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Effect of Different Amino Acids on the Activity of Amylase Produced by Selected Members of Enterobacteriaceae as Well as Other Pathogenic Bacteria

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ABSTRACT. The effect of 24 different amino acids on 40 selected members of Enterobacteriaceae and other pathogenic bacteria was studied. The results showed that *iso*-leucine and α-amino butyric acid were the least inductive amino acids for amylase production by the tested organisms (21-23 species) whereas histidine; lysine or arginine; threonine and valine were most efficient (35, 34 and 33 members respectively). These were followed by aspartic acid (32 organisms), glutamic acid, hydroxyproline, tyrosine (31 species); methionine, phenylalanine tryptophan (29 members); n-leucine, proline (28 members); cysteine, cystine, ornithine (27 organisms).

Within these amylase producing organisms, *Klebsiella* and *Pseudomonas* responded very highly, showing a very remarkable activity with 15 amino acids. These were followed by *Vibrio ogawa* (11 amino acids); *V. inaba* (10 amino acids). Furthermore, although all tested amino acids released amylase in *Yersinia* media, yet none of them was able to stimulate high potentialities comparable with those of the other organisms. In the meantime, several amino acids (from 9-18) did not allow for the release of the enzyme whereas the rest favoured a very low activity within the two species of *Erwinia* and the three strains of *E. coli*.

A scheme to differentiate the tested organisms, based on induction of high amylase activity through amino acid feeding is proposed.

Amylases are widely distributed in all members of the plant and animal kingdoms. They break down starch and glycogen to maltose and/or glucose. Kuranova *et al.* (1966) were able to isolate and purify an α-amylase, produced by a thermophilic *Bacillus subtilis* strain 110. Khan and Khan (1977) proved that *Bacillus* sp., *Pseudomonas* and *Escherichia coli,* isolated from the mid gut of adults of *Musca domestica*, exhibited amylase and invertase activity. Hamid *et al.* (1979) concluded that all *Vibrio* and *Enterobacter,* isolated from the intestine of the gray mullet, possessed a strong protease and amylase isozymal electrophoretic bands. Nakakuki *et al.* (1984) demonstrated the presence of an exoamylase, in *Pseudomonas*

stutzeri, capable of degrading water-insoluble starch. Mahmoud *et al.* (1982) suggested a synthetic medium containing a large quantity of tryptophan (15.4 g/1) for the best production of α-amylase by *Bacillus stearothermophilus*.

In this investigation, we tried to find out whether or not tryptophan as well as 23 other amino acids could be effective in releasing amylase by 40 pathogenic bacteria. In the meantime, the results may be helpful in differentiation of such organisms.

Materials and Methods

Selected identified pathogens, isolated from Bilharzia patients as well as authentic samples, kindly provided by the National Salmonella Center, Hamburg, W. Germany; and two plant pathogens (Saleh 1977) were used in this investigation. The bacteria were grown on Triple Sugar Iron medium, for 24 hr, then the biomass was harvested, washed several times and suspended in sterile saline solution at the density of 10^{12} cells/ml.

0.5 ml of this suspension was mixed with 0.5 ml of basal salt medium containing 0.5% of one of 24 tested amino acids. The mixtures were incubated, for four hr, at 28°C, then centrifuged, at 5000 rpm, for 15 min and the supernatant was tested for its amylase activity using the Automatic Clinical Analyser 60-Channel ACAII (Du Pont Instruments, Wilmington, USA) at 340 nm wave length.

Results and Discussion

Table 1 shows that *iso*-leucine and α-aminobutyric acid were the least inductive amino acids for amylase release in the media of the tested organisms (only 21-23 species) whereas histidine; lysine or arginine; threonine and valine were most effective (35, 34 and 33 members respectively). These were followed by aspartic acid (32 organisms); glutamic acid, hydroxyproline, tyrosine (31 species); glycine (30 members); methionine, phenylalanine, proline, tryptophan (29 organisms); n-leucine (28 bacteria); cysteine, cystine, leucine and ornithine (27 species).

Alanine preceeded *iso*-leucine and α-aminobutyric acid as a weak amylase producer (24 organisms); then serine (25 bacteria) and dihydroxyphenyl alanine (26 species).

