PHENOTYPICAL VARIATION IN BODY AND CELL SIZE OF *DROSOPHILA MELANOGASTER*

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I.

The purpose of this paper is to contribute to the solution of the question of the relationship of the cell size and body size, using well-known and standard material. The literature devoted to this question is very extensive, but most of the work done cannot be considered to fulfil the requirements of exact experimental investigation, in regard either to the control of conditions, or the homogeneity of the material, or the precision and accuracy of the treatment. Comparatively modern compilations of the data available have been made by Levi (1906) and Martini (1924).

Concerning the more limited problem of the correlation of body size and cell size in Diptera there have been two recently published papers. Loewenthal (1923) attacks a problem which corresponds to one part of the present investigation, namely the influence of underfeeding on the body and cell size of the blow-fly. The first criticism which may be made of Loewenthal's work is that he does not give any indication of the ages of the normal and underfed maggots. It therefore is not clear whether the observed smaller size of the hypodermis cells is due to differences in the age of larvae or in the feeding. At the same time Loewenthal does not find any difference in the cell size of the gonad rudiments, in spite of their difference in size. The following conclusion is reached (p. 91) : " Danach ist die Körpergrösse der ausgebildeten Imagostadiums unabhängig von der Zellgrösse und allein bedingt von der mehr oder minder grossen Zellanzahl." Further a totally incorrect statement is made concerning the absence of cell divisions during the larval life (p. 92) : " Mit Abschluss der Embryonalentwicklung stellen die larvalen Zellen ihre Vermehrungstätigkeit ein, das ganze Wachstum der Larve von wenigen mm Länge nach dem Schlüpfen aus dem Ei bis zur Länge von 2 cm einer verpuppungsreifen Ruhelarve beruht allein —wenn man von den während der Larvalperiode für die Gesamtgrösse nicht ins Gewicht fallenden Imaginalanlagen absieht—auf dem Grössenwachstum der Zellen." Przibram's and Megušar's (1912) investigations showed that this is not the case in the postembryonal develop-

ment of *Sphodromantis* (Orthoptera, Mantidae) and I (1929) have shown also that the metamorphosis of *Drosophila* is connected with six simultaneous divisions of the cells of the whole body.

The same subject of the relationship of the size of an organ and the size of the cell has been touched upon by Bridges (1921, 1925). In both of his papers differences in the cell structure, namely, nuclear structure, are shown to be connected with the size of the whole body and its organs. It was discovered that these intersex-producing females (triploid) could be identified by their somatic characters, namely, large coarse bristles and large roughish eyes (1921, p. 253). In the second paper it is the size of the ommatidia which is shown to be different in flies having different chromosomal complexes. "The cells of triploid individuals are readily seen to be larger than the cells of diploids, and correspondingly their facets are larger" (Bridges, 1925, p. 709).

I became interested in the problem of body size and cell size years ago while working on the oceanographic expedition of the Floating Marine Scientific Institute to the Russian arctic seas. The first expedition in 1921 gave very impressive material on the geographical variation in the dimensions of the body of different marine animals. It could be particularly easily shown on such a group of animals as Isopoda, which have a postembryonal development ending with a definite imaginal stage analogous to that of insects. Extensive biometrical data on variation of Isopoda, taken from localities with different temperatures, showed perfectly that the colder regions (for instance, the Kara Sea) are populated by races which have a larger body size than regions with warmer water temperature (Barents Sea). On the second expedition I strove to collect some material on the histology of local races of some of the species of Isopoda. But the severe conditions of navigation during this and following summers did not allow the accomplishment of this intention. During the winter of 1927–28, working at this Institute, I succeeded in working out a more or less accurate method of producing *Drosophila* imagoes of different sizes, using two factors, temperature and underfeeding. The method of counting the number of hairs on the wings of *Drosophila* as a method of estimating the number of cells on a certain surface of the wing was discovered by a friend, Dr. Th. Dobzhansky (1929), who was kind enough to explain it to me. I have the pleasure to express also my deepest gratitude to Dr. Raymond Pearl for criticism and valuable suggestions.

II.

Two factors have been used in producing flies of an abnormal size. It was shown in an earlier paper that the first of them was the low tem-

perature, which decreases the rate of development and produces flies of a larger size (see Alpatov and Pearl, 1929). The method of collecting new-born larvæ has already been described (Alpatov, 1929). Flies belonging to Wild Line 107 have been taken for parents of our experimental animals, the collected larvæ being 0–2 hours old at the moment of putting them on food. The bottles had been planted with yeast 2 hours before the putting on of larvæ, and watered with a few drops of distilled water. Electric and low temperature Hearson incubators were used for keeping the bottles with flies. Five bottles with 50 larvæ each were kept at 18° C., five others at 28° C. The development from the moment of the populating of the bottle till the moment of the pupation was more than twice as long in the cold series as in the warm. It is unnecessary to discuss here at length the question of temperature and development rate, this having been done in another paper (Alpatov and Pearl, 1929). The technique of breeding in the experiment with underfeeding was the same except for the fact that the yeast was put in the bottles with synthetic medium at the moment of populating the bottles with larvæ.

