INVERSION IN *DROSOPHILA MELANOGASTER*

MARC A. GRAUBARD
Columbia University, New York, New York

Received May 20, 1931

TABLE OF CONTENTS

INTRODUCTION

Many genetic factors suppressing crossing over in *Drosophila melanogaster* have been described and their locations in the three large chromosomes determined (MORGAN, BRIDGES and STURTEVANT 1925). The first case of this kind was encountered by A. H. STURTEVANT (1913). A genetic factor located near sooty in the third chromosome inhibited crossing over in that region when present in heterozygous condition. The factor was therefore considered dominant and was represented by the symbol *C* (crossing over suppressor). However, a fly homozygous for *C* showed normal crossing over values. MULLER found the same crossing over suppressor and analyzed its effects (MULLER 1916). STURTEVANT later reported a case (1919), found in a stock of wild flies collected in Nova Scotia, in which two separate crossover suppressors were present simultaneously, one in each limb of the second chromosome. The one in the left limb was found to suppress practically all crossing over in the half of the chromosome to the left of purple eye color and the one in the right limb had the same effect on the region to the right of purple. For this reason they were named C_{IIL} and C_{IIR} respectively. The number II designates the linkage group, L the left and R the right limb of the chromosome. As in the first case found, no suppression of crossing over occurred when these factors

were present in homozygous condition. A similar case of double crossing-over suppressors was reported in the third chromosome by PAYNE (1924). During the course of the breeding experiments with PAYNE'S suppressors (C_{IIILP} and C_{IIIRP}) a lethal arose by mutation in the left limb and another in the right limb. Several other cases have been reported where suppression of crossing over occurred in definite regions or along the entire length of the first, of the second or of the third chromosome. The crossover suppressors reported by GOWEN and GOWEN (1922) for Drosophila, and by BEADLE (1930) for maize, which inhibit crossing over in all chromosomes, are probably of a different character.

The experiments reported in this paper were conducted on suppressors found by L. WARD (1923). She found flies which showed a new character, Curly wings. They were isolated and found to breed true for the character, which proved to be a dominant, located in the second chromosome. The breeding true, however, was due to "balanced lethals," such as those found by MULLER (1918). As a rule, neither homozygous Curly nor the homozygote of the homologous chromosome survived. Curly (symbol $= C_y$) was found to be linked with cinnabar-2 ($c_n{}^2$) eye-color, an allelomorph of cinnabar, slightly duller in appearance. It soon became apparent that the presence of Curly cinnabar-2 suppressed crossing over along the entire second chromosome. High temperatures (30° C and 14° C) were found to increase markedly the crossing over above the small amount occurring at the standard temperature of 25° C. By such crossing over the Curly and the cinnabar-2 were separated. Heterozygous Curly without cinnabar-2 gave crossing over in the right half but not in the left in which it is located. Heterozygous cinnabar-2 permitted normal crossing over in the left limb but inhibited it in the right limb in which its locus is. It was assumed that the suppression of crossing over was not due directly to the C_y and $c_n{}^2$ genes but to associated crossing over factors, namely, $C_{IIL}\, C_y$ with C_y and $C_{IIR}\, C_y$ with $c_n{}^2$.

A consistent explanation of the phenomenon of crossover suppressors was advanced by STURTEVANT (1926). From work with C_{IIIB}, a factor which he showed to be allelomorphic to his C_{III} he reached the conclusion that the inhibition of crossing over was due to an inversion of a section of the chromosome. In the case of C_{III} a section at the end of the right limb of the third chromosome, beginning a few units to the right of stripe, showed an inverted order for the genes within it. STURTEVANT also suggested that the explanation of certain rare crossovers in a fly heterozygous for a crossover suppressor was that the chromosomes had synapsed with one chromosome inverted in relation to the other. In this case the genes of

the inverted section would be opposed to the corresponding genes in the normal chromosome in correct order. But only double crossovers would then survive, since singles would produce either too large or too small a chromosome with consequent derangement of development.

The experiments described below show that the same explanation of inversion holds for the C_{IIR} with cinnabar-2. The chromosome containing the different order of genic material is then utilized for obtaining information upon certain aspects of crossing over.

The author wishes to express his gratitude to Doctors J. SCHULTZ and C. B. BRIDGES of the CARNEGIE INSTITUTION OF WASHINGTON, to Professor A. H. STURTEVANT of the CALIFORNIA INSTITUTE OF TECHNOLOGY, Pasadena, California, and to Professors D. E. LANCEFIELD and L. C. DUNN of COLUMBIA UNIVERSITY for many helpful suggestions in connection with the following work.

ANALYSIS OF $C_{IIR}\ C_y$

Experimental

Flies of the constitution $C_{IIL}\ C_y\ C_y\ C_{IIR}\ C_y\ c_n^2\ s_p/a_l\ b\ p_r\ c_n\ v_g\ s_p$ (a_l equals aristaless; b equals black body color; p_r equals purple eye color; c_n equals cinnabar eye color; v_g equals vestigial wings; s_p equals speck, pigment spot at base of wings; for the remainder of this paper the symbols C_L and C_R will be used instead of the longer forms $C_{IIL}\ C_y$ and $C_{IIR}\ C_y$.) were obtained from C. B. BRIDGES and mated to black-purple-Lobe-c and black-purple-vestigial females. F_1 females $C_L\ C_y\ C_R\ c_n^2\ s_p/b\ p_r\ L^c$ (L^c equals lobe eye, piece of eye missing) and $C_L\ C_y\ C_R\ c_n^2\ s_p/b\ p_r\ v_g$ respectively were backcrossed to the second-chromosome multiple recessive "albasp" (an abbreviation for the homozygous II-chromosome multiple recessive stock: aristaless-black-purple-cinnabar-vestigial-arc-speck) males, for the purpose of studying the extent of the suppression of crossing over throughout the length of chromosome II. The results are given in tables 1 and 2. The recombination percentages obtained in experiment 1 were: $C_y-b(1)$, 0.0; $b-p_r$ (2), 0.4; $p_r-C_R(3)$, 0.04; C_R-s_p (6), 0.04. Within the C_R-inversion there were also six double crossovers (4, 5) which will be discussed below. In experiment 2 the recombination percentages were: $C_y-b(1)$, 0.0; $b-p_r$ (2), 0.5; $p_r-C_R(3)$, 0.08; C_R-s_p (6), 0.01; included doubles (4, 5), 5.0. The total crossing over was thus less than one percent of the normal amount.

Some of the crossovers obtained could not be mated successfully. Of those that produced offspring, the following flies were shown to carry C_R:

vestigial-cinnabar-2-speck, black-purple-cinnabar-2-speck, Curly-Lobe-c-cinnabar-2-speck, and Curly-vestigial-cinnabar-2-speck. In $C_y\ v_g\ c_n{}^2\ s_p$ the Curly did not show on account of the short wings of vestigial, but was revealed by testing. These four types of flies arose through rare double crossing over within the inverted section (4, 5). Curly-purple-Lobe-c, Curly-purple-vestigial and black-purple did not have C_R.

