Resumen por el autor, Stefan Kopeć.

Autodiferenciación fisiológica de los gérmenes de las alas injertados en orugas del sexo opuesto.

El gérmen de las alas imaginales injertado en ejemplares de orugas de Lymantria dispar L. del sexo opuesto continúa desarrollándose y las alas diferenciadas a sus expensas presentan su tinte dimórfico propio, en vez de exhibir el del individuo sobre el cual se desarrollan. Los pigmentos de la escama no proceden, por consiguiente, directamente de la sangre desecada, sino que son el producto de ciertos cambios químicos, los cuales, según Mayer, tienen lugar en la sangre bajo la influencia de substancias específicas contenidas en las células formadoras de las escamas.

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PHYSIOLOGICAL SELF-DIFFERENTIATION OF THE WING-GERMS GRAFTED ON CATERPILLARS OF THE OPPOSITE SEX[1]

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The development of the pigment in the wings of moths has not yet been sufficiently studied; there is, however, no longer any doubt that the chief ingredient in the formation of the pigments is the blood, or the so-called haemolymph of the animal. How this formation takes place has not yet been ascertained; some authors believe that the pigment is directly caused by the drying up of the haemolymph. On the contrary, according to Mayer ('96, '97), there are certain ferments in the scale-forming cells which render the formation of the pigment possible. On this view the pigment is the product of certain changes which occur in the insect's blood, under the influence of special ferments.

Crampton ('00) is inclined to Mayer's opinion, but I think that his notable experiments are not sufficient to demonstrate it. Crampton united various parts of the pupae of the moth Callosamia promethea by means of paraffin, so that the front part of the body obtained belonged to one sex, the hind part to the other. These bodies developed further until the stage of the adult moth, when each part of the artificially united body showed its specific and dimorphic color, different in front and behind. "We must conclude, therefore," says Crampton, "that the production of the sexually-different ground-colors of the adult moths is determined by some 'ferment' factors which differ in the two sexes, and that the difference in the adult colors is not due to a difference between

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the respective haemal fluids of the two sexes." Crampton's investigation itself, however, does not justify our drawing such a conclusion; at the same time it is quite true, as we shall soon convince ourselves. First, the united parts were relatively large bodies, very thoroughly and abundantly supplied with blood, this being different in each part; it seems to me very probable, therefore, that in the cases given there was no thorough mingling of the two haemolymphs in the artificially joined organism; since it is quite possible that these did not mix at all, Crampton's conclusion does not seem to me to be proved. Secondly, the connection of the chrysalides was possibly made at too late a stage for this supposed influence of one part on the other to be visible. I tried, therefore, to verify Mayer's theory by a somewhat different method. At the same time my experiments were intended to show the morphological and physiological self-differentiation of the insect wing still more distinctly than it has been shown in the experiments on the castration of caterpillars. (See below.)

I removed the germs of the first right wing from male caterpillars of Lymantria dispar L. after their last moult, and in their place I grafted a similar germ from a female caterpillar, and vice versa. It was necessary that both caterpillars should be of about the same age, so that the time of pupation of the one operated upon would coincide most strictly with the time at which the implanted wing of the opposite sex should attain the stage of metamorphosis. Naturally, the fulfillment of this condition was extremely difficult, and on the other hand the metamorphosis of the grafted wing always occurs quite independently of the organism on which it has been implanted. Therefore, if the condition mentioned has not been fulfilled, the implanted wing undergoes metamorphosis at a different time than the caterpillar on which it is grafted and cannot easily come forth and develop subsequently. If we take into consideration also that the delicate germ was often injured during transplantation, it is not astonishing that out of 120 specimens operated upon, the implanted germ developed into the pupal and imaginal wing only in two cases. In these two cases we had to

do with male wings developed on female organisms. The grafted wings were somewhat folded and could not be drawn out of the pupal integument by the moth alone. During the extraction of these wings the scales, especially from the upper surface, nearly all remained in the pupal skin, from which they could very easily be removed uninjured. The form and color of the scales were afterward studied exactly under the microscope.

In one case the form of these scales was quite normal, in the other the scales were less deeply dentated than in normal conditions, which seemed to show that they were not completely developed. The color of these wings corresponds to the dimorphic hue which the transplanted wing would have had if it had remained intact in the previous organism; therefore, the grafted wing was in both cases otherwise colored than the normal left wing of its foster-mother. The wings of the male, which had been transplanted on the female, had more or less dark gray or dark brown scales, in contrast to the female wings, which were for the most part white.