A method of getting undernourished larvæ by taking larvæ from the food before the normal end of feeding has been used by various workers, for instance, Ezhikov (1917, 1922), Smirnov (1926, 1927), Cousin (1926), Herms (1928) and others. Most of these authors did not attempt to determine with sufficient accuracy the moment of taking the larvæ from the food, Herms being in that respect an exception. In the present investigation, larvæ were taken from the food exactly 48 hours after the moment of populating the bottles with 0–2 hour-old larvæ. Larvæ which reached the desired age were taken from bottles and placed in half-pint bottles containing plain agar. The mouths of the bottles were covered with 40 mm. watch glasses and sealed with plastaline used in modelling. This was done in order to prevent the larvæ, which become very active, from crawling out. The day after the larvæ had turned into pupæ the watch glasses were replaced by the usual cotton stoppers.

Table I shows that the larvæ with a subnormal period of feeding pupate earlier than normally fed ones. This can be compared with Kopeč's (1924) statement that ". . . if we begin to apply starvation to older specimens during developmental stages . . . the transformation of these animals is accelerated." A little longer prepupal development of the normally fed larvæ, those which served as controls to the underfed being compared with the 28° flies of the early October experiment, cannot be very easily interpreted. It might exist in a difference in conditions—perhaps a difference in yeast growth which lengthened the duration of development of larvæ in the second set of experiments.

TABLE I

Data on the Conditions of the Development of Flies Reared for the Study of the Problem of Cell Size

	Temperature limits of variation	Average	Time of the beginning of the experiment	Time from egg until pupation, in hours	Time of feeding
Underfed flies	Kept at 28°	—	October 24, 1928	80.39±.50	48 hours
Normally fed flies	Kept at 28°	—	October 24, 1928	93.16±.74	Until normal leaving of the food
28° flies	27.1–28.9°	28.2°	October 8, 1928	87.40±.36	Until normal leaving of the food
18° flies	17.0–20.0°	18.2°	October 8, 1928	200.86±.89	Until normal leaving of the food

The flies have been collected in 70 per cent alcohol and measured in glycerine under a cover glass. The following characters on the wings of collected flies have been studied: the length and width of the wing, and the number of hairs on a surface equal to 0.1 square mm. on the lower surface of the wing. Fig. 1 represents the points of measurement and the place where the hairs have been counted.

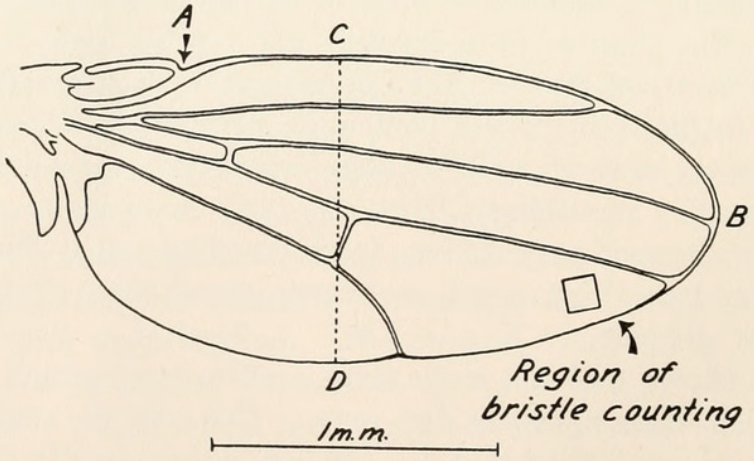

FIG. 1. Measurements of the wing. *AB*, length of the wing, *CD*, width of the wing. The square shows the area of the bristle countings.

For the measurements the following optical systems were chosen: Spencer 25.4 mm. objective and a micrometer ocular in a No. 2 ocular. The countings of the hairs of the lower surface of the wing were done in a way approved by Th. Dobzhansky. Pieces of paper with squares

representing 0.1 square mm. at a given magnification have been prepared by projecting through an Abbé camera lucida 0.1 mm. from an object micrometer placed on the microscope stage. A Spencer microscope was used with objective 4 mm. and ocular × 10. The hairs have been projected by means of the camera lucida and drawn with a sharp pencil. Only hairs whose bases happened to fall inside the square have been counted (Fig. 2).

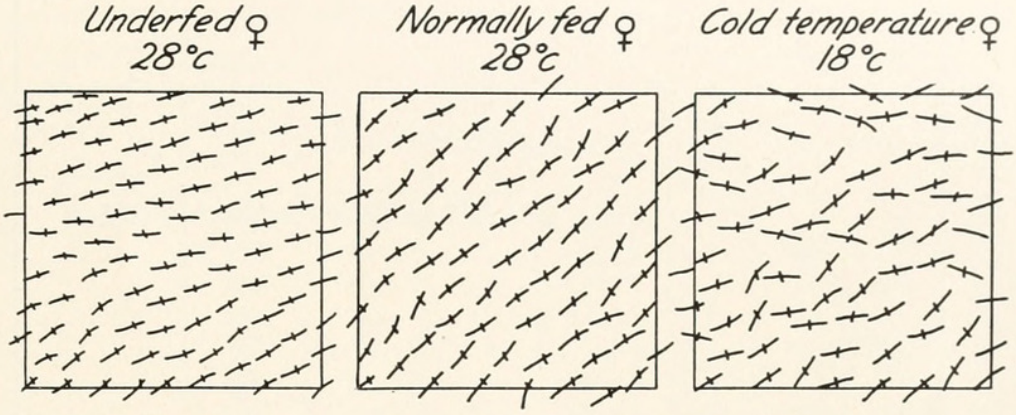

FIG. 2. This figure represents the bristles on the surface of 0.1 mm.² in the lower surface of the wings of underfed, normally fed, and cold temperature females. The bristles which have a line across their middle have been counted, those without lines had their basis outside the limits of the 0.1 mm.² and have not been counted.

