アナナスショウジョウバエの逆位と生長過程における体重増加

誌名	The Japanese journal of genetics
ISSN	0021504X
著者	蛯谷, 宗哲
巻/号	46巻5号
掲載ページ	p. 309-319
発行年月	1971年10月

JAPAN. J. GENETICS Vol. 46, No. 5: 309-319 (1971)

BODY WEIGHT AT DIFFERENT DEVELOPMENTAL STAGES OF INVERSION HOMOZYGOTES AND HETEROZYGOTES IN *DROSOPHILA ANANASSAE*

NORIAKI EBITANI

Department of Biology, Tokyo Metropolitan University, Tokyo 158

Received July 30, 1970

In *Drosophila ananassae* balanced inversion polymorphisms were reported in the cage populations which contained the following different types of gene arrangements, A and B for the second chromosome or A and B for the third chromosome (Moriwaki *et al.* 1956; Tobari 1962; Tobari and Kojima 1967). Comparing the components of fitness of three karyotypes (AA, BB and AB) for the gene arrangements of the second chromosome, Moriwaki and his coworkers (Moriwaki *et al.* 1957; Moriwaki and Tobari 1963) found that the AB heterozygotes developed faster than either AA or BB homozygotes. However, they have no information about body weight which is an important measure of growth.

The purpose of the present experiment is to see whether the faster development of inversion heterozygotes is accompanied by greater increase in body weight.

MATERIALS AND METHODS

Two lines of *Drosophila ananassae*, In2LA and In2LB homozygotes, were used in the present study. Both strains were sampled from a population cage with an established equilibrium between A and B gene arrangements of the second chromosome. They had been maintained in mass cultures for several years before the present experiment.

Equal numbers of virgin females and males were collected from each homozygous strain and four types of crosses were made between them: AA♀×AA♂, AA♀×BB♂, BB♀×AA♂ and BB♀×BB♂. Forty pairs of flies, 5 to 7 days old, were introduced into a vial with 10 ml of 20% yeast medium and allowed to lay eggs for 2 hours at 25°C. Then, as observed under a binocular microscope, the desired number of eggs was left on the food. As viability did not exhibit any difference among karyotypes under the optimum condition, the number of eggs per vial was determined according to previous data (Moriwaki *et al.* 1957).

Four sets of experiments were performed under the following conditions:

Experiment I Low density (100 eggs per vial) at 25°C
Experiment II Low density (100 eggs per vial) at 29.5°C
Experiment III Low density (100 eggs per vial) at 20°C
Experiment IV High density (400 eggs per vial) at 25°C.

In all sets, four replicate vials were prepared for each type of cross. Body weight was examined at the following various developmental stages:

Experiment	Larvae	Pupae	Adults	
I	22, 44, 95 hrs	126, 168 hrs	2 hrs	5 days
II	22, 44, 75	96, 126	2	5
III	45, 95, 123, 144, 191, 240	288	2	5
IV	52, 72, 100	—	2	5

In the larval and pupal stages, the time for sampling was expressed by the hours elapsed after the midpoint of the egg-laying period. For adults it was expressed by the hours or the days after eclosion.

When larvae or pupae were sampled, the vials were flooded with water to wash them clean off the medium. As a sample for body weight measurement, 100 larvae or pupae were randomly chosen from all individuals which had been washed out into a dish from a group of four replicate vials. Then 100 individuals were divided into five batches of equal size (20 individuals). Each batch was put in a small petri dish, dried for 24 hours at 70°C, and left in a desicator overnight at room temperature. The total weight of a batch was measured with a Mettler microbalance.

RESULTS

Body Weight

a) Low density at 25°C (Experiment I): The mean and standard error of body weight are presented in Table 1. The results of t-test for the difference between mean weight are also shown. Among four genotypes (AA and BB homozygotes, AB heterozygotes drived from AA mothers and BA heterozygotes from BB mothers), no significant difference was found in the egg stage. On the other hand the heterozygotes were significantly heavier than the homozygotes in both larval and pupal stages, though the superiority was not obvious in but a few exceptional cases (22 hours old BA heterozygotes and 126 hours old pupae). Heterozygote superiority is equally true of adult flies, except for the 5 days old male. The ratio of mean heterozygote to mean homozygote is also given in Table 1. For larval stages the ratio is 1.088 on the average, and the average ratio is 1.041 and 1.091 for pupal and adult stages respectively.

