

side of it allows the air to circulate and prevents overheating. The asbestos is perforated for wiring, then slipped into place and the lamps are mounted upon it with the cords carried out behind as indicated.

The box shown in back view at the right (B) is made like the other except that in it there are three strips of asbestos with open spaces between them. Each strip is three inches wide and has one lamp mounted at the center of it. Several specimens may be lighted at one time by this box or six to eight lantern slides may be shown. A cheap support for the lantern slides can be made from a sheet of galvanized tin of the same size as the front of the box. When the windows are cut in this the tin at their upper and lower edges is folded back so as to make a groove. Above and below the openings in these grooves the slides may be moved and held in place. When finished the upper edge of the sheet is clipped to the top of the box for support. Exhibits intended to be at all permanent are improved by masks which conceal the extraneous accessories and extra light. Such masks are made of medium-weight black paper in which openings are easily cut. The paper is then lightly pasted to a sheet of window-glass a little larger than the front of the box against which it is supported. Labels receive light enough to be easily read if they are placed flat on the table in front of the openings in the mask.

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CONTROLLING DAMPING-OFF WITH ELECTRIC LAMPS

THOSE who have had difficulty in growing seedlings because of the damping-off fungus during dull, moist weather may be interested in a simple method that has given complete control on pure lines of cucumber seedlings which are very susceptible to the fungus. Pure lines which have been weakened by selfing are slow to germinate during the autumn and winter months because the soil does not receive the sun's heat as it does during the summer. Also, old seed will not germinate well unless optimum conditions are supplied. Second generation seedlings which segregate for certain characters are worthless if damping-off takes the weaker ones.

Previously the soil has been sterilized by chemicals or heat, with varying degrees of success. Where seedlings are germinated every month this method becomes laborious, and considerable time must elapse before the seed can be planted. The sterilized soil becomes infested in a short time so that not more than two lots of seedlings can be grown for each sterilization.

A method has been in use whereby seed is germinated under 200-watt Mazda lamps suspended two feet above the seedlings. A dome reflector concentrates the light and heat so that two hundred seedlings can be grown under one lamp. A mixture of half sand and garden soil is used because it affords good drainage and reduces nitrification. This soil, without the lamps, controls the damping-off if sunlight is not reduced too much and the air does not remain too moist, but the lamps are needed after autumn begins. The seedlings become spindly if a rich garden soil is used.

The lamps are lighted as soon as the seed is planted and are not turned off except on bright sunny days. After the cotyledons have unfolded, the lights may be discontinued if the weather is bright and the surface soil is kept dry. It is preferable, however, to use the lamps until the seedlings are transplanted. With unfavorable growing conditions the potted plants may be exposed to the lamps several hours in the evening until the plants are large enough so that there is no further danger of damping-off.

The lamps have been used to advantage on selections that produce only pistillate flowers during the winter months. These selections may be selfed if the potted seedlings are exposed to the electric light until four or five true leaves are formed. Sufficient staminate flowers will be produced so that the first few pistillate flowers may be self-pollinated. An extra generation can thus be grown for those characters that are not influenced by environment.

Corn and lettuce grown by this method produce sufficient seed for an extra generation. The method has considerable application in northern latitudes where only the vegetative stage of adapted greenhouse plants can be grown during the winter months.

The advantages of the lamps, in addition to controlling damping-off, are that the soil is warmed so that weak or old seed germinates better in a shorter time, and the germinating can be done in a cool greenhouse without increasing the temperature of the greenhouse.

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SPECIAL ARTICLES

CINEMATOGRAPHS OF LIVING DEVELOPING RABBIT-EGGS¹

RABBITS' eggs of definite ages after mating; 21²/₃, 22, 23¹/₂, 67, 69 and 71 hours, were washed from

¹The rabbits for these experiments were supplied by Professor W. E. Castle, of Harvard University.

the tubes with Locke solution, mounted in blood plasma or modified plasma on flat slides with supports under the cover-glass, sealed with paraffin and kept on the microscope stage in a warm box under the camera. Autoplasma and homoplasma were better than chicken plasma. The preparation of the mounts (cutting out the tubes, washing the eggs out with Locke solution at room temperature, picking up the five or six eggs of a litter with a small pipette and mounting each egg separately in plasma) took from one half hour to one hour. During most of this period the eggs were at room temperature, hence their development was probably somewhat delayed. The magnification on the cinematographic film was forty-four diameters. Exposures were made at the rate of two and one half per minute.