We did not consider it wise to count the hairs exactly at a certain point (in so many parts of a millimeter from a certain vein) as has been done by Dobzhansky. There are two reasons for not doing so. First of all the distribution of hair on that part of the wing is more or less uniform. On the other hand, the wings of underfed and normal are so different in size that a distance expressed in absolute measurement would show morphologically quite different regions. Fifty specimens of each set of underfed, normal fed and 18° flies were studied in regard to the density of the hairs. Dr. Th. Dobzhansky succeeded in finding that on the wings each hair corresponds to a separate cell. This can be seen on specimens of flies just emerged from the pupæ. The wings look opaque and the cells can be distinctly seen. It is very likely that the tiny hair covering the thorax of *Drosophila* corresponds also to hypodermis cells, and their density may also be used as a method of studying the size of the hypodermal cells.

III.

It is desirable at this stage to digress briefly to consider a matter which arose as an extension of the original problem. It is the question

of functional relation between the time of larval feeding and the final size of the flies. First of all I reinvestigated the data published by Herms (1928) and found that when plotted on a diagram they reveal a very interesting picture.

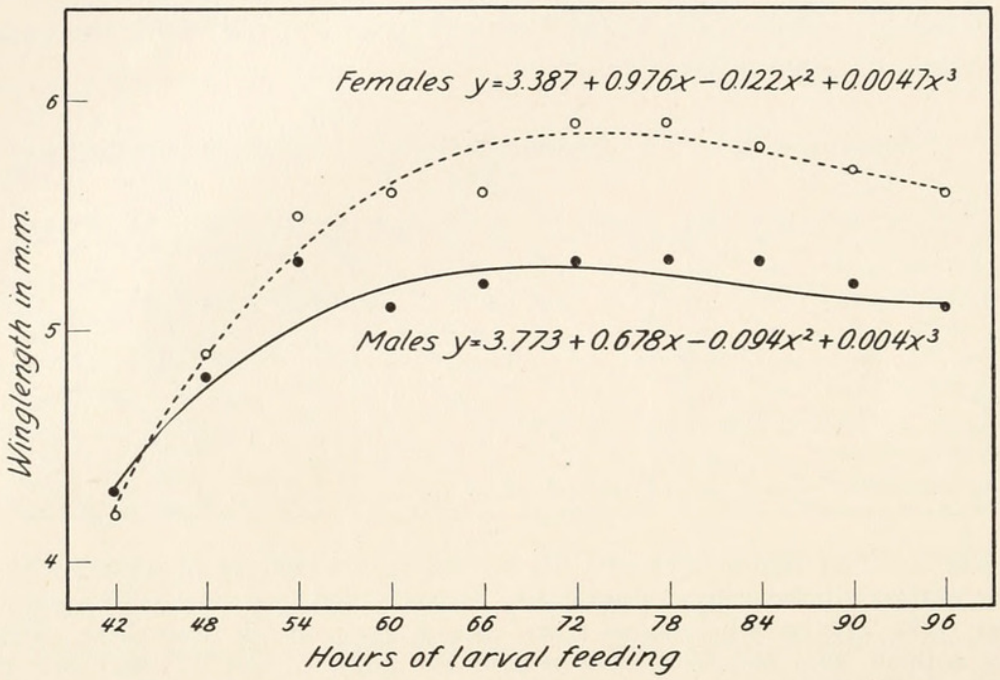

FIG. 3. The relation between the wing length and the length of the feeding period in *Lucilia sericata*. Data from Herms (1928).

Fig. 3 represents Herms' data and two cubic parabolas which I fitted to the observed points. Up to the 78 hour point the trend of the curves represents the upper part of a typical growth curve. There cannot be any doubt that this trend corresponds exactly to the upper branch of the logistic curve which can be fitted to the growth of *Drosophila* larvæ of the third instar (see Alpatov, 1929). But the decline after 78 hours is quite remarkable. Going back to my paper on larval growth in *Drosophila* I was able to find in Fig. 13 particularly a slight indication as to an analogous decline of the size of the larvæ killed at the latter end of the life of the culture. It was therefore decided to clear up this question on specially collected material. This was done in April 1929. Forty bottles containing 0.500 grams of Magic yeast with 25 drops distilled water were populated by 80 larvæ each. Five drops of water were added every day during the larval growth. The experiment was run at a temperature of 25° C.

