmonella newport	5.0	–	–	–
Salmonella offa	7.5	–	–	–
Salmonella paratyphi B	7.6	–	–	–
Salmonella pomona	6.2	–	–	–
Salmonella st.paul	7.6	–	–	–
Salmonella sofia	7.5	–	–	–
Salmonella typhi	6.3	–	–	–
Salmonella typhimurium	5.0	–	2.0	–
Salmonella wassenaar	5.1	1.5	–	–
Shigella boydii	5.0	–	–	1.5
Shigella dysenteriae 8	5.0	–	–	1.0
Shigella dysenteriae 10	7.5	–	–	1.0
Shigella flexneri 1	5.1	–	–	–

Valine decarboxylase

Organism	At R_f
	0.27
Proteus morganii	4.0
Proteus vulgaris	1.0

Scheme for Identification of Members of Enterobacteriaceae and Other Pathogenic Genera Based on Their Amino Acid Decarboxylase Activity

1. **Tyrosine** decarboxylase

A.	**Positive**	B.	**Negative**
	Erwinia carotovora V. citrullis		(39 members)

2. **Tryptophan or Valine** decarboxylase (for 1.B.)

A.	**Positive**	B.	**Negative**
	Proteus morganii *Proteus vulgaris*		(37 members)

A.1. Ornithine decarboxylase

Positive	**Negative**
Proteus morganii	*Proteus vulgaris*

3. **Proline** decarboxylase (for 2.B.)

A.	**Positive**	B.	**Negative**
	Salmonella saint paul *Salmonella wayne*		(35 members)

A.1. Histidine decarboxylase

Positive	**Negative**
Salmonella saint paul	*Salmonella wayne*

A.2. Ornithine decarboxylase

Positive	**Negative**
Salmonella wayne	*Salmonella saint paul*

4. **Alanine** decarboxylase (for 3.B.)

A.	**Positive**	B.	**Negative**
	Escherichia coli DM 3219 *E. coli* K12 *E. coli* 018 *Shigella flexneri* 6		(31 members)

A.1. Hydroxyproline, iso-leucine or nor-leucine decarboxylase

A.1.a.	**Positive**	A.1.b.	**Negative**
	E.coli K12 *E.coli* 018		*E.coli* DM 3219 *Shigella flexneri* 6

A.1.b.1. Lysine or ornithine decarboxylase

Positive	**Negative**
E.coli DM 3219	*Shigella flexneri* 6

5. **Cysteine** decarboxylase (for 4.B.)

A.	**Positive**	B.	**Negative**
	Arizona (5),29:33-21 *Klebsiella* *Yersinia*		(28 members)

A.1. Methionine decarboxylase

Positive	**Negative**
Yersinia	*Arizona* (5),29:33-21 *Klebsiella*

A.2. Glutamic decarboxylase

Positive	**Negative**
Klebsiella	*Arizona* (5), 29:33-21

A.3. Ornithine decarboxylase

Positive	**Negative**
Arizona (5), 29:33-21	*Klebsiella*

6. α-**Aminobutyric Acid** decarboxylase (for 5.B.)

A. **Positive**	B. **Negative**
Arizona 30:32-25 *Arizona* 6:13-14:- *Erwinia toxica* *Shigella boydii* *Vibrio inaba*	(23 members)

A.1. Arginine decarboxylase

A.1.a. **Positive**	A.1.b. Negative
Arizona 30: 32-25 *Vibrio inaba*	*Arizona* 6:13-14:- *Erwinia toxica* *Shigella boydii*

A.1.a.1. Threonine decarboxylase

Positive	**Negative**
Arizona 30:32-25	*Vibrio inaba*

A.1.b.1. Glutamic decarboxylase

Positive	**Negative**
Shigella boydii	*Arizona* 6:13-14:- *Erwinia toxica*

A.1.b.2. Histidine decarboxylase

Positive	**Negative**
Arizona 6:13-14:-	*Erwinia toxica*

7. **Glutamic** decarboxylase (for 6.B.)

A. **Positive**	B. **Negative**
Shigella dysenteriae 8 *Shigella dysenteriae* 10 *Shigella flexneri* 1 *Shigella sonnei* *Vibrio ogawa*	(18 members)

A.1. Aspartic acid decarboxylase

Positive	Negative
Shigella sonnei	*Shigella dysenteriae* 8
	Shigella dysenteriae 10
	Shigella flexneri 1
	Vibrio ogawa

A.2. Arginine decarboxylase

Positive	Negative
Shigella dysenteriae 8	*Shigella flexneri* 1
Shigella dysenteriae 10	
Vibrio ogawa	

A.3. Histidine decarboxylase

Positive	Negative
Vibrio ogawa	*Shigella dysenteriae* 8
	Shigella dysenteriae 10

A.4. Ornithine decarboxylase

Positive	Negative
Shigella dysenteriae 10	*Shigella dysenteriae* 8

8. **Serine** decarboxylase (for 7.B.)

