FRACTIONATION OF DROSOPHILA POPULATIONS ACCORDING TO OPTOMOTOR TRAITS

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(Received 15 October 1969)

Behavioural traits of the fruitfly Drosophila are studied as regards their neuronal aspects by appropriate investigation of individual flies, and as regards their genetic aspects by appropriate investigation of population samples. A countercurrent distribution method and a selection-by-inspection method have been developed in order to isolate phototactically deficient mutants from the progeny of the chemically mutagenized homogeneous wild type populations (Benzer, 1967; Pak, Grossfield & White, 1969). Multiple T-mazes and related devices have been used to fractionate heterogeneous populations according to their geotactic or phototactic reactions (Hadler, 1964a, b; Hirsch, 1967; Hostetter & Hirsch, 1967; Dobzhansky & Spassky, 1967; Dobzhansky, Spassky & Sved, 1969; Murphey & Hall, 1969), according to their spontaneous activity (Levine & Kessler, 1965; Connolly, 1966, 1967) and according to their optomotor response (Siegel & Hirsch, 1963; Siegel, 1967).

The term optomotor response denotes the tendency to follow the angular movements of the visual environment. This tendency is common to various insects and is readily established in both normal and mutant fruitflies (Hecht & Wald, 1934; Gavel, 1939; Kalmus, 1943; Götz, 1964). The visual perception of the moving environment involves the comparison of signals from different light receptors. Neuronal interconnexions between the nerve endings of at least two photoreceptors are essential in a movement detector. The integration of the signals from the movement detectors in the different eye regions requires another set of neuronal interconnexions. Moreover, the visual control of orientation and locomotion cannot be achieved without the neuronal conveyance of the movement information to particular sites of the motor system.

Different levels of information processing in Drosophila have already been studied by the methods of stimulus-response analysis. A tentative diagram of the flow of information from the movement detectors to the movement effectors in the normal fruitfly can be devised on the basis of the present results. The diagram describes in some detail the optomotor control reactions in response to the movements of the visual environment (Götz, 1965b, 1968, 1969). An investigation of behavioural mutants would facilitate the identification and localization of the neuronal correlates. Deviations from ordinary optomotor behaviour have been investigated in some of the existing eye mutants. However, these deviations almost exclusively reflect modifications of light reception (e.g. abnormal light screening of the pigment cells between the facets of the eyes; Hengstenberg & Götz, 1967; Wehner, Gartenmann & Jungi, 1969). Most desirable is the generation and isolation of mutants involving modifications of the optomotor control system. The objective of the present report is the investigation of an appropriate selection technique with inbred populations of different optomotor behaviour.
FRACTIONATION TECHNIQUE

The fractionation of Drosophila populations is achieved with a representative number of individuals simultaneously subjected to a sequence of trial and separation processes. The procedure resembles the countercurrent distribution method used for fractionating mixtures of molecules according to their partition between two solvent phases. The efficiency of this principle in the separation of behavioural mutants of Drosophila has been demonstrated by Benzer (1967), who succeeded in isolating phototactic mutants from inbred strains.

The distribution of Drosophila populations in classes of different optomotor responses can be achieved with the cyclically operating machine outlined in Text-fig. 1. The visual stimulus is provided by the helical stripe pattern on the inner surface of

Text-fig. 1. Selection machine for fractionating Drosophila populations according to their responses to stripe movement. The upper diagram shows a translucent Plexiglass cylinder with a helical pattern on the inner surface. The stripes of the rotating pattern apparently proceed in axial direction. The movement is perceived by a number of test flies in a tube system inside of the cylinder. The transparent Plexiglass tube system is shown in the lower diagram. It consists of five stationary tubes (1–5). Each of the tubes joins on either side one of the six end tubes which are mounted in a pivoted disk. Flies which have entered the end tubes can be translocated to the openings of the neighbouring stationary tubes by pulling or pushing a central shaft. Countercurrent distribution of the flies in the tube system results from a repetitive self-acting programme for altering the position of the shaft as well as the direction of the stimulus movement according to the operation cycle shown in Text-fig. 2.
the introverted 'barber pole' in the upper diagrams. The rotating pattern apparently moves in axial direction. The pole consists of a $200 \times 620$ mm. translucent Plexiglass cylinder. Patterns of different stripe width are mounted on transparent films which can alternatively be attached to the inner surface of the cylinder. The transparent Plexiglass tube system in the lower diagram comprises a stationary set of five $12 \times 400$ mm. tubes which are joined on either side to a pivoted set of six $12 \times 60$ mm. end tubes. The tube system contains the test flies and can be brought to a fixed position inside of the illuminated cylinder. Plate 1 shows the arrangement with the tube system partially extracted from the 'barber pole'.