Table II contains data on the sex relations in the material studied. Let us first compare the percentage of males emerged from larvæ taken from the food at 48–80 hours, which is equal to 102.6, with that of

males emerged from larvæ taken from food at the age of 84–96 hours, in which case the percentage is 89.9. This difference finds its explanation in the fact that male larvæ in our case started pupation earlier than females, which is shown by the very high percentage of males among

TABLE II

Absolute and Relative Numbers of Larvæ, Pupæ and Adult Flies in the Experiment on Underfeeding of Larvæ

Hours from the beginning of feeding	Number of larvæ taken from the food	Number of the pupæ observed	Number of pupæ unable to produce flies	Number of flies emerged				
				Total	In per cent of the larvæ	Male	Female	Male in per cent of female
48	151	—	17	30	19	18	12	150.0
52	170	—	7	80	47.1	44	36	122.2
56	152	—	11	87	57.2	37	50	74.0
60	158	—	12	123	77.8	62	61	101.6
64	162	—	2	154	95.1	72	82	87.8
68	170	—	7	153	90.0	85	68	125.0
72	151	—	12	117	77.6	69	48	143.8
76	135	—	3	127	94.1	56	71	78.9
80	150	—	1	142	94.7	70	72	97.2
Total 48–80	—	—	—	—	—	513	500	102.6
84	86	36	2	78	90.7	39	39	100.0
88	137	12	4	115	83.9	62	53	117.0
90	111	30	11	100	90.1	56	44	127.3
92	110	46	3	100	90.1	45	55	81.8
94	95	61	10	79	83.2	30	49	61.2
96	85	73	6	75	88.2	27	48	56.3
Total 84–96	—	—	—	—	—	259	288	89.9
Flies emerged from pupæ at 84–96 hours								
84	—	—	—	—	—	25	11	227.3
88	—	—	—	—	—	6	5	120.0
90	—	—	—	—	—	23	8	287.5
92	—	—	—	—	—	30	12	250.0
94	—	—	—	—	—	32	23	139.1
96	—	—	—	—	—	45	27	166.1
Total 84–96	—	—	—	—	—	161	86	187.2

larvæ pupated naturally at the age of 84–96 hours—187.2 per cent. On the whole the group of bottles which was taken to get larvæ fed 84–96 hours shows a percentage of males equal to 109.1. Comparing it with the sex proportion in normal undisturbed bottles where we had

356 males and 415 females, we find that the normal percentage of males is 85.8. We can therefore draw the conclusion that there is a definite preponderance of males among flies emerged from the underfed larvæ. In other words it seems that a selective process makes the male more resistant to underfeeding.

TABLE III

Wing Length, Width and Relative Width of the Flies Emerged from Larvæ taken from the Food at Different Hours

Hours	Males				Females			
	Length	Width	Index	Number	Length	Width	Index	Number
48	1.107	.6490	58.6	17	1.207	.6972	57.8	12
52	1.164	.6847	58.8	25	1.239	.7154	57.7	25
56	1.331	.7847	59.0	25	1.413	.8034	56.9	25
60	1.321	.7697	58.3	25	1.493	.8459	56.7	25
64	1.394	.8145	58.4	25	1.572	.8898	56.6	25
68	1.409	.8289	58.8	25	1.588	.9102	57.3	25
72	1.406	.8428	59.9	25	1.561	.9083	58.2	25
76	1.371	.7983	58.2	25	1.511	.8493	56.2	25
80	1.412	.8261	58.5	25	1.586	.8938	56.4	25
84	1.476	.8833	59.8	25	1.673	.9349	55.9	25
88	1.440	.8516	59.1	25	1.641	.9321	56.8	25
90	1.472	.8777	59.6	25	1.646	.9255	56.2	25
92	1.423	.8468	59.5	25	1.614	.9032	56.0	25
94	1.426	.8457	59.3	25	1.613	.9077	56.3	25
96	1.444	.8686	60.2	25	1.608	.9083	56.4	25

TABLE IV

Wing Length, Width and Relative Width of the Flies Emerged from Pupæ Pupated at a Given Hour, and of Those Emerged from Pupæ Pupated during the Whole Pupation Period

Hour	Males				Females			
	Length	Width	Index	Number	Length	Width	Index	Number
84	1.484	.8805	59.3	24	1.709	.9901	57.9	11
88	1.490	.8887	59.6	6	1.728	.9944	57.5	4
90	1.493	.8876	59.5	15	1.715	1.007	56.7	8
92	1.456	.8516	58.5	25	1.649	.9312	56.5	12
94	1.455	.8499	58.4	25	1.644	.9389	57.1	23
96	1.463	.8544	58.4	25	1.672	.9536	57.0	25
Normal pupation	1.475	.8745	59.3	40	1.673	.9668	57.8	40

Tables III and IV give the average length and width of wings of our material. The wing length is graphically represented in Fig. 4.

With the exception of some cases (72, 76 and 80 hours) the material confirms what could be seen on curves based on Herms' data. The most interesting thing is the declining slope of the curves toward the end. It is not only with underfed flies that this decline is noticeable,

