

are unsatisfactory to use, because of the uncertain factor of variable climatic conditions from year to year; but some comparisons might have been made between the yields obtained during recent years from land that has long grown wheat and the yields on virgin or nearly new fields on similar soils. Or, lacking such data, it would have been helpful to the reader had there been given some statement as to the present wheat yielding capacity of the fields from which the samples were obtained.

Unless it is shown definitely that the maintenance of the nitrogen and humus content of these Cache Valley soils is correlated with the maintenance of their wheat yielding capacity, these investigations lose much of their possible value.

As to the matter of the farming methods for wheat production on this Cache Valley land, it is the general practise to harvest the grain with a header or with a combined harvester and thresher, either of which implements leaves on the land the major portion of the grain straw, which is subsequently plowed under.² Mr. Stewart makes incidental reference to this feature of the agricultural practise in the Cache Valley, but he does not make it clear that in this respect that practise is essentially different from what it is in the dry-land wheat regions of the Great Plains and eastward, where it is the custom to harvest the grain with a binder and remove the larger part of the straw. This omission seems particularly unfortunate, in view of the general, and possibly misleading, inferences that may be drawn from Mr. Stewart's otherwise valuable contribution to knowledge. If, as it seems reasonable to believe, the true explanation of the observed humus maintenance lies in the practise of plowing under each year the large amount of wheat straw, it becomes apparent that similar results are not to be expected where a similar practise is not followed.

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² See Bulletin No. 103, Bureau of Plant Industry, U. S. Department of Agriculture, pp. 31-35, issued May 31, 1907.

SPECIAL ARTICLES

SOME EXPERIMENTS ON THE PRODUCTION OF MUTANTS IN DROSOPHILA

MACDOUGALL has reported the successful production of mutations by treating the ovaries of certain plants chemically or osmotically. As long as the full account of his results is not available, it is not easy to judge to what extent it is possible to produce mutations at desire with his method. Tower has apparently succeeded in producing in various species of *Leptinotarsa* certain color mutations at desire by submitting the beetles, during the period of the growth of the eggs, to different degrees of temperature and moisture from those in which they usually live. Gager mentions that by treating the pollen or ovaries of *Oenothera* with radium, some of the new plants were entirely different from the mother plant. Morgan has published the statement that a number of the interesting mutations of *Drosophila*, which he has recently described, came from a culture which had been treated with radium.

The following experiments were undertaken for the purpose of forming a conception concerning the degree of certainty with which mutations can be produced experimentally. We tried the effects of a constant and comparatively high temperature, of radium and of Röntgen rays. The stock of *Drosophila* which we used in these experiments was given us kindly by Dr. Lutz, to whom we wish to express our thanks.

1. *Effects of High Temperature.*—Several culture dishes with *Drosophila* were put into a thermostat, the temperature of which remained constant within 1° around 30.5° C. We found that at higher temperatures we lost a large number of cultures. In the fifth generation of flies, kept in the thermostat, on February 16, a number of dark flies appeared. They were mated with normal ones of the same culture. Some of these cultures were kept in the thermostat and others were brought into room temperature, to see whether at a lower temperature they would continue to breed true. This has now been the case for five

generations. Darkness is recessive to the normal yellow and is not sex limited. Our dark mutation is possibly identical with Morgan's "melanotic" mutant.

On the seventh of March we began to repeat this experiment with the necessary control at room temperature. On April 10, we found in the first filial generation of the control culture kept at normal temperature a dark specimen. None of eleven new cultures kept in the thermostat have thus far given rise to a dark or any other type of mutant. Since then dark individuals were found in another control culture.

From these experiments we must draw the conclusion that a constant temperature of 30.5° does not necessarily produce mutations in *Drosophila*, and second, that a dark form of *Drosophila* may arise "spontaneously," that means by forces at present unknown.

2. *Experiments with Radium*.—A very large number of experiments with radium were undertaken, because it happened that the first culture which we treated with radium chanced to give us mutants. We succeeded in producing short-winged specimens in two different cultures by treating them with radium; while thus far we have not yet observed this mutation in cultures not treated with radium. The manner of appearance of this short-winged mutation was in both cases the same. In the second filial generation of the flies treated with radium, one or more short-winged males appeared. The various forms of mating were tried and yielded the result that the short-winged condition is a sex-limited character. The wild normal males were found to be heterozygous in regard to short wingedness. Thus our short-winged mutant behaved like, and is probably identical with the "miniature"-winged mutant discovered by Morgan. We have now bred the short-winged males and females for five generations and find that they remain constant.