b) Low density at 29.5°C and 20°C (Experiment II and III): The results are shown in Tables 2 and 3. As well known, larval growth was remarkably influenced by temperature. The mean length of developmental time from egg to eclosion is about 168 hours at 29.5°C and 360 hours at 20°C, while it is 192 hours under the standard condition of 25°C. The body weight of newly emerged flies also varies with temperature. Flies are heavier at 20°C and lighter at 29.5°C. In both the high and low temperature series, however, heterozygote superiority in body weight was generally found in most of the developmental stages examined. Average heterozygote/homozygote ratio is 1.225 (larvae), 1.019 (pupae) and 1.093 (adults) at 29.5°C and 1.177 (larvae), 1.123 (pupae) and 1.065 (adults) at 20°C.

c) High density at 25°C (Experiment IV): In this experiment the amount of food

Table 1. Body weight of 20 individuals (in micrograms) at different developmental stages under low density at 25°C (Experiment I)

Stages	Genotypes								Significant differences	$\frac{AB+BA}{AA+BB}$
	AA		AB		BA		BB			
	mean	S. E.	mean	S. E.	mean	S. E.	mean	S. E.		
2 hrs eggs	61.4 ±	4.3	66.0 ±	2.7	67.0 ±	7.6	64.0 ±	2.4	BA=AB=BB=AA	1.061
22 hrs larvae	76.2 ±	6.7	83.8 ±	3.7	71.2 ±	6.7	80.0 ±	4.5	AB≫BB≫AA≫BA	0.992
44 hrs larvae	307.4 ±	10.0	354.4 ±	12.2	347.4 ±	9.4	302.2 ±	7.8	AB>BA≫AA>BB	1.151
72 hrs larvae	3142.2 ±	54.4	3560.8 ±	84.3	3510.0 ±	80.9	3015.5 ±	49.4	AB=BA≫AA≫BB	1.148
95 hrs larvae	5392.6 ±	41.0	5860.0 ±	98.9	5839.4 ±	42.8	5625.6 ±	114.3	AB=BA≫BB>AA	1.062
126 hrs pupae	6304.6 ±	132.6	6422.2 ±	122.9	6503.8 ±	79.0	6653.4 ±	84.2	BB=BA=AB=AA	0.998
168 hrs pupae	6217.4 ±	81.1	6612.8 ±	35.4	6666.2 ±	101.5	6047.0 ±	105.8	BA=AB≫AA=BB	1.083
2 hrs adults♀	4153.8 ±	25.7	5406.0 ±	65.6	5328.6 ±	75.4	4216.6 ±	214.1	AB=BA≫BB≫AA	1.282
2 hrs adults♂	3481.8 ±	31.6	3621.2 ±	40.7	4123.6 ±	32.0	3551.8 ±	18.9	BA≫AB=BB=AA	1.101
5 days adults♀	7621.2 ±	76.8	8084.5 ±	43.0	8059.6 ±	56.0	7678.6 ±	52.5	AB=BA≫BB≫AA	1.055
5 days adults♂	4892.0 ±	86.7	4132.0 ±	28.0	4454.5 ±	26.1	4368.0 ±	96.1	AA≫BA=BB≫AB	0.927

>: Indicates the difference significant at 0.05 level of probability.

≫: Indicates the difference significant at 0.01 level of probability.

Table 2. Body weight of 20 individuals (in micrograms) at different developmental stages under low density at 29.5°C (Experiment II)