Within these amylase - producing organisms, *Pseudomonas, Salmonella cilbek, S. typhi, S. wassenaar, Vibrio inaba* and *Yersinia* were the only organisms

Table. 1. Effect of various amino acid feeding on amylase activity produced by selected members of Enterobacteriaceae as well as other pathogenic bacteria.
(International unit per 10^Y cells)

Organism	Alanine	α-amino butyric	Argi-nine	Aspar-tic	Cys-teine	Cys-tine
Arizona 30:32-25	25.1	0.0	0.1	0.0	0.2	0.0
Ar. 6:13,14:-	22.1	0.0	0.1	0.0	0.1	0.0
Ar. (5),29:33-21	24.8	0.0	0.1	0.1	0.0	0.0
Ar. 20:29-25	37.4	0.1	0.1	0.1	0.0	0.0
Citrobacter 0 48_1, 48_3, 48_4	0.0	25.9	0.0	0.1	0.0	0.1
Erwinia carotovora var. citrullis	0.0	0.0	0.0	0.1	0.1	0.0
E. toxica	0.0	0.0	0.1	0.2	0.1	0.0
Esclierichia coli DM 3219	0.0	0.0	0.0	0.1	0.0	0.1
E. coli k12	0.0	0.0	0.0	0.1	0.0	0.1
E. coli 018	0.0	0.1	0.0	0.1	0.0	0.1
Klebsiella	0.0	35.9	31.9	18.9	34.8	30.7
Proteus morganii	0.0	0.0	14.2	15.3	12.5	0.0
P. vulgaris	0.0	0.0	14.8	14.4	11.4	0.0
Pseudomonas	10.9	10.5	9.0	10.2	10.2	8.1
Salmonella anatum	0.0	0.0	0.2	0.0	0.0	0.0
S. cilbek	0.1	0.1	0.1	0.1	0.2	47.5
S. dar-es-salaam	0.0	0.0	23.6	0.1	0.0	0.0
S. enteritidis	0.0	0.0	0.0	0.1	0.0	0.1
S. farmsen	0.0	0.0	17.0	0.0	0.0	0.0
S. Kralendyk	0.0	26.2	0.1	0.1	0.1	0.0
S. montevideo	0.0	18.9	16.7	0.0	0.0	0.0
S. newport	0.1	0.0	0.1	0.0	0.0	0.1
S. offa	30.0	0.0	0.0	0.0	0.1	0.1
S. paratyphi B	0.2	0.2	0.4	0.2	0.3	0.2
S. pomona	0.1	0.0	31.6	0.1	0.1	0.1
S. saint-paul	0.0	0.0	29.3	33.2	0.1	0.1
S. sofia	0.1	43.2	0.1	39.6	0.0	0.1
S. typhi	0.2	0.4	111.6	0.2	0.1	0.2
S. typhimurium	0.1	0.1	0.1	39.2	31.6	38.4
S. wassenaar	0.1	0.3	87.3	0.1	0.2	0.2
S. wayne	0.1	0.1	27.1	0.0	0.1	0.1
Shigella boydii	50.0	0.1	0.1	0.2	49.5	54.8
S. dysenteriac 8	77.5	0.1	0.1	0.2	80.8	68.3
S. dysenteriac 10	88.2	0.2	0.2	0.3	109.4	114.1
S. flexneri 1	73.0	0.1	0.2	0.2	80.9	73.9
S. flexneri 6	38.7	0.1	0.1	0.1	42.2	39.6
S. sonnei	83.2	0.2	0.2	0.3	100.0	77.9
Vibrio inaba	118.8	0.1	116.3	116.3	122.5	0.1
V. ogawa	91.4	0.1	93.3	90.5	90.5	0.1
Yersinia	0.2	0.4	0.6	0.1	0.2	0.3
Arizona 30:32-25	0.1	21.4	23.2	26.8	0.1	0.0
Ar. 6:13,14:-	0.0	18.6	22.3	18.4	0.0	0.0
Ar. (5),29:33-21	0.0	22.0	23.5	24.0	0.0	0.0