Text-fig. 2. Principle of the selection machine demonstrated by the separation of two idealized flies according to their property to follow the stripe movement (+) or to move in the opposite direction (−). The axial sections through a simplified 3-tube system and the surrounding 'barber pole' represent four subsequent steps in a single cycle after which the separation is accomplished. (a) Partition of the flies when the stripes move to the right. (b) Selective translocation of the flies when the end tubes attain an alternate position. (c) Partition of the flies when the stripes move to the left. (d) Selective translocation of the flies when the end tubes return to their initial position.

The test flies are placed in the central tube No. 3 of the five-tube system and are allowed to spread out in the stationary tube and the two adjoining end tubes. Flies with different optomotor behaviour may partially segregate during this process. Although the segregation effect is diminished by random walk and random flight, two possible reactions of a fly can always be distinguished. Either the locomotion in the direction of the pattern movement outweighs the locomotion in the opposite direction or vice versa. The fly therefore attains either the 'follower's group' on one side, or the 'opponent's group' on the other side of the tube system.

After a given time interval a servomotor acts upon an axial shaft to bring the movable end tubes to an alternate position. The end tubes on one side are shifted to join the openings of the higher ranking stationary tubes and the end tubes on the other side are shifted to join the openings of the lower ranking stationary tubes. The end tubes translocate flies of the 'follower's group' to the next higher ranking position and flies of the 'opponent's group' to the next lower ranking position. During the subsequent time interval the flies spread out in the joining tubes while the stimulus movement is reversed. When the end tubes finally return to their initial position they again translocate flies of the 'follower's group' to higher ranking positions and flies of the 'opponent's group' to lower ranking positions. Continuous repetition of the separation
cycle finally generates a steady-state countercurrent distribution of the test flies in the five-tube system.

The described process is elucidated in Text-fig. 2 by the separation of two idealized flies in a simplified schematic three-tube system. *Positive* responses or the property to follow the stripe movement are attributed to one of the flies (+), while *negative* responses or the property to move in the opposite direction are attributed to the other fly (—). The axial sections (a–d) denote the four subsequent steps of a single separation cycle. White arrows below the diagrams (a) and (c) indicate the actual directions of stripe movement. Black arrows refer to the locomotor responses of the idealized flies in the tube system. The separation of the two flies is accomplished at the end of the cycle when the system returns to its initial state.

Fractionations in the five-tube system are usually performed with about 100 females—sometimes with about 100 males—from one of the inbred strains of *Drosophila melanogaster*. At the beginning of the experiment the flies are placed in the central tube. During the next 24 h they are subjected to about 120 separation cycles. The distribution of the flies in the tube system attains an equilibrium which is finally evaluated by counting the flies in the different tubes and end tubes. Let \( n_+ \) be the number of flies which have reached the higher ranking tubes and end tubes by following the moving stripes, and \( n_- \) the number of flies which have reached the lower ranking tubes and end tubes by moving in the opposite direction. The quotient derived from the difference and the sum of these values,

\[
q = \frac{(n_+ - n_-)}{(n_+ + n_-)},
\]

defines a certain value between \(-1\) and \(+1\) which characterizes in magnitude and sign the *optomotor response* of the inbred population under the selected conditions. To obtain unbiased results one has merely to reverse the rank of the tubes and the sequence of the stimuli in the separation cycle after each experiment.