Stages	Genotypes								Significant differences	$\frac{AB+BA}{AA+BB}$
	AA		AB		BA		BB			
	mean	S. E.	mean	S. E.	mean	S. E.	mean	S. E.		
22 hrs larvae	174.2	± 6.3	225.8	± 6.6	220.4	± 22.3	185.4	± 13.0	AB=BA≫BB≫AA	1.241
44 hrs larvae	673.2	± 33.4	1159.0	± 52.6	1099.6	± 44.2	1014.8	± 10.6	AB=BA>BB≫AA	1.338
75 hrs larvae	5664.4	± 50.0	6076.8	± 78.2	6307.8	± 9.1	5648.2	± 94.6	BA>AB≫AA=BB	1.095
96 hrs pupae	5895.6	± 32.3	6003.4	± 91.3	6174.6	± 78.1	5980.0	± 24.1	BA=AB=BB=AA	1.025
126 hrs pupae	5070.6	± 40.7	5581.6	± 40.5	5345.2	± 96.5	5725.4	± 34.6	BB>AB=BA>AA	1.012
2 hrs adults♀	4138.0	± 70.2	4420.6	± 81.4	4380.5	± 90.0	3990.0	± 13.6	AB=BA=AA>BB	1.083
2 hrs adults♂	3305.7	± 56.2	3519.3	± 12.3	3545.0	± 45.5	3224.4	± 17.9	BA=AB>AA=BB	1.082
5 days adults♀	5954.5	± 82.5	6672.0	± 74.8	6369.2	± 56.8	5511.0	± 129.3	AB=BA>AA=BB	1.136
5 days adults♂	3640.5	± 10.5	3739.7	± 35.1	3769.0	± 27.1	3420.5	± 67.0	BA=AB=AA=BB	1.064

>: Indicates the difference significant at 0.05 level of probability.

≫: Indicates the difference significant at 0.01 level of probability.

Table 3. Body weight of 20 individuals (in micrograms) at different developmental stages under low density at 20°C (Experiment III)

Stages	Genotypes								Significant differences	$\frac{AB+BA}{AA+BB}$
	AA		AB		BA		BB			
	mean	S. E.	mean	S. E.	mean	S. E.	mean	S. E.		
45 hrs larvae	83.2	± 6.7	98.2	± 2.8	101.6	± 2.8	113.4	± 9.9	BB>BA=AB>AA	1.016
96 hrs larvae	490.6	± 28.2	903.6	± 20.9	827.6	± 16.1	859.8	± 34.0	AB=BB=BA≫AA	1.282
123 hrs larvae	2660.6	± 60.4	3560.2	± 49.1	3421.8	± 89.6	3219.0	± 41.6	AB=BA>BB≫AA	1.187
144 hrs larvae	3015.6	± 75.6	4718.2	± 148.0	5965.0	± 104.5	4318.2	± 136.6	BA≫AB≫BB≫AA	1.457
191 hrs larvae	6760.0	± 49.7	7078.0	± 5.2	7155.2	± 89.6	6585.4	± 34.0	BA=AB≫AA=BB	1.067
240 hrs larvae	5631.2	± 110.9	6421.8	± 109.4	6530.6	± 112.8	6643.6	± 48.5	BB=BA=AB≫AA	1.055
288 hrs pupae	4620.4	± 26.5	6196.0	± 59.6	6248.6	± 34.9	6456.3	± 45.5	BB>BA=AB≫AA	1.123
2 hrs adults♀	4571.0	± 73.5	5465.8	± 86.4	6100.0	± 47.7	5048.6	± 45.4	BA≫AB≫BB≫AA	1.202
2 hrs adults♂	3795.5	± 35.4	4136.8	± 12.1	4340.4	± 45.0	3858.0	± 35.4	BA=AB>BB=AA	1.108
5 days adults♀	7242.6	± 227.6	8111.3	± 223.6	8525.8	± 145.5	8200.8	± 70.0	BA≫BB=AB≫AA	1.077
5 days adults♂	4849.5	± 87.2	4606.5	± 42.8	4913.8	± 72.8	4929.8	± 76.5	BB=BA=AA>AB	0.974

>: Indicates the difference significant at 0.05 level of probability.

≫: Indicates the difference significant at 0.01 level of probability.

Table 4. Body weight of 20 individuals (in micrograms) at different developmental stages under high density at 25°C (Experiment IV)

Stages	Genotypes								Significant differences	$\frac{AB+BA}{AA+BB}$
	AA		AB		BA		BB			
	mean	S. E.	mean	S. E.	mean	S. E.	mean	S. E.		
52 hrs larvae	401.6	± 4.7	414.8	± 15.5	502.2	± 30.3	389.8	± 14.1	BA≫AB=AA=BB	1.159
72 hrs larvae	2475.8	± 29.2	3202.2	± 68.7	3466.4	± 70.4	2308.0	± 91.9	BA>AB≫AA=BB	1.394
100 hrs larvae	5458.6	± 121.0	5664.8	± 83.2	6076.6	± 99.7	5143.0	± 67.4	AB≫AB=AA>BB	1.108
2 hrs adults♀	3863.6	± 68.0	4439.6	± 136.3	4306.6	± 70.6	3282.4	± 56.4	AB=BA≫AA≫BB	1.224
2 hrs adults♂	3115.8	± 120.6	3114.8	± 81.7	3230.2	± 61.2	2617.4	± 77.0	BA=AA=AB≫BB	1.107
5 days adults♀	6994.6	± 89.9	6371.6	± 35.6	6526.4	± 154.6	6566.4	± 74.8	AA>BB=BA=AB	0.951
5 days adults♂	3887.6	± 65.4	3871.8	± 73.6	3655.6	± 50.8	3766.8	± 60.5	AA=AB=BB=BA	0.983