TABLE 1*

$\lceil C_y \rceil$ $\lceil c_n{}^2 \rceil$ s_p ♀ × "albasp" ♂

1 b 2 p_r 3 4 L^c 5 6

0	2	3	6	4,5	T
3926+3911	10+20	1+2	2+1	3+3	7879

* Explanation of tables: In the diagrams heading each table the chromosome bearing the leftmost mutant gene is placed above its homologue, with a horizontal line between them. The horizontal bracket for the upper or lower chromosome signifies presence of inverted section throughout the region covered by the bracket. Two such bars opposite each other, one for the upper, one for the lower chromosome, mean that the fly is homozygous for the inversion. Small Arabic numerals, placed below the dividing line and between the symbols for the genes, designate the section in which crossing over is followed. In the table the symbol 0 stands for the non-cross-over classes. Similarly the numerals in the table headings designate the type of crossing over of the flies entered below. Thus, "4, 5" indicates double crossovers—one crossover in region 4, the other in region 5. Of the two complementary classes for each crossing-over section, the class which carries the leftmost gene is given first or above. T at end of table stands for total number of flies counted in the experiment. In those tables in which no temperature is stated the flies were in an incubator at 25°C±0.5.

TABLE 2

$\lceil C_y \rceil$ $\lceil c_n{}^2 \rceil$ s_p ♀ × "albasp" ♂

1 b 2 p_r 3 4 V_g 5 6

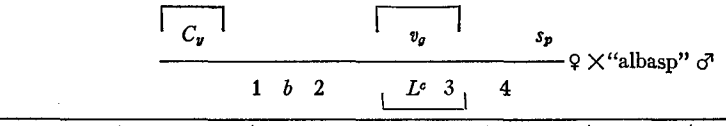
0	2	3	3,6	4,5	T
3843+3830	19+20	0+5	0+1	3+2	7723

Experiments were then undertaken to find which genes lay within and which outside of the inverted section, if such was the nature of the suppressor, as assumed. By crossing over in the $\overline{|C_y|}\ \overline{|L^c\ c_n{}^2|}^1\ s_p/+$ stock a male was obtained which was $\overline{|L^c\ c_n{}^2|}\ s_p/+$. This male was mated to a $\overline{|C_y|}\ \overline{|c_n{}^2|}/+$ female (stock) and the F_1 ♀ $\overline{|C_y|}\ \overline{|c_n{}^2|}/\overline{|L^c\ c_n{}^2|}\ s_p$ females were crossed to black-cinnabar-2-speck males. Crossovers appeared in this cross, because the

[1] The square horizontal bracket is used to designate loci within an inversion.

mother was homozygous for the crossover suppressor linked with cinnabar-2. A double-crossover fly of the composition $\overline{|C_y|}\ \overline{|L^c\ c_n{}^2|}/b\ \overline{|c_n{}^2|}\ s_p$ was then mated to black-cinnabar-2-speck. This cross produced $b\ \overline{|L^c\ c_n{}^2|}/b\ \overline{|c_n{}^2|}\ s_p$ males, which were mated to $\overline{|C_y|}\ \overline{|v_g\ c_n{}^2|}\ s_p/b\ \overline{|L^c\ c_n{}^2|}$ females from table 2. Daughters from this cross were backcrossed to "albasp" males and gave the offspring recorded in table 3. In the diagrams in the headings of tables 3, 4, 5, 6 and 8 the mothers were homozygous for $c_n{}^2$ and hence the symbols have been omitted from both homologs.

TABLE 3



0	2	3	4	2,3	2,4	3,4	T
1726+1836	226+220	121+116	1009+997	1+6	54+59	9+7	6387

When the data of table 3 are calculated by the ordinary methods, the order of genes is as follows: C_y, b L^c, v_g, s_p. The corresponding recombination percentages are: C_y-b, 0.0; $b-L^c$, 8.9; L^c-v_g, 4.1; v_g-s_p, 33.4. This order definitely proves that $C_R(C_{IIR}\ C_y)$ is an inversion, for the normal order is b, v_g, L^c, s_p (BRIDGES 1921). Both L^c and vestigial are included in the inversion, as shown by the fact that vestigial, which is normally to the left of Lobe-c, is now to the right; that is, the region containing both has been reversed.

In table 1 two flies are listed as black-purple-cinnabar-2-speck. When they appeared their genetic constitution could not be ascertained at first, and they were temporarily classified as having a new eye-color mutation because the combination of purple-cinnabar-2 was not then known to have a different eye color from purple-cinnabar, resembling purple more closely than the light orange color of $p_r\ c_n$. Further tests, and backcrosses to cinnabar-2 and purple, showed these two genes to be present. As a result of having obtained the crossover suppressor with purple the above experiments were repeated with this additional locus followed. Flies homozygous for black-purple-$\overline{|\text{vestigial-cinnabar-2}|}$ were obtained and crossed to $\overline{|\text{Lobe-c-cinnabar-2}|}$-speck. F_1 females, homozygous for C_R and for $c_n{}^2$, were backcrossed to "albasp" males. The results are recorded in table 4. The recombination percentages are: $b-p_r$, 2.6; p_r-L^c, 7.9; L^c-v_g, 4.4; v_g-s_p, 30.4. The distance of 2.6 for black-purple may appear to be significantly below its standard value of 6, but later experiments conducted on

a larger scale gave consistently values between 4 and 5. None of the other values deviate beyond the usual amounts.

TABLE 4

b p_r | v_g | ♀ × "albasp" ♂

1 2 | L^c 3 | 4 s_p

0	1	2	3	4	1,2	1,3	1,4	2,3	2,4	3,4	1,2,4	1,3,4	2,5,4	T
1895	48	226	139	933	6	3	23	5	38	8	3	1	0	
2155	67	230	144	1076	5	3	21	3	44	4	1	0	1	7082

While it is obvious that the C_R inversion included vestigial and Lobe-c, and to some extent genes outside this section, it is not certain that the break occurred far enough to the left to include the cinnabar locus. However, if the locus of cinnabar lies outside the inversion, then some crossing over ought to occur between it and the inversion. None has been detected in the large scale experiments presented in this paper nor in the large volume of miscellaneous work carried out with the Curly stock by BRIDGES and others. It is known, from the work of STURTEVANT on C_{IIIB}, that crossing over can occur, and in considerable amount, between the locus of the spindle fiber and the inversion. In the case of the second chromosome, the locus of the spindle fiber has been found to be very close to purple (BRIDGES 1919, STURTEVANT 1919) and to the right (DOBZHANSKY 1930). Furthermore, in the high-temperature experiment reported later in table 7, the amount of crossing over between purple and the C_R inversion rose to the high value of 2.4 percent. The total absence of crossing over between c_n^2 and C_R under this and all circumstances makes it very probable that the break came to the left of cinnabar. Hence the locus of c_n^2 in the inversion is transposed to a position to the right of Lobe-c and vestigial and near the intact terminal section of normal chromosome.

On the assumption that the break came to the left of the cinnabar locus, we may evaluate the probable length of the inverted section. Assuming that the break did not occur to the left of the middle of the purple-cinnabar interval, we may assign 1.5 as the probable maximum length of the section normally to the left of cinnabar. The cinnabar-vestigial and the vestigial-Lobe-c intervals of 9.5 and 5.0 (MORGAN, BRIDGES and STURTEVANT 1925) are both included. In the homozygous inversion data (tables 3,5) the purple-Lobe-c interval is about 10.5. All of this is within the inversion except the amount by which the break came to the left of cinnabar.