The *optomotor stimulation* of the test flies in the selection machine is matched to the requirements of the visual system of *Drosophila melanogaster* (Götz, 1965a, b; Hengstenberg & Götz, 1967). The translucent cylinder is illuminated from the outside by the white light of four DC-powered fluorescent lamps. The average *luminance* \( I \) of the pattern is in order of 380 cd/m.\(^2\) or 1200 asb, and the angular distribution approximates to Lambert's law. This level of brightness is sufficient to keep the *Drosophila* compound eye in the light-adapted state. The *contrast* \( \Delta I/2I \) between two neighbouring stripes in the pattern is approximately 0.95. The width of such a pair of stripes is seen by the test flies in the tube system under an average angle \( \bar{\lambda} \). The magnitude of this angle determines the spatial resolution of the pattern and the transfer of its contrast by the optic system of a given facet eye. Unless otherwise stated the pattern used in the present experiments has an average *spatial period* \( \bar{\lambda} = 28^\circ \), which is large enough for appropriate perception in the visual system of *Drosophila*. The *temporal period* of the stimulus movement is set to 1 sec. per pair of stripes. This period is most effective in torque experiments with fixed flying individuals (Götz, 1964). The temperature in the tube system is kept in the range of \( 25 \pm 3^\circ \text{C} \). Humidified air is provided inside of the cylinder in order to sustain the activity of the starved animals during the experiments. Sex, age, the state of nutrition of the flies, the temperature within the given limits and the time of the day at the final counts proved to have little if any effect
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on the scores obtained with the selection machine (T. Davidson & K. G. Götz, unpublished results).

MATERIAL

The following stocks of Drosophila melanogaster were used in the present work. (1) The wild stock 'Berlin' which is designated here as wild type (w+) or (+). (2) The mutant In (1) sc8, sc8 wR, which is designated here as white-apricot (wR) according to the phenotypically predominant eye-colour mutation on the X-chromosome. Less conspicuous in this strain is the scute-eight (sc8) mutation, which is associated with the inversion of the sequence of genes on the X-chromosome (Lindsley & Grell, 1967). Phenotypically, certain bristles on the fly's scutum are missing (Dubinin, 1933). Both strains are maintained in the Max-Planck-Institut für biologische Kybernetik in Tübingen. (3) The mutant sine oculis (so). The ocelli are absent and the eyes are reduced to small clusters of ommatidia in this mutant.

OPTOMOTOR RESPONSES OF INDIVIDUAL FLIES

The movement perception by the visual system of the test flies has been studied in previous investigations. Text-fig. 3 gives the average torque response R (dyne x cm.) of fixed flying individuals from the two strains + and wR when a vertically striped pattern of the spatial period λ is rotated at constant speed around the fly's vertical axis. The temporal period, the average luminance, and the contrast of the stimulus are held at constant values approximating to those given in a previous section.

Text-fig. 3. Optomotor torque responses of 116 normal (+) and nine mutant (wR) females from the Drosophila strains investigated in the present report. The flies were individually mounted on a torque meter and surrounded by rotating periodic patterns consisting of black and white stripes. The averages from the recordings during stationary flight in still air are given as a function of the spatial period λ of the patterns. The positive sign of the responses at sufficient stripe widths indicates in both cases the tendency of the flies to turn in the direction of the stimulus movement. The inversion of this tendency in the range below λ = 9.2° can be attributed to the inversion of the apparent movement by incomplete resolution of the periodic patterns in the facet eyes of the two strains (Götz, 1965b).
Torque responses with a positive sign indicate the tendency of the flies to turn in the direction of the stimulus movement. This tendency is unequivocally found in both strains, provided that the spatial period $\lambda$ of the periodic pattern is well above the limit set by the resolving power of the periodic array of sampling points in the facet eye. Reduction of $\lambda$ below this limit causes, among other phenomena of interference between the two periodic structures, an inversion of the apparent stripe movement in the array of visual elements. The inversion effect is necessarily accompanied by an inversion of the optomotor torque responses. It can be seen from Text-fig. 3 that in both strains the optic resolution is limited at about $\lambda = 9.2^\circ$. This value corresponds to an average angular spacing $\Delta \phi = 4.6^\circ$ between neighbouring visual elements of the movement detectors, which in turn is consistent with the array of the 1400 facets in the two eyes of the fruitfly. The previous results support the evidence provided by other authors that the facet-associated neurommatidia constitute the resolution-determining array of sampling elements in the visual system of flies. The acuity-determining effective angular widths of the visual fields of these elements have been derived from optomotor responses and about the same value $\Delta \phi = 3.5^\circ$ was found in both strains. The ratio of the two parameters $\Delta \rho$ and $\Delta \phi$ approximates to the condition $\Delta \rho / \Delta \phi = 0.88$ for optimum imaging (Götz, 1965a). The different behaviour of the flies in the lower range of $\lambda$ can be attributed to the increased translucency of screening pigment cells in the ommatidia of the pigment-deficient mutant. Independent methods indicate that the photoreceptors of the equally illuminated eyes of the normal (+), and the mutant (w°) fruitfly receive light in a ratio of about 1:8 (Hengstenberg & Götz, 1967).