>: Indicates the difference significant at 0.05 level of probability.

≫: Indicates the difference significant at 0.01 level of probability.

available to individual larvae is one forth of those in Experiments I-III. As a result of poor nutritional conditions, larval development was slightly slower than in the conditions of low density at 25°C. The body weight of newly emerged flies was also affected, being lighter by 17-18% than corresponding flies in Experiment I. As to the difference between heterozygotes and homozygotes, the same tendency as those in Experiments I-III was apparent. Heterozygous larvae were always significantly heavier than homozygous ones. The average heterozygote/homozygote ratio was 1.140 throughout larval stages. Although heterozygote superiority is not clear in older fllies, the ratio is still 1.066 on the average for adult stages.

d) Rate of increase in body weight As a measure of the rate of growth, the increment in larval body weight per hour was calculated from Tables 1-4 and compared among different genotypes (Table 5). As expected from the heterozygote superiority in larval body weight, heterozygous larvae usually showed a large weight increment per hour. It follows that the mean heterozygote/mean homozygote ratio in body weight increment is mostly larger than unity. In fact, the ratio is always much higher than unity in early developmental stages, being 1.203, 1.344, 1.327 and 1.441 in Experiments I-IV respectively. As clearly seen in Table 5, however, the ratio decreases gradually with larval development, being around unity at the end of larval stages.

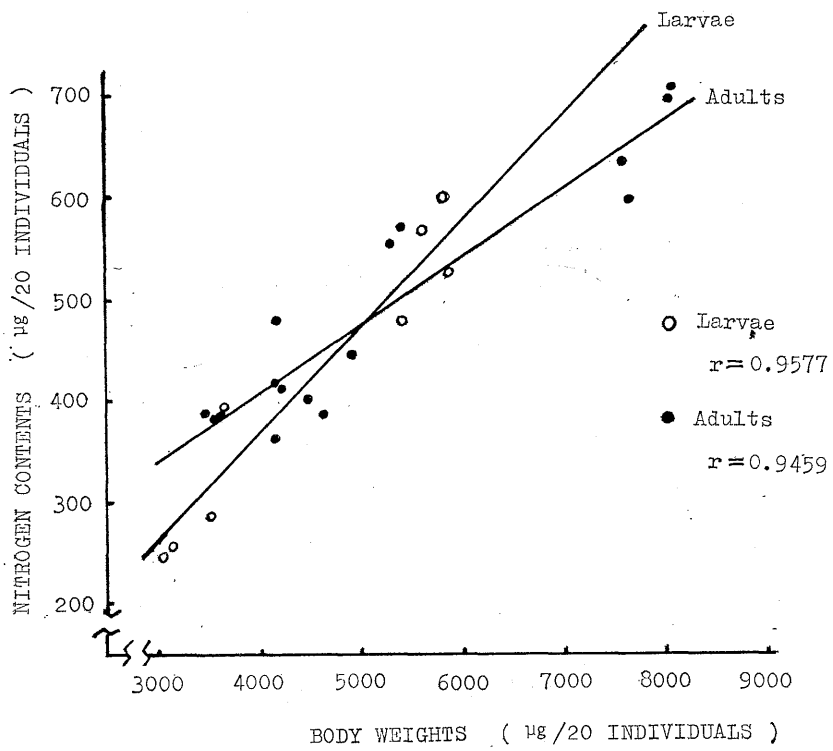

Fig. 1. The regression of nitrogen contents on the body weight of larvae and adult flies

Table 5. The rates of increase of larval body weight in Experiment I, II, III and IV