If we assume this to have been 1.5 units, then 9.0 units are included. This gives a total of $1.5+9.5+5.0+9.0=25$ units. In view of what was said above, the actual length of the inversion may be smaller than this figure, although the difference cannot be great.

STURTEVANT'S hypothesis provides a simple explanation of the way in which the genes for vestigial and Lobe-c crossed into the inverted section. When C_R is present in heterozygous condition the following situation occurs at synapsis:

$$\begin{array}{rl} & b\ p_r\ c_n\ v_g\ L^c \quad\quad s_p \\ \text{Normal} & \text{———————} \\ C_R & \text{———————} \\ & b\ p_r\ L^c\ v_g \quad\quad c_n{}^2 \end{array}$$

This type of synapsis does not allow crossing over to take place in the inverted section of the chromosome because the genes in that region are not opposite their homologs. But occasionally synapsis occurs in such a way that the genes in the inverted section are opposite their allelomorphs in the other chromosome while the remainder of the chromosome is mated inversely, as represented by the following diagram:

$$\begin{array}{rl} & b\ p_r\ c_n\ v_g\ L^c \quad\quad s_p \\ & \text{———————normal} \\ C_R & \text{———————} \\ & s_p\ c_n{}^2\ v_g\ L^c\ p_r\ b \end{array}$$

The presence of an inversion in one chromosome at synapsis means that when one section is opposed by its allelomorphic genes the remainder is not. It seems that chromosomes tend to synapse in such a manner as to have the larger part opposed by allelomorphic genes. Occasionally, however, the inverted fraction synapses homologously, causing the remainder to be inverted. The frequency of such synapses would probably be higher the longer the inversion or the larger the ratio of the inverted section to the remainder. With this type of synapsis it is apparent that single crossovers within the inversion would produce two irregular chromosomes, one too short and the other too long in relation to the normal second chromosome. Each of these long chromosomes would be a "deficiency" for the region to the right of the inversion and a "duplication" for the region to the left. The short chromosomes would be deficient for the left region and duplicated for the right region. There is reason to believe that a fly possessing either of these chromosomes would not survive, and none have been found. Every crossover into the inverted section was either Curly-

Lobe-c-cinnabar-2-speck or Curly-purple-vestigial-cinnabar-2, both double crossovers.

The data show that the number of included double crossovers was 11 in a total of 15,607 flies (tables 1 and 2), a frequency of one in 1420. Since these flies are double crossovers, the frequency of the corresponding single crossovers which perish should be much higher. For this section of 25 units the number of single crossovers would be expected to be about 50 times as frequent as the number of double crossovers. This means that the expected frequency of single crossovers is one in 28 eggs, or 3.5 percent. Such a figure would indicate that the number of cases of inverse synapsis is very high, roughly one-seventh of the total cases, since 3.5 is about one-seventh of the normal frequency of singles. An accurate evaluation of the frequency is impossible because inverse synapsis, by causing the original inversion to be opposed by allelomorphic genes, produces on either side of that section non-allelomorphic sections of genes. Such sections would behave like inversions and therefore interfere with crossing over even in the inverted section where synapsis is in correct sequence. Such a condition would necessitate assigning still greater values to the frequency of inverse synapsis.

Conclusions and discussion

The above data show that we are dealing here with a chromosome about a quarter of which is inverted and which therefore presents a different order of genic content. That such inversion does not affect the external appearances of the fly had been demonstrated in many other cases as well as in this instance. The genes are all represented in the normal ratio, or in the normal "genic balance."

The C_R inversion happens to be situated near the center of the chromosome. It differs in that respect from the inversion reported by STURTEVANT which is situated at the end of the third chromosome. A central inversion has been found by SHLAER (unpublished) in the third chromosome, but it contained only one known gene, c_d^2, and was therefore less useful than the present inversion.

Experiments reported by BRIDGES (1915, 1919), PLOUGH (1917, 1921), and by BRIDGES and MORGAN (1919, 1923) show that the centrally located regions behave differently with respect to the influence of age and temperature on crossing over than the regions located at the ends. DOBZHANSKY (1930) presents evidence corroborating the view that the relation between the genetic and cytological maps of these central positions in one of the V-shaped chromosomes is also different. STERN'S study of the age

effect upon crossing over in chromosome I confirms the view of BRIDGES that the presence of the spindle-fiber attachment is correlated with this special sensitivity of the central regions of chromosomes II and III. It therefore seemed likely that a study of a chromosome containing an inversion near the spindle fiber in the center would supply some information about crossing over in relation to the above mentioned phenomena.

1. A study was made of the possible modification of crossing over in one arm of the second chromosome, while that in the other arm is inhibited. STURTEVANT (1919) and PAYNE (1924) had shown that in general there was no effect of one arm upon the other. But it seemed desirable to see whether high or low temperatures would have any effect upon this relationship. Some of the data of PAYNE and of DOBZHANSKY (1930) seemed to indicate a rise in crossing over values in one arm when none occurred in the other, or, as in DOBZHANSKY'S case, when some interference occurred in the other.

2. Both high and low temperatures, as mentioned above, greatly increase crossing over in regions that lie in the middle of the second chromosome. The more distant a region is from the center, the less is its crossover value affected by the temperature change. In fact, beyond fifteen units to the right and probably also to the left of purple, only slight influence of temperature on crossing over values has been observed. It is therefore of interest to find how a section, which is normally so far removed from the spindle fiber that its crossover values are practically unaffected by high and low temperatures, would react to the same change of environment when transferred by inversion to a point nearer the spindle fiber. Such a section is the part of the chromosome lying normally to the right of Lobe-*c* but lying between purple and Lobe-*c* in the inverted chromosome. Temperature effects were therefore studied at 25° C, 30° C, 16.5° C and 14° C. Controls were also run simultaneously upon the normal chromosome. These experiments should decide whether the effect of temperature on crossing over is a function only of the genes between the two points involved, or of the proximity of a section to the point of spindle-fiber attachment.

3. Although the published data on coincidence in the second or third chromosome are very scanty they seem to show that the general mechanism shown by WEINSTEIN (1918) to be at work in the first chromosome cannot be applied to either of the V-shaped ones as a whole. In fact, the statement made by BRIDGES and MORGAN (1923) that the symmetrical behavior of crossing over in each arm of the two large autosomes seems to indicate that synapsis and crossing over begin either at both ends of the

arms and proceed from there to the center, or that they start at the center and proceed simultaneously to both ends, necessitates an analogy between the X chromosome and only one arm of the V.

EFFECT OF CROSSING-OVER IN ONE ARM UPON ITS OCCURRENCE IN THE OTHER AT 25°, 30° AND 16° C

Experimental

The gene for Curly is linked with a genetic factor which suppresses practically all crossing over to the left of purple. For this reason it was used in heterozygous condition in flies which were also heterozygous for the genes, black, purple, Lobe-*c*, vestigial and speck. All of these except black are located in the right arm of the second chromosome. The flies were homozygous for the right arm inversion and therefore gave free crossing over.

Females of the constitution black-purple-vestigial-cinnabar-2 were mated to $C_y\ L^c\ c_n^2\ s_p/+$ males. The F_1 Curly-Lobe-*c* females, homozygous for C_R, were backcrossed to "albasp" males. The results obtained at 25° C are given in table 5. A year later this experiment was repeated using

TABLE 5.