Table 1.—Continued

Organism	*Dihydroxy ph.Al.*	*Glutamic*	*Glycine*	*Histi-dine*	*Hydroxy-proiine*	*Leucine*
Ar. 20:29-25	0.0	22.7	28.8	27.1	0.0	0.0
Citrobacter 0 48_1, 48_3, 48_4	30.8	31.8	0.0	0.1	0.1	0.0
Erwinia carotovora var. citrullis	0.0	0.1	0.1	0.1	0.1	0.0
E. toxica	0.0	0.0	0.1	0.1	0.2	0.0
Escherichia coli DM 3219	0.0	0.0	0.0	0.1	0.1	0.0
E. coli k12	0.0	0.0	0.0	0.1	0.1	0.0
E. coli 018	0.0	0.0	0.0	0.0	0.0	0.0
Klebsiella	0.0	0.1	0.1	35.2	23.7	34.4
Proteus morganii	0.0	0.1	15.3	15.3	0.1	0.1
P. vulgaris	0.0	0.0	14.8	14.1	0.1	0.1
Pseudomonas	6.6	7.1	5.8	8.2	11.3	8.1
Salmonella anatum	0.7	0.0	0.0	0.0	0.0	0.1
S. cilbek	48.5	46.5	0.1	0.1	0.2	0.1
S. dar-es-salaam	23.1	22.4	0.0	23.1	0.1	0.0
S. enteritidis	0.0	0.0	0.0	0.1	25.6	26.6
S. farmsen	16.0	0.0	17.3	0.0	0.0	17.0
S. Kralendyk	0.0	0.1	20.8	0.0	0.1	24.1
S. montevideo	14.4	0.0	0.0	0.1	0.2	0.0
S. newport	0.1	32.0	0.0	29.7	30.0	32.0
S. offa	0.1	0.1	0.0	0.1	31.0	29.7
S. paratyphi B	0.1	0.3	0.2	0.3	102.1	0.2
S. pomona	0.1	38.7	30.7	31.9	31.3	0.0
S. saint-paul	0.1	30.4	0.1	0.1	0.0	0.0
S. sofia	0.1	35.9	36.4	0.1	0.1	0.2
S. typhi	103.5	10.1	0.2	0.2	0.1	0.3
S. typhimurium	0.0	0.1	0.1	0.1	0.1	0.1
S. wassenaar	0.2	74.6	0.2	0.3	0.1	0.1
S. wayne	0.1	29.6	0.1	0.1	34.6	0.0
Shigella boydii	0.1	0.1	0.1	0.1	0.0	0.1
S. dysenteriac 8	0.1	0.5	0.2	0.2	0.1	0.2
S. dysenteriac 10	0.1	0.6	0.3	0.2	0.1	0.3
S. flexneri 1	0.1	0.4	0.2	0.2	0.1	0.2
S. flexneri 6	0.1	0.2	0.1	0.1	0.0	0.1
S. sonnei	0.1	0.7	0.2	0.2	0.2	0.3
Vibrio inaba	116.3	0.1	110.0	122.5	0.1	0.2
V. ogawa	98.5	0.1	90.5	90.5	0.1	91.4
Yersinia	0.4	0.1	0.6	0.2	0.2	0.2
Arizona 30:32-25	0.1	0.1	26.5	0.1	0.0	0.0
Ar. 6:13,14:-	0.1	0.1	23.0	0.1	0.0	0.0
Ar. (5),29:33-21	0.0	0.1	24.5	0.1	0.0	0.1
Ar. 20:29-25	0.0	0.1	28.8	0.1	0.0	0.0
Citrobacter 0 48_1, 48_3, 48_4	0.1	25.9	0.0	0.1	0.0	32.1
Erwinia carotovora var. citrullis	0.0	0.0	0.2	0.1	0.1	0.1
E. toxica	0.0	0.0	0.1	0.1	0.1	0.0
Escherichia coli DM 3219	0.0	0.0	0.0	0.1	0.0	0.1
E. coli k12	0.0	0.0	0.0	0.0	0.0	0.0

Table. 1. Contd.