Recent unpublished investigations of individual flies walking on top of a tread compensator (Götz & Gambke, 1968) proved that the tendency to turn under the influence of the moving stripe patterns is independent of the mode of locomotion.

**Optomotor responses of population samples**

The tendency to turn with the visual environment of appropriate structure and angular velocity is obviously a general property of both normal (+) and mutant (w°) individuals. If this property determines the behaviour of test flies in the selection machine, it is likely to find similar responses in the two different populations. The responses should be slightly positive for the following reasons. The flies in the tube system of Text-fig. 1 may take random orientations when the surrounding helical stripe pattern is at rest. However, the axial movement of the stripes simulates a clockwise rotation of the environment in one part, and a counterclockwise rotation of the environment in another part of the visual system. By following the predominant stimulus the flies are compelled toward an orientation in which the two antagonistic influences balance each other. The balance is established when the longitudinal axes of the flies point in the direction of the pattern movement. In this position the moving stripes display equivalent back-to-front movements in equivalent regions of the left and the right eye. Random locomotion of flies with increased probability for this particular orientation will necessarily lead to an accumulation of flies in the higher ranking tubes and end tubes. The final distribution of the flies in the tube system should, therefore, correspond to a quotient $q > 0$.

However, the opposite behaviour is found in the experiments with the *Drosophila*
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The histograms in Text-fig. 4 illustrate the distributions of wild type females over the five tubes of the machine in the initial state (white bars), and in the final state (black bars) of the selection process. Each of the five histograms is representative for a series of unbiased experiments at a certain average spatial period $\lambda$ of the stripe pattern in the 'barber pole'. The total number of flies under investigation is specified on top of the white bars and the histograms are normalized with respect to these numbers. The $q$-values below the histograms characterize the final distribution of the investigated samples of the population. Unexpectedly, these values are negative whenever the parameter $\lambda$ is large enough to ensure the appropriate perception of the stimulus. Instead of following the axial movement of the pattern the animals tend to proceed in the opposite direction.

Text-fig. 4. Initial (white) and final (black) distributions of wild type female fruitflies in the five-tube system of the selection machine. The five histograms refer to different spatial periods $\lambda$ of the stimulating patterns. The distributions are normalized with respect to the total number of flies per sample which is given on top of the white bars. A trend to proceed against the axial pattern movement is established by the negative $q$-values in the experiments on the right. The decrease of the magnitude and the inversion of the sign in the lower range of $\lambda$ are attributed to well known deficiencies in the visual system of Drosopkila. The responses of the flies are obviously evoked by the visual perception of the moving patterns.

The well known limitations for contrast transfer and pattern resolution of the Drosophila eyes are expected to show up in the lower range of $\lambda$ if visual perception of the pattern movement is prerequisite for the responses. The histograms on the left side of Text-fig. 4 indicate both the decrease of the magnitude, and the inversion of the sign of the responses in analogy to the behaviour of the fixed flying individuals (Text-fig. 3). The $q$-value of the sample at $\lambda = 7^\circ$ is in fact slightly positive. The probability that a negative mean value of $q$ holds for the population is $P < 0.02$ in the $7^\circ$-experiment whereas it is $P > 0.9999$ in the $28^\circ$-experiment. It can be concluded that the responses are actually evoked by the perception of the moving stripes. Helical patterns with an average spatial period $\lambda = 28^\circ$ are appropriately perceived, and therefore used throughout the subsequent experiments.
It remains to be shown that the *axial* component of the stripe movement in the 'barber pole' is in fact the response determining stimulus. This is done by comparison of the responses obtained with a right-handed and a left-handed helix. Only the axial component of the stripe movement is invariant to simultaneous exchange and anti-cyclic rotation of the two patterns. The coincidence of the $q$-values is established by a number of test experiments. The results prove that the *non-axial* components of the stripe movement do not significantly contribute to the responses of the flies.

![Text-fig. 5. The negative optomotor reactions of *Drosophila* wild type (+) in the selection machine. Upper histogram: distribution of the $q$-scores from 159 experiments. Lower histograms: distributions of the $q$-scores from 82 experiments with the substrains Pos. and Neg. The scores of the substrains were collected through eight generations of progressive selection and assortative mating of the most positively reacting flies and of the most negatively reacting flies. No significant effect of selection pressure on the reactions of the inbred strain can be observed.

