end, that irritability was retained by the posterior dissected portion of the cord for more than 45 minutes. Since it is possible to complete a respiration experiment in 20 minutes, the tissue must have been irritable throughout each experiment.

The procedure consisted first in determining the rate of CO_2 output for the resting cord. The effect of activity on the respiratory rate was found by suspending the tissue in the indicator tube on platinum electrodes which were passed through a paraffined stopper, and then stimulating the cord with induction shocks while the reading was being made. The current used for this purpose was not sufficient of itself to cause any change in the tint of the indicator, no matter how long continued. A third reading was now made with the tissue at rest. The relative rates of respiration in the three cases were determined by calculating the second and third readings as per cents. of the first. Averages of ten experiments made on the nerve cords of as many different animals yielded the following result: Resting 100 per cent., stimulated 89 per cent., resting 86 per cent. Electrical stimulation, therefore, not only did not increase the rate of CO_2 production of the nerve cord of *Cambarus*, but failed to interrupt the normal fall in rate. The question may thus rightly be raised whether functional activity of the cells of the central nervous system of the cryfish is accompanied by increased metabolic activity.

160 (1907)

The value of intratracheal route of immunization with pneumococcus.

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The ease which with gases diffuse through the respiratory mucosa is well known and is widely utilized in the practice of anesthesia. The absorption from the trachea, however, is not limited to gases. Thus, Mayer[1] found potassium ferrocyanide in

[1] Muller, Manual de physiologie, Paris, 1851, p. 186.

the blood two to five minutes after its introduction into the trachea of animals. Moreover, even such substances which do not easily diffuse through other tissues may sometimes readily pass through the mucosa of the respiratory system. Thus, for instance, Colin introduced intratracheally 10 c.c. of 1 per cent. solution of curare (which is not absorbed from the intestine) killing the dog in 15 minutes. The rate at which fluids may be absorbed from the trachea is remarkable. Colin[2] describes an experiment in which he introduced intratracheally into a tracheotomized horse a continuous stream of warm (30°–35°) water at a rate of six liters per hour for 3½ hours in succession without causing any noticeable discomfort to the horse. When at the end of the experiment the horse was sacrificed the observer could not detect any water in the trachea or bronchi, all water having been thoroughly absorbed.

In spite of this remarkable power of absorption, trachea was not generally employed as the route for the parenteral introduction of foreign substances in the experimental work until recently when Besredka[3] called attention to the fact that trachea constitutes as good and perhaps even a better site of introduction of antigen into the experimental animals than any other employed. During the summer of 1920 one of us had the privilege of personal acquaintance with the work of Besredka and it is this experience that suggested the possibility of utilizing the tracheal route for the purpose of production of immunity to pneumococcus. This investigation was undertaken with the hope that intratracheal introduction of pneumococcus antigen might be particularly advantageous in view of the fact that it suggested the possibility of increasing the local resistance of the tissues of the respiratory system to pneumococcus in addition to creating the state of humoral immunity. The experiments which will be reported later will bear on the question of successful production of increased local tissue-resistance. In this paper we wish to report on the relative value of *intratracheal* route for parenteral introduction of pneumococcus antigen as compared with subcutaneous, intravenous and intrapleural routes and as measured by the concentration of circu-

[1] Colin, Traite de physiologie comparee des animeau, Paris, 1873, t. 88, p. 112.

[2] Colin, loc. cit., pages 109–110.

[3] Besredka, *Ann. Inst. Pasteur*, 1920, xxxiv, 51 and 361.

lating antibodies in animals treated respectively by the four above-mentioned methods. A series of normal rabbits were divided into four groups of two animals each and immunized by the repeated introduction of a suspension of Type I pneumococcus (heated for 30 minutes at 60° C.) by the four above-mentioned routes. In all the animals received 9 injections each, over a period from October 1 to November 17. Special care was taken to maintain the uniformity of the amount of bacterial protein injected in respective series. Ten days after the last injection the blood of all the animals was tested for its bactericidal power, for its opsonic power and for its power to protect mice against infection with virulent culture.

Bactericidal power was tested by the technique of Heist and Solis-Cohen.[1] Bacterial suspension containing 1,600,000 pneumococci per cubic centimeter was diluted as indicated on the chart below.

Rabbit No.	Route of Immunization.	Dilutions of Bacterial Suspension.		
		2:25	1:125	1:625
328.....	Intravenous	++	++	++
329.....	Intravenous	+	+	+
330.....	Intratracheal	−	−	−
331.....	Intratracheal	+	−	−
332.....	Subcutaneous	+++	++	++
333.....	Subcutaneous	++	+	+
334.....	Intrapleural	++	+	−
335.....	Intrapleural	++	−	+
336.....	Normal controls	+++	+++	+++
337.....		++	++	+++

The bactericidal power of the blood is of course inversely proportional to the extent of growth.

Opsonizing power of the respective sera was determined by using mouse leucocytes and was expressed in terms of percentage relation of the number of leucocytes in the state of active phagocytosis to the total number of leucocytes counted.

[1] Heist, George D. and Cohen-Solis, Solomon, *Journal of Immunology*, 1919, iv, No. 4.

+++ Very good growth.
++ Medium growth.
+ Poor growth.
− No growth.

The relative opsonizing power of various sera was found to be as follows:

Rabbit No.	Route of Immunization.	
328	Intravenous	2% to 8%
329	Intravenous	
330	Intratracheal	10% to 20%
331	Intratracheal	
332	Subcutaneous	2% to 4%
333	Subcutaneous	
334	Intrapleural	10% to 20%
335	Intrapleural	
336	Normal (control)	below 1%
337	Normal (control)	

In the test for protection three series of mice received intraperitoneally 1/2 c.c. of the respective sera each. Immediately following the serum mice of respective series received intraperitoneally virulent culture in the amount representing from 12,500 to 50,000 M.L.D.

Route of Immunization.	Number of M.L.D. of Pneumococcus.		
	12,500	25,000	50,000
Intravenous	living	living	+ 24 hrs.
Intratracheal	living	living	living
Subcutaneous	+ 24 hrs.	+ 24 hrs.	+ 24 hrs.
Intrapleural	living	living	living
Normal (control)	+ 24 hrs.	+ 24 hrs.	+ 24 hrs.

161 (1908)

Racial variations in Blepharisma undulans.

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Studies on *Blepharisma undulans* and *Blepharisma lateritia* Stein, have emphasized the impossibility or great difficulty of observing the micronuclei, and also the inevitable death of exconjugants without dividing. Thus Bütschli[1] was unable to identify micronuclei in vegetative specimens of *B. lateritia* though he found them in conjugants. Exconjugants invariably died. Calkins[2] did not find micronuclei in non-dividing *B. undulans*, but

[1] Bütschli, *Abhandl. d. Senckenb. naturf. Gesellsch. Frankfurt a/M.*, 1876, Bd. x.

[2] Calkins, *Journ. Morph.*, 1912, xxiii.