Organism	Iso-Leucine	n-Leucine	Lysince	Methio-nine	Orni-thine	Phenyl-alanin
E. coli 018	0.0	0.0	0.1	0.0	0.0	0.0
Klebsiella	0.0	35.9	36.3	0.0	35.6	0.0
Proteus morganii	0.0	0.0	0.1	0.0	16.5	14.2
P. vulgaris	0.0	0.0	0.1	0.0	15.0	15.2
Pseudomonas	7.1	11.0	11.1	7.1	10.8	10.1
Salmonella anatum	0.0	19.0	18.8	0.0	0.0	19.2
S. cilbek	48.5	0.1	0.2	0.1	0.2	49.0
S. dar-es-salaam	20.5	0.0	0.1	0.0	0.0	0.1
S. enteritidis	0.0	0.0	28.1	0.0	0.1	0.0
S. farmsen	0.0	17.1	15.5	0.0	17.7	0.0
S. Kralendyk	0.0	0.1	0.0	0.0	0.1	0.0
S. montevideo	0.0	15.6	0.0	0.1	0.0	0.1
S. newport	0.0	0.1	0.1	31.7	0.1	0.1
S. offa	0.7	0.0	0.1	0.0	0.1	0.1
S. paratyphi B	0.3	0.2	85.3	76.8	0.4	0.1
S. pomona	0.0	0.1	0.0	0.1	0.1	31.3
S. saint-paul	0.1	0.0	0.1	0.1	27.1	0.0
S. sofia	0.0	0.1	0.1	0.2	0.1	0.1
S. typhi	0.2	0.1	0.2	0.2	110.5	0.3
S. typhimurium	0.0	0.1	39.6	0.1	0.1	0.1
S. wassenaar	0.2	0.3	0.1	0.4	0.1	0.2
S. wayne	0.2	0.1	0.1	0.1	0.0	0.1
Shigella boydii	0.2	0.3	0.2	0.5	0.1	44.7
S. dysenteriac 8	0.3	0.3	0.3	0.2	0.1	77.5
S. dysenteriac 10	0.4	0.5	0.4	0.2	0.1	112.9
S. flexneri 1	0.3	0.4	0.2	0.1	0.1	83.5
S. flexneri 6	0.2	0.2	0.1	0.1	0.0	43.1
S. sonnei	0.4	0.4	0.3	0.2	0.1	101.1
Vibrio inaba	0.2	0.2	122.5	0.1	0.3	0.1
V. ogawa	0.1	0.0	90.5	0.1	0.1	0.1
Yersinia	0.4	0.1	0.4	0.3	0.2	0.3
Arizona 30:32-25	0.1	0.0	0.1	22.4	25.7	0.1
Ar. 6:13,14:-	0.1	0.0	0.1	21.2	22.8	0.1
Ar. (5),29:33-21	0.1	0.0	0.1	22.8	22.0	0.1
Ar. 20:29-25	0.1	0.1	0.1	28.2	26.2	0.1
Citrobacter 0 48_1, 48_3, 48_4	28.5	0.1	0.1	0.0	0.1	0.1
Erwinia carotovora var. citrullis	0.1	0.2	0.2	0.0	0.2	0.2
E. toxica	0.1	0.2	0.2	0.0	0.2	0.2
Escherichia coli DM 3219	0.0	0.1	0.1	0.1	0.0	0.0
E. coli k12	0.1	0.1	0.0	0.0	0.0	0.0
E. coli 018	0.2	0.1	0.0	0.0	0.0	0.0
Klebsiella	34.8	0.0	0.0	35.2	34.1	35.2
Proteus morganii	0.0	0.0	15.5	16.5	0.0	0.0
P. vulgaris	0.0	0.0	14.5	15.2	0.0	0.0
Pseudomonas	10.2	10.7	10.5	10.7	10.5	10.8

Table 1.—Continued

Organism	*Proline*	*Serine*	*Threo-nine*	*Trypto-phan*	*Tyro-sine*	*Valine*
Salmonella anatum	0.0	0.1	18.4	0.0	0.0	0.0
S. cilbek	49.5	0.1	0.1	0.1	48.5	0.1
S. dar-es-salaam	0.0	0.0	0.1	0.0	23.1	20.2
S. enteritidis	0.0	0.1	0.1	26.9	0.0	0.0
S. farmsen	0.0	0.0	0.0	17.3	0.0	15.7
S. Kralendyk	0.0	0.1	0.0	0.1	0.0	0.1
S. montevideo	0.0	0.1	0.0	0.0	18.5	0.1
S. newport	0.1	0.1	0.0	32.0	0.1	0.1
S. offa	0.1	30.7	33.1	31.4	0.1	0.1
S. paratyphi B	0.3	0.3	104.2	0.3	0.2	0.2
S. pomona	0.0	0.1	0.1	0.0	31.3	30.6
S. saint-paul	0.0	0.0	0.1	0.0	32.5	29.6
S. sofia	0.1	0.2	0.1	45.0	0.1	0.1
S. typhi	0.2	0.2	0.4	0.2	112.8	102.3
S. typhimurium	0.1	38.4	38.8	0.0	0.1	0.1
S. wassenaar	0.3	0.2	0.2	0.2	0.2	0.2
S. wayne	0.1	0.1	35.3	0.1	28.6	0.1
Shigella boydii	51.1	0.0	44.2	49.0	52.1	44.7
S. dysenteriac 8	80.8	0.0	80.0	81.7	78.3	82.5
S. dysenteriac 10	102.4	0.0	92.9	115.3	110.6	116.5
S. flexneri 1	74.8	0.0	82.6	80.9	77.4	73.0
S. flexneri 6	41.8	0.0	44.0	37.8	41.8	39.1
S. sonnei	0.9	0.0	104.2	1.0	101.1	91.6
Vibrio inaba	0.1	0.1	0.1	121.3	120.0	0.2
V. ogawa	0.1	0.1	0.1	90.5	90.5	0.1
Yersinia	0.9	0.9	0.2	0.2	0.4	0.2

