is of interest from several standpoints. It establishes an additional locality record for this rickettsial species which had been positively identified previously only in New York and Boston. It is to be expected that additional strain recoveries will extend the known distribution of *R. akari*. The present strain recovery provides evidence that the patients diagnosed as having rickettsialpox were exposed in an environment where the hazard of infection was present. Finally, this strain recovery, indicating the presence of the causative agent in mites collected in a natural environment, provides additional evidence for the vector role of the house mouse mite, the species previously incriminated by the studies of Huebner, Jellison, and Pomerantz(9) in New York.

The recovery of the present strain by inoculation of mice was supplemented by isolation accomplished by direct inoculation of developing chick embryos with mite suspension which has been appropriately treated. This simple method of strain isolation may be applicable to other problems involving recovery of rickettsiae or viruses from potentially infected arthropods.

Summary. The recovery of a strain of *Rickettsia akari* from mouse mites, *Allodermanyssus sanguineus* (Hirst), collected in West Hartford, Connecticut, is described. The evidence for its identification as *R. akari* is presented, and certain implications of these findings are discussed.

1. Rindge, M. E., *Conn. Health Bull.*, 1952, v66, 73.
2. Fuller, H. S., Murray, E. S., Ayres, J. C., Snyder, J. C., and Potash, L., *Am. J. Hyg.*, 1951, v54, 82.
3. Bovarnick, M. R., Miller, J. C., and Snyder, J. C., *J. Bact.*, 1950, v59, 509.
4. Huebner, R. J., Stamps, P., and Armstrong, C., *Pub. Health Rep.*, 1946, v61, 1605.
5. Plotz, H., Bennett, B. L., Wertman, K., Snyder, M. J., and Gauld, R. L., *Am. J. Hyg.*, 1948, v47, 150.
6. Mayer, M. M., Osler, A. G., Bier, O. G., and Heidelberger, M., *J. Exp. Med.*, 1946, v84, 535.
7. Topping, N. H., and Shepard, C. C., *Pub. Health Rep.*, 1946, v61, 701.
8. Boyd, H. M., *J. Lab. Clin. Med.*, 1951, v38, 313.
9. Huebner, R. J., Jellison, W. L., and Pomerantz, C., *Pub. Health Rep.*, 1946, v61, 1677.

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Effect of Glucose on Oxygen Consumption and Respiratory Quotient of Normal and Denervated Muscle.* (19686)

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The oxygen consumption of denervated muscle was first studied by Langley and Itagki(1). They compared the oxygen content of arterial and venous blood from innervated and denervated extremities of anesthetized cats. Since that time several workers have studied the oxygen consumption of denervated muscle *in vivo*(2,3) and *in vitro* (4.5). These various reports are not in agreement with each other as to the extent of the increase in oxygen consumption by the denervated muscle. Changes in various respiratory enzyme systems following denervation have been reported by Knowlton and Hines (4) and Humoller *et al.*(6,7). The experiments upon which this report is based were done with the hope of clarifying the effect of denervation on the oxygen consumption and respiratory quotient of muscle tissues. The influence of glucose in the perfusate on the oxygen consumption and the respiratory quotient of muscle was also investigated.

Method. One hundred young white rats weighing between 100 and 120 g were used. Unilateral phrenectomy was performed under ether anesthesia by destroying one phrenic nerve in the neck. Ten to 14 days after sec-

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TABLE I. Average Oxygen Consumption and Respiratory Quotient (with standard errors) of Muscle. The intervals represent half hour periods.

	With glucose		Without glucose	
	Normal	Denervated	Normal	Denervated
$Q O_2$ (1st interval)	8.39 ± .18	8.18 ± .17	6.31 ± .18	7.73 ± .23
$Q O_2$ (2nd interval)	7.36 ± .19	7.21 ± .17	5.53 ± .16	6.87 ± .22
$Q O_2$ (3rd interval)	6.61 ± .19	6.42 ± .18	5.05 ± .16	6.16 ± .21
R. Q. (1 hr)	.94 ± .05	.93 ± .04	.95 ± .05	.93 ± .06

tioning the nerve, the animals were sacrificed by cerebral concussion, the majority of them at 11 days. The domes of the diaphragms were placed in Warburg vessels within 10 minutes after the animals were killed. The oxygen consumption was measured by the Warburg manometric method. Krebs-Ringer's solution with phosphate buffer was used. In 96 experiments involving normal muscle and an equal number for denervated muscle, no glucose was present in the solution. In 64 experiments with each type of muscle the solution contained 0.2% glucose. Oxygen was allowed to pass through the flasks for 10 minutes. The flasks were shaken at a rate of 110 per minute in a water bath at 38°C. Before the initial manometric reading, a 15-minute period was allowed for stabilization. Readings were made at 30-minute intervals. The oxygen consumption was calculated from the tissue dry weight. The respiratory quotient was measured by the Dickens and Simer second method according to the procedure described by Dixon(8). Thirteen experiments were performed on denervated and normal muscle without glucose, and 7 with glucose present in the medium.

Results. The averages and standard errors of the oxygen consumption and respiratory quotient determinations are presented in Table I. The oxygen consumption of normal muscle in the absence of glucose is significantly lower (20%) than that of normal muscle with glucose and that of denervated muscle without glucose. A parallel decrease in the oxygen consumption for the 4 muscle-glucose combinations over the 1½-hour period is seen when these results are expressed graphically. No statistically significant difference was observed in the values obtained for the respiratory quotient of the various muscle-glucose combinations.