$\overline{|\ C_y\ |}$ $\overline{|\ L^c\ |}$ s_p

1 *b* 2 p_r 3 $|\ 4v_g\ |$ 5 ♀ × "albasp" ♂

0	2	3	4	5	2,4	2,5	3,4	3,5	4,5	3,4,5	T
1567	3	290	143	915	0	0	2	76	11	0	
1518	4	243	149	873	1	1	1	67	11	1	5876

different stocks. Females $C_y\ p_r\ v_g\ c_n^2$/"albasp" were mated to $a_l\ b\ L^c\ c_n^2\ s_p$/"albasp" males and F_1 Curly-Lobe-*c* females, homozygous for C_R, backcrossed to "albasp" males. This cross was conducted also at 30° and at 14.5° C. The results are given in table 6. The recombination percents obtained from these two experiments at 25° C show very good agreement, as can be observed from table 10, where the values at both high and low temperatures are also given.

A control experiment with normal left arms and with crossing over prevented in the right arms by heterozygous C_R was also performed. Flies homozygous for black-purple-cinnabar-2-speck were mated to aristaless-dumpy males (dumpy equals dumpy wings, symbol d_p), and the F_1 females backcrossed to males homozygous for aristaless-dumpy-black-

TABLE 6

a_l b | L^c | s_p ♀ × "albasp" ♂

1 | C_v | 2 $3p_r$ 4 | $5v_g$ | 6

T°C	0	3	4	5	6	3,4	3,5	3,6	4,5	4,6	5,6	3,4,6	4,5,6	T
14.5°	1009	9	203	84	464	0	1	1	3	30	3	0	0	
	1033	10	187	80	481	1	1	7	2	42	5	1	0	3657
25°	296	1	47	12	175	0	0	0	1	12	2	0	0	
	305	0	48	34	178	0	0	2	0	14	3	0	1	1131
30°	523	5	94	36	362	0	0	9	3	45	12	0	3	
	516	7	95	48	295	0	0	2	3	56	15	1	0	2130

purple-cinnabar-vestigial-speck ("aldpbasp"). The results are given in table 7 and the recombination percentages based on them are shown in table 10.

TABLE 7

a_l d_p ♀ × "aldpbasp" ♂

1 2 b 3 p_r 4 | c_n^2 | 5 s_p

T°C	0	1	2	3	4	5	1,2	1,3	1,4	2,3	2,4	3,4	1,2,3	1,2,4	2,3,4	T
16°	1093	220	501	69	3	0	17	9	0	15	1	0	0	0	0	
	1085	216	653	95	0	0	13	9	0	15	0	0	0	0	0	4014
25°	1088	190	433	63	2	0	10	7	1	6	0	0	0	0	0	
	1137	213	465	66	5	0	8	2	0	14	3	0	0	0	0	3713
30°	429	63	182	79	10	1	9	10	1	15	4	0	0	1	0	
	434	80	226	65	19	0	11	9	0	14	3	1	1	1	1	1669

Conclusions and discussion

The data supplied by these experiments indicate that the presence or absence of crossing over in one arm of the V-shaped chromosome exerted only negligible influence upon its occurrence in the other. Females bred at high and low temperatures showed the usual temperature effects. The black-purple recombination was about normal (4.2) in the presence of C_R and it rose normally to 11.7 at 30° C and to 5.0 at 16.5° C. Similarly aristaless-dumpy had a recombination percentage of 11.6 at 25° C (in the presence of C_R), 11.1 at 30° C, and 12.0 at 16.5° C. Other similar relations

TABLE 8

a_l t_w | L^c | s_p

1 2 b 3 p_r 4 | $5v_g$ | 6 ♀ × "aldpbasp" ♂

	14°C	16.5°	25°	30°		14°C	16.5°	25°	30°
0	525+569	813+862	1545+1676	648+671	1,4,5	1+0	0+1	..	1+0
1	63+88	129+172	228+282	129+132	1,4,6	5+5	8+6	9+15	12+10
2	213+273	356+390	544+661	275+312	1,5,6	1+0	2+2	1+3	2+1
3	117+114	98+111	98+97	92+89	2,3,4	2+3	1+3	1+6	1+3
4	138+149	138+152	259+234	170+188	2,3,5	1+1	1+1	0+1	1+0
5	26+43	56+59	101+127	49+57	2,3,6,	9+8	7+7	3+5	11+8
6	214+248	437+410	864+894	334+343	2,4,5	..	1+5	1+2	3+3
1,2	7+8	11+6	13+21	19+14	2,4,6	10+4	13+10	18+22	18+28
1,3	12+8	12+14	10+8	12+24	2,5,6	1+0	1+2	5+4	5+6
1,4	18+28	26+19	34+42	31+35	3,4,5	0+1	0+1	1+2	2+2
1,5	5+6	17+10	27+17	9+10	3,4,6	8+4	1+1	3+1	5+4
1,6	25+41	73+84	107+139	70+90	3,5,6	0+1	..	1+0	2+0
2,3	14+17	11+13	13+7	22+28	4,5,6	1+0	..	0+1	4+2
2,4	44+49	66+82	99+92	59+56	1,2,3,4	..	..	..	1+0
2,5	16+14	26+30	44+41	22+15	1,2,3,5	1+0	..	..	..
2,6	75+98	155+191	288+345	157+165	1,2,3,6	..	2+0	1+0	..
3,4	24+22	14+16	16+11	25+20	1,2,4,5	..	1+0	..	0+1
3,5	10+11	3+10	8+2	6+7	1,2,4,6	..	1+0	1+0	2+1
3,6	29+37	48+55	49+52	57+53	1,2,5,6	..	..	..	1+0
4,5	1+5	5+5	7+5	8+8	1,3,4,6	..	0+1	1+0	0+2
4,6	18+27	16+28	50+66	67+66	1,4,5,6	..	..	..	1+1
5,6	4+4	3+4	10+12	11+14	2,3,4,5	1+0	..	..	..
1,2,3	0+1	1+1	2+0	4+1	2,3,4,6	2+1	..	1+1	2+0
1,2,4	1+0	0+2	1+4	3+8	2,3,5,6	..	..	0+1	0+1
1,2,5	0+0	0+1	1+1	2+1	2,4,5,6	..	..	0+1	2+0
1,2,6	3+1	3+3	5+14	14+15	1,2,3,4,5	..	..	..	1+0
1,3,4	0+3	5+2	2+0	3+2	1,2,3,4,6	..	..	..	2+0
1,3,5	1+1	0+1	1+0	1+2	1,3,4,5,6	..	..	..	1+0
1,3,6	4+6	4+4	5+5	9+13					
					T	3549	5339	9398	4910

are observed in table 10. The values obtained from these experiments demonstrate that high or low temperatures were inoperative as factors bringing about crossing over where it was inhibited by an inversion. The effect of temperature is seen on the regions very close to the spindle fiber which are outside of the inversion, but the cinnabar-2-speck region remained unaffected.

Unfortunately, from the matings made, I cannot draw any conclusions about the possible increase of the frequency of inverse synapsis at the temperatures used.