Experiments	Genotypes	Increments of body weight (μg/20 individuals/Hour)		
		22 — 44 hrs	44 — 72 hrs	72 — 95 hrs
I	AA	10.6 ± 0.5	101.3 ± 1.9	97.7 ± 2.0
	AB	12.3 ± 0.6	114.5 ± 3.0	100.0 ± 4.3
	BA	12.6 ± 0.4	112.9 ± 2.9	101.3 ± 1.7
	BB	10.1 ± 0.4	96.9 ± 5.6	113.5 ± 3.0
		BA=AB>AA≫BB	AB=BA≫AA>BB	BB>BA=AB=AA
	$\frac{AB+BA}{AA+BB}$	1.203	1.147	0.953
		22 — 44 hrs	44 — 75 hrs	
II	AA	22.7 ± 1.5	161.0 ± 1.6	
	AB	42.4 ± 2.4	158.6 ± 2.5	
	BA	40.0 ± 2.0	168.0 ± 0.9	
	BB	38.6 ± 1.0	149.5 ± 3.1	
		AB=BA=BB≫AA	BA≫AA=AB=BB	
	$\frac{AB+BA}{AA+BB}$	1.344	1.037	
		45 — 96 hrs	96 — 123 hrs	123 — 191 hrs
III	AA	8.0 ± 0.8	80.4 ± 2.2	67.4 ± 0.4
	AB	15.8 ± 0.1	96.4 ± 2.8	51.7 ± 0.5
	BA	14.2 ± 0.3	96.1 ± 3.3	54.9 ± 1.4
	BB	14.6 ± 0.7	87.4 ± 1.4	49.5 ± 0.7
		AB=BB=BA≫AA	AB=BA>BB>AA	AA≫BA=AB=BB
	$\frac{AB+BA}{AA+BB}$	1.327	1.147	0.910
		52 — 72 hrs	72 — 100 hrs	
IV	AA	103.7 ± 4.6	106.5 ± 3.8	
	AB	139.4 ± 3.4	88.0 ± 1.3	
	BA	148.4 ± 3.5	93.2 ± 2.5	
	BB	95.9 ± 4.5	101.3 ± 1.4	
		BA=AB≫AA=BB	AA=BB>BA=AB	
	$\frac{AB+BA}{AA+BB}$	1.442	0.872	

Table 6. Nitrogen content of 20 individuals (in micrograms) at different developmental stages under low density at 25°C

Stages	Genotypes								Significant difference
	AA		AB		BA		BB		
	mean	S. E.	mean	S. E.	mean	S. E.	mean	S. E.	
72 hrs larvae	255.5	± 12.3	292.6	± 6.9	285.6	± 4.0	245.0	± 1.6	AB=BA>AA=BB
95 hrs larvae	479.3	± 25.2	526.4	± 54.8	607.1	± 11.4	567.9	± 20.8	BA=BB=AB=AA
168 hrs pupae	470.4	± 3.5	494.7	± 35.6	487.7	± 43.6	516.1	± 8.1	BB=AB=BA=AA
2 hrs adults♀	477.8	± 4.2	567.4	± 2.6	551.6	± 9.8	413.4	± 20.4	AB=BA≫AA>BB
2 hrs adults♂	389.2	± 1.6	386.4	± 8.6	418.0	± 10.8	383.6	± 14.5	BA>AA=AB=BB
5 days adults♀	633.8	± 5.2	701.0	± 8.9	691.6	± 15.4	599.2	± 27.3	AB=BA≫AA=BB
5 days adults♂	446.2	± 11.7	363.0	± 3.4	405.0	± 8.9	383.6	± 1.6	AA>BA>BB≫AB

>: Indicates the difference significant at 0.05 level of probability.
≫: Indicates the difference significant at 0.01 level of probability.

Nitrogen content

In the Experiment I, parallel with body weight measurement, the nitrogen content of 20 individuals was examined by the usual semimicro-Kjeldahl method. Three measurements were repeated at five developmental stages (72 and 96 hours larvae, 168 hours pupae and 2 hours and 5 days flies). The results are summarized in Table 6. In most cases heterozygotes exceed homozygotes in nitrogen content. The general tendency is similar to that found in body weight (Table I). This is a natural result of the fact that there is strong correlation between nitrogen content and dry weight as seen in Fig. 1. The correlation coefficient is $r=0.9577$ for larval stages and $r=0.9459$ for adults. From these relationships, it may be said that body weight is useful as a rough measure of the amount of protein.

