MEMOIRS OF THE COLLEGE OF SCIENCE, UNIVERSITY OF KYOTO, SERIES B,
Vol. XXV, No. 3, Article 2 (Biology), 1958

Experimental Studies on the Developmental Mechanism of Bar Eye in *Drosophila melanogaster*

V. Effect of Metallic Compounds on the Facet Increase of the Bar Eyed Mutant[1)]

By

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(Received Nov. 17, 1958)

Among organic substances there are not a few which have been shown by the previous experiments of the author (KAJI, 1954, 1955a, b, 1956, 1958) to have the effect in increasing the facet number of the Bar eye of *Drosophila melanogaster*. Besides these organic substances, it has been noted by the former experiment of the author (1955b) that some of the metallic compounds also exert the facet-increasing effect. But the effect is not so great, in so far the author's experience goes. Recently, however, OGAKI (1956, 1957) has pointed out that the effect of several kinds of metallic compounds especially magnesium salts is conspicuous. Therefore, it seems necessary to undertake further investigation on the effect of metallic compounds. The following is the results of our experiments carried out with a large number of metallic compounds including those which have been shown by OGAKI to exert the conspicuous effect. Tests of the substances were made by the same experimental procedure as described in the previous paper of the author (KAJI, 1954). Larvae of *Drosophila melanogaster* of the Bar (*B*) strain, kept at 25°C and aged from hatching to fifty hours in the routine raising bottle, were transferred to a glass tube containing agar molasses medium plus the chemical substance to be tested.

The author wishes to express his cordial thanks to Dr. H. TAKAYA, Dr. M. KATO and Dr. Z. YOSHIDA for their kind encouragement. He is also indebted to Prof. M. ICHIKAWA of the University of Kyoto, for the criticisms of the manuscript.

Experiments

Tests were made with 18 kinds of metallic compounds. These were as follows: sodium chloride, sodium acetate, potassium chloride, magnesium acetate, magnesium chloride, magnesium sulfate, calcium chloride, cupric sulfate, zinc sulfate, chromium chloride, ammonium phosphomolybdate, manganese chloride, iodine, potassium iodide,

1) This work was supported by a grant for the Scientific Research from the Ministry of Education.

ferrous chloride, ferric chloride, cobalt sulfate and nickel sulfate.

The results obtained are summarized in Table 1.

Table 1. Effect of the metallic compounds on the increase of facet number in *B* strain.*

Substances	Conc. (%)	Facet numbers	Substances	Conc. (%)	Facet numbers
NaCl	2.0	180.9	$CrCl_3 \cdot 6H_2O$	1.0	97.4
	1.0	191.5		0.5	108.0
$Na(CH_3CO_2) \cdot 3H_2O$	12.0	201.7	$(NH_4)_3PO_4 12MoO_3 \cdot 6H_2O$	0.2	109.7
	5.0	101.3	$MnCl_2 \cdot 4H_2O$	0.5	127.2
KCl	3.0	236.9		0.2	135.4
$Mg(CH_3CO_2)_2 \cdot 4H_2O$	5.0	216.1	I	0.2	151.1
	3.0	121.5	KI	1.0	198.1
$MgCl_2 \cdot 6H_2O$	3.0	212.3		0.5	160.8
	2.0	152.5	$FeCl_2 \cdot 4H_2O$	1.5	168.2
$MgSO_4 \cdot 7H_2O$	4.0	209.8		1.0	212.1
	2.0	127.6	$FeCl_3 \cdot 6H_2O$	1.5	134.0
$CaCl_2 \cdot 5H_2O$	3.0	118.7		1.0	140.8
	2.0	101.2	$CoSO_4 \cdot 7H_2O$	0.5	69.1
$CuSO_4 \cdot 5H_2O$	0.5	85.0		0.2	216.7
	0.2	105.0	$NiSO_4 \cdot 6H_2O$	0.5	107.8
$ZnSO_4 \cdot 7H_2O$	0.5	104.0		0.3	108.2
	0.2	144.5			

* Facet number represents an average in males. Facet numbers of the control *B* eye and the wild flies are 74 and 680 respectively on the average.

As shown in the table, all the substances tested proved to be more or less effective in increasing the facet number. Among these metals, potassium chloride, cobalt sulfate, magnesium acetate, magnesium chloride, ferrous chloride, magnesium sulfate, sodium acetate, potassium iodide and sodium chloride were found to exert rather stronger effect. The average number of the facets of the treated male flies was found to be 236.9 in the case of 3.0 per cent solution of potassium chloride, 216.7 in the case of 0.2 per cent of cobalt sulfate, 216.1 in the case of 5.0 per cent of magnesium acetate, 212.3 in the case of 3.0 per cent of magnesium chloride, 212.1 in the case of 1.0 per cent of ferrous chloride, 209.8 in the case of 4.0 per cent of magnesium sulfate, 201.7 in the case of 12.0 per cent of sodium acetate, 198.1 in the case of 1.0 per cent of potassium iodide and 191.5 in the case of 1.0 per cent of sodium chloride. The effect of these substances was nearly the same in degree. As far as the present experiment is concerned, other metallic compounds are less effective than the above mentioned compounds, although they have some effect in increasing the facet number, i. e., the augmentation of facets of the flies tested was always within the range from 1.5 to 2.5 times the facet number of the untreated Bar eye (74.2 on the average in male), and their effectiveness can be arranged in the following order; iodine $\geqq$ zinc sulfate $\geqq$ ferric chloride $\geqq$ manganese chloride $>$ calcium chloride $>$ ammonium phosphomolybdate $\geqq$ nickel sulfate $\geqq$ chromium chloride $\geqq$ cupric sulfate.