The $q$-values of the *Drosophila* wild type (+) population have been determined in 159 experiments over a period of about one year. The upper histogram in Text-fig. 5 illustrates the frequency of occurrence of the different results. Each of the bars represents by its length, position and width the number of flies that made the scores in a certain interval of $q$. The distribution is characterized by the following parameters:

- Sample mean of the + -population $\bar{q} = -0.37$
- Standard deviation $s = \pm 0.23$
- Degrees of freedom $f = 158$
- Standard error of the mean $\text{S.E.} = \pm 0.02$
The reliability of the negative effect of the wild type (+) population in the selection machine is demonstrated by these data.

About half of the experiments were devoted to the question whether or not the considerably inbred wild type strain would still be capable of hereditary changes in the optomotor behaviour. The results of the 82 experiments allow one to compare the behaviour of the substrains Pos. and Neg., which were obtained by progressive selection and assortative mating of the most positively or most negatively reacting females and males. The investigation was extended to eight subsequent generations under continuous selection pressure. Text-fig. 6 shows the sample means and the standard deviations of the q-values of the two substrains as a function of the generation number. The last column refers to the integral responses of the two substrains. The data are based on the q-values from generations 1 to 8 inclusive. The lower histograms in Text-fig. 5 illustrate the distributions of these values.

The results in Text-fig. 6 suggest that there are no significant behavioural differences between the substrains Pos. and Neg. Discrepancies between the means and between the standard deviations of any two corresponding substrain samples can be attributed to the sampling error. Statistical tests have been applied in order to calculate the probabilities that differences greater than those observed occur by chance in a uniform population. Unless indicated by an asterisk, the probabilities for appropriate sampling...
errors of the means \((P_t)\) and of the standard deviations \((P_F)\) are well above the 0.05 level.

An entirely different optomotor behaviour was found with the inbred population of the *Drosophila* mutant *white-apricot* \((w^a)\) in the selection machine. The \(q\)-values of this mutant have been determined in 196 experiments. The frequency of occurrence of the different results is shown in the upper histogram of Text-fig. 7. Neglecting

The positive optomotor reactions of the *Drosophila* mutant *white-apricot* \((w^a)\) in the selection machine. *Upper histogram:* distribution of the \(q\)-scores from 196 experiments. *Lower histograms:* distributions of the \(q\)-scores from 139 experiments with the substrains Pos. and Neg. The scores of the substrains were collected through eight generations of progressive selection and assortative mating of the most positively reacting flies and of the most negatively reacting flies. A slight effect of the selection pressure on the reactions of the inbred strain can be observed.

The asymmetry of the histogram the distribution can be characterized by the following parameters:

- Sample mean of the \(w^a\)-population: \(\bar{q} = +0.44\)
- Standard deviation: \(s = \pm 0.31\)
- Degrees of freedom: \(f = 195\)
- Standard error of the mean: \(\text{S.E.} = \pm 0.02\)

The effect of the selection pressure on the *white-apricot* strain has likewise been studied through eight generations. The \(q\)-values of the substrains Pos. and Neg. are
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derived from 139 of the experiments with this mutant. Text-fig. 8 shows the sample means and the standard deviations for each generation as well as for the entire sequence of eight generations. The distributions of the $q$-values obtained during this period are presented in the lower histograms of Text-fig. 7. The results of the t-test in the last column of Text-fig. 8 suggests a slight disparity of the two substrains in response to the selection pressure. Deviations greater than those observed between the means of the two substrain samples may still occur by chance in a uniform population. However, the probability of occurrence $P_t = 0.003$ is below the 0.01 level.

DISCUSSION

The difference between the responses of normal and mutant population samples in the selection machine (Text-figs. 5–8) contrasts remarkably with the similarity of the optomotor reactions of normal and mutant flies in the torque experiment (Text-fig. 3). To reconcile the differing observations we must re-examine the arguments given at the beginning of the last section.

Two postulates were used in order to predict unequivocally positive $q$-values of the population samples from the positive torque responses of the individual flies. The first postulate was concerned with the orientation of the flies in the selection machine. It was expected that the moving stripes would stimulate the flies to orientate their longitudinal axes in the direction of the pattern movement. This behaviour is actually
observed in the tube system. At rest, the flies are preferentially aligned in the direction of the moving stripes. The second postulate was concerned with the locomotion of the flies in the tube system. The predominance of locomotor activities in the flies' forward directions is easily established by observation. However, it was assumed that locomotion in contrast to orientation is essentially independent of the particular visual stimulation. If this is true, we can predict the optomotor behaviour in the selection machine. The majority of the flies should follow the moving stripes and accumulate in the higher ranking tubes and end tubes, thus producing positive \( q \)-values.