capable of releasing the enzyme in all tested amino acids media, though to different extents. The type of amino acid seemed of minor effect with either the low producer (*Yersinia*) or the moderate amylase producer (*Pseudomonas*). The activity of the former ranged between 0.1-0.9 units whereas that of the latter reached 6-11 units per ml. On the other hand, the type of amino acid played a significant role in enzyme induction by these salmonellas or *V. inaba*. Glutamic acid favoured high amylase activity by the three salmonellas but suppressed that of *V. inaba*. The reverse was observed with administration of alanine.

Generally speaking, *V. inaba* seemed to be the best amylase producer, giving a range of 100-120 units in 10 amino acids. Such a range was not reached by any of the tested arizonas, *Citrobacter*, escherichias, proteii or *Pseudomonas*. Within the tested salmonellas, only *S. paratyphi B* could release such a range on hydroxypro-

line or threonine; and *S. Typhi* on arginine, dihydroxyphenyl alanine, ornithine, tyrosine or valine. Within the tested shigellas, *S. dysenteriae 10* or *S. sonnei* could release such range of amylase activity on 7 and 4 amino acids respectively, of which cysteine, phenylalanine and tyrosine were common inducers.

The second best amylase producer seemed to be *Vibrio ogawa* since it released the enzyme in all amino acids but n-leucine; at the range of 90-100 units in 11 of its media. The shigellas occupy the third rank, since the high activity was only apparent in 9 amino acids, at a range that dropped to 40-50 units in several of the species. The salmonellas release a range of 30-50 units that may drop to 20 units or increase to 70-80 units in few cases. Such ranges were reached in a maximum of 9 amino acids (*S. farmsen*) and a minimum of 2 acids (*S. wassenaar*). The remaining amino acids were mostly weak inducers, if not at all.

The escherichias and erwinias are the least enzyme producers, since 8 (*Erwinia*) and 12 (*Escherichia*) amino acids were completely unable to induce the enzyme whereas 3 (*Erwinia*) and 5 (*Escherichia*) amino acids were capable of inducing the enzyme in one or two of the organisms. Under all conditions, the amylase activity was very weak (0.1-0.2 units).

The arizonas were better amylase producers than *Citrobacter*. This is evident from the fact that 3 amino acids (arizonas) and 8 amino acids (*Citrobacter*) were incapable of releasing the enzyme. Furthermore, 7 other amino acids were unable to do so for one to three of the tested arizonas. Still, both genera were capable of releasing almost similar amounts (20-30 units) in 6 (*Citrobacter*) and 7 (arizonas) amino acids.

Furthermore, the results show that the release of the enzyme under various amino acids feeding, is of a limited diagnostic value for differentiation between the forty-tested organisms. This is due to the fact that all tested *Arizona* species showed the same high amylolytic activity when fed with either alanine, glutamic acid, cysteine, glycine, histidine, ornithine, phenyl alanine. Threonine or tryptophan highly induced amylase formation by both *Proteus* species. Similarly, all tested *Shigella* species were highly active amylase producers when cultured on alanine, cysteine, cystine, phenylalanine, proline, threonine, tryptophan, tyrosine or valine. Still, alanine, arginine, aspartic acid, cysteine, dihydroxyphenyl-alanine glycine, histidine, lysine, tryptophan or tyrosine highly stimulated amylase production by both *Vibrio* species.