TABLE 9

	a_l t_x					L^c s_p			
	1	2 b	3 p_r	4 c_n	5 v_g 6 7	♀ × "aldpbasp" ♂			
	14°C	16.5°	25°	30°		14°C	16.5°	25°	30°
0	416+446	707+710	828+795	589+629	1,6,7	2+0	3+1	1+2	2+3
1	51+66	109+149	126+175	75+120	2,3,4	1+1	1+1	..	3+1
2	207+305	345+464	333+418	267+318	2,3,5	6+5	3+7	1+1	4+3
3	93+90	97+142	77+66	90+91	2,3,6	2+2	1+2	1+0	1+1
4	14+37	30+35	21+33	37+49	2,3,7	12+8	10+4	2+1	4+7
5	103+102	149+174	114+166	96+109	2,4,5	..	1+0	1+0	1+2
6	40+48	74+66	75+50	54+30	2,4,6	0+1	0+2	1+0	1+0
7	215+237	397+398	434+440	317+355	2,4,7	8+3	4+9	2+3	10+6
1,2	8+3	19+16	9+9	12+13	2,5,6	..	1+0	2+1	0+1
1,3	7+8	11+12	6+6	9+14	2,5,7	13+18	13+22	10+6	15+20
1,4	4+7	1+10	6+2	9+8	2,6,7	1+5	2+7	2+2	7+4
1,5	14+14	24+33	18+26	20+19	3,4,5	..	0+1	1+0	0+2
1,6	7+13	13+17	10+11	8+10	3,4,6	..	1+0	..	..
1,7	28+57	48+80	44+86	53+72	3,4,7	0+2	0+2	0+1	2+2
2,3	13+19	26+20	5+5	16+14	3,5,6	..	1+0	..	0+1
2,4	12+16	24+18	10+6	20+24	3,5,7	3+8	9+7	2+3	8+4
2,5	49+84	93+99	56+85	45+67	3,6,7	3+1	1+0	0+1	5+2
2,6	20+29	36+60	24+28	17+29	4,5,6	..	0+1	..	..
2,7	92+115	204+298	168+217	124+195	4,5,7	0+2	1+2	0+1	..
3,4	0+2	4+4	3+0	2+3	4,6,7	..	2+0	..	1+0
3,5	15+15	21+19	14+10	18+16	5,6,7	..	0+1	..	1+0
3,6	8+7	13+10	6+5	7+4	1,2,3,4	..	..	..	1+1
3,7	42+60	58+74	40+39	38+37	1,2,3,6	..	..	..	1+0
4,5	0+3	4+1	3+0	1+6	1,2,3,7	..	1+1	..	..
4,6	2+2	0+1	0+2	1+5	1,2,4,5	..	..	..	1+0
4,7	7+10	10+14	7+7	21+14	1,2,5,7	2+1	3+0	2+1	..
5,6	1+0	2+1	1+6	2+6	1,3,4,5	..	1+0	..	..
5,7	28+41	44+33	25+22	33+34	1,3,4,7	..	..	..	1+1
6,7	2+2	10+6	11+5	8+13	1,3,5,7	0+1	1+0	..	..
1,2,3	..	1+0	2+0	4+0	1,3,6,7	1+0	1+0	..	..
1,2,4	1+1	1+0	0+1	0+1	1,4,6,7	..	..	..	0+1
1,2,5	0+3	1+2	1+1	4+5	1,5,6,7	..	..	..	0+1
1,2,6	2+0	2+2	2+2	0+1	2,3,4,5	..	..	..	0+1
1,2,7	3+5	7+8	0+4	10+11	2,3,4,6	..	..	1+0	..
1,3,4	..	..	1+1	..	2,3,4,7	..	..	..	1+0
1,3,5	4+3	2+5	1+2	5+2	2,3,5,6	..	..	..	0+1
1,3,6	1+0	0+3	0+2	1+1	2,3,5,7	0+3	1+1	..	1+2
1,3,7	7+8	6+9	1+2	2+9	2,3,6,7	..	1+0	..	0+1
1,4,5	1+0	1+0	..	1+0	2,4,5,7	..	1+0	..	0+1
1,4,6	1+0	2+1	..	2+1	2,5,6,7	..	..	1+0	..
1,4,7	..	2+2	2+1	6+2	3,4,6,7	..	..	..	0+1
1,5,6	0+1	..	0+1	1+0	1,2,3,4,7	..	..	..	1+1
1,5,7	6+3	5+5	5+5	2+6	1,2,4,5,6,7	..	..	..	0+1
					T	3521	5739	5284	4514

TABLE 10

Crossing-over values for sections studied.

RECTION	HOMOZYGOUS C_R				NORMAL CHROMOSOME II				HETEROZYGOUS C_R			HOMOZYGOUS C_R, HETER. C_L		
	14°C	16.5°	25°	30°	14°C	16.5°	25°	30°	16.5°	25°	30°	14.5°	25°	30°
a_l—t_x	9.7	10.2 13.1 Average11.7	10.6	15.6 11.3 14.3 Average14.1	9.8	10.8	10.9	11.8 11.8 Average11.8	12.1	11.6	11.1	..	..	..
t_x-b	24.7	25.6 27.0 Average26.3	24.1	27.1 25.1 26.4 Average26.5	30.6	32.2	27.0	29.1 28.3 Average28.8	30.3	25.3	28.0	..	..	..
b-p_r	13.4	9.1 7.7 Average 8.4	4.4	10.8 9.6 10.2 Average10.4	13.0	10.4	5.8	10.3 9.2 Average 9.9	5.3	4.2	11.1	0.8	0.17 0.26 Average 0.2	1.1
p_r-c_n	..	..	..	..	3.9	3.4	2.2	5.8 5.5 Average 5.7	..	..	..	..	..	..
c_n-v_g	..	..	..	..	15.7	13.9	11.2	12.2 13.7 Average12.6	..	..	..	..	..	..
v_g-L^c	..	..	..	..	5.6	6.0	4.8	5.2 5.3 Average 5.3	..	..	..	..	..	..

TABLE 10 (continued)

SECTION	HOMOZYGOUS C_R				NORMAL CHROMOSOME II				HETEROZYGOUS C_R			HOMOZYGOUS C_R, HETER. C_L		
	14°C	16.5°	25°	30°	14°C	16.5°	25°	30°	16.5°	25°	30°	14.5°	25°	30°
L^c-s_p	..	..	..	..	30.2	31.9	30.4	33.4 31.5 Average32.8	..	..	..	..	..	..
p_r-L^c	16.2	12.3 11.4 Average11.9	10.7	19.5 15.2 16.0 Average17.5	..	..	..	..	..	..	..	12.8	11.7 10.9 Average11.4	14.1
L^c-v_g	4.4	4.5 4.7 Average 4.7	4.6	6.2 5.4 4.6 Average 5.5	..	..	..	..	..	..	..	4.9	4.7 5.4 Average 5.3	5.6
v_g-s_p	25.2	28.5 30.7 Average29.6	32.0	32.3 33.4 32.4 Average32.6	..	..	..	..	..	..	..	28.3	33.3 34.2 Average33.4	37.5
p_r-c_n^2 or p_r-inversion	..	..	..	..	..	..	..	..	0.1	0.3	2.5	..	..	..
c_n^2-s_p or inversion-s_p	..	..	..	..	..	..	..	..	0	0	0.1	..	..	..
a_t-C_y	..	..	..	..	..	..	..	..	..	..	..	0	0	0
C_y-b	..	..	..	..	..	..	..	..	..	..	..	0	0	0

We may compare the inhibiting effects of inversions on crossing over in proximal and in distal regions with respect to the spindle fiber. Since the C_R inversion includes about 25 units, the extent of the normal chromosome between the right end of the inversion and speck is approximately 27.5 units. Nevertheless, only negligible crossing over occurs in this region when C_R is heterozygous, while in the black-purple section, which is only 6 units long, but which differs apparently from the former by its nearness to the spindle fiber, relatively high crossing over occurs. The same is observed for the distance between purple and C_R. To summarize, regions situated near the spindle fiber show crossing over, though reduced, in the presence of an inversion, while much larger regions distal to the inversion show practically none. These facts would lead one to conclude that crossing over (or synapsis) begins at the spindle fiber and not at the ends of the chromosome.