On the other hand, it was noted that the effect of these metallic compounds varied in considerable degree when tested in different concentrations. At the same time, it was found that higher concentrations were not necessarily more effective than lower concentrations. On the contrary, some of the compounds such as zinc sulfate, cobalt sulfate, ferrous chloride and cupric sulfate became gradually less effective, from certain point, in inverse proportion to the increase of the concentration (see Fig. 1). Under these conditions, it is likely that these metallic compounds have each optimum concentration in which the facet-increasing effect is maximum.

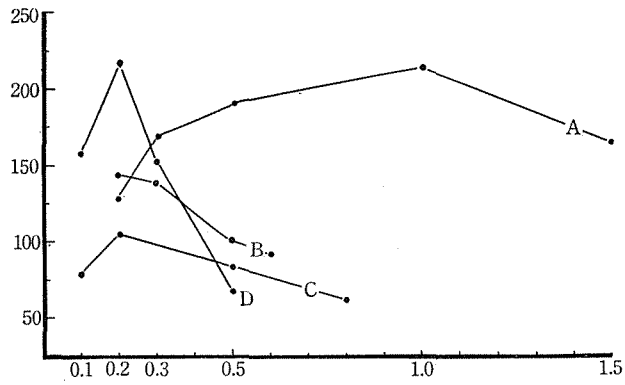

Fig. 1. Optimum concentrations in some of the transition metals. Abscissae: concentrations of the substances; ordinates: number of facets; A: ferrous chloride, B: zinc sulfate, C: cupric sulfate, D: cobalt sulfate.

Discussion

The results of the present experiments have disclosed that all the metallic compounds tested exert influence on increasing the facets of the developing compound eye of the Bar (*B*) strain. But the degrees of the effectiveness were not necessarily the same in different substances. Among them, potassium chloride, cobalt sulfate, magnesium acetate, magnesium chloride, ferrous chloride, magnesium sulfate, sodium acetate, potassium iodide and sodium chloride were found to be more effective, but the actual increase of facets brought about by these substances was not so great, that the average number was counted about 220. Namely, under the influence of these compounds the facet number becomes threefold as many as that of the untreated fly. But the increase in such degree cannot be considered as conspicuous, at least, in comparison with the increase influenced by such acid amides as acetamide and lactamide. The effect of the latter has been found to increase the Bar eye as large as or even larger than the wild type eye (KAJI, 1954, 1955a, b). Moreover, the effect of the other metallic compounds tested was always inferior to that of the above mentioned compounds, i. e., the facet number of the Bar eye treated with these chemicals increased only 1.5 to 2.5 times that of the control Bar eye.

It was the characteristic of most of the metallic compounds that their effect did not become stronger in proportion with the increase of the concentration of the substance employed. As is pointed out above, they have an optimum concentration each in which the greatest effect is produced. This point differs from the previous

result obtained with the organic substances (KAJI, 1954, 1955a, b, 1956, 1958). In that case it was a rule that the higher the concentration of the organic substances, the stronger the effect in increasing the facet number of the Bar eye.

Recently, OGAKI (1956, 1957) pointed out with *Bar ebony* (B; e^{11}) and *coisogenic Bar* (*B coiso*) that the effect of the magnesium salts was the most marked among the metallic compounds used, the increase of the facets influenced by magnesium acetate being 541.3 in the case of B; e^{11} and 332.0 in the case of *B* (*coiso*) strain. Based on this fact, he seems to assume that magnesium bears a special significance upon the manifestation of the Bar character. Unfortunately, however, such marked effect as he found with the magnesium salts was not evidenced by the present experiment. In our experiment also the magnesium salts were found to be effective to some extent, but their effect was by no means special, it being the same as that of the other metallic compounds. There is, therefore, an apparent discrepancy between the result of OGAKI and ours. At present, there is no elucidation to reconcile the discrepancy. However, it seems probable that the difference of the fly strains used by the two authors may be one of the factors responsible for the difference of the results in question.

So far as the Bar (*B*) fly used in the present experiments is concerned, however, it should be emphasized that the facet-increasing effect of the metallic compounds is generally far inferior to that of the organic compounds. None of the former was found to have influence to make the Bar eye as large as the wild type.

Summary

Effect of the metallic compounds upon the developing Bar eye of *Drosophila melanogaster* was examined. Eighteen kinds of compounds tested have proved to have more or less influence by increasing the facets number of the Bar eye. But their effect was far less than the effect of acetamide and lactamide. There was none which increased the Bar eye to the size of the wild type.

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