Still, it may merit the trial to differentiate a genus or few species from one or more genera, from each other, on the basis of the high amylolytic activity according to the proposed scheme.

The results further show that substitution of one hydrogen atom from glycine by a methyl group (nonpolar alkyl radical) increased the potentiality for large amount release of amylase (40% and 56% of the producing organisms on glycine and alanine respectively). The potentiality of alanine derivatives depended entirely on the type of substituting radical. Methylation (non-polar alkyl radical) of the β-carbon [α-aminobutyric acid] reduced the percentage of highly potential organisms from 65% to 25%. Benzyl (non-polar aryl radical) or imidazole (non-polar heterocyclic radical) substitution was less suppressive (40% and 34% for phenylalanine and histidine respectively); whereas indole substitution (non-polar aromatic heterocyclic radical) was stimulatory (68% with tryptophan).

On the other hand, carboxylation or hydroxylation of the β-carbon (highly polar alkyl radicals) drastically reduced the number of potentially high amylase producers (28% and 12% highly active organisms on aspartic acid and serine respectively, compared with 56% on alanine). Acetylation of alanine was less suppressive (43% with glutamic acid). Methylation of serine partially counteracted the suppressive effects of the hydroxyl group (compare 43% of the total producers in threonine with 12% on serine).

These observations suggest that increasing the polarity in substituted alanines lowered their inductivity of potentially high amylase producers. This is further evidenced from the fact that the basic amino acids (lysine, arginine, ornithine) though increased the number of amylase producers yet the percentage of potentially high producers was far less than that of alanine. (40% in case of lysine or arginine and 25% for ornithine). Similarly, the increased polarity of tyrosine was very suppressive to the number of high amylase producers (62% and 35% for tyrosine and dihydroxyphenyl alanine respectively).

Although the sulphydryl group (moderately polar radical) is essential for amylase activity (Srivastava *et al.* 1981), yet cysteine was less promotive to potentially high amylase producers (48% compared with 56% with alanine). The other sulphur amino acids were more suppressive particularly methionine, which reduced the highly active organisms to 10% of the producers.

Diethylation of the β-carbon of alanine was less suppressive than the *mono-* or *tri-* methylation whereas *tri*-methylation or *iso-* propyl substituion were almost equally effective (compare valine with n-leucine or leucine; initiating high production in 40, 23 and 29% of the enzyme producers respectively). The most suppressive substitution was that of *iso-* leucine (methyl- ethyl- disubstitution) where only 2 organisms out of the 21 producers (10%) were highly active.

According to Srivastava (1984) the protein digest of bacterial amylase contained 16 amino acids of which aspartic acid showed the highest value, followed

by glutamic acid and leucine + *iso* − leucine. In this experiment, aspartic or glutamic acid though increased the number of amylase producers (still less than histidine) yet the ratio of the potentially active organisms was far less than in presence of the aromatic amino acids or alanine. On the other hand *iso*-leucine and α-amino butyric acid were the least inductive to amylase producers whether qualitatively or quantitatively.

These observations might be attributed to repression of enzyme synthesis by these amino acids; a phenomenon already observed by Srivastava and Mathur (1984) when dealing with amylase induction by carbohydrates. They noticed that glucose and maltose repressed whereas starch induced the synthesis of amylase by *Bacillus stearothermophilus*.

Scheme for Identification of Members of Enterobacteriaceae and Other Pathogenic Genera Based on Their Amino High Amylase Activity

