then used in lantern slides, four pictures or less to a slide, or prints can be made from the negatives. Since the latitude of moving picture film is usually better than ordinary film, sharper results can be obtained.

In photographing pages of loaned articles, either enlargements can be made or a magnifying glass used to read the matter. Each roll of film gives 30–40 exposures; the prints are not expensive and the usual equipment can be used in developing the negatives.

If one is accustomed to a camera, using the adapter does not call for relearning the peculiarities of a new machine, and since the adapter plate is instantly removable, no alterations of the camera itself are necessary.

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ON THE PREPARATION OF HEMOLYTIC AND PRECIPITATING SERA

THE usual directions for the preparation of hemolytic and precipitating sera call for the use of washed red blood cells and serum respectively as antigens. Therefore, the majority of students in immunology leave their classes with the impression that red blood cells are required for the preparation of hemolysins and that serum is essential for the preparation of precipitins. The fact is, however, that an immune serum prepared by using clear serum as antigen will function perfectly as a hemolytic serum and that a hemolytic serum prepared by using washed red blood cells as antigen will do very well as a precipitating serum. One might also use whole blood as antigen in the preparation of these kinds of antibodies. Such a procedure may not always be successful, because rabbits are rather sensitive to fresh sheep serum. It is, therefore, necessary to heat sheep serum for one half hour at 56° C. and to remove fresh serum from the red blood cells by repeated washing in saline solution. Heating whole blood is not desirable. Neither whole blood nor washed red blood cells can be kept for any great length of time. Sterile serum or plasma, on the other hand, can be kept for months or years, thus offering a saving in time and effort, especially to those who may wish to work with hemolysins and precipitins without having ready access to sheep or goats.

Giving rabbits 2, 3, 4 and 4 cc of sheep serum intravenously, allowing 3 to 4 day intervals between injections and 8 to 10 days between the last injection and the bleeding, I have produced sera which, when used as precipitins, showed titers of better than 1:12800. The same sera used as hemolysins dissolved 0.5 cc of a 2 per cent. suspension of sheep erythrocytes in quantities of 0.01 cc of a 1 per cent. dilution and of 0.05 cc of a 0.5 per cent. dilution in the presence of 2 units of complement in 30 minutes at 37° C.

Hemolytic sera produced by injecting rabbits intravenously with 2.5, 3, 3.5 and 4 cc of washed sheep erythrocytes at 3 to 4 day intervals, allowing 8 to 10 days to elapse between the last injection and the drawing of the blood, have given titers as follows, when used as precipitins: slight precipitation in dilutions of 1:6400 in 10 minutes, marked precipitation in the same dilution in 20 minutes, and slight precipitation in dilutions of 1:12800 in 30 minutes. All precipitation tests ("ring tests") were made at room temperature. The hemolytic titers of these sera were almost exactly the same as those given above for the precipitating sera.

Both kinds of sera produce marked hemagglutination when inactivated and mixed with washed sheep erythrocytes.

Rather strong hemolytic and precipitating sera have also been obtained by using as antigen the clear saline solution in which blood cells had been washed, the solution from the fourth washing being about as effective antigenically as that from the first one.

Suspensions of liver and spleen tissue, as nearly as possible washed free from blood, proved inferior to the supernatant saline solution from washed sheep cells when used as antigens for the production of hemolytic and precipitating sera.

Some of the sera tested seemed to retain their hemolytic power longer than the precipitating properties. In others there was no difference in this respect.

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SPECIAL ARTICLES

AN EFFECT OF THE RECENT SOLAR ECLIPSE ON THE IONIZED LAYERS OF THE UPPER ATMOSPHERE

MEASUREMENTS of the virtual heights of the ionized layers of the atmosphere during the recent solar eclipse of August 31 indicate strongly that at least one ionizing agency effective in the lower layer comes from the sun and travels with a speed approaching that of light. It would seem then that ultra-violet light rather than neutral particles was the ionizing agency which caused the phenomena observed in these tests.

The measurements were made[1] at Deal, N. J., using three transmitters and receivers adjusted to the fol-

[1] For description of method and equipment used see J. P. Schafer and W. M. Goodall, *Proc. I. R. E.*, July, 1932.

lowing frequencies: 2,398 kc, 3,492.5 kc and 4,797.5 kc. Tests were conducted during the day of the eclipse and for several days before and after, in order that the data of the eclipse day might be compared with that obtained on other days at this time of the year.

