the non-irradiated ones with regard to their properties in the complement-fixation test, namely, anticomplementary power, specificity, and antigenicity; their stability, when kept in the icebox for several weeks, is being tested.

The results of these tests are shown in Table I and can be summarized as follows:

With our special set-up, periods of time of 90 minutes are required to destroy the virulence of the Eastern and Western equine encephalomyelitis antigens, and 40 minutes with rabies fixed, lymphocytic choriomeningitis, and St. Louis encephalitis antigens.

The irradiated antigens are not anticomplementary. The titer of the complement in the presence of irradiated and of non-irradiated antigen is the same.

As shown by the highest dilution of serum exhibiting a 2 plus or better reaction, the titer of a mouse hyperimmune serum against the irradiated and the non-irradiated antigen is the same. The specificity of the reaction with the irradiated antigen is as complete as with the non-irradiated one, as shown in both instances by the absence of any crossing with the heterologous antigens.

And finally, the titer of the antigen following irradiation is the same, or only slightly lower than the titer of the non-irradiated antigen; these titers were determined by testing serial twofold dilutions of antigen with a constant amount of immune serum.

In conclusion, by exposing virus suspensions of Eastern and Western equine encephalomyelitis, rabies, lymphocytic choriomeningitis, and St. Louis encephalitis to ultraviolet light, complement-fixing antigens can be obtained that are non-infective and specific, that lack an anticomplementary effect, and have a high antigenic titer.

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Effect of 2-4 Dinitrophenol on Rat Brain Respiration at 25, and 37.5°C.*

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Measurements of the effects of graded concentrations of 2-4 dinitrophenol (DNP), at equilibrium, on oxygen consumption (concen-

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tration-action curves),[1] have been made on the intact rat[2] and on yeast.[3] However, satisfactory data of this sort do not appear to be available for mammalian tissue *in vitro*. In the present communication such data are furnished for cerebral cortex slices from adult male albino rats. Oxygen consumption was measured at 25° (±0.01°) and 37.5° (±0.01°) C. The methods used have been described previously,[4] therefore only certain features of the procedure, particularly relevant to the problem in hand, will be presented here. Conventional manometric methods (Warburg) were used.[5] The suspension medium was Ringer's-phosphate-glucose, pH 7.35, and all solutions added were of the same pH. The gas phase was oxygen. In each set of 14 manometers there was one control vessel receiving no DNP as well as a thermobarometer. DNP was added to the experimental vessels 30 minutes after the end of the thermoequilibration period. Thus in each case a 30-minute control period (used in the calculation of the stimulation ratio) preceded addition of the drug. Readings were taken every 10 minutes. The rate of oxygen consumption was constant again within 15 minutes after addition of DNP in concentrations evoking increased oxygen consumption and 30 minutes after addition of concentrations which inhibited oxygen consumption.

Respiration was measured in μl, N.P.T., per mg wet weight per hour (Q_{O_2}). In order to express the intensity of action of DNP on oxygen consumption, the stimulation ratio[6] was calculated for each concentration of DNP. This is the ratio of the metabolic rate under the influence of DNP (Q_{O_2} DNP) to that which would exist if DNP were not present (Q_{O_2} C). The values of Q_{O_2} C used were taken from measurements of respiration preceding addition of DNP (control experiments showed that the respiration of rat brain cortex, under the conditions of these experiments, remains constant for several hours when no DNP is added). Thus the stimulation ratio may be written

$$\text{Stimulation ratio} = \frac{Q_{O_2}\ \text{DNP (after becoming constant)}}{Q_{O_2}\ \text{C}}$$