Under normal conditions *in vivo* the rabbit's egg segments into two cells at 22 to 25½ hours after mating, into four cells at 25½ to 32 hours, into eight cells at 32 to 41 hours, and into sixteen cells at 41 to 47 hours. The cleft which forms the beginning of the segmentation cavity appears at 68 to 76 hours after mating, and a small cavity develops within an hour or two. The inner cell mass begins to show at about the same time, *viz.*, 70 to 76 hours. The segmentation cavity increases slowly in size, the trophoblast walls become thinner and thinner, and the whole egg also increases slowly in size until about 90 hours after mating when the egg rapidly increases in size and the zona pellucida becomes stretched and thin.

I. BEGINNING WITH THE ONE-CELL STAGE

Several eggs in the one-cell stage were obtained from three animals that were killed 21²/₃, 22 and 23½ hours after mating. Those that were followed divided into two cells from one to three hours after they were taken from the tubes, or from 24½ to 24¾ hours after mating. If one hour delay due to cooling be deducted the divisions fall well within the normal period (22 to 25½ hours) after mating. Three of the eggs were in the four-cell stage at 28 to 30 hours after mating and were also well within the normal period of 25½ to 32 hours. Two were in the eight-cell stage at 39 and 42:05 hours after mating. They also indicate that the progress of cell division was about normal.

The cleavage of egg C50-2 which was taken from the tube 23 hours and 30 minutes after mating was as follows: at 24:45 it was in the two-cell stage, at 29:06 in the three-cell stage, and at 29:45 in the four-cell stage, each of the first two blastomeres divided once thus giving rise to two pairs. One cell of the first pair of blastomeres of the four-cell stage

divided at 35:50 and the other at 36 hours after mating. One cell of the second pair of the four-cell stage divided at 38 and the other at 39 hours after mating. The eight-cell stage was thus brought about by dichotomous division. There was an interval of about four hours between the two- and the three-cell stage and of one hour between the three- and the four-cell stage. Between the four- and the five-cell stage there was an interval of about six hours. The intervals between the five- and the six-, the six- and the seven-, the seven- and the eight-cell stages were from ten minutes to two hours each, the longest interval occurring between the six- and the seven-cell stages. Are such differences in the time of division significant and do they indicate differences in the cells? The fact that one of the first two blastomeres divided about an hour before the other may indicate that already at the first cleavage a segregation of materials has taken place.

There are other indications that the cells differ from one another. Four eggs in the two-cell stage were cinematographed and in each case the two cells were of unequal size throughout this stage. The cinematographs of two of the eggs show that the largest cell in each egg was the first one to divide. The other two eggs were not recorded. In the four-cell stage the first pair of cells are usually larger than the second pair. In the eight-cell stage the cells of the first quadruplet, that is the four cells derived from the first pair of the four-cell stage and the largest cell of the two-cell stage, are larger than those of the second quadruplet.

During the eight-cell stage of egg C50-2 the cells of the first quadruplet were nearly spherical in form and occupied more than half the space within the zona pellucida. They remained as a distinct group with triangular spaces between them and the zona pellucida. The second quadruplet massed themselves more closely together, lost their spherical forms and as a result the cell limits were not clear. This mass became somewhat flattened out against the zona pellucida.

The cinematographs also reveal a certain amount of shifting of the cells, especially after each division. The movements of the granules within the cells became more pronounced shortly before the cell division. The cells also seem to undergo peculiar pulsations or changes in size.

Some of the eggs that were started in the one-cell stage continued to live for about two days after they were taken from the tubes. We have not yet determined just how far it will be possible to carry them or how much of the growth is normal.