We expected that we might succeed in producing short-winged mutants at desire, but in this we failed. Although we treated more than two hundred different cultures with radium we only observed the appearance of the

short-winged mutation in the two cultures, although we repeated the conditions of our successful experiments quite frequently. In both successful cases we submitted the animals only for one or two hours to the action of radium. In one of the two cases the newly-hatched imago alone, males and females were treated for two hours with a weak radium preparation (10,000 units) which was coated with collodium. It is possible that the alpha rays may have affected the animals. In the second successful case a strong radium bromide preparation (over 1,000,000 units) in a glass tube was applied for one hour to a mixture of imago, eggs and young larvæ.

In five different cultures of flies treated with radium the dark mutation appeared, but, while the short-winged mutants appeared in both cases in the second filial generation, there was no regularity in regard to the appearance of the dark mutants.

In one culture treated with radium a white-eyed female appeared in the first filial generation; it is possible that the existence of a white-eyed male in a previous generation may have escaped our notice. In two radium cultures we observed the pink-eyed mutants, but this was also found in cultures not treated with radium.

3. *Experiments with Röntgen Rays* have given us thus far no mutants.

Our results can be summarized as follows:

1. A large number of cultures of *Drosophila* were treated with high and constant temperature, with radium, and with Röntgen rays. Four types of mutations were observed; a dark form (which was the most common), a pink-eyed, white-eyed and short-winged form.

2. In the control cultures, which had not been treated, the dark and the pink-eyed mutations were also observed. As far as the white-eyed mutation is concerned, it is probable that it originated before the treatment of the culture with radium.

3. The short-winged mutants have appeared thus far only in the cultures treated with radium, namely in two cultures out of several hundred. We did not succeed in producing the short-winged mutation at desire by treat-

ing the cultures with radium.

We wish to express our thanks to Mr. Berlinicke, of the firm of Hugo Lieber & Co., who was kind enough to loan us the radium used in these experiments, and to Mr. Bagg, who assisted us in our observations.

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AN EXPERIMENT IN DOUBLE MATING

IN my "Inheritance in Silkworms, I," (1908)¹ I called attention (pp. 37-39) to the beginnings of an experiment in double mating. Only the F_1 generations following a few matings had been obtained at that time, but they gave such promise of interest that I determined to continue the experiment and to widen it. I have now in hand the notes on 85 silkworm broods belonging to this double mating experiment series of 1910. Some of these broods are the F_1 generation from the original 1907 double matings, while others are F_2 or F_1 generations from the original 1908 or 1909 double matings. Taken together the notes of these various 1907-10 rearings from double matings are sufficient to pose some suggestive queries.

By the double mating of silkworms I mean the mating of a female of one race with two males representing different races, one of them usually of the same race of the female, the other of another race. Races are chosen which are readily distinguished by a difference in cocoon color, as yellow or white, or in larval pattern, as banded and unbanded. The silkworm is polygamous and polyandrous, both males and females usually mating more than once before egg-laying begins. Or this repeated mating may continue after egg-laying has begun.

Moths to be experimentally double-mated are reared from carefully isolated cocoons, and

¹"Inheritance in Silkworms, I.," *Leland Stanford Junior University Publications*, University Series, No. 1, 89 pp., 4 plates, 2 text figs., 1908. Address Librarian, Stanford University, California.

the two matings are made to take place immediately following one another for equal or definitely determined unequal periods of coupling, and always before any egg-laying by the female. The young produced from the eggs of each double-mated female are reared isolated in separate trays, which are covered over during the later larval life (possible straggling time).

In any consideration of the results of such repeated mating the unusual way in which the eggs of insects (at least of the silkworm moth and hosts of others) are fertilized must be remembered. This way is, simply, that the male fertilizing cells, the spermatozoa, are received by the female at mating into a special sac or receptacle, the spermatheca (there may be several spermathecae, as in flies) in which the spermatozoa remain alive and active. This spermatheca, a diverticulum of the oviduct, is situated near its external opening, the vagina. As the unfertilized eggs of the moth pass slowly down from the ovarian tubes into the oviduct they lack only fertilization to be entirely ready for development. They have already their full supply of yolk, they are already enclosed in their protecting envelopes (vitelline membrane and outer, firmer chorion). But these envelopes do not completely enclose the egg-mass; there is, at one pole of the egg, one or more small openings, the micropyle, through which the spermatozoa, issuing from the duct of the spermatheca as the eggs pass, enter the eggs. As soon as a single spermatozoan has entered, a jelly-like substance closes the micropyle and prevents polyfertilization.

Thus when the silkworm moth first mates she receives in her spermatheca, and holds there, a considerable number of spermatozoa representing the heritable characters of the male involved. When she couples again she receives another lot of spermatozoa, and if the second coupling is with a male of different race from the first these spermatozoa represent a new set of characters. What is going to be the result of this double mating as exhibited in the offspring?

It seems, at first thought, that this result