When this ratio is greater than unity, DNP is increasing oxygen

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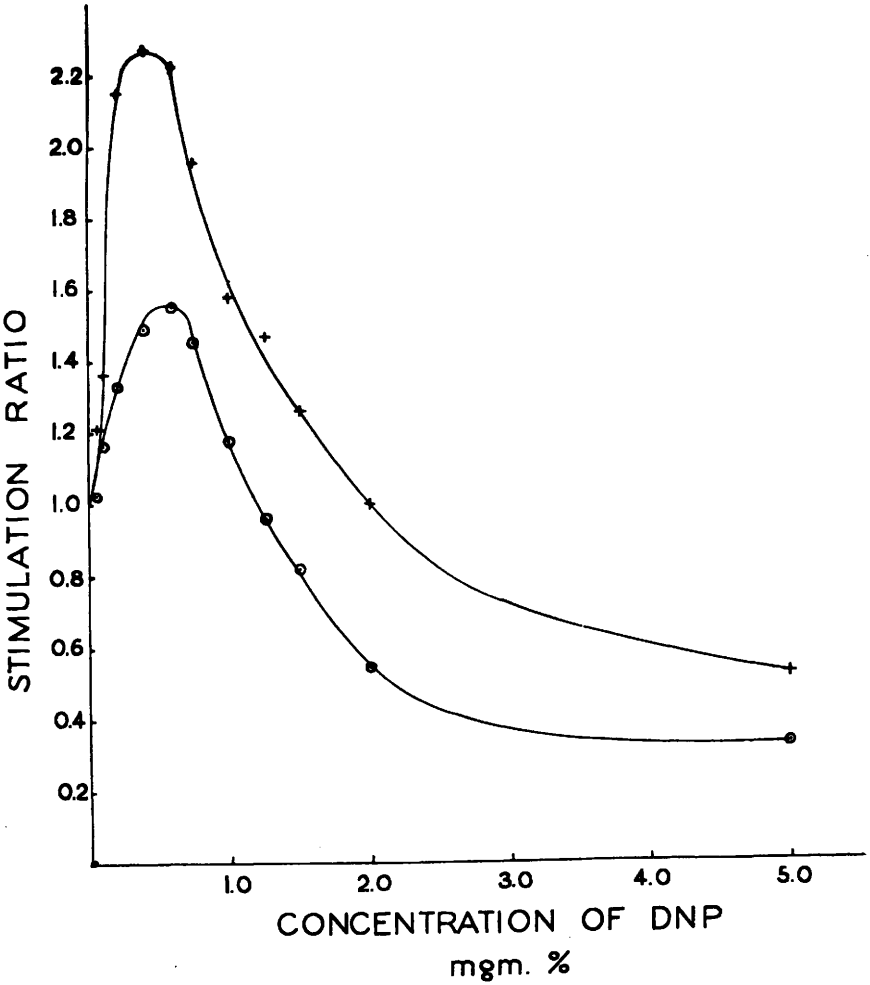

FIG. 1.

Graph showing the intensity of stimulation and inhibition (as the stimulation ratio) of oxygen consumption of excised rat cerebral cortex by graded concentrations of DNP. Crosses give values of the stimulation ratio at 25°C, circles at 37.5°C.

consumption, when less, oxygen consumption is diminished by the drug.

The results are given in Fig. 1, where the stimulation ratios at 25°C (crosses) and at 37.5°C (circles) are plotted as functions of DNP concentration (1.0 mg % sodium 2-4 dinitrophenoxide = 4.46×10^{-5} M DNP). The fact that Q_{O_2} DNP quickly became constant indicates that during this period of constancy equilibrium conditions obtained with respect to the distribution of DNP between the tissue slices and the suspension medium. The partition coefficient

was not determined and for present purposes uniform distribution was assumed.

It is shown in Fig. 1 that the concentration of DNP corresponding to the maximum value of the stimulation ratio is nearly the same at 25°C and 37.5°C (0.4 mg % and 0.6 mg % respectively). However, the optimum concentration of DNP for stimulation of respiration in excised rat cerebral cortex at 37.5°C is much lower than the optimum concentration reported by Tainter[2] for stimulation of oxygen consumption in the intact rat (*ca* 6 mg %). This indicates either that the cerebral cortex is an atypical organ in this respect or that the experimental conditions were so different that direct comparison of the two findings cannot be made. Studies are in progress on other organs to clarify this point.

It is also shown in Fig. 1 that the maximum value of the stimulation ratio and the minimum inhibitory concentration of DNP are both considerably greater at 25°C than at 37.5°C. This is in concordance with our earlier observation[4] that the stimulation ratio corresponding to a single stimulating concentration of DNP (0.75 mg %) varies with temperature and increases with diminution in temperature over a wide range. It is also in accord with the findings of Hall, Crismon and Chamberlin[6] on the intact anesthetized cat.

Conclusions. The optimum concentration of DNP for the stimulation of oxygen consumption in excised rat cerebral cortex is approximately the same at 37.5°C and 25°C. However, the maximum value of the stimulation ratio and the minimum inhibitory concentration of DNP are considerably greater at 25° than at 37.5°C.

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Oxygen Uptake of Blood Serum.*

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In connection with experiments in which solutions containing normal human blood serum were used, it was found that at pH values slightly below 7.0 the serum consumed O_2. This report is concerned with results obtained from experiments performed in an attempt to obtain further information concerning this phenomenon.

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