DISCUSSION

The relationship between two measures of growth, speed of development and increase in body weight, varies with environmental conditions, such as culture medium, density and temperature.

On *Drosophila melanogaster*, Robertson (1960) found that the larval period is prolonged but final body size is not necessarily reduced, provided the diet is sub-optimal, but when the diet is further reduced development time is lengthened and body size is reduced as well.

Lints (1963) studied the relation between size and duration of development at two different egg-densities and at three different temperatures. He observed that at low egg-density size decreases regularly as development time increases, at high density there is no relationship between prolongation of the development time and size. As to the effect of temperature, at higher temperature the development time was shorter but flies were smaller than at lower temperatures. The same relationship was found in the present experiments at 20°, 25° and 29.5°C (Experiment I, II and III). These results indicate that speed of development and increase in body weight represent different two measurements of growth.

The relationship between the two measurements may vary with differential environmental effects on different genotypes. On *D. ananassae* larvae grown at 19° and 25°C, Moriwaki *et al.* (1956) found faster development of inversion heterozygotes than homozygotes. The present experiments clearly revealed that faster development of heterozygotes was always accompanied with a faster increase in body weight. In all experiments which were performed under different nutritional and temperature conditions, the body weight of inversion heterozygotes was heavier than that of homozygotes throughout all developmental stages. In the larval stages, the body weight increment per hour was larger in heterozygotes than in homozygotes. This may indicate that heterozygotes exceed homozygotes in the speed of protein synthesis. Since heterozygote/homozygote ratio in body weight increment is much larger than unity in early stages of larval development, and since it is almost unity in late larvae, it is clear that heterozygote superiority over homozygotes in larval growth is more pronounced in earlier stages.

Moriwaki *et al.* (1957) and Moriwaki and Tobari (1963) reported the clear maternal

effects in larval development. In2LA/2LB, heterozygotes (AB) from AA mothers grow slightly faster than BA heterozygotes from BB mothers. In the present experiments, however, no systematic differences were found between body weight of AB and BA heterozygotes. That is, no maternal effect was confirmed as to the body weight increase.

SUMARRY

Body weight of *Drosophila ananassae* at different developmental stages was measured under different nutritional and temperature conditions. Comparison of body weight was made among four different karyotypes, derived from a subterminal inversion In2L.

Body weight of inversion heterozygotes was significantly heavier than that of homozygotes throughout all developmental stages under most of the experimental conditions. Larval weight increment per hour was larger in heterozygotes than homozygotes, especially in earlier larval stages.

Heterozygote superiority was also true for nitrogen content, which was strongly correlated with body weight.

ACKNOWLEDGMENTS

The author is indebted to Drs. S. Ohba, Y.N. Tobari and D. Moriwaki for their valuable suggestions and criticisms in the course of this study. He also thanks Professor Daniel McCoy of the Science English Center of Sophia University Biological Laboratory for reading manuscript.

LITERATURE CITED

Lints, F. A., 1963 Size in relation to development time and egg density in *Drosophila melanogaster*. Nature **197**: 1128-1130.

Moriwaki, D., M. Ohnishi (Shirai), and Y. Nakajima, 1957 Analysis of heterosis in populations of *Drosophila ananassae*. Proc. Intern. Genet. Symp. **1956**: 370-379.

Moriwaki, D., M. Shirai, Y. Yoshida, and M. Tsusue, 1956 Frequency changes of two inversion arrangements in artificial populations of *D. ananassae*. In "Syudan Idengaku" Edited by T. Komai and K. Sakai. Baifukan, Tokyo, p. 95 (in Japanese).

Moriwaki, D., and Y. N. Tobari, 1963 Maternal effects and heterosis in *Drosophila ananassae*. Genetics **48**: 171-176.

Robertson, F. W., 1960 The ecological genetics of growth in *Drosophila*. I. Body size and development time on different diets. Genet. Res. I: 288-304.

Tobari, Y. N., 1962 Heterosis relating to a terminal inversion in artificial population of *Drosophila ananassae*. Japan. J. Genetics **37**: 302-309.

Tobari, Y. N., and K. Kojima, 1967 Selective modes associated with inversion karyotypes in *Drosophila ananassae*. I. Frequency dependent selection. Genetics **57**: 179-188.