I think the results of these experiments certainly speak in favor of Mayer's theory, as it turned out that the cells of the wing germs were able to collect the respective dimorphic pigments in their scales, forming them from the heterogeneous blood of the other sex in the new surroundings. Thus the pigments of the scales are not directly derived from the desiccation of the haemolymph, but must be the outcome of certain chemical processes occurring in the insect blood under the influence of substances formed in the scale-producing cells, and considered by Mayer to be ferments. Owing to these substances, which are different in different parts of the imaginal wing, various pigments are formed in the blood of the insect, and this results in the production of complicated figures on the wings. These substances develop by means of physiological self-differentiation, and their formation is outside the influence of the haemolymph of the other sex. This fact is unexpected, since the investigations made by Dewitz ('09–'16b), Steche ('12 a, b,) and Geyer ('13, '14) have demonstrated that the blood in insects of one and the other sex is not the same, but shows great differences in the

chemical qualities as well as in its coloring. It happens that the differences are not important enough to have a decisive influence on the hue of the insect wing. Consequently, the real cause of the color of the wings of dimorphic moths, being distinct for the two sexes, is not the difference of the blood, but the difference of the substances which are present in the cells of the germs of the wings. The physiological self-differentiation of the corresponding cells takes place early, even before the pupation of the caterpillar. In this stage the germ of the yet unformed wing seems to resemble in some degree an exposed but as yet undeveloped photographic plate.

The self-differentiation of the wing of moths, as I have stated it, is sufficient to interpret the known results on the castration of caterpillars, which does not lead to even the slightest changes in the dimorphic coloring of the wings of the adult moth (compare the experiments of Oudemans, '99; Kellogg, '04; Meisenheimer, '07, '09; Kopeć, '08, '11, '13, and Geyer, '13). Prell ('15 a), having castrated caterpillars of the moth Cosmotriche potatoria L., observed, on the contrary, a much larger variability in the direction of a lighter hue in the wings of castrated males than in normal specimens. Prell does not question the negative results of the experiments on the castration of moths hitherto performed, and obtained chiefly on the species Bombyx mori L. and Lymantria dispar L., but he believes that the results ought not to be extended to all forms of moths. It is possible that further investigations made in this direction would lead us to distinguish between those forms of moths which react and those which do not react after castration by showing a change of color of the wings. But the experiments hitherto made by Prell are too small in number to draw any certain conclusion from them. In Prell's investigations we also miss standard experiments, such as might exclude the possibility of the influence of the operation itself, whether the sexual glands were removed from the insect body or not. According to Prell's own words, the castrated caterpillars refused food, and they had also to be far more abundantly sprinkled with water. If we bear in mind that they were subjected to a powerful ether narcosis and that they often lost a

large quantity of blood during the operation, we can readily understand that they might have been much weakened by the operation and have responded more readily to all changes of external conditions, such as temperature, moisture, etc., even when these were imperceptible to us. In contrast to Lymantria dispar, Cosmotriche potatoria belongs to those species of moths whose dimorphic wing-colors undergo distinct changes under the influence of cold; to this difference Prell ascribed his results, which he considers different. The later investigations of Prell ('15 b) on the castration of various Vanessae, the classical material for the study of the influence of temperature, do not seem to support this opinion. Castrated Vanessae as well as castrated females of Cosmotriche potatoria undergo no changes after castration, in contrast to males of the latter species. In this behavior of the Vanessae, Prell ('15 b) sees a proof that in the experiments on males of Cosmotriche the change in hue was not excited by the operation itself, but that we have here to do with the effect of the removal of the sexual glands; for if the lighter hue of the wings of the males of Cosmotriche were to depend on the operation itself, it would have to be admitted, according to Prell, that similar changes would appear also among the Vanessae operated upon, as they are even more sensitive to the influence of external conditions. I believe, however, that we might just as well suppose a different reaction power of the moth to castration, possibly different in the two sexes or in various forms of moths, as a different behavior of various organisms, in respect to their change of color, affected by debility and supersensitivity resulting from the operation. In this way the results obtained by Prell may be made to accord with the results of the researches of Oudemans, Kellogg, Meisenheimer, and of my own.

While some authors have seen certain contradictions in the results of different investigations, which, according to them, prove the influence of castration on the dimorphism of the moth, others have drawn the conclusion, from all these experiments, that the germ of the wings is already differentiated early in the larval life, and hence the removal of the gonad or the implantation of glands of the other sex cannot change anything in the

coloring of the insect wing. The results of my experiments on the transplantation of the wing-germs confirm the latter conclusion, showing at the same time the principle in virtue of which these animals have quite a different position in regard to the development of their dimorphic secondary characters from that of vertebrates. Steche ('12, a, b) and Geyer ('13), who relied on their experiments on the dimorphic differences of the insect blood, expressed the opinion that the fundamentally different behavior of insects and vertebrates after castration is caused by the fact that the whole body (soma) of the former undergoes sexually dimorphic differentiation from the beginning of life. In the light of my own experiments described in this paper this hypothesis gains a new and important confirmation.

SUMMARY

The germ of the imaginal wings grafted on specimens of caterpillars of Lymantria dispar L. of the opposite sex continues developing, and the differentiated wings have the dimorphic hue proper to them, and not to the specimen on which they develop. The pigments of the scale therefore do not proceed directly from the desiccated blood, but are the product of certain chemical changes which, according to Mayer, occur in the blood under the influence of specific substances contained in the scale-forming cells.

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