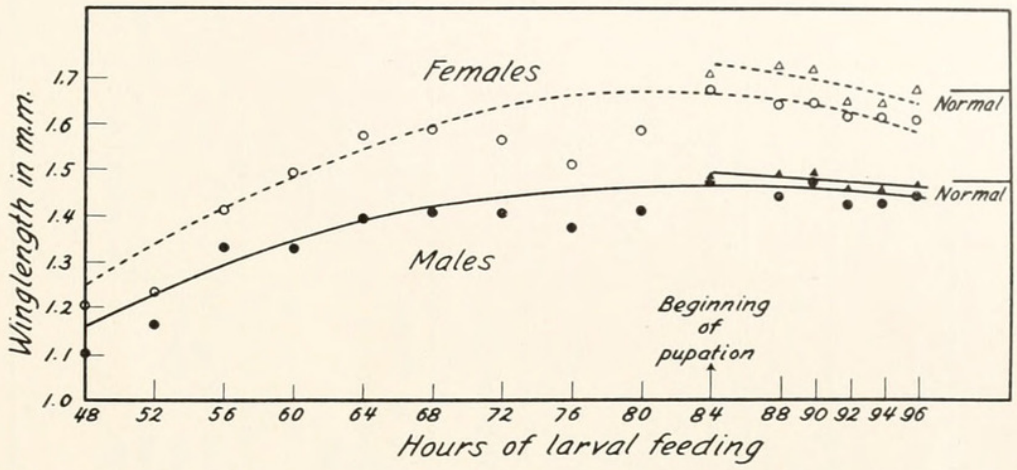

FIG. 4. This figure represents the relation of the length of the wing and the length of the larval feeding in *Drosophila melanogaster*. The triangles indicate the length of wings of flies pupated at certain hours.

but the flies normally pupated in the beginning of the pupation period had longer wings (*i.e.*, larger bodies) than flies in which pupation has been delayed.

Table V gives the statistical proof of this conclusion. It can be definitely seen that in males and females without regard to whether the pupation is going naturally or the flies emerge from larvæ taken from the food, those which pupate first are larger than those which pupate later. We may express the observed phenomenon in a little different form. There is a negative correlation between the duration of larval life and the size reached during growth. The faster the larva grows the sooner it reaches the pupal stage. We take the liberty of comparing our case with the experiments on *Cucumis melo* described by Pearl in his book, *The Rate of Living,* (1928). The larvæ which reach a larger size in a short time have naturally a higher rate of growth than larvæ which remain small for a longer time. Therefore the statement brought forward by Pearl (p. 139) that "between growth rate and duration of life to the beginning of death the correlation is negative and significant in degree" can be perfectly well applied to our case.

We do not know whether these differences arise really as a result of inherent vitality or are the result of differences of treatment of larvæ during the population of the bottle. Further experiments have to solve this question. Our results are very close to Kopeč's discovery (1924) of the negative correlation between the duration of larval period

TABLE V

Length of the Wings (in mm.) of Flies Emerged from Larvæ Taken at a Given Hour from the Food, from Pupæ Formed at a Certain Hour, and from Control Bottles

Males

			Difference and ratio			
Larvæ taken from the food at 84–88 and 90 hours. $N = 75$	Mean	1.458±.004	0.27±.005	Larvæ taken from the food at 92–94–96 hours. $N = 74$	Mean	1.431±.003
	Standard deviation	.0450	$R = 5.4$		Standard deviation	.0405
	C. of V.	3.08±.17			C. of V.	2.83±.16
Difference and ratio	—	.025±.005 $R = 5.0$	—	Difference and ratio	—	.026±.004 $R = 6.5$
Pupated at 84–88–90 hours. $N = 45$	Mean	1.433±.003	.022±.004	Pupated at 92–94–96 hours. $N = 74$	Mean	1.457±.002
	Standard deviation	.0265	$R = 5.5$		Standard deviation	.0277
	C. of V.	1.79±.13			C. of V.	1.90±.11
—	—	—	—	Pupated during the whole pupation period. $N = 40$	Mean	1.469±.004
					Standard deviation.	.0393
					C. of V.	2.68±.20

TABLE V—*Continued*

Females

			Difference and ratio			
Larvæ taken from the food at 84–88–90 hours.	Mean	1.650±.004	.043±.005	Larvæ taken from the food at 92–94–96 hours.	Mean	1.607±.003
N = —	Standard deviation	.0545		N = 75	Standard deviation	.0432
	C. of V.	3.30±.18			C. of V.	2.69±.15
Difference and ratio	—	.064±.006 R=10.7	—	Difference and ratio	—	.047±.004 R=11.75
Pupated at 84–88–90 hours.	Mean	1.714±.004	.060±.005	Pupated at 92–94–96 hours.	Mean	1.654±.003
N = —	Standard deviation	.0312		N = —	Standard deviation	.0339
	C. of V.	1.82±.18			C. of V.	2.05±.13
—	—	—	—	Pupated during the whole pupation period.	Mean	1.672±.005
				N = —	Standard deviation	.0507
					C. of V.	3.03±.23

and the weight of the chrysalids in *Lymantria dispar* (L.). This negative correlation found in twelve experimental groups out of sixteen is particularly well expressed in males.