Table 10 shows the purple-cinnabar-2 or purple-C_R recombination percentage to be between 0.1 and 0.27 and to rise at 30° C to 2.4. The black-purple distance rises to 1.1 at 30° C. It is probable that the nearer a section is to the spindle fiber, the greater is its relative rise at high or low temperatures.

EFFECT OF INVERSION UPON THE TEMPERATURE EFFECT AND UPON COINCIDENCE VALUES

Experimental

One object of the following experiments was to observe the behavior of a distal section, normally situated far enough from the spindle fiber so as to show no temperature effect, when it is placed near the spindle fiber by an inversion. The second problem was to investigate variations, if any, in coincidence, that might be produced if the new order of the genes affects the behavior of the chromosomes at crossing over.

In the first series of experiments, females of the constitution black-purple-C_R-vestigial-cinnabar were crossed to males $a_l\ t_x\ L^c\ s_p/a_l\ d_p$. Thoraxate ($t_x$), an allelomorph of dumpy, was used here because homozygous dumpy would fail to show in the presence of vestigial. Although thoraxate is lethal when homozygous, t_x/d_p was found by BRIDGES to be viable. It was easily classifiable under the conditions of these experiments. F_1 females obtained from the above cross, of the constitution $a_l\ t_x\ L^c\ c_n^2\ s_p/b\ p_r\ v_g\ c_n^2$, were backcrossed to males homozygous aristaless-dumpy-black-purple-cinnabar-vestigial-speck ("aldpbasp"). These multiple recessive males were found to be very poorly viable. They mated with difficulty with the F_1 females, which were wild-type in appearance, except for the dominant

Lobe-*c* eye. Large males from first hatches of pair cultures were therefore selected for use. They were placed with the females in $2\times\frac{1}{2}$ inch vials for two days, after which interval the mated flies were transferred to bottles. On an average one mating in seven was successful. To obtain recombination percentages for the temperatures higher or lower than 25° C, the P_1 flies were allowed to lay eggs at 25° C until larvae appeared in the bottles, which were then transferred to the respective incubators. The F_1 virgin females were isolated from the bottles, which had been kept at the desired temperatures, and were then mated at 25° C, at which temperature they remained.

The results for the first series of experiments, where F_1 females, homozygous for C_R, were mated to aristaless-dumpy-black-purple-cinnabar-vestigial-speck males, are given in table 8. The recombination percentages computed on the basis of the flies obtained in this series of experiments are recorded in table 10.

For the second series of experiments, which were to serve as controls, flies were synthesized to contain the same genes in both normal chromosomes, as in the experimental chromosomes. Females homozygous for black-purple-cinnabar-vestigial were mated to males $a_l\ t_x\ L^2\ s_p/a_l\ d_p$ and the F_1 females were backcrossed to aristaless-dumpy-black-purple-cinnabar-vestigial-speck ("aldpbasp") males. The experiments were performed at the same temperatures as the previous series in which the F_1 females were homozygous for the inversion. In fact the experimental cultures of both series were always kept together in the same incubator at the respective temperatures. The range of variation of all incubators was ± 0.5° C. The results obtained are presented in table 9. In this series, as well as in the previous one, males and females were classified separately because it was believed by Doctor BRIDGES that males having the genes t_x/d_p occasionally fail to show the character, while females show the character strongly. In neither of these experiments did the t_x/d_p male class overlap the wild-type class. Overlap would have given very asymmetrical "1, 2" double crossover classes, which did not occur; nor was the total of the 1, 2 doubles abnormally high. The male and female data have accordingly been combined. The recombination percentages obtained from the different experiments are given in table 10. The standard map of MORGAN, BRIDGES and STURTEVANT in the *Genetics of Drosophila* is reproduced in figure 1 for the purpose of comparison with the values obtained from our experiments. Withal the agreement is quite satisfactory. The fact that the two large sections thoraxate-black and Lobe-*c*-speck do not show close agreement is to be expected, since these distances on the

standard map are built up from their constituents when intermediate genes are used.

Conclusions and discussion of the temperature effect

On comparing the recombination percentages of the flies recorded in tables 8 and 9 which carry inversion bearing and normal chromosomes respectively, we see that both high and low temperatures produce an increase in crossing over which is about twofold for the regions immediately to the right and left of purple, that is, black-purple and purple-cinnabar. The further away a section is from purple the smaller is the influence of high and low temperatures upon crossing over in that section. The most central sections black-purple and purple-cinnabar show an average rise of about 100 percent. In the normal chromosome the distance vestigial-Lobe-*c* is increased only slightly, if at all, and this section is only about 13 units to the right of purple. This gradient of effect by temperature is shown by the normal chromosome and also by the one containing the C_R inversion. (See standard map of chromosome II, Morgan, Bridges and Sturtevant, 1925.)

The rise in the recombination percentage for cinnabar-vestigial in the normal chromosome is variable in quantity though always present. It happens that the value for that interval at 25° C obtained in our experiment (11.2) is higher than the one (9.0) given by MORGAN, BRIDGES and STURTEVANT (1925). The increase in crossing over for c_n–v_g, based on our normal value at 25° C, is 16 percent at 30° C, 27 percent at 16° C and 41 percent at 14° C. The increases for the similarly placed section in the inversion-bearing chromosome, that is, in the purple-Lobe-*c* distance, are 70 percent at 30° C, 11 percent at 16° C and 50 percent at 14° C. The purple-Lobe-*c* distance in the C_R-chromosome consists mostly of genes which were brought into this position by the inversion. Normally this section of the chromosome does not show any special increase in its crossing over at 30° C or 14° C. But in the inversion, purple-Lobe-*c* increases markedly, as stated above. In the normal chromosome the section between purple and vestigial, consisting of purple cinnabar and cinnabar vestigial, increases 45 percent at 30° C, 35 percent at 16° C and 50 percent at 14° C. These figures are quite comparable with those given above for the percentage increase in the identically situated section within the inversion-bearing chromosome.

Attention should be drawn to the fact that the Lobe-*c*-vestigial distance does not show any significant difference in its increase at the effective temperatures in the two chromosomes in which it was studied, the normal and the one bearing the inversion. A comparison of the diagram on page

87 shows that the vestigial-Lobe-*c* section is only slightly nearer to the point of spindle-fiber attachment in the inversion-bearing chromosome than in the normal. The difference in position, about 3 units, was not sufficient to produce a measurable difference in increase in crossing over, especially since the section happens to be situated about eleven units to the right of the spindle fiber, where temperature is not as effective as a modifying agent of crossing over.

