Possible anomalous communication in *Drosophila melanogaster.*

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Summary

This is a submission of work exploring the possibility of an anomalous communication between individuals of a *Drosophila melanogaster* population, obtained by selection along 50 generations, with a suitable method for that purpose. The results, statistically significant, show that possibility.

Introduction

We understand that an anomalous communication is – for the purpose of this work – any communication among the individuals of the *Drosophila melanogaster* fly not made by means of sight, sound, smell, chemical or vibratory reception.

The reasons for this work were to some extent accidental, but need not be explained now. The purpose of this work is to explore the possibility to obtain, selecting among many generations, a *Drosophila melanogaster* population having a significant proportion of individuals with such anomalous communication form among them.

Method and Material

In brief, the work consisted in observing the existence of a communication between a group of flies with food, in the light or in the dark, towards another group of flies without food, under a long lasting fast, so the latter would be induced towards zones preferably lighted or in the dark, depending if the group of flies with food is in the light or in the dark.

Initially, the work was performed with a wild population of the City of Buenos Aires, Argentina. After many tests with different elements, the following quantitative experimental design was adopted. This design makes it possible to select flies and complete a statistical analysis of the results' meaning.

Basically, a special tray is used, on which 50 hemolysis small tubes are fitted up to the half of their length into the tray crevise in complete darkness.

In each one of these small tubes, with a cotton cover, a fly is introduced, resulting from the crossing of one or more couples formerly selected.

The flies in this tray will be the communication receivers – hereinafter (R). These are left in a room, featuring an even lighting from above. In another room, sufficiently distant and isolated to avoid a normal communication, the emitter flies are left – hereinafter (E). The experiment is carried out separately with (R) males and (R) females. The latter must be virgin for a further crossing with selected males.

For the offsprings of the selected couple we used standard test tubes. A 2 ml watery solution is added consisting in 5% acetic acid and 5 ‰ propionic acid, then an excess of a mixture consisting in 79% "instant" corn flour, 7% skimmed powder milk, 7% standard sugar, and 7% powder yeast.

The bottom of the tube is slightly tapped to deposit the mixture, and any excess not absorbed by the solution is poured out.

The same procedure is followed with hemolysis tubes, with food, but only 1 ml of the solution. Additionally, test tubes are prepared following the same process as with the test tubes for the offsprings, to maintain male and female flies separated as these eclose. The separation of these, passing from the maintenance tubes and then into the hemolysis tubes with food is performed immobilizing the flies with chloroform. After each separation, the tubes containing the flies are left in the refrigerator at 15-17 °C to retard ageing until the amount needed for the tests is obtained.

The (E) flies consisted – in the case of the (R) males – in all the selected female flies available, the male flies which did not participate in the test as well as the tubes containing the offspring of the selected couples, where both male and female flies eclose. In the case of the (R) females, the same applies, but replacing the corresponding flies for others of the opposite sex.

Upon completing the test with the (R) flies without food, these are passed into the hemolysis tubes with food, individually, and these are also kept in the refrigerator until the test is completed, and then choose the (R) males and the (R) females, with the best results in order to carry out the new crossing of selected flies.

Experiment

The work, the result of which is submitted hereby, was done with 160 flies obtained from the crossing of selected flies, after almost 50 generations. Only in the last 8 generations, the selection was followed as per the experimental design we mentioned above: half males, half females. Moreover, 20 male flies and 20 female flies were added. These descended from the original "wild" population – hereinafter (O.P.). This was done to control the efficiency of the selection. Such (O.P.) was maintained without any type of selection also during 50 generations, approximately.

Therefore, the work consisted in studying the behaviour of flies in two trays, with 40 selected flies, and 10 (O.P.) males in each, and two similar trays with females. On each tray, to avoid any effect resulting from the position, the (O.P.) were distributed lengthwise, 1 (O.P.) flies every 4 selected flies.

The day before the test, 50 flies are passed from the maintenance tubes to the hemolysis tubes with food, one fly in each tube; and these are maintained in refrigerator that way until the following day, the day of the test. On that day the flies pass from the hemolysis tubes with food to the hemolysis tubes without food, without need to immobilize them. The fast period is counted since then. Such period will be variable, depending on the room temperature. At a higher temperature the fly metabolism accelerates and its feeding needs to increase.

In this experiment submitted by us, the room temperature was 22 °C, and the total fast length was of about 14 hours. Observations started when there had been a fast of about 8-9 hours. Until then, in general, the flies were relatively motionless in the small tubes. When these are hungry, they move from the lighted part to the dark part and vice versa. The 50 small tubes holding 1 fly each, numbered from 1 to 50, are placed on the tray. On the tray front side there is a numbered line from 1 to 50, holding the bottom of the small tube, with its opening covered with cotton, inside the tray dark gap.