Discussion. The presence of glucose produced a significant difference in the oxygen consumption of normal muscle, but no significant difference in the oxygen consumption of denervated muscle. Also no significant difference occurred in the oxygen consumption of normal and denervated muscle in the presence of glucose when the oxygen consumption was determined on a dry weight basis. This suggests that in the absence of glucose it is possible for the oxygen consumption of normal muscle to be reduced, whereas it is not possible to reduce that of denervated muscle. Such a loss of the ability of muscle to respond to a reduction in glucose content of the surrounding medium may express a change in the enzyme system of the muscle after denervation.

Assuming that the oxygen consumption of muscle and the dry weight of the muscle decrease proportionately after denervation without change in the total number of cells, on a cellular basis the oxygen consumption of normal and denervated muscle in the absence of glucose should show no significant difference. Then the hypothesis that the enzyme system of normal muscle is able to utilize oxygen to a greater extent in the presence of glucose than can denervated muscle, becomes tenable.

The theory that the normal muscle metabolism is more adaptable to increases in glucose content of the medium seems as logical as the one that denervated muscle metabolism is less adaptable to decreases in glucose. Since there is no significant difference in the respiratory quotient of the glucose muscle combination we may assume that the difference in the oxygen consumption is not the result of change in the metabolic pattern.

Summary. 1. The oxygen consumption of innervated muscle *in vitro* is significantly greater in the presence of glucose than in the

absence of glucose. 2. The presence or absence of glucose produces no significant difference in the oxygen consumption of denervated muscle *in vitro*. 3. No statistically significant difference in the respiratory quotients of innervated and denervated muscle with and without glucose in the medium was found.

1. Langley, J. N., and Itagki, M., *J. Physiol.*, 1917, v51. 202.

2. Hines, H. M., Leese, C. E., and Knowlton, G. C., *Am. J. Physiol.*, 1931, v98, 50.

3. Sato, T., und Kasugai, F., *Tohoku J. Exp. Med.*, 1935, v26, 336.

4. Knowlton, G. C., and Hines, H. M., *Am. J. Physiol.*, 1934, v109, 200.

5. Califano, L., *Riv. di Pat. Sper.*, 1927, v2, 48.

6. Humoller, F. L., Griswold, B., and McIntyre, A. R., *Am. J. Physiol.*, 1950, v161, 406.

7. ———, *Am. J. Physiol.*, 1951, v164, 742.

8. Dixon, M., *Manometric Methods*, p94, The Macmillan Co., 1943.

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Microdetermination of Carbon Dioxide Content of Serum or Plasma with a Glass Electrode. (19687)

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The estimation of carbon dioxide content of serum by titration(1) has been little used in the past, chiefly because gasometric estimation is usually more convenient. Titrimetric micromethods(2), however, have been adopted recently. It occurred to us that the titration curves of different sera would be quite similar in the region of pH 5 to 7 and would enable one to estimate the carbon dioxide content from the pH after the addition of a known amount of acid to low pH, followed by a known amount of alkali to this region of pH. In the present paper we describe a rapid method which has been developed for the determination of carbon dioxide in this way on samples of 0.1 and 0.2 ml of serum or plasma.

Experimental. Titration curves were determined in 2 ml samples of serum or plasma obtained under oil from patients in various states of acid-base balance. The samples were diluted with 0.9% saline, and the pH was determined with a Cambridge Research model glass electrode after interval additions of about 0.02 ml of standard 0.2 N hydrochloric acid until the pH was in the region of 4.0. Then standard 0.2 N sodium hydroxide was added in the same way and the pH was determined until the initial pH of the diluted serum was reached. Fig. 1 shows a representative curve drawn on cross-section paper with pH on one axis and 0.2 N acid or alkali on the other. Only the curve for the back titration with alkali, curve B, would be required to calculate the content of carbon dioxide from the difference in the amount of acid and alkali added and the initial pH. It may be seen in Fig. 1 that the carbon dioxide content is equivalent to the amount of acid or alkali between curves

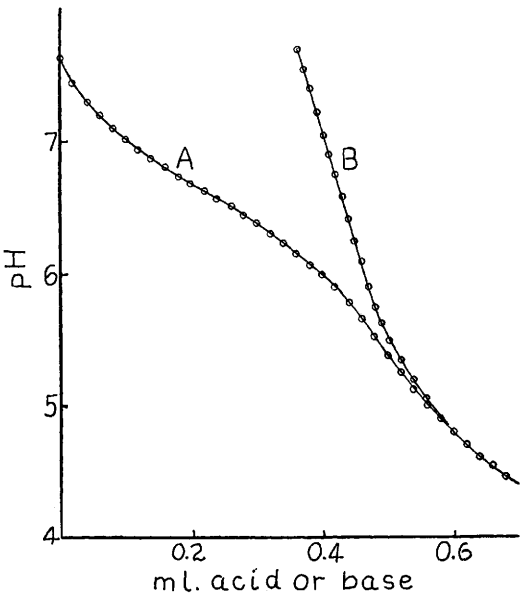

FIG. 1. Representative curve for titration of 2 ml of plasma with 0.2 N hydrochloride and 0.2 N sodium hydroxide.

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