II. BEGINNING WITH THE LATE MORULA STAGE

The eggs taken out from 67 to 71 hours after mating lived for a much longer time than those that started in the one-cell stage. Two .67-hour eggs (C52-1 and C52-2) continued to live and increase in size for about eight days after they were taken from the tubes. Egg C52-2 remained in the morula stage for several hours. The number of cells seemed to increase. At 76 hours and 20 minutes after mating, or 9 hours after removal from the tube, part of its periphery was surrounded by columnar epithelial-like cells with a cleft at one place along their inner border. A small cavity was visible. This is about the normal time for the appearance of the segmentation cavity especially if one hour be deducted for the delay when the egg was at room temperature. During the next hour the extent of the columnar epithelial-like cells, the trophoblast, increased and the segmentation cavity enlarged. The inner cell mass was not in a favorable position for observation. During the next 1½ hours ending at 79 hours after mating the segmentation cavity increased in size and the epithelial-like cells of its walls became cuboidal. During the next two hours the cavity increased still more in size, the trophoblast wall became thin and the entire egg had enlarged somewhat. During the next seven hours the egg continued to expand and the zona pellucida became thin. During this period the egg occasionally contracted rapidly in the course of two or three minutes to more nearly its original size and then slowly expanded again. This phenomenon was probably due to the stretching of the rather tough elastic zona pellucida by the accumulation of fluid under pressure within the segmentation cavity, until a small break occurred sufficient to allow the escape of fluid from the cavity, and as the tension was relieved the zona pellucida contracted down to about its original size. The cells of the trophoblast then apparently healed over the break and the process was repeated again and again. During contraction the thin trophoblast became thicker only to thin out again as the egg expanded. At 98 hours a small cellular hernia was noted. This slowly increased in size as it bulged out through a distinct break in the zona. As the hernia increased in size the tension on the zona pellucida was relieved and it returned to about its original size and thickness.

From now on during the next six days the hernia continued to expand, and reached its greatest extent (.555 mm diameter) about ten days after mating, or between seven and eight days after the egg was taken from the tube. The segmentation cavity was early observed to extend into the hernia or exovate.

Its walls consisted partly of thin trophoblast and partly of thicker material, probably the inner cell mass which had apparently also escaped out into the hernia. The old zona pellucida with its lining of thin cells and small segmentation cavity formed a very insignificant part of the whole mass at the end of the period.

During this period of expansion of the hernia there were rapid variations in the size of the sac. An explanation similar to that given for the expansions and contractions of the zona during the earlier stages probably applies here also. Fluid was evidently secreted under pressure into the segmentation cavity by the trophoblast. When the pressure became too great a small break probably occurred and the elastic recoil of the elastic and tense trophoblast cells expelled fluid until an equilibrium was reached. The cells then healed over the break and the expansion began again. The expansions and contractions were revealed by the cinematograph. Such variations would hardly have been discovered as readily by any other method. It was not possible to follow with the eye any differentiation of the inner cell mass and the cinematograph did not help in this respect.

After the eighth day of incubation the egg took on an unhealthy aspect and decreased considerably in size. We are not quite sure just how long it continued to live.

A second egg (C51-1) from the same rabbit was carried along at the same time on a separate mount and filmed at intervals. It behaved very much as did the first egg. The inner cell mass, however, was located in a more favorable position and could be followed until it passed out into the hernia, where it was lost from view. The cells of the inner cell mass of the second egg appeared to be in a constant state of agitation as long as it was visible. The hernial sac did not become quite as large as the first one but the egg continued to live about as long.

Both films showed what appeared to be the ameboid cells, probably endodermal cells migrating about on the inner surface of the thin trophoblast.

Under normal conditions the eggs enter the uterus 72 to 80 hours after mating and soon undergo rapid expansion. In order that the latter may take place it is probable that some of the secretions of the uterus alter the character of the zona pellucida so that the expanding egg can stretch it into a thin membrane without exerting undue tension and producing rupture. Without such a softening of the zona pellucida normal development probably can not take place.

The great advantage of the combination of the