Meanwhile, in another room, featuring the above mentioned conditions, the (E) flies were left, with food. These, since the (R) flies are initially observed, are maintained in light or darkness, alternatively, every 45 minutes, and the (R) observer did not know the light or darkness state in which the (E) flies were.

Table 1. Selected population flies

Emitter	Mean	Standard deviation	Degree of freedom	t value	1 – tail prob.
Light	62.22	21.32	159	2.23	.014
Darkness	58.07	19.12			

Table 2. Original population flies

Emitter	Mean	Standard deviation	Degree of freedom	t value	2 – tail prob.
Light	48.50	20.41	39	-1.01	.320
Darkness	51.80	19.99			

Table 3. Wild population flies

Emitter	Mean	Standard deviation	Degree of freedom	t value	2 – tail prob.
Light	52.52	19.52	99	-1.12	.264
Darkness	54.21	17.27			

Table 4. Difference between selected and original population

Population	Mean	Standard deviation	Number of cases	t value	1 – tail prob.
Selected	4.13	23.54	160	1.83	.035
Original	-3.30	20.74	40		

Table 5. Difference between selected and wild population

Population	Mean	Standard deviation	Number of cases	t value	1 – tail prob.
Selected	4.13	23.54	160	2.21	.014
Wild	-1.69	15.03	100		

The observer of the (R) flies in front of the tray, put down "/" on a sheet, with columns numbered from 1 to 50, when observing the tubes and the fly was in the light; or "o" if it was not, that is, if the fly remained in the dark side of the tray. This observation and noting procedure was performed 7 times during each change of light of the (E) flies. This may be carried out in 10-15 minutes.

In this experiment the (E) flies were 4 times in the light, and 4 times in the dark, alternatively, with the corresponding (R) observation, except in two occassions – the least number of times, and due to causes beyond personal control. Therefore, the Student "t" analysis had to be made, so the results could be compared against the proportion of the resulting figures. It is understood that the results on behalf of an anomalous communication are obtained when the fasting (R) flies, are the higher number of times in the light, when the (E) flies with food are also in the light, rather than when the (E) flies are in the dark. The same criterion applies to chose the best male and female flies, to obtain with their offsprings a new selected fly population.

Later on, also for comparison purposes, another test was carried out, applying the same conditions as the ones of the depicted tests, with the first generation of wild flies recently caught in the City of Buenos Aires. The test comprised 50 males and 50 females. The (E) flies were similar to the former tests, including wild males and females.

Results

For the statistical analysis 2 values were obtained for each (R) fly, estimating the percentage after an observation in the light, with the emitter in the light and the emitter in the dark. The difference between these values was calculated with a "paired" Student "t", for the population selected, the original population and the wild population.

Also, the selected population was compared with the original and the wild populations, with a "t" for independent groups, taking for this purpose the difference value between the 2 values above mentioned (percentage of observations in the light, with the emitter in the light, minus percentage of observations in the light with the emitter in the dark). The significance level was fixed as .05. The results are presented in Tables 1 to 5. In the case of Tables 2 and 3, the probability was based on 2 tails, because the highest value was not the one expected (emitter in the light), but the opposite one (emitter in the dark).

As it was expected, there was a significant difference in the selected population but not in the original and wild populations. Additionally, there were significant differences among the selected and the original and the wild populations, as above mentioned, means to say, more observations in the light (population selected) when the emitter was in the light.

Conclusions

Based on the results obtained, it would seem that there is the possibility that, by means of a guided selection through 50 generations, a population of *Drosophila melanogaster* may be obtained, whose individuals feature an anomalous communication, which it is understood has not been by means of sight, sound, smell, chemical or vibratory reception. The possibility of a communication by means of some form of electromagnetic energy should be investigated.

References: Ashburner, M., and T.R.F. Wright (editors). 1978, The *Genetics and Biology of* Drosophila. Volume 2a and 2b, Academic Press, New York;

List of Drosophilid species so far described and recorded from Kumaon region, India.

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Genus	Subgenus	Species
Amiota Loew		
	Phortica Schiner	
		bandes Singh and Neigi, 1992
		biprotrusa Chen and Toda, 1998 New record (this paper)
Dettopsomyia Lamb		
		nigrovittata (Malloch, 1924c) New record (this paper)
Drosophila Fallén		
	Dorsilopha Sturtevant	
		busckii Coquillett, 1901
	Drosophila Fallén Str.	
		analspinai Singh and Neigi, 1995
		bageshwarensis Singh and Dash, 1999 (in press)
		bishti Singh and Neigi, 1995
		bizonata Kikkawa and Peng, 1938, new record (this paper)
		elongata Singh and Dash, 1999 (in press)