1. **Activity in *Iso*-Leucine Media**

A. **Positive**	B. **Negative**
Salmonella cilbek	(38 members)
Salmonella dar-es-salaam	

A.1. Activity in Arginine, Histidine or Valine Media

Positive	**Negative**
Salmonella dar-es-salaam	*almonella cilbek*

A.2. Activity in Cystine, Phenylalanine or Proline Media

Positive	**Negative**
Salmonella cilbek	*Salmonella dar es-salaam*

2. **Activity in Serine Media (for 1.B.)**

A. **Positive**	B. **Negative**
Pseudomonas	(35 members)
Salmonella offa	
	Salmonella typhimurium

A.1. Activity in Alanine Media

Positive	**Negative**
Pseudomonas	*Salmonella typhimurium*
Salmonella offa	

A.2. Activity on α-aminobutyric, Aspartic, Cysteine or Ornithine Media

Positive	**Negative**
Pseudomonas	*Salmonella offa*

3. **Activity in Methionine Media (for 2.B.)**

A.	**Positive**	B.	**Negative**
	Salmonella newport		(33 members)
	Salmonella paratyphi B		

A.1. Activity in Glutamic, Histidine, Leucine, or Tryptophan Media

Positive	**Negative**
Salmonella newport	*Salmonella paratyphi B*

A.2. Activity in Lysine Media

Positive	**Negative**
Salmonella paratyphi B	*Salmonella newport*

4. **Activity on α-Aminobutyric Acid Media (for 3.B)**

A.	**Positive**	B.	**Negative**
	Citrobacter $048_1,48_3,48_4$		(28 members)
	Klebsiella		
	Salmonella kralendyk		
	Salmonella montevideo		
	Salmonella sofia		

A.1. Activity in Arginine Media

A.1.a.	**Positive**	A.1.b.	**Negative**
	Klebsiella		*Citrobacter* $048_1,48_3,48_4$
	Salmonella montevideo		*Salmonella kralendyk*
			Salmonella sofia

A.1.a.1. Activity on Cysteine, Cystine, Histidine or Hydroxyproline Media

Positive	**Negative**
Klebsiella	*Salmonella montevideo*

A.1.a.2. Activity in Dihydroxyphenyl Alanine Media

Positive	**Negative**
Salmonella montevideo	*Klebsiella*

A.1.b.1. Activity in Aspartic Acid Media

Positive	**Negative**
Salmonella sofia	*Citrobacter* $048_1,48_3,48_4$
	Salmonella kralendyk

A.1.b.2. Activity in Dihydroxyphenyl Alanine, Glutamic, n-Leucine, Phenyl Alanine or Proline Media

Positive	**Negative**
Citrobacter $048_1,48_3,48_4$	
Salmonella kralendyk	

5. **Activity in n-Leucine (for 4.B.)**

A.	**Positive**	B.	**Negative**
	Salmonella anatum		(26 members)
	Salmonella farmsen		

A.1. Activity in Arginine, Dihydroxyphenyl alanine, Glycine, Leucine, Ornithine, Tryptophan or Valine Media.

Positive	**Negative**
Salmonella farmsen	*Salmonella anatum*

A.2. Activity in Phenylalanine or Threonine Media

Positive	**Negative**
Salmonella anatum	*Salmonella farmsen*

6. **Activity in Leucine Media (for 5.B.)**

A. **Positive**	B. **Negative**
Salmonella enteritidis	(24 members)
Vibrio ogawa	

A.1. Activity in Alanine, Arginine, Aspartic, Cysteine, Dihydroxyphenyl alanine, Glycine, Histidine or Tyrosine Media.

Positive	**Negative**
Vibrio ogawa	*Salmonella enteritidis*

A.2. Activity in Hydroxyproline Media

Positive	**Negative**
Salmonella enteritidis	*Vibrio ogawa*

7. **Activity in Ornithine Media (for 6.B.)**

A. **Positive**	B. **Negative**
Proteus morganii	(20 members)
Proteus vulgaris	
Salmonella saint paul	
Salmonella typhi	

A.1. Activity in Cysteine, Glycine, Histidine, Phenylalanine, Threonine or Tryptophan Media.

A.1.a. **Positive**	A.1.b. **Negative**
Proteus morganii	*Salmonella saint paul*
Proteus vulgaris	*Salmonella typhi*

A.1.b.1. Activity on Dihydroxyphenyl Alanine Media

Positive	**Negative**
Salmonella typhi	*Salmonella saint paul*

8. **Activity in Dihydroxyphenyl Alanine Media (for 7.B.)**

A. **Positive**	B. **Negative**
Vibrio inaba	(19 members)

9. **Activity in Hydroxyproline Media (for 8.B.)**

A. **Positive**	**Negative**
Salmonella pomona	(17 members)
Salmonella wayne	

A.1. Activity in Glycine, Histidine, Phenylalanine or Valine Media

Positive	Negative
Salmonella pommona	*Salmonella wayne*

A.2. Activity in Threonine Media

Positive	Negative
Salmonella wayne	*Salmonella pomona*

10. **Activity in Alanine (for 9-B.)**

A. Positive	B. Negative
Arizona 30: 32-25	(7 members)
Arizona 6: 13,14:-	
Arizona (5), 29:33,21	
Arizona 20: 29-25	
Shigella boydii	
Shigella dysenteriae 8	
Shigella dysenteriae 10	
Shigella flexneri 1	
Shigella flexneri 6	
Shigella sonnei	

