When the animals were sacrificed small bits of the synovial membrane were planted in culture media. Of 90 joints, thus cultured, 25 were positive and 65 were negative. Nine of the positive cultures were contaminations, and in the remaining 16, organisms apparently identical with those injected were obtained.

We have thus produced the lesions characteristic of chronic arthritis in a series of rabbits by injecting organisms obtained from the joints of patients with the disease, and in a certain number of cases we have grown the organisms from the joints of the rabbits. We have injected these organisms into the circulation of rabbits with negative results. The staphylococci produced chronic arthritis in 66 of 84 instances (78%) and minute bacilli produced chronic arthritis in 34 of 50 instances (66%).

These results are open to 3 interpretations. (1) We are dealing with contaminations which are capable of producing chronic low grade arthritis, in animals. (2) We are dealing with non-specific organisms which are present in the tissues of patients with chronic arthritis, but which have no causative relationship to the disease. (3) Chronic arthritis is an infectious disease which may be due to any one of several organisms and is most often due to a low grade staphylococcus. It is probable that there is some as yet unknown abnormal susceptibility in those individuals who develop the disease.

4562

Oxidation of Lactic Acid by Dog Erythrocytes.

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The blood of certain species converts glucose to lactic acid but lacks the power of oxidizing either, and of oxygen consumption. The observation by Fürth and Lieben[1] of lactic acid destruction by horse erythrocytes was not confirmed by Warkany;[2] while Koechig[3] in this laboratory using the method described by Ray[4] failed to detect loss of lactate or formation of acetaldehyde. It appears that

[1] Fürth and Lieben, *Biochem. Z.*, 1922, cxxviii, 144.

[2] Warkany, *Biochem. Z.*, 1927, clxxxiv, 480.

[3] Koechig, unpublished results.

[4] Ray, *Am. J. Physiol.*, 1927, lxxxii, 405.

such "activation" of glucose as occurs in glycolysis does not alone suffice to accomplish the oxidation either of glucose or lactic acid—or an intermediate—by molecular oxygen. The same inference can be drawn from the similar behavior of cancer tissue or muscle extracts, the glycolyzing power of which is little affected by oxygen (Warburg and Meyerhof).

Recently Barron and Harrop have shown that the addition of methylene blue to blood undergoing glycolysis causes a marked change in its behavior. There results an increase of O_2 consumption and of CO_2 production,[5] and a large part of the sugar which disappears fails to appear as lactic acid,[6] thus resembling reactions observed with muscle and other tissues. In the presence of the dye the blood respires with the oxidation, presumably, of either glucose or lactic acid or both. The rate of sugar disappearance is only slightly affected. The dye appears to supply an oxidation catalyst of a sort which is normally absent.

We have confirmed the observations of Barron and Harrop cited above and have attempted to decide whether it is the glucose which is oxidized and thus prevented from appearing as lactic acid, or whether lactic acid may be first formed and subsequently oxidized.

Our results show that dl-sodium lactate added to washed blood erythrocytes disappears on incubation in air in the presence, but not in the absence of methylene blue. The fact that O_2 consumption is increased indicates that the lactic acid is oxidized. It seems unlikely that it could be converted to some form of carbohydrate; no evidence for such possibility has been found. The action of methylene blue in bringing about the oxidation of lactate is not prevented by cyanide or fluoride. Since cyanide abolishes metal catalysis and fluoride prevents glycolysis, neither of these factors is necessarily concerned. In serum but under otherwise identical conditions neither lactate nor glucose is oxidized. The facts stated seem to indicate that corpuscles "activate" ("dehydrogenate") lactate, but not to a degree allowing oxidation by molecular oxygen or other components or catalysts (glutathione) present in blood. Methylene blue serves as a more efficient catalyst, presumably of a higher oxidation intensity, and perhaps resembling the iron-bearing catalysts which are supposed to participate in the more active oxidations in other tissues.

The red corpuscles of freshly drawn defibrinated dog blood were washed once with saline and suspended in an equal volume of an

[5] Harrop and Barron, *J. Exp. Med.*, 1928, xlviii, 207.

[6] Barron and Harrop, *J. Biol. Chem.*, 1928, lxxix, 65.

isotonic solution containing NaCl, sodium lactate and phosphate buffer (pH 7.3). One portion was precipitated immediately by the $ZnSO_4$ method of Somogyi.[7] Two other samples, one containing 0.005% methylene blue, were incubated in stoppered 300 cc. Erlenmeyer flasks. During incubation the flasks were rocked mechanically by motor to maintain uniform aeration. The short periods of incubation preclude errors from bacterial growth. Sugar ("true sugar") was determined on zinc filtrates, using a modified Shaffer-Hartmann reagent (unpublished No. 30 by Shaffer and Somogyi) which permits accurate determinations at very low sugar concentrations. Lactic acid was determined on copper-lime filtrates by the oxidation method.[8]

TABLE I.

Description	Sugar in 100 cc. corpuscle suspension.		Lactic acid in 100 cc. corpuscle suspension.	
	Present mg.	Change mg.	Present mg.	Change mg.
Control (not incubated)	8	—	158	—
Control (incubated 3 hrs. 37° C.)	0	—8	167	+9
Incubated with methylene blue	5	—3	131	—27

The data given in the table illustrate the results obtained. Without methylene blue there is no evidence of loss of lactate, but rather in increase equal to the loss of glucose. In presence of the dye 20 to 35 mg. % of lactate disappears. Although the amount lost is not as great as with unwashed corpuscles the results are well beyond analytical errors and have been obtained in a series of such experiments.

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Anatomical Evidence for Existence of Enteric Reflex Arcs Following Degeneration of Extrinsic Nerves.

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Dogs were subjected to an operation in which a large mesenteric artery and the nerves accompanying it were divided in order to

[7] Somogyi, M., PROC. SOC. EXP. BIOL. AND MED., 1929, xxvi, 353.

[8] Friedemann, Cotonio and Shaffer, *J. Biol. Chem.*, 1927, lxxiii, 335.