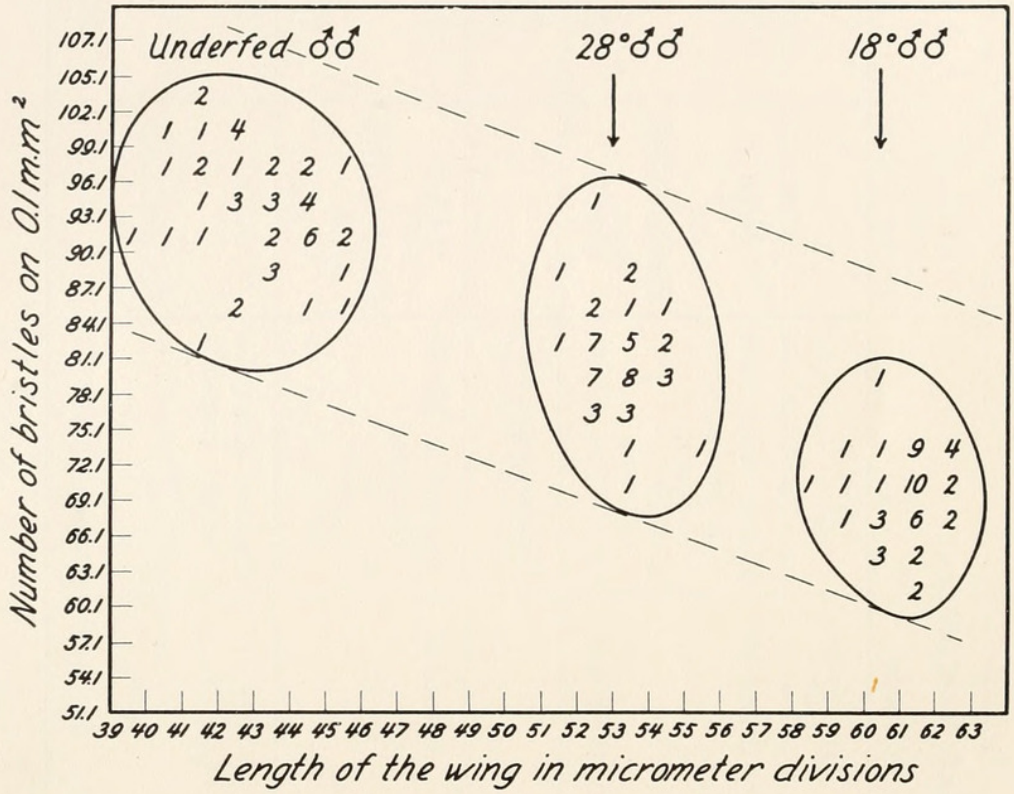

FIG. 5. Correlation between the length of the wings and the number of bristles per 0.1 mm.² on the lower surface of the wings of the male.

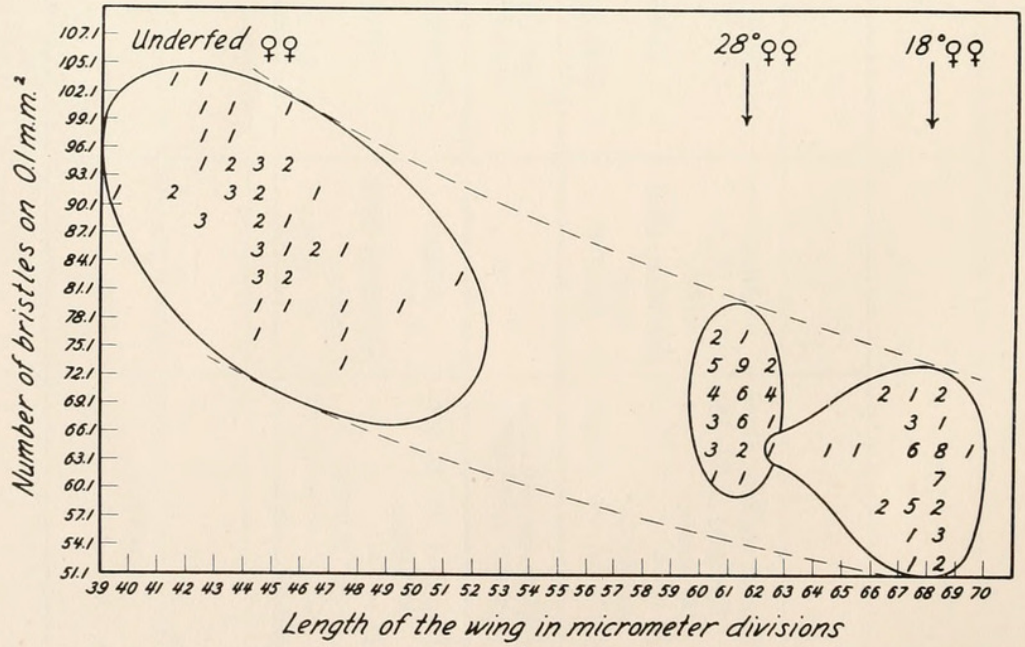

FIG. 6. Correlation between the length of the wings and the number of the bristles per 0.1 mm.² on the lower surface of the wings of the females.

TABLE VI

Biometrical Constants for the Length of the Wings and the Number of the Bristles per 0.1 mm.² of the Lower Wing Surface, as Well as the Coefficient of Correlation between These Characteristics

Males

	Underfed flies		Normally fed flies		28° flies		18° flies	
	Number of the bristles per 0.1 mm.²	Length of the wing in mm.	Number of the bristles per 0.1 mm.²	Length of the wing in mm.	Number of the bristles per 0.1 mm.²	Length of the wing in mm.	Number of the bristles per 0.1 mm.²	Length of the wing in mm.
Mean	94.00±.46	1.224±.004	81.04±.39	1.507±.002	—	1.488±.002	70.00±.34	1.737±.002
Standard deviation	4.836	.0456	4.125	.0230	—	.0227	3.600	.0240
C. of V.	5.14±.35	3.73±.25	5.09±.34	1.53±.10	—	1.53±.10	5.14±.35	1.38±.09
r	−0.299±.086 $R = -3.5$		−0.024±.094				+0.098±.093	

Females

Mean	89.14±.68	1.283±.006	70.36±.35	1.737±.002	—	1.716±.002	63.50±.46	1.917±.005
Standard deviation	7.089	.0590	3.691	.0190	—	.0215	4.773	.0498
C. of V.	7.95±.54	4.59±.31	5.25±.35	1.09±.07	—	1.25±.08	7.52±.51	2.60±.18
r	−0.563±.064 $R = -8.8$		+0.075±.094				+0.101±.093	

IV.