Reciprocally, the genic material situated just to the right of the spindle fiber ($p_r - c_n$, $c_n - v_g$) gives a marked increase with the effective temperature when in the normal chromosome. But this same material, when carried by the inversion to a point in the middle of the right arm, gives only negligible increase (part of the $v_g - s_p$ section in the C_R chromosome). Thus three comparisons have been made: (1) Genic material that normally is situated far from the spindle fiber and which there gives only negligible temperature effect gives marked effect when carried near the spindle fiber by the inversion. (2) Genic material that is normally situated near the spindle fiber and there gives a marked temperature effect no longer gives this effect when those genes are placed more distal by the inversion. (3) The genes near the center of the inversion (the $v_g - L^c$ section) are not much displaced nearer to or farther from the spindle fiber by the inversion, and both in the normal chromosome and in the C_R chromosome this section gives the same, not-marked, temperature effect. The conclusion to be drawn from these observations is that the sequence of the genic materials within the chromosome studied is not an essential part of the reactions or mechanisms which are affected by the temperature. The temperature effect is determined by the position of the genes in relation to the spindle fiber and not by the specific nature of the section of the chromosome itself. This confirms the conclusion reached by BRIDGES from study of the relation of the spindle-fiber position to the intensity of the age effect in normal chromosomes.

The next point upon which these experiments were expected to shed some light was the problem of coincidence. By coincidence is meant the ratio of the number of double crossovers obtained to the number expected if one crossover does not influence the position of the next. When a given distance undergoes 10 percent of crossing over and another one 15, it is expected that simultaneous or coincident crossovers occur in a total of 10 percent $\times$ 15 percent = 1.5 percent. Actually it is found that the number obtained is less than that expected. It was stated above that it would be of interest to know whether the values of coincidence were modified by the change of genic content within the chromosome. Coincidence has been held

TABLE 11

Coincidence values for sections in Normal and C_{IIR} chromosome.

GROUP 1. *Both sections situated in same arm. Sections in right arm whose coincidence values are here compared are identically situated in two chromosomes with respect to distance from p_r.*

	1-2		1-3		2-3		4-5 (C_{IIR}) 5-6 (N)		4-6 (C_{IIR}) 5-7 (N)		5-6 (C_{IIR}) 6-7 (N)	
	LEFT-C_{IIR}	LEFT-N	LEFT-C_{IIR}	LEFT-N	LEFT-C_{IIR}	LEFT-N	RIGHT-C_{IIR}	RIGHT-N	RIGHT-C_{IIR}	RIGHT-N	RIGHT-C_{IIR}	RIGHT-N
14°C	0.19	0.10	0.56	0.34	0.31	0.23	0.24	0.13	0.31	0.41	0.20	0.06
16°C	0.10	0.17	0.50	0.36	0.19	0.24	0.35	0.16	0.23	0.30	0.09	0.14
25°C	0.14	0.11	0.39	0.35	0.20	0.12	0.27	0.26	0.36	0.26	0.16	0.20
30°C	0.18	0.16	0.50	0.45	0.37	0.24	0.33	0.35	0.47	0.35	0.28	0.27
Average	0.15	0.13	0.49	0.38	0.27	0.21	0.30	0.22	0.34	0.33	0.18	0.17

GROUP 2. *Coincidence for sections not situated in the same arm.*

	C_{IIR} CHROMOSOME									NORMAL CHROMOSOME								
	1-4	1-5	1-6	2-4	2-5	2-6	3-4	3-5	3-6	1-5	1-6	1-7	2-5	2-6	2-7	3-5	3-6	3-7
14°C	0.92	0.81	0.84	0.66	0.78	0.79	0.71	1.18	0.65	0.52	1.02	0.81	0.79	0.79	0.64	0.42	0.57	0.74
16°C	0.61	0.92	0.85	0.90	0.87	0.83	0.56	0.63	0.78	0.63	0.80	0.64	0.74	0.85	0.84	0.48	0.64	0.68
25°C	0.70	0.96	0.75	0.79	0.83	0.87	0.62	0.58	0.76	0.68	0.75	0.74	0.89	0.76	0.90	0.71	0.75	0.86
30°C	0.55	0.51	0.79	0.50	0.63	0.76	0.50	0.46	0.60	0.67	0.65	0.72	0.67	0.67	0.75	0.60	0.48	0.52
Average	0.69	0.80	0.80	0.71	0.78	0.81	0.60	0.71	0.70	0.60	0.80	0.73	0.77	0.77	0.78	0.55	0.61	0.70

to arise from the assumed internode formation by chromosomes preceding crossing over. If the postulated internode section, or actual length of block of genes transferred by a double crossover, is a purely physical phenomenon, then its size should not be modified by changes in genic content. The relative internode length can be inferred from coincidence values. Thus, the coincidence for all distances in the two types of chromosomes, normal and "inverted," was computed and is recorded in table 11.

Analysis of coincidence data

Coincidence of sections located in different arms of the same chromosome

The data of the four experiments with homozygous C_R and homozygous normal chromosomes seem to indicate, as found in the past, that the further apart two sections are on the chromosome map the greater are their coincidence values. This behavior is observed regardless of whether both sections lie in one arm of the chromosome or in different arms. For our purposes we may compare sections that are relatively near to each other but do not lie within the same arm. Such sections in the C_R chromosome are black-purple in the left and purple-Lobe-*c* or Lobe-*c*-vestigial in the right limb. Similar sections are black-purple and cinnabar-vestigial in the normal one. It is to be expected that if crossing over in one arm is entirely independent of crossing over in the other, the coincidence of two sections located in different arms should vary about a mean of one. Table 11 shows that this is not the case; the values rarely reach one. Even if we take two sections at opposite ends of the chromosome, as far from each other as possible, the coincidence values obtained, while fairly high, do not consistently reach the value of one.

If we divide all coincidence data into two groups (table 11), one consisting of values of sections within the same arm, either right or left, and the other of sections in both arms simultaneously, we notice that the first group gives fairly uniform values, all low. The only temperature which gives consistently higher values is that of 30° C, while 14° C, which has the same effect on crossing over, does not show the same uniform rise. No rise of coincidence values with temperatures is observed in the second group. It can also be seen that group 2 presents on the whole greater variations than group 1. Another point which the data bring out is that for equivalent lengths of map the intervals near the center show higher values than those at the ends. Unfortunately, these data do not contain small sections at the end of the chromosome, so that no exact comparison of coincidence for identical lengths at the outer and inner ends of an arm

can be made on the basis of small sections. It is apparent, nevertheless, that sections 5–6 of group 1 normal chromosome which includes 14 units in one arm very close to the center give a much smaller value than sections 3–5 group 2 (normal) which is 17 units long and includes both arms.

From the data on coincidence of sections located in both arms of the chromosome it is apparent that crossing over in the two arms is not entirely independent. If crossing over in one arm were entirely independent of that of the other, then, theoretically, coincidence should be approximately one, which was not the case. We must therefore assume that although crossing over in one arm is largely independent of the other, some mechanism exists, connected perhaps with the spindle fiber, which interferes with simultaneous crossing over in both.