Over Deal the optical or light eclipse was total in the upper-layer, and about 98 per cent. total in the lower-layer. The light eclipse began at 2:24 P. M., was maximum at 3:34 P. M. and ended at 4:40 P. M. (E. S. T.). If the ionization in the lower-layer (E region) were due to neutral particles and if the speed of the particles were 1,600 kilometers per second, the nearest approach of the "corpuscular" eclipse discussed by Chapman[2] would have been much higher than the upper-layer (F region). If the particle speed were, say, 3,000 kilometers per second, this location would have been in the path of the corpuscular as well as the optical eclipse.

Unfortunately, a magnetic disturbance began several days previous to August 31 and was still in progress during the day of the eclipse. For this reason special care must be taken in interpreting the results, since previous experience shows that the ionization of the lower layer tends to increase and the virtual height of the upper layer changes in an erratic manner, when conditions are magnetically disturbed.

For example, while there were variations in the upper-layer reflections which appear to have been due to the light eclipse, we hesitate definitely to attribute these variations to the eclipse because of the disturbed magnetic conditions. For this reason these effects are not discussed in these preliminary notes.

There were, however, some phenomena observed which, on the basis of previous experience, were not due to the magnetic disturbance. The most striking effect was found for the 2,398 kc. frequency. From 8:00 A. M. until 2:00 P. M. nothing unusual was observed. When the eclipse began, lower-layer reflections were being received for the 2,398 kc. frequency. This was the usual condition for the other days of this series of tests. At about 3:08 P. M. the reflections from the lower layer on 2,398 kc. disappeared and weak upper-layer reflections were received. These upper-layer reflections had become strong by 3:19 P. M. and except for occasional periods of short duration remained strong until about 4:10 P. M. At about 4:07 P. M. weak lower-layer reflections returned and increased in strength. At 4:18 P. M. only lower-layer reflections were received and conditions had become similar to those which prevailed before 3:08 P. M.

[2] S. Chapman, *Monthly Notices*, R. A. S., Vol. 92, No. 5, March, 1932; E. V. Appleton and S. Chapman, *Nature*, May 21, 1932.

At no time during this series of tests did the ionization in the lower layer increase sufficiently to return strong reflections for 3,492.5 kc. and 4,797.5 kc. Upper-layer reflections were received during the eclipse period as well as at all other times during the daylight periods of the tests.

The upper-layer reflections on all three frequencies were much stronger during the central portion of the eclipse. As these reflections became stronger, static also increased. This increase in the signal strength of the reflections is normally found when the ionization decreases as sunset approaches. This same effect normally leads to an increase in the static received on these frequencies at sunset, and the increase actually observed during the eclipse seemed to be of the same nature. A complication is introduced, however, by the fact that an approaching thunder storm provided a simultaneous increasing source of atmospheric disturbance.

Everything taken together, the phenomena observed during the eclipse were suggestive of the changes which occur in the lower layer during the late afternoon from about two hours before sunset until sunset, followed by the changes which occur during the corresponding period after sunrise.

If the ionization in the lower layer were due to ultra-violet light, the minimum in the ionic density and hence the most pronounced effect on the lower-layer reflections should have occurred a few minutes after the maximum of the light eclipse. If, on the other hand, the lower layer is produced, as has been suggested by Chapman, by neutral particles emitted from the sun, the minimum in the ionic density should have been observed near the end of the corpuscular eclipse.

Owing to the difference in the speed of the neutral particles and that of light, and to the relative motion of the moon and earth, the center of the corpuscular eclipse should occur before the center of the light eclipse. For a particle speed of 1,600 km. per second, this time difference, which with good approximation varies inversely with the speed, would be two hours.

The actual ionization minimum was observed at approximately 3:45 P. M., which is consistent with the assumption that the ionization is due to ultra-violet light. In order for the corpuscular eclipse to have caused an ionization minimum at this same time, it would have been necessary for the center of the corpuscular eclipse to have passed over Deal at 3:15 P. M., since the duration of this type of eclipse would be about one hour in this latitude. Thus the center of the corpuscular eclipse would have occurred about 19 minutes before the center of the optical eclipse. It follows, then, that if the neutral particles are responsible for the ionization in the lower layer, their speed

would have to be of the order of 10,000 km. per second.