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<tr>
<th>Model I</th>
<th>Model II</th>
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<tr>
<td><img src="image1" alt="Torque response vs. Stimulus Inclination" /></td>
<td><img src="image2" alt="Torque response vs. Stimulus Inclination" /></td>
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<tr>
<td><img src="image3" alt="Thrust response vs. Stimulus Inclination" /></td>
<td><img src="image4" alt="Thrust response vs. Stimulus Inclination" /></td>
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Text-fig. 9. Insect models in which movement information is exclusively conveyed from the stimulated eye to the contralateral motor system (I) and to the ipsilateral motor system (II). The thrust components on either side of the insects are given by the straight arrows at the ends of the baselines. The dashed lines refer to the thrust in absence of visual stimulation. The curved arrows in the periphery of the eyes denote actual directions of the stimulus movement which may either increase (black) or decrease (white) the corresponding thrust component. The difference and the sum of the thrust effects on either side represent the rotatory and the translatory responses of the insect. The models cannot be distinguished from each other by the rotatory responses in situation \( a \) or in the equivalent torque experiments. Yet they produce translatory responses of the opposite sign in situation \( b \) which is encountered in the selection machine.

The negative \( q \)-values of the wild type (Text-fig. 5) disprove the independence of the locomotor events. Frequency, strength and duration of these events are obviously not altogether invariant to movements of the environment. At least one of these parameters must be under control of the visual system.

This result has led to the search for possible linkages between the optomotor effects of orientation and locomotion in flies. Actually, there exist strong functional relations between the rotatory and the translatory movements of the fruitfly. The torque produced in order to follow the rotation of the visual environment results from
the unbalance of the thrust components on either side of the motor systems. It is
the optomotor control of the thrust components which entails the adjustment of the
torque during the flight of Drosophila (Götz, 1968, 1969). The conclusions suggest
an attempt to derive the locomotor behaviour of the flying or walking fruitflies from
the well known torque responses in the two modes of locomotion. This attempt is of
particular interest in respect of the discrepancy mentioned at the beginning of the
discussion. The following considerations will show that the rotatory responses of the
flies in the torque experiment are in fact compatible with either of the different trans-
latory responses in the selection machine.

Optomotor reactions depend on the conveyance of visual information from the
stimulus detectors to the response effectors. This conveyance is achieved by neuronal
interconnexions which may extend from one side of the visual system to either the
contralateral motor system, or the ipsilateral motor system, or both systems. Text-
fig. 9 shows two different models of an insect which is endowed with the capability
to follow the rotation of the visual environment. For simplicity the models comprise
exclusively contralateral interconnexions (I) and ipsilateral interconnexions (II) of the
eyes and the motor system on either side. Actual directions of the stimulus movement
are indicated by the curved arrows in the periphery of the schematic eyes. The thrust
components of the motor system on either side of the insect models are given by the
length and direction of the straight arrows at the ends of the baselines. The dashed
lines denote the magnitude of the thrust components in absence of visual stimulation.
The shade of the arrows indicates the increase (black) or the decrease (white) of the
thrust components under the influence of stimulus movements in actual directions
(e.g. back-to-front or front-to-back).

The upper diagram (a) illustrates the rotatory responses by which the schematic
insects follow a counterclockwise rotating environment. The two models obviously
cannot be distinguished from each other by torque measurements such as shown in
Text-fig. 3. However, translatory responses of different sign result from the equally
stimulated models in the lower diagram (b). Front-to-back movement of the visual
environment on either side of the insect increases the thrust of model I but decreases
the thrust of model II. Both torque and thrust responses undergo inversions if the
stimulus movement across the eye surface is tilted in the reverse direction. The direc-
tion at right angles to the orientation of the movement-detecting units is specified by
the inversion points or zeros in the response curves. An essential property of the models
in Text-fig. 9 is the coincidence of the zeros for both torque and thrust responses.

