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Table 1. Fermentation products of *E. Coli* under static conditions.

	Glucose consumed	Lactid acid formed	Acetic acid formed	Pyruvic acid formed	Ethanol formed
mg/dl	2000.0	900.0	205.7	20.0	128.0
% on consumed glucose		45.0	10.3	1.0	6.4

Table 2. Fermentation products by shaking culture.

Glucose used (%)		2.0	4.8
(NH ₄) ₂ HPO ₄ used (%)		0.05	0.1
(NH ₄) ₂ SO ₄ used (%)		0	0.1
Glucose consumed (%)		100.0	100.0
Lactic acid formed (%)		0	0
Acetic acid formed (%)		0	0
α -Ketoglutaric acid formed (%)		40.0	45.0
Pyruvic acid formed (%)		0	0

Microbiological Studies of *Coli-aerogenes* Bacteria. (II)

Oxidative Fermentation of Glucose

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In the previous paper, it has already been found by us that the various species of the genus *Esherichia* and the genus *Aerobacter* reveal their ability of producing a large amount of α -ketoglutaric acid from glucose, when they are cultivated under aerobic conditions such as shaking culture. From the results of experiments, it is observed that some strains of the bacteria of *coli-aerogenes* types accumulated none of the metabolic intermediates except α -ketoglutaric acid, while so many products such as acetic, pyruvic and α -ketoglutaric acids were obtained with other strains of the same types of the bacteria. In other words, so many forms of oxidative fermentation of glucose exist in the metabolisms of the bacteria of *coli-aerogenes* types, as given below:

- 1) α -Ketoglutaric acid fermentation,
- 2) Acetic, pyruvic and α -ketoglutaric acids fermentation,
- 3) Acetic and α -ketoglutaric acids fermentation,
- 4) Pyruvic acid fermentation,
- 5) complete oxidation to CO₂ and H₂O.

It is worth to note that a lower yield of α -ketoglutaric acid was generally observed, when the production of acetic acid was increasing. Lockwood *et al.* found 2-ketogluconic acid, which in the early stage of fermentation was produced from glucose and that 2-ketogluconic acid was then changed into α -

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ketoglutaric acid by *Ps. fluorescens*. As for *Serratia marsecens*, similar results were obtained by Asai and Aida *et al.* On the contrary, it was demonstrated by us, that 2-ketogluconic acid could not be an intermediate product of α -ketoglutaric acid with *Escherichia coli*, since 2-ketogluconic acid was never detected at any stage of fermentation. It was also found by us that a remarkable amount of α -ketoglutaric acid was already produced in the early stage of fermentation (within 14 hours' culture) by some species of *coli-aerogenes* types.

Phosphoric acid ester of vitamin B₁ has already been pointed out to be the principal component of the coenzyme of the oxidizing systems of pyruvic and α -ketoglutaric acids by Gunsalus *et al.* Therefore, they concluded that vitamin B₁ would reveal such a noticeable effect on the decomposition of α -ketoglutaric acid as it has been already ascertained on the decomposition of pyruvic acid. In our experiments with the bacteria of *coli-aerogenes* types, vitamin B₁ was found to diminish the formation of pyruvic acid, however no effect of vitamin B₁ was observed on the decomposition of α -ketoglutaric acid throughout the fermentation.

Table 1. The production of α -ketoglutaric acid.

Exp. No.	Strain No.	Time of incubation (hrs.)	Initial conc. of glucose (%)	Glucose consumed (g.)	α -Ketoglutaric acid produced (% on consumed glucose)
1	2C	84	5.0	4.94	42.9
2	2C	46	5.0	4.50	42.3
3	B24	60	5.0	4.70	43.6
4	B25	60	6.0	5.58	39.8
5	B24	60	5.0	4.70	44.7
6	B24	120	8.0	7.61	48.8
7	B24	120	8.0	6.70	45.0
8	B25	120	8.0	6.48	51.5
9	2C	140	9.0	8.33	51.0
10	2C	140	9.0	8.83	45.6
11	2C	140	9.0	7.01	49.0
12	5E	115	9.0	9.00	19.5
13	6E	115	9.0	8.88	32.4
14	7E	115	9.0	9.00	33.2
15	8E	115	9.0	8.73	34.4
16	9E	115	9.0	9.00	41.8

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Oxidations of Glucose and Pyruvic Acid

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