Genet. Res., Camb. (1983), **41**, *pp.* 17–28
With 1 text-figure
Printed in Great Britain

The effect of an unusual chromosome architecture on disjunction and non-disjunction in Drosophila

By RAPHAEL FALK

Department of Genetics, The Hebrew University, Jerusalem 91904, Israel

(*Received 19 November 1981 and in revised form 28 April 1982*)

SUMMARY

Two homologous autosomes of Drosophila that were attached to form a single entire compound autosome II were found to affect the segregation of the sex chromosomes in both males and females. The compound segregated nearly always from an attached *X*. *Y* chromosome in males with no other sex chromosome. When two sex chromosomes were present together with the compound they differed in their tendency to segregate from the compound. In males the *X*. *Y* chromosome segregated more often from the compound than did the *Y* chromosome, and the *Y* chromosome segregated more often from the compound than did the regular *X* chromosome. In females the *X*. *Y* segregated more often from the compound than did the regular *X* chromosome. This preferential segregation in females was observed for exchange *X* chromosomes as well as for the non-exchange chromosomes.

In the presence of the compound the frequency of primary non-disjunction of the sex chromosomes was elevated in both females and males; usually both sex chromosomes segregated *from* the compound and only rarely *with* it.

Flies devoid of most of the proximal heterochromatin of the sex chromosomes die. However, when the compound autosome was present some such flies survived. This indicates that a segment of the proximal heterochromatin of the sex chromosomes was intercalated into the compound when it was constructed. It was concluded that the segment intercalated into the compound carries specific sites for sex chromosome disjunction. Specific sites determine sex chromosome disjunction in males. In females they determine the disjunction of the sex chromosomes in cooperation with exchange pairing.

(1) INTRODUCTION

One of the classical problems of Genetics that has not yet been settled satisfactorily is the mechanism by which homologous chromosomes interact in preparation for disjunction during meiosis. The possibility of manipulating the chromosomes of *Drosophila melanogaster* practically at will made the disjunction of homologues in the oocytes of Drosophila a favourable system for the study of this problem.

Although it is well documented that exchange pairing is a significant factor in determining disjunction of homologues, it is clear that this alone cannot explain regular disjunction in Drosophila. In about 5% of the oocytes non-exchange tetrads of regular *X* chromosomes disjoin normally, and the frequency of non-exchange tetrads increases dramatically in heterozygotes for inversions, without being accompanied by a corresponding increase in the frequencies of non-disjunction. The small fourth chromosomes disjoin regularly in spite of forming practically always non-exchange tetrads. This is all in addition to the regular disjunction of the chromosomes in male meiosis, where no exchange whatever takes place.

On the basis of extensive experimental work Grell (1962*a*, 1976) suggested that the regular disjunction of non-exchange chromosomes in Drosophila oocytes was determined by a 'distributive pairing'. The most important premises of the distributive pairing hypothesis were that non-exchange chromosomes were not determined to disjoin unless they paired a second time, after the exchange pairing; that second pairing is not on the basis of homology between the chromosomes. To ensure that such a distributive pairing would, as a rule, occur between homologues, it was argued that pairing was based on similarity of chromosome length. This takes care, for example, of the *X* chromosomes of Drosophila that would not regularly pair with the tiny fourth chromosomes.

The model of distributive pairing following the exchange pairing has been challenged mainly by Novitski, both on theoretical grounds (Novitski, 1964) and on experimental grounds. One of the predictions of the model is that chromosomes that participated in exchange pairing would not participate in heterologous disjunction, which should be strictly distributive. This prediction was not found to be borne out by appropriate experiments (Novitski, 1975, 1978; Portin, 1975).

The entire compound chromosome II, C(2)EN, constructed by Novitski (1976), offered a new opportunity to investigate the factors operative in chromosome disjunction. In this chromosome both homologues are attached to a single centrometer. It has no homologous partner in the genome and it is a specially long chromosomal element – twice the size of a regular autosome. In the present study I will show that the compound autosome disjoins non-randomly from the sex chromosomes in both males and females. Apparently a segment of the proximal heterochromatin of the sex chromosomes had been intercalated into the compound during its construction. The experiments confirm the findings of Gershenson (1940) and of Lindsley & Sandler (1958) that disjunction determinants for the sex chromosomes are located in the proximal heterochromatin of these chromosomes.

(2) MATERIALS AND METHODS

The entire compound chromosome II is a chromosome in which the left arms of both homologues are attached to a single centromere, while the right arms are attached to the distal end of each of the left arms via a heterochromatic segment (Novitski, 1976) (Fig. 1).

Half the gametes of flies with the compound are expected to carry the two

complete homologues of the autosome, and half none. Therefore, when flies with the compound are mated to flies with regular, unattached autosomes, the only progeny that survive are those in which autosomal non-disjunction occurred in the regular parent. When females with the compound are mated to males with the compound, half the progeny should carry the maternal compound and half the

Fig. 1. The entire compound chromosome II. The two homologues are rearranged and attached to a single centromere.

paternal compound. In order to distinguish which parent contributed the compound to a specific progeny, we used throughout this study one compound that had no markers, C(2)EN, +, and another that had two markers, C(2)EN, *b bw* (black body colour, brown eyes). For brevity they will be referred to by C, + and C, *b bw*. Both compounds were provided by E. Novitski, Eugene, Oregon.