The pattern movement in Text-fig. 9 resembles the visual stimulation of the flies
in the selection machine. The behaviour of the two schematic insects in the tube
system may therefore be derived by analogy with the examples of this figure. The
orientation of the two insects is obviously achieved by equivalent optomotor torque
responses. However, random locomotion against the movement of the stripes is enhanced
in model I and diminished in model II by optomotor thrust responses of the appropriate
sign. Random locomotion with the moving stripes is modified by the inverse effects.
The optomotor thrust responses thus compel type-I populations into the lower
ranking tubes and type-II populations into the higher ranking tubes of the machine. The
type-I populations produce negative q-values, provided that the locomotor responses outweigh the torque-induced accumulation in the higher ranking tubes.
The type-II populations, however, produce positive $q$-values. The locomotor responses of these populations reinforce the torque-induced accumulation in the higher ranking tubes.

The results of these considerations may be summarized as follows: (1) Optomotor thrust responses are most probably involved in the orientation-control reaction of the flies. (2) Negative as well as positive $q$-values of the populations in the selection machine are already expected from the possible locomotor by-products of the orientation-control reaction.

The negative $q$-values of the wild-type population in Text-fig. 5 may be tentatively attributed to the locomotor by-products of the orientation-control reaction. However, this imposes certain restrictions on the nervous interconnexions of the stimulus detectors in the visual system and the response effectors in the motor system of the flies. The interconnexions must resemble the wiring diagram of model I in Text-fig. 9 or its more complex derivatives. This condition is apparently not fulfilled in Drosophila. Unpublished results indicate that both torque and thrust responses of stationary walking fruitflies are appropriately described by a ‘minimum model’ of type II rather than of type I. More complex relations exist between torque and thrust responses under conditions of stationary flight in still air. The stimulus inclinations for zero-torque and zero-thrust responses are approximately orthogonal to each other. The appropriate description of the responses is given by a hybrid ‘minimum model’ in which the movement detectors of a type-I subsystem are directed to the lower frontal region whereas the movement detectors of a type-II subsystem are directed to the upper frontal region of the fruitfly. The subsystems control exclusively the wingbeat amplitude on either side of the fly. The optomotor responses appear suitable to counteract involuntary changes of both direction and altitude in the free flight of Drosophila (Götz, 1968, 1969).

It is evident that the negative $q$-values of the wild-type population cannot simply be derived from the optomotor reflex by which the normal and the mutant flies follow the rotation of the visual environment. Independent locomotor responses are probably involved in the behaviour of the flies in the selection machine. The failure to identify these responses on the steady-state torque and thrust recordings may be ascribed to the complex nature of this particular behaviour. The recordings were made under ‘open loop’ conditions in order to prevent the inherent feedback in the stimulus-response system. ‘Closed-loop’ stimulus-response analysis of the optomotor behaviour of the fly Musca is now in progress (Reichardt & Wenking, 1969). The approach appears most promising for revealing the missing link in the locomotor reactions of the free-moving flies.

It has been shown in the experiments of Text-fig. 4 that the prominent deficiencies of the locomotor responses of the fruitflies in the selection machine correlate with well established deficiencies of visual acuity and resolving power. The locomotor responses are obviously evoked or triggered by visual perception of the pattern movement. It is of particular interest to know whether or not the different optomotor behaviour of the normal and the mutant populations is correlated with permanent disparities in the wiring pattern or synaptic contacts of the nervous network. Evidence against this possibility has been more recently accumulated in co-operation with T. Davidson and M. Heisenberg by investigation of the constancy and the herita-
Ability of optomotor behaviour in the selection machine. A report on the results is in preparation. It has been shown that the optomotor responses are not entirely invariant properties of the populations. The responses depend on various parameters of the stimulus and the environment. Moreover, the magnitude as well as the sign of the \( q \) -values may change with habituation or adaptation to the pattern movement. The positively reacting \( w^a \) -population, for instance, produced significant negative \( q \) -values in the initial phase of the selection process if the velocity of the pattern movement and the temperature in the tube system are chosen appropriately.

![Graph showing optomotor torque responses of two different females from the strain sine oculis (so). The flies possess only about 1% of the normal number of facets. Yet they perceive the rotation of surrounding periodic patterns, provided that the spatial period \( \lambda \) exceeds the optical limits for pattern resolution and contrast transfer. The torque responses during stationary flight in still air, \( R \), are in general positive (upper diagram). The most unusual negative responses in the lower diagram are probably due to a spontaneous modification of the optomotor control system.]