A.1. Activity in Cysteine, Cystine, Phenylalanine, Threonine or Valine Media

Positive	Negative
Shigella boydii	*Arizona* 30: 32-25
Shigella dysenteriae 8	*Arizona* 6: 13,14:-
Shigella dysenteriae 10	*Arizona* (5), 29:33,21
Shigella flexneri 1	*Arizona* 20: 29-25
Shigella flexneri 6	
Shigella sonnei	

A.2. Activity in Glycine, Glutamic, Histidine or Lysine Media

Positive	Negative
Arizona 30: 32-25	*Shigella boydii*
Arizona 6: 13,14:-	*Shigella dysenteriae* 8
Arizona (5), 29:33,21	*Shigella dysenteriae* 10
Arizona 20: 29-25	*Shigella flexneri* 1
	Shigella flexneri 6
	Shigella sonnei

11. **Activity in Arginine Media (for 10.B.)**

Positive	Negative
Salmonella wassenaar	*Erwinia carotovora v. citrullis*
	Erwinia toxica
	Escherichia coli DM 3219
	Escherichia coli K12
	Escherichia coli O18
	Yersinia

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تـأثير الأنـواع المختلفة لـلأحماض الأمينيـة على نشـاط أنـزيم الأميليز المتكون في بعض أفراد عائلة البكتريا المعـوية وبعض الأنواع الأخرى من البكتريا المُمْرِضة

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قسم البنات ـ كلية العلوم ـ جامعة القاهرة [1]ومعهد الأبحاث لطب المناطق الاستوائية
القاهرة ـ مصر

من خلال دراسة تأثير ٢٤ نوعاً مختلفاً من الأحماض الأمينية على تكوين أنزيم الأميليز في ٤٠ نوعاً من البكتريا المعوية وأنواع أخرى من البكـتريا المُمْـرِضة إتضـح أن حمض الايزوليوسين وحمض الفا امينوبيوتيرك هما أقـل الأحماض الأمينيـة تأثيـراً على تكـوين أنـزيم الأميليز في بعض الأنـواع المستخدمـة (٢١ ـ ٢٣ نوعـاً)، في حين أن أحمـاض الهستيدين، الليسين، الأرجينين، الثريونين والغالين كـانت ذا تأثـير أقوى عـلى أنواع البكتريا (٣٥، ٣٤، و٣٣ نوعاً على التوالي).

ويتبع هذه الأحماض حمض الاسبرتيـك (٣٢ نوعـاً) ثم الجلوتاميك والهيدركسي برولين ثم التيروسين (٣١ نوعاً) والمثيونين والفينيل ألانين والتربتوفـان (٢٩ نوعـاً) ثم النورليرسين والبرولين (٢٨ نوعاً) ثم السيستئين والسيستين والاورنيثين (٢٧ نوعاً).

وقد اتضح ان بكتريا الكلبسييلا والبسيدوموناس تستجيب بشـدة لتكوين هـذا الأنزيم في وجود ١٥ نوعاً من الأحمـاض الأمينية، يتبـع هذه الأنـواع الفيبريـو اوجاوا (١١ حمض آميني) والفيبروابنابا (١٠ أحماض أمينية).

كذلك اتضح ان اليرسينيا تستطيـع تكوين أنـزيم الاميليز في وجـود جميع أنـواع الأحمـاض الأمينية المستخـدمة التي لم تعطِ أي تـأثير عـالٍ عـلى الأنـواع الأخـرى من البكتريا. وفي نفس الوقت كانت بعض الأحمـاض الأمينية (٩ ـ ١٨ نـوعاً) غـير قادرةٍ على تنشيط البكتريا لتكوين أنزيم الأميليز، في حين ان بقية الأحماض الأمينية أعـطت تأثيراً ضعيفاً في نوعي الاروينيا وفي ثلاثة أنواع من الايشيريشيا كولاى.

وقد وُضِعَ جدولٌ ارشاديٌّ لتمييز الأنواع المختلفة من البكتريا المستخدمة إعتماداً على نشاطها في تكوين أنزيم الاميليز.