Summary. Oral administration of 100 mg 1-3-butyl-p-tolylsulfonylurea per kg body weight to normal rats results in a marked hypoglycemia within one hour which persists for 4 hours. No significant change occurs in blood sugar of similarly treated alloxan diabetic rats.

1. Franke, H., and Fuchs, J., *Deutsche Med. Wochnschn.*, 1955, v80, 1449.

2. Bertram, F., Benfeldt, E., and Otto, H., *ibid*, 1955, v80, 1455.

3. Achelis, J. D., and Hardebeck, K., *ibid.*, 1955, v80, 1455.

4. Holt, C. v., Holt, L. v., Kroner, B., and Kuhnau, J., *Arch. exp. Path. Berlin*, 1955, v224, 66.

5. ———, *Die Naturwissenschaften*, 1954, v41, 166.

6. Nelson, N., *J. Biol. Chem.*, 1944, v153, 375.

7. Mirsky, I. A., Perisutti, G., and Diengott, D., *Metabolism*, 1956, v5, 156.

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Effect of CO_2 on Blood Lactic Acid in Cats.* (22298)

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Fenn and Asano(1) have observed in cats an increased acidity or fall in the alkali reserve of the blood during CO_2 inhalation, which could not be explained by movements of sodium, potassium, or chloride between tissues and plasma, and Shaw and Messer(2) have similarly observed a fall in the bicarbonate level during CO_2 breathing.

These experiments were undertaken to help clarify these findings and to identify if possible the acid mobilized by CO_2.

Procedure. The effect of high CO_2 tension on blood lactic acid was investigated in cats. Each animal was anesthetized by intraperitoneal injection of dial with urethane. Urethane has been found to raise the blood sugar, but not the lactic acid and allows for a large and rapid effect of adrenalin on the blood lactic acid(3). Four different gas mixtures were used, 10%, 20%, 30%, and 40% CO_2 in oxygen. Thus in every case the experimental animal was assured of a sufficient supply of oxygen to avoid any possibility of anoxic lactic acid formation. The desired gas mixture was inhaled by the cat from a spirometer through respiratory valves and a tracheal cannula. Blood samples were taken from the right carotid artery and were immediately deproteinized with trichloroacetic acid to stop glycolysis. After the tracheal and arterial cannulation, heparin was injected (10 mg/kg of body weight). One sample was always taken just before the onset of CO_2 inhalation. The lactic acid was determined colorimetrically by the method described by Barker and Summerson(4).

Results. The findings of these experiments are illustrated in the accompanying graphs. Increments of lactic acid in m. eq./liter above the rather variable initial blood level are plotted against time after the beginning of CO_2 inhalation. To save space, a logarithmic time scale has been used. In the graphs on the left, the different concentrations of CO_2 were inhaled for 10 minutes, (solid lines) after which the cat breathed air, (dotted lines) while in those of the right, the CO_2 was inhaled for one hour or more and a few curves are continued as dotted lines during recovery breathing air. The initial levels of lactic acid corresponding to each curve are indicated by points with appropriate symbols plotted just to the right of each graph.

With 10% CO_2 there is often a slight fall, but never a definite increase in lactic acid. The increase with 30% and 40% CO_2 is on the average somewhat greater than with 20%. The cats were unable to take 40% CO_2 for

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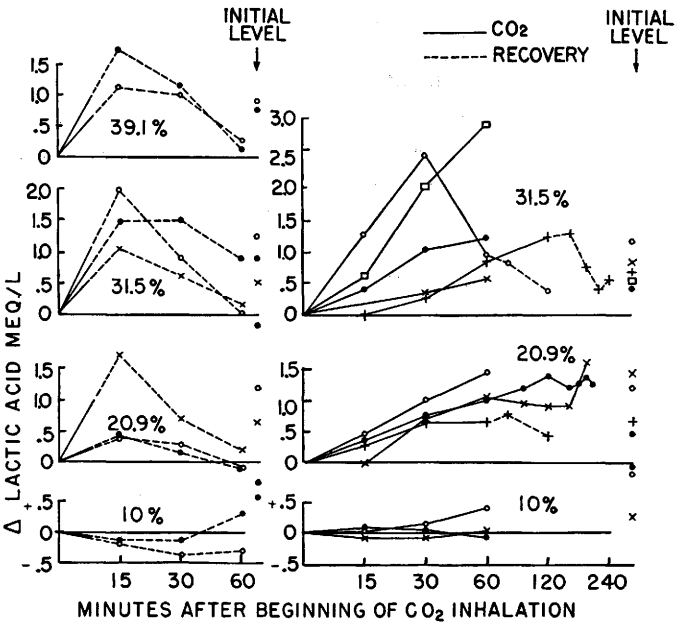

FIG. 1. Changes in lactic acid concentration of blood resulting from inhalation of different concentrations of CO_2, as indicated on the graph. Abscissae represent time after beginning of CO_2 inhalation in logarithmic units. Duration of CO_2 period was uniformly 10 min. in the graphs on the left; in graphs on the right the duration is longer as indicated by length of solid line. Broken lines represent recovery periods breathing air. The initial level of lactic acid before CO_2 is indicated by the appropriate symbol for each experiment on the right of the graph.