Correlation tables shown in Figs. 5 and 6 contain the basic data on the number of hairs on 0.1 mm.^{2} and length of the wings. The horizontal axis gives the wing length in divisions of the ocular micrometer, each division being equal to 28.333 microns. Table VI represents constants derived from Figs. 5 and 6 with the addition of wing length of 28° flies. The wing length is expressed in millimeters.

TABLE VII

Average Width of the Wings and Width Index, i.e., Width Expressed in Per Cent of the Length

	Underfed flies		Normally fed flies		28° flies		18° flies	
	Width of the wing	Index	Width of the wing	Index	Width of the wing	Index	Width of the wing	Index
Males . . .	.7151	58.23	.8970	59.33	.8871 ±.0021	59.62 ±.11	1.024	58.85
Females .	.7253	57.29	1.015	58.30	1.004 ±.002	58.51 ±.12	1.102	57.31

Let us discuss the influence of the factor under consideration on the wing as a whole. Table VII gives us the constants for the width in millimeters as well as the width in percentage of the length. There is a pronounced sex difference in the size of the wing, the females in all groups being larger than the males. The relative width of the wing is larger in the males, as can be seen by comparing males and females in all groups, and particularly those of the 28° group. The difference is 6.9 times larger than its probable error. (The indices in this case have been calculated by the use of Pearson's formula.) Another point of interest concerning the relative width of the wing is that in the females as well as in the males the underfed and 18° flies seem to have narrower wings than the "normal" 28° flies. The sex difference is also influenced by abnormal conditions. Table VIII shows that in "normal" 28° conditions the sex difference is the greatest, while underfeeding and low temperature reduce the difference. The lower line in Table VIII contains recalculated data from the experiment described in a former paper (Alpatov and Pearl, 1929). The effect of low temperature and consequently of the slow development can be seen in this case also. It is difficult to find an adequate explanation of this phenomenon, which very likely is connected with certain differences in male and female postembryonal development, that is, with different time of the manifestations of different characters during the larval or pupal life.

Turning our attention to the main problem of our investigation, one glance at the correlation tables shows that the larger the size of the wing of the corresponding group of flies, the smaller the number of cells on the area of 0.1 mm.^{2} In other words, the larger flies, considering *inter-group* variation, have also larger cells. The coefficients of correlation for each of the six groups of flies have been calculated separately. They are given in Table VI. Only in the case of underfed males and females is the correlation significant and negative. The conclusion is that in underfed flies the size of the body is negatively correlated with the number of cells on a definite surface of the wing. A possible but very dubious explanation of the absence of such correlation in the case of normally fed and cold temperature flies might be that the variation in the wing length of *Drosophila* developed from normally fed larvæ is so small that the correlation could not manifest itself.

TABLE VIII

Sex-Index of the Wing Length, i.e., Male Wing Length Expressed in Per Cent of the Female

When studied	Underfed flies	Normally fed flies (28°)	28° flies	18° flies
1928	95.33±.54	86.76±.15	86.71±.15	90.61±.26
1927	—	—	88.18±.16	91.93±.18

So far as the variation of the flies belonging to different groups is concerned, it can be seen that the coefficient of variation of the number of cells does not show any definite difference in different groups. At the same time the variation of underfed flies in the length of the wing is much greater than that of the flies which had a normal feeding, no matter at what temperature. Previous investigators who have worked on variation of flies under conditions of under-feeding have also described the increasing variation of experimental animals (see Smirnov and Zhelochovtsev, 1926).

We have now to approach the problem of the actual surface-size of the cells and its relationship to the size of the whole organ. Table IX represents all the calculations relating to this question. The surface of a cell in square microns was determined by dividing 10,000 microns (0.1 mm.^{2}) by the number of hairs on that surface. It can easily be seen that the larger the flies the greater the surface of the cell. Another point of interest is the pronounced sex difference in the size of the cells, the females having much larger cells than the males. This has been pointed out by Dobzhansky (1929). The next step was to

TABLE IX

Biometrical Constants and Indices for Different Characteristics of the Experimental and Control Flies. The last three columns show these characteristics expressed in percentages of the values for the 18° flies.

