problem one must appreciate the fact that the convertibility of prothrombin deserves attention equally with the problem of the actual prothrombin level. By combined study with one-stage and 2-stage methods it is possible to evaluate both factors.

The cause of variation in convertibility has not been determined. In rabbit plasma the prothrombin is much more readily converted than in the plasma of man,[4] despite the fact that the actual prothrombin levels in the 2 are almost identical. We have also seen human cases in which a lowering of the prothrombin level was compensated by increased convertibility. Whether the variable convertibility represents differences in the prothrombin itself, or in the amount of "antiprothrombin" or in other factors must be answered by future research.

Summary. A study of 38 normal infants confirms previous work from this laboratory that the plasma prothrombin level is low in early infancy. It is also shown that an additional fall, not previously recognized, occurs between the second and sixth days of life. Evidence is presented to indicate that the rate of thrombin formation during coagulation depends upon convertibility of prothrombin as well as upon the amount of the latter. In newborn infants rapid convertibility of prothrombin compensates for deficient quantity of prothrombin. Evidence also suggests that variations in thromboplastin serve, in some circumstances, to compensate for a deficiency in the amount of prothrombin.

10613 P

Oxidation of Citric Acid by *Coli-aërogenes* Bacteria.

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Species of *Aërobacter* and *Citrobacter* utilize citric acid as a sole source of carbon in contrast to *Escherichia.* Previous work[1] has established the nature of the anaërobic dissimilation of citrate by *Aërobacter.* The present communication reports the results of an investigation of the aërobic breakdown of citric acid by *Aërobacter indologenes* and *Citrobacter freundii.*

1 Brewer, C. R., and Werkman, C. H., 1939, accepted for publication in *Enzymologia.*

TABLE I.
Serial Aërobic Dissimilation of Citric Acid by *Aërobacter indologenes* and *Citrobacter freundii*.
Products expressed as millimoles per 100 mM citrate fermented.

Organism	Sample No.	Time, hr	Citrate fermented, mM	H_2 mM	CO_2 mM	Formic acid mM	Acetic acid mM	Succinic acid mM	Non-reducing carbohydrate as glucose mM	O_2 uptake mM	Carbon recovery %	Redox index
A. indologenes	1	39	50	14.5	162.9	7.0	159.5	14.4	—	4.0	91.1	1.08
,, ,,	2	119	0	32.5	259.9	0	121.2	3.1	—	107.7	85.8	0.96
,, ,,	3	322	0	37.2	418.4	0	51.8	0	2.6	264.5	89.6 98.4*	0.97
C. freundii	1	39	36.8	0	194.9	0	79.2	42.0	—	69.9	86.9	0.98
,, ,,	2	202	13.2	0	516.0	0	2.2	0.3	—	352.0	86.9	1.03
,, ,,	3	322	0	0	521.7	0	1.0	0	2.2	378.7	89.5 98.7*	0.99
A. indologenes†				31.9	173.1	4.3	152.5	13.1	—	—	97.1‡	1.04

*Plus carbon in residue from ether extraction.
†Anaërobic dissimilation presented for comparison.
‡3.5 mM acetylmethylcarbinol, 3.9 mM 2,3-butylene glycol, and 6.2 mM lactic acid were produced but are not shown.

The course of citrate-oxidation was followed by serial analysis of media containing 0.1 M citrate as a sole carbon source. After inoculation, the media were kept saturated with air and the gaseous products removed continuously by forced circulation of the gas in a macrorespirometer. Results are presented in Table I.

During the first 39 hours only 4 mM O_2 were consumed by *A. indologenes* although the medium was saturated with air. By comparison with the anaërobic dissimilation shown in the bottom line of the table, the products during the initial stages of aërobic citrate breakdown appear to be those of a normal fermentation. The second and third analyses demonstrate that the action of oxygen is on acetic, succinic, and formic acids rather than citric acid itself. It is noteworthy that hydrogen was produced throughout the experiment even when its only apparent source was the aërobic dissimilation of acetic or succinic acid.

The experiment with *C. freundii* shows that in contrast to *A. indologenes* the former attacks citrate less vigorously but oxidizes the products of citric acid more completely. Hydrogen is not produced. Accumulation and subsequent oxidation of acetic and succinic acids is apparent as with *Aërobacter.*

Carbon recoveries were consistently lower in aërobic than in anaërobic experiments. The undetermined carbon was found to be non-volatile and non-ether-soluble. The properties of the unidentified material resemble those of complex carbohydrates, suggesting an oxidative assimilation of a part of the organic acids.

Respirometric experiments using the Barcroft-Warburg apparatus, on the effect of sodium azide on citrate-oxidation revealed increases in O_2 uptake and CO_2 production by cell-suspensions in the presence of M/1200 NaN_3 and by proliferating cells with M/4000 NaN_3 (Table II) (*cf.* Clifton[2, 3]). Similar effects were obtained with cell-

TABLE II.
Effect of NaN_3 on Aërobic Citrate Dissimilation by Proliferating *Aërobacter indologenes.*
200 μl citrate/flask. 41 hours.

Molarity NaN_3	CO_2 μl	O_2 μl
0	839	549
0.000125	900	608
0.0001875	970	645
0.00025	1001	701
0.000375	97	17

[2] Clifton, C. E., *Enzymologia,* 1937, **4**, 246.

[3] Clifton, C. E., and Logan, W. A., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 619.

suspensions of *A. indologenes* on *l*-malate, succinate, pyruvate, acetate, and aconitate. Aconitate was attacked aërobically only after an induction-period and anaërobically not at all. Citraconic, itaconic, tricarballylic, and α-OH isobutyric were not dissimilated either aërobically or anaërobically.

Other microrespirometric studies have shown that the respiratory quotients of oxidations of citric, aconitic, oxaloacetic, *l*-malic, fumaric, succinic, and pyruvic acids by *A. indologenes* decrease with time, indicating that the first steps in the oxidation of these acids are anaërobic.

Schemes for oxidation of citric acid by animal tissue[4] in which the citrate is oxidized stepwise through α-ketoglutaric, succinic and oxaloacetic acids cannot apply to the bacterial oxidation because the latter produces more acetate and less CO_2 in the early stages (Table I) than required by the former.

The oxidation of citric acid by *coli-aërogenes* bacteria proceeds through the normal anaërobic fermentation, to products that are dehydrogenated to CO_2, H_2O and assimilated to complex carbohydrate-like materials. The synthesis of "carbohydrate" is inhibited by NaN_3 as evidenced by increased O_2 uptake and CO_2 production.

10614 P

Some Effects of Low Choline Diets.

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The importance of choline in the prevention of "fatty livers" has been previously demonstrated by the investigations of Best[1] and of Channon.[1] The following experiments indicate that the production of a fatty liver on a low choline diet is only one manifestation of a more fundamental deficiency condition.

Male rats, 40 g in weight and 24 days of age, were used in groups of ten. The basal diet consisted of fibrin-4, casein-8, dried egg white-3, salt mixture[2]-4, calcium carbonate-1, codliver oil-5, lard-35, agar-2, and sucrose-38. The water soluble vitamins were supplied

4 Martius, C., *Z. physiol. Chem.*, 1937, **247**, 104.

1 Best, C. H., and Channon, H. J., *Biochem. J.*, 1935, **29**, 2651.

2 Hawk, P. B., and Oser, B. L., *Science*, 1931, **74**, 369.