periods much longer than 10 minutes without cessation or serious depression of respiration and the possibility of anoxia. With 10 minute exposures to CO_2 the lactic acid reached a maximum at 15 minutes and then returned to normal in about one hour. With longer exposures the lactic acid continued to rise as long as the CO_2 was breathed or at least for one hour. In 3 out of 4 cases where recovery was followed after a prolonged CO_2 exposure, there was a slight additional rise of lactic acid when the CO_2 was discontinued. A similar "off effect" may account for the fact that the 15 minute values in short term exposures (taken after 5 minutes of recovery) seem to be on the average higher than the 15 minute values during the prolonged breathing of CO_2. No correlation was found between the initial level of lactic acid and the magnitude of the increase under similar CO_2 concentrations and no other single factor seems to account for the wide variation in response that occurs in different cats with identical CO_2 mixtures. It is likely that a combination of experimental factors and individual differences is involved.

Discussion. The release of adrenalin by CO_2 inhalation in cats has been observed by the resulting contraction of a previously denervated nictitating membrane by Fenn and Asano(1). It is assumed that the rise in blood lactate in these experiments is at least partially due to the action of the liberated adrenalin(5).

The rise in lactic acid found in these experiments after inhalation of 30% CO_2 for 60 minutes was 0.5 to 2.9 m. eq. per liter. Fenn and Asano(1) under comparable conditions found a decrease in bicarbonate (measured at constant pCO_2) of 4.16 m. eq. per liter. The lactic acid therefore is only partially sufficient to explain the acidity previously observed. In the experiments of Shaw and Messer(2) it was reported that inhalation of 10-11% CO_2 for 40 minutes caused a decrease of bicarbonate (at constant pCO_2) of 1.9 m. eq. per liter (4.2 vol %). This figure is in good agreement with the results of the present ex-

periments although there seems to be some discrepancy in the threshold CO_2 concentration required for lactic acid formation. In the experiments reported here 10% CO_2 never caused a rise in lactic acid concentration, but this is close to the threshold and a deeper anesthesia or a slightly larger respiratory dead space in the experiments of Shaw and Messer might have increased the alveolar CO_2 sufficiently to cause an effect.

In contrast to the results of these experiments, high CO_2 tensions have been reported to cause a fall in lactate in dogs(6), as well as an increased breakdown of phosphocreatine in isolated muscle(7). Further an increase in lactate has been reported during a respiratory alkalosis in dogs(6) and in a heart-lung preparation(8), and also following the injection of an alkaline solution in dogs(9,10). This suggests a metabolic buffering mechanism whereby more lactic acid is produced when the pH rises, and less lactic acid when the pH falls. In consideration of such data, the results of the present experiments, showing an increase in lactic acid on inhalation of CO_2, require some explanation, possibly on the basis of a species difference. The data of Giebisch, Berger, and Pitts(6) indicate a fall of approximately 1.0 mMol of lactate per liter in dogs that had been breathing 20% CO_2-80% O_2 for 75 minutes, while in the experiments on cats presented in this paper, there is never any evidence of a drop in lactate at any time up to nearly 3 hours with 20%, 30%, or 40% CO_2 in the inspired air.

This apparent contradiction in the results with cats and dogs may be explainable by a greater dominance or influence of the sympathetic nerves in cats(11). It is possible that the metabolic buffering mechanism seen in dogs tends to occur also in cats during CO_2 inhalation, but is masked by a release of adrenalin, which enhances the breakdown of muscle glycogen to lactic acid. If there is any purposefulness to be discerned in the reaction observed in cats, it must depend upon some features of the response other than the change in pH since lactic acid enhances the acidity caused by CO_2. Guinea pigs apparently behave like cats in this respect since Schaefer, King, Mego, and Williams(12) have reported an increase of 5.3 m. eq./liter of lactic acid 1 hour after breathing 30% CO_2 in O_2.

Summary. Inhalation of 20-30% CO_2 in oxygen by cats causes a progressive increase in the lactic acid concentration of the blood, due in part to the liberation of adrenalin.

1. Fenn, W. O., and Asano, T., *Am. J. Physiol.*, in press.

2. Shaw, L. A., and Messer, A. C., *ibid.*, 1932, v100, 122.

3. Goldblatt, M. W., *J. Physiol.*, 1933, v78, 96.

4. Barker, S. B., and Summerson, W. H., *J. Biol. Chem.*, 1941, v138, 535.

5. Griffith, F. R., Lockwood, J. E., and Emery, F. E., *Am. J. Physiol.*, 1939, v127, 415.

6. Giebisch, G., Berger, L., and Pitts, R. F., *J. Clin. Invest.*, 1955, v34, 231.

7. Lipmann, F., and Meyerhof, O., *Biochem. Z.*, 1930, v227, 84.

8. Anrep, G. V., and Cannan, R. K., *J. Physiol.*, 1923, v58, 244.

9. MacLeod, J. J. R., and Hoover, D. H., *Am. J. Physiol.*, 1917, v42, 460.

10. Haldi, J., *ibid.*, 1933, v106, 134.

11. Hodes, R., *ibid.*, 1939, v126, 171.

12. Schaefer, K. E., King, C. T. G., Mego, J. L., and Williams, E. E., *Am. J. Physiol.*, 1955, v183, 53.

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