

rum has been observed to occur as early as the second transfer. The "R" colonies stand out in marked contrast to the "S" forms and can be easily detected. Tests carried out with the "S" cultures grown on the dog blood agar medium have shown that they fully retain their original biological characteristics such as in regard to specific agglutination* and high virulence, while "R" colonies invariably give the properties of those of "Rough" cultures.

Summary. A highly satisfactory and differential medium for the study of "S" and "R" forms of pneumococci is described. The medium consists of ordinary beef infusion agar pH 7.8 to which 5 to 10% defibrinated blood of the dog is added. To obtain the optimum result, the organisms are plated on such a medium and incubated for 16 to 18 hours. On examination with reflected light the "R" colonies present an extremely rough surface in distinct contrast to the smoothness of "S" forms.

Evidences point to the dog blood cells as furnishing the important constituent of such a medium.

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Permeability of Muscle to Sodium Ion.

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It is well known that muscles, like blood corpuscles, contain more potassium than sodium, whereas blood plasma contains more sodium than potassium. The unequal distribution of the cations between the corpuscles and the plasma has been shown experimentally¹ and theoretically² to be due to the impermeability of the corpuscle membrane to cations. Although a similar explanation may be offered in the case of muscle, no conclusive proof is available. Mauriac, Aubel and Boutiron³ found that muscle immersed in experimental edema fluid gained Na but lost K, indicating that the muscle membrane is permeable both to Na and K. The per-

* We are indebted to Dr. A. B. Wadsworth of the New York State Department of Health for the antipneumococcus horse serum employed in this investigation.

¹ Doisy, E. A., and Eaton, E. P., *J. Biol. Chem.*, 1921, **47**, 377.

² Wu, H., *J. Biol. Chem.*, 1926, **70**, 203.

³ Mauriac, P., Aubel, E., et Boutiron, *Compt. rend. Soc. Biol.*, 1927, **97**, 78.

meability to K was confirmed by Wojtczak⁴ who found that muscle in nutrient solution containing no glucose lost K. Mond and Amson⁵ also found that resting uninjured muscle was permeable to K but impermeable to Na. Callison⁶ in a study on the so-called "bound potassium" of muscle found that varying amounts of potassium went into the Ringer solution in which cut pieces of muscle were placed. while there is general agreement as to the permeability of muscle to K, the evidence is conflicting with regard to Na. Monauni⁷ had reason to believe that even K does not really penetrate through the cells but only through the interstices.

A general criticism which may be raised against all the previous work on the subject is the fact that they are *in vitro* experiments. In the course of a study on the swelling of muscle we had occasion to test the permeability of muscle *in vivo* to sodium, and we have obtained results which, we believe, are conclusive.

Rabbits were anesthetized with morphine hydrochloride. The abdominal wall was slit open, the ureters were tied, and the abdominal wall closed. 20 cc. of 25% NaCl solution were injected into the pleural cavity. After 30 minutes about 40 cc. of blood were drawn from the heart, and pieces of abdominal muscle freed from fat and connective tissue were taken. Determination of H₂O, total base, and chloride were made on the muscle and blood plasma. Water was determined by drying at 110°C. for 12 hours. Chloride was determined by the method of Wilson and Ball,⁸ total base by the method of Stadie and Ross.⁹

As controls 20 cc. of 0.8% NaCl were injected into rabbits instead of the 25% solution. The results of 3 controls and 4 experiments are shown in the accompanying table.

It will be seen that the total base, Cl and H₂O contents of the muscle in the control were all normal. In the rabbits receiving injections of strong NaCl solution, the concentration of total base and chloride increased in the muscle as well as in the plasma. The water content is not appreciably changed, showing that the increase of base in the muscle is real and not due to dehydration. It is noteworthy that in the muscle the total base increased by

⁴ Wojtczak, A., *Bull. Intern. Acad. Polonaise*, 1927, B, 1253.

⁵ Mond, R., and Amson, K., *Arch. Ges. Physiol.*, 1928, **220**, 69.

⁶ Callison, W. E., *J. Biol. Chem.*, 1931, **90**, 665.

⁷ Monauni, J., *Arch. Ges. Physiol.*, 1929, **221**, 800.

⁸ Wilson, D. W., and Ball, E. G., *J. Biol. Chem.*, 1928, **79**, 221.

⁹ Stadie, W. C., and Ross, E. C., *J. Biol. Chem.*, 1925, **65**, 735.

TABLE I.
Water, base and chloride in plasma and muscle of rabbit.
(Figures in milliequivalents per kg.)

Rabbit No.	Concentration of NaCl injected	Plasma			Muscle		
		H ₂ O	Base	Cl	H ₂ O	Base	Cl
1	0.8	93.1	165.2	94.5	74.2	124.1	13.7
2		92.7	166.3	101.7	73.9	131.7	11.3
3		92.4	163.3	109.0	75.3	130.7	15.5
Average		92.7	164.9	101.7	74.5	128.8	13.5
4	25.0	93.4	181.5	132.3	75.7	158.0	18.5
5		94.0	188.1	123.2	74.7	153.5	17.8
6		92.1	183.2	133.8	74.6	156.7	17.5
7		93.1	180.7	118.7	73.7	150.8	17.6
Average		93.2	183.4	127.0	74.7	154.8	17.8

26 mM per kg. while the chloride increased only by 4 mM per kg. In the plasma the increases in base and in Cl are not very different. We may conclude, therefore, that the increase of base in the muscle of these rabbits is due to the migration of base from the plasma into the muscle cells and not merely due to increase of the base content in the intercellular fluid. Since plasma contains practically no K, the base which migrates into the muscle must be Na.

The unequal increases of base and chloride in the muscle and plasma suggest a membrane equilibrium and the problem is being studied further. For the present we wish only to conclude that the membrane of the muscle cells *in vivo* is permeable to sodium ion.

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Nutritional Edema. I. Effect of Level and Quality of Protein Intake on Nitrogen Balance, Plasma Proteins and Edema.

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Nutritional edema is apparently another type of edema in which the mechanism of its production is related to the low level of plasma proteins, similar to that of nephrosis and plasmapheresis. In nutritional edema, however, the lowered plasma proteins are brought