The inbred \( w^a \) -strain also allows a certain latitude of the optomotor responses. A slight disparity in the average behaviour of the repeatedly selected substrains Pos. and Neg. is seen in the last column of Text-fig. 8. It is therefore more reasonable to assume that the different behaviour of the normal and mutant populations originates from similar nervous networks which are held in different modes of operation.

It is not yet known how the organization of the nervous system is encoded in the genome of Drosophila. Consequently, there are no reliable estimates of the chance to generate by mutagenesis, and to separate by selection processes, behavioural mutants with defects or modifications of the optomotor control system. There already exist visually defective mutants of Drosophila which have been successfully isolated from the progeny of the mutagen-treated wild type by various methods of selection (Benzer, 1967; Pak, Grossfield & White, 1969). The loss of the phototactic response in at least one of the mutants is most probably due to functional defects of the underlying nervous system (Pack et al. 1969; Hotta & Benzer, 1969). We have tried to induce by chemical
mutagenesis mutations affecting the optomotor control system. However, our attempt to isolate the corresponding mutants in the present selection machine has not yet been successful.

Nevertheless, there is possibly one manifestation of a functional change in the optomotor system. The modification occurred spontaneously in the *Drosophila* strain *sine oculis* (*so*) in which the eyes are not completely suppressed. The predominant *so*-phenotype is a single-eyed fly with a reduced number of facets. The upper diagram in Text-fig. 10 gives the average torque responses $R$ (dyne $\times$ cm.) of a single-eyed individual with a total of 12 instead of 1400 facets. The responses are evoked by vertically striped patterns of the spatial period $\lambda$ rotating at constant speed around the fly’s vertical axis. The temporal period, the average luminance and the contrast of the stimulus are chosen according to previous specifications. The optomotor response of the *so*-mutants obviously resemble the behaviour of the normal and the mutant flies in Text-fig. 3. The positive sign of the responses reflects the general tendency to follow the rotating environment, provided that the spatial period $\lambda$ is well above the optical limits for pattern resolution and contrast transfer. The normal optomotor behaviour has been found in almost all *so*-mutants investigated thus far (Götz, 1965 b). However, three females with the inverse behaviour were encountered in one generation. The three flies showed no deviations from the average *so*-phenotype. Optical reasons for inverse movement perception (e.g. distal intersection of the facet axes) have been ruled out in subsequent tests. The lower diagram in Text-fig. 10 gives the negative torque responses of one single-eyed fly with a total of 19 facets. Countercurrent selection was not possible at this time. Thus we were not able to confirm the hereditary character of the spontaneous modification in the *so*-strain.

It appears worthwhile to search for heritable modifications of the optomotor control system which can be identified electrophysiologically or localized histologically in the nervous network of *Drosophila*.

**SUMMARY**

1. The optomotor control of orientation and locomotion in the fruitfly *Drosophila melanogaster* requires the conveyance of information from distinct movement detectors in the visual system to distinct movement effectors in the motor system. Abnormalities of the optomotor control system have been found occasionally in *Drosophila*.

2. The abnormal flies can be isolated from population samples by appropriate fractionation according to the magnitude and the sign of the optomotor responses. A cyclically operating machine was used to fractionate two inbred strains, $w^+$ and $w^a$, which possess different alleles on the white-locus of their X-chromosomes.

3. Movements of an artificial visual environment elicit similar orientation-control responses, but antagonistic locomotion-control responses in the two strains. The responses depend on various parameters and may even change with habituation to the stimulus. However, the application of selection pressure through eight generations has little if any effect on the different optomotor behaviour of the inbred strains.

Valuable assistance was received in the different stages of the present work. The mechanical design was carefully executed by Mr H. Braun and coworkers in the institute workshop. Miss B. Köhler and Miss C. Stroebel participated in the main-
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tenance and testing of the strains and substrains. Stimulating suggestions came from Mr T. Davidson, Professor M. M. Green, Dr M. Heisenberg, Professor W. Reichardt and Dr G. Weidel. Also Dr J. Thorson has contributed interesting comments. Mr B. Boschek, Mr E. Freiberg, Mrs I. Geiss and Mrs K. Schwertfeger helped with the preparation of the manuscript and the figures.

REFERENCES


EXPLANATION OF PLATE

Selection machine consisting of an introverted ‘barber pole’ (1) providing the stimulus and a tube system (2) containing the test flies. The latter is partially extracted from a fixed position in the centre of the rotating cylinder in order to show the details of the machine and the relative dimensions of its components.