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Effect of certain electrolytes on the buffering power of bacterium coli.[1]

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In the present studies we have developed a technique for the measurement of the power of the bacterial cell to combine with hydrogen and hydroxyl ions. The bacteria show a general tendency to simulate amphoteric colloids and we have therefore measured their amphoteric property by the use of titration curves. A bacterial suspension in water or in a particular salt solution is titrated electrometrically with hydrochloric acid and with sodium hydroxide and similar titration curves are obtained for the menstruum alone. From these two sets of curves it is possible to calculate a series of "buffer ratios" by taking the quotients of the amounts of acid or alkali added to shift the P_H one unit in the bacterial suspension and in the menstruum alone. Such buffer ratios have been calculated for each P_H unit zone between P_H 2 and P_H 12. The titration cells which have been developed for these experiments are illustrated in the accompany figure. The buffer ratios for *Bact. coli* in distilled water, 0.725, M NaCl, 0.145 M $CaCl_2$ and in 0.580 M NaCl + 0.145 M $CaCl_2$ solutions are presented in Table I.

TABLE I.

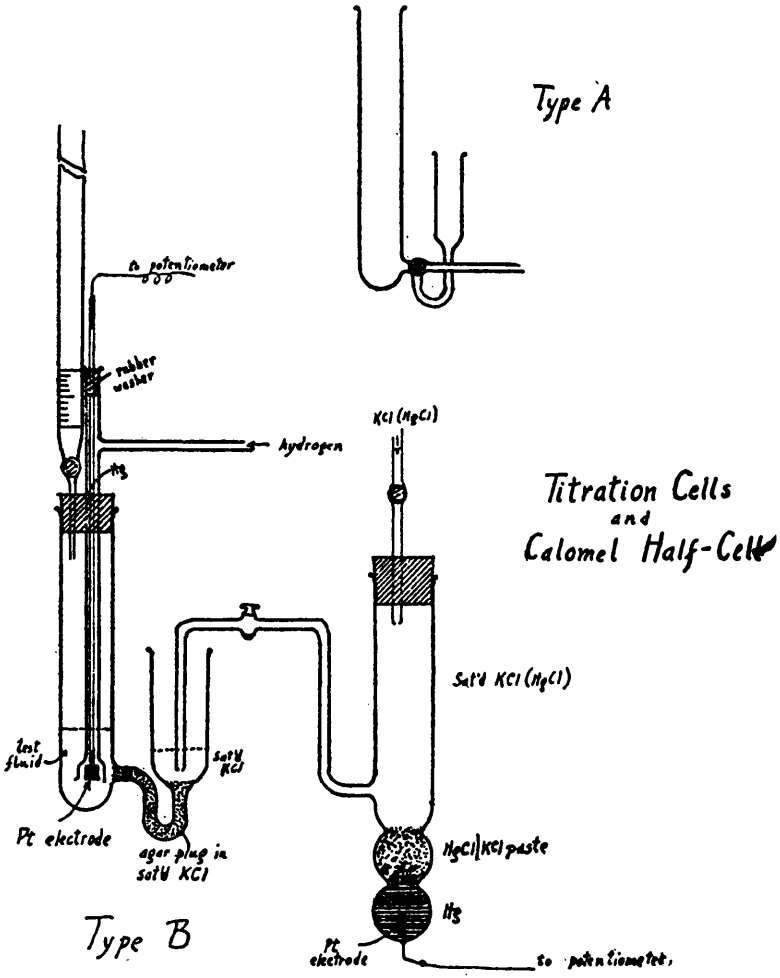
Average Buffer Ratios for Bacterium coli in Water and in Salt Solutions.

P_H Zone.	Ratio for *Bact. coli* in:			
	Water.	0.725 M NaCl	0.145 M $CaCl_2$	0.580 M NaCl + 0.145 M $CaCl_2$
3-2	0.85	1.5	1.0	1.2
4-3	0.77	1.0	1.0	1.3
5-4	1.7	1.8	0.75	1.4
6-5	.....	.....	1.0	1.0
7-6	8.2	1.9	.....	1.3
7-8	4.7	1.4	.....	.....
8-9	1.7	0.75	1.2	1.3
9-10	2.4	0.88	1.5	0.75
10-11	0.59	0.92	0.92	0.91
11-12	0.93	0.68	0.91	1.0
Number of experiments	7	3	3	4

[1] Studies reported here were aided by a grant from the Loomis Research Fund of the Yale School of Medicine.

Our findings may be summarized in the following categorical manner:

1. In distilled water suspensions of *Bacterium coli* show a marked tendency to resist a change in hydrogen-ion concentration in the zones of P_H which are of physiological interest, *i. e.*, P_H 4-10. In more acid and more alkaline solution this buffering power is weaker and the buffer ratios approximate unity at P_H 3-4 and at P_H 10-11.



The cells used in conducting electrometric titrations of bacterial suspensions.

2. Sodium and calcium chlorides, singly and in combination in the concentrations used depress the buffer ratios, particularly in the physiological zones of P_H.

3. The acidic P_H values at which the buffering power of *Bact. coli* becomes insignificant are approximately those at which this organism is known to be spontaneously agglutinable and to be isoelectric with the menstruum.[1] It is therefore significant to note that a similar reduction in the buffer ratio is attained at alkaline as well as at acidic reactions. This observation suggests the existence of a second, an alkaline isoelectric point for bacteria.

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The influence of certain electrolytes upon the electrical charge of bacteria.[2]

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In connection with an extensive series of studies on the effect of electrolytes upon the various properties of the bacterial cell, we have measured the electrical charge of vegetative cells of *B. cereus* (chosen on account of its large size) by the direct microscopic method described by Northrop.[3] In conducting these experiments a voltage of known magnitude (112 v.) is applied to non-polarizing zinc-zinc sulphate electrodes and the direction and velocity of migration of the bacteria in unbuffered suspensions determined by observing through the microscope the time taken by the bacterial cells to cross a definite space on the

[1] Michaelis, *Deut. med. Wochensch*, 1911, xxxvii, 969; Eisenberg, *Centr. Bakt.*, 1919, lxxxiii, 70, 472, 561; Northrop and DeKruif, *J. Gen. Physiol.*, 1922, iv, 639.

[2] Studies here reported were aided by a grant from the Loomis Research Fund of the Yale School of Medicine.

[3] *J. Gen. Physiol.*, 1922, iv, 629.