A. Positive	B. Negative
Salmonella anatum	(10 members)
Salmonella enteritidis	
Salmonella farmsen	
Salmonella kralendyk	
Salmonella pomona	
Salmonella sofia	
Salmonella typhimurium	
Salmonella wassenaar	

A.1. Phenylalanine decarboxylase

Positive	Negative
Salmonella kralendyk	*Salmonella anatum*
	Salmonella enteritidis
	Salmonella farmsen
	Salmonella pomona
	Salmonella sofia
	Salmonella typhimurium
	Salmonella wassenaar

A.2. Lysine decarboxylase

Positive	Negative
Salmonella anatum	*Salmonella sofia*
Salmonella enteritidis	
Salmonella farmsen	
Salmonella pomona	
Salmonella typhimurium	
Salmonella wassenaar	

9. **Ornithine** decarboxylase (for 8.B.)

A. **Positive**	B. **Negative**
Arizona 20: 29-25	*Citrobacter* $048_1,48_3,48_4$
Salmonella dar-es-salaam	*Pseudomonas*
Salmonella montevideo	*Salmonella cilbek*
Salmonella newport	*Salmonella typhi*
Salmonella offa	
Salmonella paratyphi B	

A.1. Arginine decarboxylase

A.1.a. **Positive**	A.1.b. Negative
Salmonella dar-es-salaam	*Arizona* 20: 29-25
Salmonella montevideo	Salmonella offa
Salmonella newport	
Salmonella paratyphi B	

A.1.a.1. Threonine decarboxylase

Positive	**Negative**
Salmonella montevideo	*Salmonella dar-es-salaam*
Salmonella newport	
Salmonella paratyphi B	

B.1 Arginine decarboxylase

Positive	**Negative**
Citrobacter $048_1,48_3,48_4$	Pseudomonas
	Salmonella cilbek
	Salmonella typhi

B.2 Histidine decarboxylase

Positive	**Negative**
Salmonella cilbek	*Pseudomonas*
	Salmonella typhi

B.3 Threonine decarboxylase

Positive	**Negative**
Salmonella typhi	*Pseudomonas*

دور أنزيم الديكربوكسيليز كوسيلة للتقسيم الكيميائي في البكتريا

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في محاولة لعمل دراسة تقسيمية لأربعين نوعاً من البكتريا، يتبع معظمها عائلة البكتريا المعوية (Enterobacteriacea)، باستخدام نشاط هذه الأنواع في نزع مجموعة الكربوكسيل من ٢٤ نوعاً مختلفاً من الحموض الآمينية. أثبتت التجارب الآتي:

٢٩ نوعاً من البكتريا يستطيع نزع مجموعة الكربوكسيل من حمض الثريونين.

٢٥ نوعاً من البكتريا يستطيع نزع مجموعة الكربوكسيل من حمض أورنيثين.

٢٤ نوعاً من البكتريا يستطيع نزع مجموعة الكربوكسيل من حمض الليسين.

٢٢ نوعاً من البكتريا يستطيع نزع مجموعة الكربوكسيل من حمض الهستيدين.

٢١ نوعاً من البكتريا يستطيع نزع مجموعة الكربوكسيل من حمض الارجنين.

وعدد قليل من البكتريا (٤ أنواع) يستطيع نزع مجموعة الكربوكسيل من الالآنين ـ الاسبرتيك والمثيونين، وثلاثة أنواع فقط من البكتريا تستطيع نزع هذه المجموعة من السيستين ـ الجليسين ـ الليوسين ـ ونوعان فقط من البكتريا يستطيعان نزعه من هيدروكسيبرولين ـ ايزوليوسين ـ نورليوسين ـ برولين ـ تربتوفان ـ فالين.

ولم يستطع غير نوع واحد فقط من البكتريا نزع مجموعة الكربوكسيل من حمض التيروسين، في حين أنه لم يستطع أي نوع أن ينزع هذه المجموعة من حمض الداى هيدروكسيفينيل الآنين.

وقد أمكن استنباط جدول للتعرف على كل نوع أو مجموعة متشابهة من البكتريا المدروسة.