Since there is little or no evidence to justify such a high speed for the particles, and since the observed results agree with those predicted for the light eclipse, it seems that ultra-violet light, rather than neutral particles, is the ionizing agency which caused the phenomena observed during the eclipse.

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THE STABILITY OF FUNCTIONALLY DISTINCT RACES OF FIBROBLASTS

IT has already been shown[1] that common connective tissue cells, or fibroblasts, do not occur throughout the organism as a separate and distinct type. Fibroblasts, as a group, comprise many different cell types which, according to their origin, can be distinguished by the physiological properties manifested under conditions of cultivation *in vitro*. They differ with respect to the growth energy that they exhibit in a given medium, the amount of acid liberated into that medium, their ability to digest fibrin, etc. Thus, although morphologically similar, a fibroblast from skeletal muscle is as different from a heart muscle fibroblast as a pigment cell of iris epithelium is different from a colloid-producing cell from thyroid epithelium. Furthermore, it has been found[2] that fibroblasts originating from embryos of different ages show different nutritional properties, even when the cells are removed from the same part of the embryo. For example, cell strains derived from a 20-day-old embryo may show, under the same experimental conditions, a rate of cell multiplication that is either higher or lower than that exhibited by similar strains isolated from a 10-day-old embryo, depending upon the particular organ or tissue from which the strains are obtained. In the present communication, it will be shown that, after separation from the body, each cell type retains indefinitely the properties possessed at the moment of isolation.

Two groups of experiments have been made. The first group was designed to test the permanence of differences due to spatial relationships, that is, those determined by the particular organ or tissue supplying the strains; the second, to test the permanence of differences of a temporal nature, or those conditioned by the age of the embryo from which the tissues are taken. The material used in connection with the first set of experiments consisted of two strains of fibroblasts derived from a 13-day-old chick embryo, one being obtained from the ventricle of the heart, the other from the periosteum of bone. Both strains were subjected to the same treatment for 18 passages (97 days), during which time they were cultivated in an appropriate mixture of chick plasma, chick embryonic tissue juice and Tyrode solution. At the 19th passage, the two strains were placed in media of different composition, the heart fibroblasts being cultivated in a medium containing much more than the usual amount of embryonic tissue juice, while the fibroblasts from the periosteum were subjected to a medium in which the percentage of tissue juice was greatly diminished. This treatment, which was continued for 4 passages (26 days), resulted in an increased rate of cell multiplication on the part of the heart fibroblasts with a corresponding retardation in the case of the bone fibroblasts. At the end of four passages, when the heart fibroblasts were dividing at a much higher rate than the starved bone fibroblasts, the two strains were again placed under the same nutritional régime. Almost immediately, they reverted to their original rate of division, and after three passages on a common medium they both displayed the same characteristics that had distinguished them before it was attempted to alter their properties by subjecting them to diverse media.

The second group of experiments, being designed to test the permanent nature of nutritional properties determined by the age of the embryo from which the fibroblasts are derived, were carried out in much the same manner as those described above. Two cell strains were isolated simultaneously from skeletal muscle removed from the lower limbs of 8- and 17-day-old chick embryos. Both strains were cultivated under the same environmental conditions. The cell strain derived from the 17-day-old embryo showed a rate of cell multiplication that was consistently higher than that manifested by the strain derived from the younger embryo. The degree of difference was as great as that found to exist between the heart and bone fibroblasts referred to above. The accompanying diagram shows the division rate of the two strains in their fourth passage (Fig. 1). At the previous transfer, one half of each culture comprising the two strains was placed in an experiment in which the 8-day strain was given access to a larger quantity of embryonic tissue juice than it had previously received, whereas the 17-day strain was placed in a medium containing much less than the usual amount. The result was similar to that obtained with the heart and bone fibroblasts. The rate of cell multiplication of the 8-day strain, that had originally been much lower than that of the 17-day strain, now became the greater of the two (Fig. 2). After this treatment had been

[1] R. C. Parker, SCIENCE, 76, 219, 1932.
[2] Unpublished experiments.