Males

Character	Underfed flies	28° flies	18° flies	Underfed flies	28° flies	18° flies
Surface of a cell	106.32±.52	123.40±.59	142.86±.69			
Square root of the surface = "length" of a cell	10.31±.07	11.09±.08	11.95±.08	86.3	92.8	100
Calculated length of a cell	8.42±.08	10.37±.08	11.95±.08	70.5	86.8	100
Length of the wing	1.224±.004	1.507±.002	1.737±.002	70.5	86.8	100
Females						
Surface of a cell	112.18±.86	142.13±.71	157.48±1.14			
Square root of the surface = "length" of a cell	10.59±.09	11.92±.08	12.55±.11	88.8	95.0	100
Calculated length of a cell	8.40±.10	11.37±.08	12.55±.11	66.9	90.6	100
Length of the wing	1.283±.006	1.737±.002	1.917±.005	66.9	90.6	100

come from the surface values to linear values which has been done by calculating the length of the cell, which was obtained by taking a square root of the surface of the cell. The data on wing length gave the possibility to calculate the percental decrease in the wing length taking the wing length of 18° flies as a basis. Multiplying by this percental decrease in wing length the number giving the " length " of cell in cold temperature (18°) flies, we obtained the figures represented in our table under the heading " Calculated length of cells." Comparing them with the dimensions obtained by taking the square root, we can easily see that the assumption that the wing length varies proportionally to the length of its constituents does not hold true. The three columns on the right of Table VII represent the changes in wing size and cell size expressed in per cent of 18° (cold) flies. The same relationship between these two characteristics is shown in a percental scale on Fig. 7, the diagonal line represents the relationship in case of a proportional change in wing length and cell length; the dotted line shows the actual percental decrease in cell size in different groups of our flies.

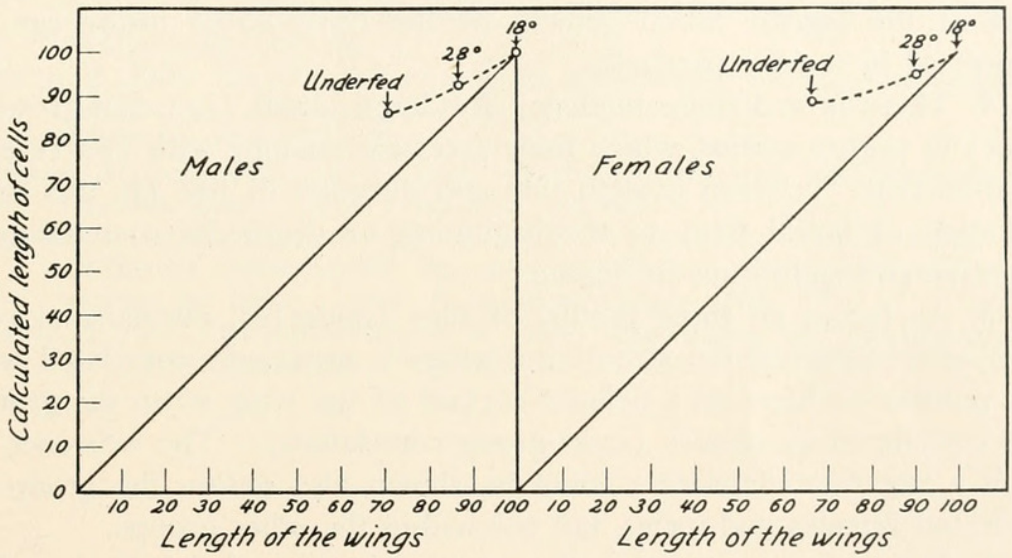

FIG. 7. The dotted lines represent the relationship between the percentage of decrease of the wing length and the percental decrease in the corresponding percental length of the cells calculated by taking the square root of the surface of the cells.

The general conclusion of all these calculations is that the reduced size of cells alone cannot explain the reduction of the organ. The only possible way to explain it is the assumption that the decrease in the organ size—in our case in wing size—is not the result of a decreased size of its cells alone, but also of a reduced number of cells. This last conclusion has a certain bearing upon the problem of the cell constancy in the organism.

If our discussion is correct, the organism can evidently react to the factor decreasing in size not only by decreasing the size of the cells but also the number of cell divisions. The present limited material does not warrant further discussion, but it may be hoped that other investigations in the field of cell-biometry may create a similar basis for understanding the variation of the whole organism as *Die Zelluläre Pathologie* of Virchow did for the interpretation of the pathology of the whole organism.

Summary

1. Dobzhansky's method to determine the number of cells under the surface of the wing membrane of *Drosophila melanogaster* by counting the number of hairs has been used in the present investigation of the relationship of the organ size to the size of its cells.

2. Underfeeding and development at low temperature have been the factors to produce flies under and above the normal size.

3. The functional relation between the time of feeding of larvæ and the size of the wings of larvæ being the expression of the upper part of the logistic larval growth of the third larval instar can be expressed by a cubic parabola.

4. There is a definite tendency for large larvæ (*i.e.*, fast-growing ones) to pupate earlier, which finds a certain analogy with Pearl's correlation that "between growth rate and duration of life (in this case, duration of larval life) to the beginning of death the correlation is negative and significant in degree."

5. As far as all three groups of flies (underfed, normal and cold flies) are concerned the size of the wings is negatively correlated with the number of hairs on a definite surface of the wing when the groups are considered *as wholes* (inter-group correlation). The existence of such a negative correlation could be shown also *within* the group of underfed females and males, but not within the other groups.

6. Expressing in per cent the increase in size of the whole organ and the increase of the linear dimensions of the cells there is a discrepancy in the rate of changes. This leads to the conclusion that the changes in size of the wing cannot be accounted solely by the changes in the size of the cells. The number of cells must play also a certain rôle in this process.

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