

9224

A Note on the Respiration of *Bacillus coli*.*

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The respiration of prepared aliquots of a suspension of *Bacillus coli* has been used extensively in this laboratory for comparing the calibration-constants of different Barcroft-Haldane differential manometers and their culture-vessels. The great accuracy of the observations has brought to light an hitherto unreported phenomenon in the oxygen-absorption of this organism.

The suspension of bacteria grown on agar for 18 hours was washed off with 10 cc. of broth, filtered through sterile cotton, and 0.1 cm. of the resulting suspension was placed in the vessel of the respirometer after further diluting it with 10 cc. of broth. The oxygen-uptake was observed at this final dilution at 37°C. Plating showed the cultures to be pure at the end of the experiment.

It was found that the rate of oxygen-uptake increased logarithmically, as would be expected, until the culture reached the age of about 3 hours and 40 minutes. At this time a new rate was initiated. The course and magnitude of this change is seen in Table I and plotted on Fig. 1. Many similar experiments were carried out, all of which showed the break in the curve at very nearly the same age (Table III).

The results tabulated in Table III were obtained with different manometers and in various types of culture-vessels. These experi-

TABLE I.
Manometric Data for Fig. 1.

Time, min.	—O ₂ h*	Time, min.	—O ₂ h*
95	0.0	240	87.9
105	.5	245	93.0
115	1.6	255	104.5
190	34.5	265	116.2
197	42.6	275	128.8
205	52.7	285	141.1
215	67.7	295	152.4
220	71.7	305	156.1
225	74.8	315	158.6

*—O₂h = difference in height in millimeters of the meniscus in the 2 arms of the manometer. Observations begun after 95 min.

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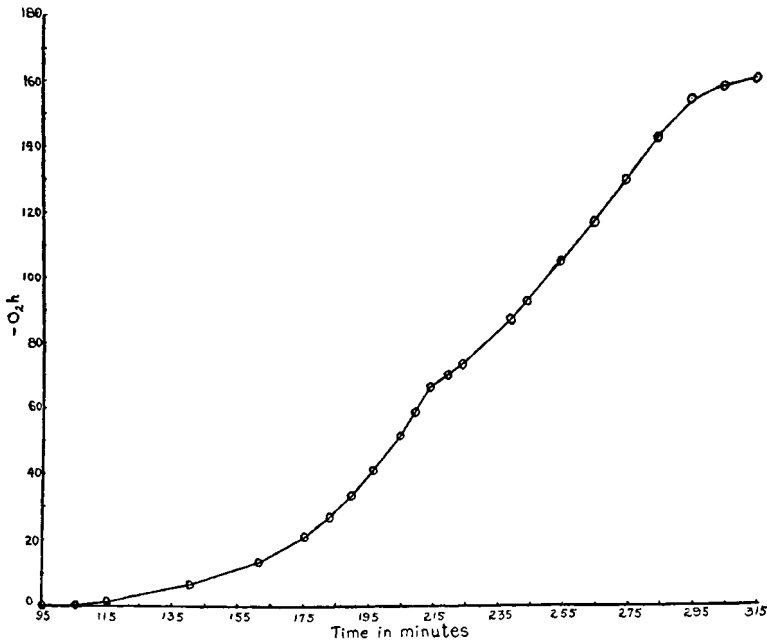


FIG. 1.

This figure shows the time of the break in the rate of oxygen use of *Bacillus coli*. See Table I for data.

TABLE II.
Manometric Data for Fig. 2.

Time, min.	$-O_2h^*$	Time, min.	$-O_2h^*$
120	0.0	186	56.9
130	4.7	188	60.0
135	5.9	190	62.9
140	6.7	192	65.4
145	9.0	194	66.8
150	13.4	196	68.3
160	24.4	198	69.8
165	28.7	200	71.4
175	40.9	210	80.0
182	50.9	220	89.9
184	53.8		

* $-O_2h$ = difference in height in millimeters of the meniscus in the 2 arms of the manometer. Observations begun after 120 min.

ments were carried out at irregular intervals over a period of several months during which time many different batches of broth were used.

In order to make sure that the break in the curve would be as sudden as manometric readings at 10-minute intervals indicated, the readings were made at intervals of 2 minutes. From data presented in Table II and plotted on Fig. 2, it may be seen that the transition in rates surprisingly occurred within a 2-minute interval.

TABLE III.
The Age of Various Cultures of *Bacillus coli* at which the New Rate of Oxygen-use is Initiated. All Cultures Carried at 37° C.

Hours	Minutes	Hours	Minutes
4	0	3	15
3	48	3	26
3	25	3	40
3	45	3	30
3	30	3	45
3	40	3	45
3	55	4	0
3	45	3	30
3	27	3	40
3	50		

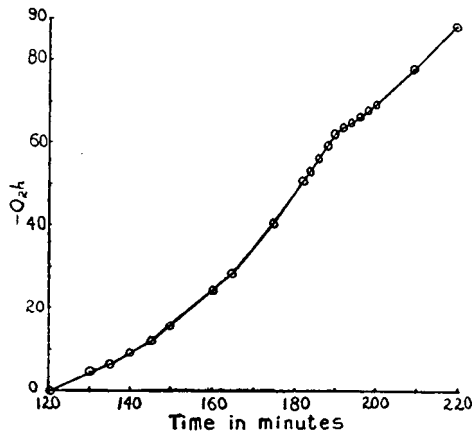


FIG. 2.

The break in the curve occurs within a 2-minute interval. See Table II for data.

It is obvious from the small magnitude of the change in the rate of oxygen-uptake that the highest accuracy in procedure is necessary for its observation. Errors as small as 2 to 3% would effectively mask it. The apparatus used was the Warburg waterbath, vigorously stirred and the temperature controlled to $\pm 0.001^\circ\text{C}$. by a vacuum-tube relay. The manometers were made from selected glass capillaries of a smooth and even bore. The manometer-fluid was triply distilled kerosene stained with Sudan III. Repeated use of the present equipment by several workers in this laboratory has shown that the oxygen-uptake of a culture may be determined with an experimental variation of less than $\pm 0.5\%$.

The fact that the culture of *Bacillus coli* exhibits these 2 cycles of respiratory activity should be of interest to the students of bacterial respiration. Since conditions do not permit immediate investigation of the nature of this phenomenon by the author, it is here reported in the hope that others will attempt its elucidation.