Comparison of coincidence in normal and inversion-bearing chromosomes

The figures recorded in table 11, group 1, bring out another point about the behavior of the arms of the C_R and the normal chromosomes. The regions involved in the right arm are so arranged that they correspond to regions analogous with respect to their distance from purple, but genetically different. Thus, 4–5 in the C_R chromosome represents coincidence of the purple-Lobe-*c* and Lobe-*c*-vestigial sections, while its analog 5–6 in normal chromosome represents values for the purple-vestigial and vestigial-Lobe-*c* distances. As demonstrated above, purple-Lobe-*c* in the inversion and purple-vestigial in the normal chromosome involve partly different chromosomal material, and the Lobe-*c*-vestigial and vestigial-Lobe-*c* regions in their respective chromosomes involve the same material but in different order. The inversion occurred so that the Lobe-*c*-vestigial fraction remained in about the same position in respect to the spindle fiber, because purple-Lobe-*c* in C_R and purple-vestigial in the normal chromosome are about equal in length, namely 15 and 17 units each, respectively. Thus, a comparison of the coincidence values in the part of the chromosome between purple and speck should demonstrate whether any significant difference in the presumable loop formation of the two chromosomes has occurred. As far as the coincidence data go, they seem to show good agreement between the two chromosomes, section by section. The values for the left end also agree quite well. On the basis of such general comparison it seems permissible to conclude that the underlying mechanism at work in this case is not affected by the sequence of the genic content of the chromosomes.

SUMMARY

1. It is demonstrated that the suppression of crossing over in the right arm of the second chromosome, which was considered to be due to a genetic factor $C_{IIR}C_v$, is the result of an inversion of a part of the chromosome.

2. The inverted section includes the genes for Lobe-*c* and vestigial, and probably for cinnabar, but not purple. It is therefore located near the center of the chromosome. Its maximum length is estimated to be 25 units.

3. The frequency of inverse synapsis of the chromosomes as calculated is unexpectedly high, approximately one-seventh of the cases.

4. No effect was found for the absence or presence of crossing over in one arm of the second chromosome upon its occurrence in the other arm at various temperatures.

5. The increase in crossing over at 30° C, 14° C and 16° C for sections in one arm, when crossing over is inhibited in the other, is the same as in the normal chromosome.

6. A gradient of size of increase in crossing over values, produced by breeding F_1 females at 30° C and 14° C, extends from purple to the right and probably to the left for about 15 units. This gradient operates to the same degree in the chromosomes homozygous for an inversion as in the normal chromosome.

7. When a piece of chromosome situated normally in a section not affected by temperature (the part of the right of Lobe-*c*) is brought near to the spindle fiber by an inversion, it shows the temperature effect characteristic of its new location. When a piece of chromosome normally situated in a part affected by temperature is removed by inversion and relocated in a more distal region, it loses its sensitivity to temperature and behaves as does genic material that is normally situated at that distance from the spindle fiber. The central genes of an inversion are not displaced toward or from the spindle fiber and correspondingly do not change their reaction to temperature. Increase of crossing over with temperature is thus a property of the position of the genes in relation to the spindle fiber and is not a function of the genes.

8. In the presence of an inversion in one arm (heterozygous), crossing over is much more frequent in a section situated between it and the spindle fiber than in one of equal length situated between it and the distal end of the chromosome. On that basis the probable point of the beginning of crossing over (or synapsis) in this V-shaped chromosome is at the spindle fiber.

9. That coincidence of sections located respectively in the two arms of the second chromosome is uniformly below one, shows that crossing over in the two arms is not completely independent.

10. A comparison between the coincidence values obtained for chromosomes homozygous for an inversion and for the normal chromosomes shows that no significant disturbances are created in the mechanism responsible for coincidence upon inverting part of the chromosome. Sections situated similarly with respect to the spindle fiber give the same coincidence values regardless of change in genic material.

11. The coincidence values of two sections located in opposite arms of the second chromosome are higher for equivalent distances than those of sections located in the same arm.

12. When a section of chromosome from the center is introduced into a terminal region no measurable change in crossing over is observed for the transposed section.

LITERATURE CITED

ANDERSON, E. G., 1925 Crossing over in a case of attached X-chromosomes in *Drosophila melanogaster*. Genetics **10**: 403–417.

BEADLE, G. W., 1930 Genetical and cytological studies of Mendelian synapsis in *Zea mays*. Cornell Agric. Expt. Sta. Mem. **129**:

BRIDGES, C. B., 1915 A linkage variation in Drosophila. J. Exp. Zool. **19**: 1–21.

1919 The genetics of purple eye color in Drosophila. J. Exp. Zool. **28**: 265–305.

1921 Current maps of the location of the mutant genes of *Drosophila melanogaster*. Proc. Nat. Acad. Sci. Washington **7**: 127–132.

BRIDGES, C. B., and MORGAN, T. H., 1919 The second chromosome group of mutant characters. Pub. Carnegie Instn. **278**: 123–304.

1923 The third chromosome group of mutant characters of *Drosophila melanogaster*. Pub. Carnegie Instn. **327**:

DOBZHANSKY, T., 1930 Translocation involving the third and fourth chromosomes of *Drosophila melanogaster*. Genetics **15**: 347–399.

GOWEN, M. S., and GOWEN, J. W., 1922 Complete linkage in *Drosophila melanogaster*. Amer. Nat. **56**: 286–288.

MORGAN, L. V., 1925 Polyploidy in *Drosophila melanogaster* with two attached X chromosomes. Genetics **10**: 148–178.

MORGAN, T. H., BRIDGES, C. B., and STURTEVANT, A. H., 1925 The genetics of Drosophila. Bibl. genet. **2**: 1–262.

MULLER, H. J., 1916 The mechanism of crossing over. Amer. Nat. **50**: 193–221, 284–305.

1918 Genetic variability, twin hybrids and constant hybrids in a case of balanced lethal factors. Genetics **3**: 422–499.

PAYNE, F., 1924 Crossover modifiers in the third chromosome of *Drosophila melanogaster*. Genetics **9**: 327–342.

PLOUGH, H. H., 1917 The effect of temperature on crossing over in Drosophila. J. Exp. Zool. **24**: 147–209.

1921 Further studies on the effect of temperature on crossing over. J. Exp. Zool. **32**: 187–202.

STERN, C., 1926 An effect of temperature and age on crossing over in the first chromosome of *Drosophila melanogaster*. Proc. Nat. Acad. Sci. Washington **12**: 530–533.

STURTEVANT, A. H., 1917 Genetic factors affecting the strength of linkage in Drosophila. Proc. Nat. Acad. Sci. Washington **3**: 555–558.

1919 Inherited linkage variations in the second chromosome. Pub. Carnegie Instn. **278**: 305–387.

1921 A case of rearrangement of genes in Drosophila. Proc. Nat. Acad. Sci. **7**: 235–237.

1926 A crossover reducer in *Drosophila melanogaster* due to an inversion of a section of the third chromosome. Biol. Zbl. **46**: 697–703.

WARD, L., 1923 The genetics of curly wing in Drosophila. Another case of balanced lethal factors. Genetics **8**: 276–300.

WEINSTEIN, A., 1918 Coincidence of crossing over in *Drosophila melanogaster*. Genetics **3**: 135–159.