The *X* chromosome of one compound was wild type, the other was marked y^2 *su-*$w^a w^a$ (i.e. yellow body colour, a partial suppressor of w^a and white-apricot eye colour). Other sex chromosomes were introduced into the compound stocks by mating flies with the compound to flies with appropriately marked *X* chromosomes and with free autosomes. The few progeny produced by autosomal non-disjunction in the free autosome stocks were maintained and propagated. Three additional sex chromosomes were introduced into compound stocks: (1)$Y^S X . Y^L$, In(1)EN,*y* (hereafter: *X*. *Y*); in this attached *X*. *Y* chromosome the short arm of the *Y* chromosome is attached distally to the *X* chromosome and the long arm of the *Y*, as a second arm, on the other side of the centromere. The *X* chromosome itself is inverted throughout and carries a yellow bristle and body colour marker. (2) In(1)$sc^{4L} sc^{8R}$ + S, *y* w^a *B* (hereafter: $sc^4 sc^8$); a chromosome with two inversions, one of which comprises nearly its whole length. A segment of the proximal heterochromatin, including the nucleolus organizer, was deleted. This chromosome is marked by yellow bristle and body colour, white-apricot eye colour and Bar eye form. (3) A regular *X* chromosome, marked with *sn*, a bristle mutant. From females heterozygous for the inverted chromosomes and the normal chromosomes, only non-recombinant and double recombinant progeny may be recovered. For further details on markers and chromosomes, see Lindsley & Grell (1968).

All flies were grown on a standard cornmeal–agar–yeast medium at 25 °C. All matings were transferred twice to new culture bottles or vials. As a rule 10–15 pairs of parents were mated in bottles. In those matings where only few exceptional progeny were expected (such as matings of compound autosome flies with free autosome flies) 30–40 pairs of flies were mated in each culture bottle. In matings where primary non-disjunction of the sex chromosomes or recombination between homologues could affect the results, single females were mated in vials, and the mothers' genotype was verified before the progeny were included in the reported results.

Table 1. *Relative recovery of maternal and paternal entire compound chromosome II in reciprocal matings*

Maternal compound	Paternal compound	No. expts	Number of progeny		Ratio of maternal to paternal compounds (mean ± stand. deviat.)
			+	*b bw*	
C, +	C, *b bw*	21	8510	2201	1:0·273 ± 0·233
C, *b bw*	C, +	29	6426	10432	1:1·307 ± 0·436

3. RESULTS

(*a*) *Recovery of compound autosomes*

Marker genes were used to distinguish maternal and paternal sex chromosomes and the compound II chromosomes. Matings were made between females carrying C, + and C, *b bw* bearing males, or vice versa. Each parent should contribute equal numbers of gametes with the compound and without it. Since progeny that obtained either both the maternal and paternal compounds or none die, only half the zygotes of such a mating are expected to survive. These should include equal numbers of progeny with the maternal and with the paternal compound. In reality such results were never obtained (Table 1). Novitski *et al.* (1981) discuss in detail the possible reasons for this. For the present study this inequality in the recovery of the complementary types of gametes must be kept in mind both in the design of the experiments and in the overall interpretation of the data. For this reason all experiments were carried out so that a given compound was maternal in some matings and paternal in others. Also, all the results will be presented separately for those progeny that obtained the compound from a given parent and for those that did not get it from that parent.

(*b*) *Sex chromosome segregation in males with compound II*

When males with a compound autosome and an *X*. *Y* chromosome, but with no free *Y* chromosome, were mated to females with a differently marked compound (irrespective of the females' *X* chromosome), nearly all daughters carried the maternal compound and nearly all sons carried the paternal compound (Table 2). Thus the compound II appears to segregate regularly from the *X*. *Y* chromosome in these males, i.e. segregation is of the *X*. *Y* ↔ C type rather than of the *X*. *Y*; C ↔ O type.

As will be shown later, non-disjunction of the *X* chromosomes in compound II females was high. In some experiments progeny of maternal *X*/*X* ↔ C non-disjunction could not be distinguished from those of paternal *X*. *Y*; C ↔ O segregation. In spite of these ambiguities the results are clear. Correcting the results by assuming all ambiguous progeny were due to maternal non-disjunction (Tables 2–4, in parentheses) probably inflates the true ratio of *X*. *Y* ↔ C to *X*. *Y*; C ↔ O segregation in males.

Table 2. *Segregation of the entire compound chromosome II from the* X . Y *chromosome in males with no free* Y *chromosome (20 experiments)*

Maternal gametes recovered*	Paternal gametes recovered				Ratio of $X.Y$; C↔O to $X.Y$↔C
	$X.Y$; C	O	$X.Y$	C	
X; O	130	—	—	4994	0·0260:1
	(2)†	—	—	(5148)	(0·0004:1)
X; C	—	190	5387	—	0·0353:1
	—	(30)	(5436)	—	(0·0055:1)

* Maternal X chromosomes were either both y^2su–w^aw^a or y^2su–w^aw^a and $X.Y$.

† In some experiments ambiguities arose from the fact that non-disjunction in the females could mimic paternal $X.Y$; C↔O segregation; the numbers in parentheses represent the values obtained upon assuming that all such ambiguous progeny were due to maternal non-disjunction (see text).

Table 3. *Segregation of sex chromosome from entire compound chromosome II in males*

Maternal gametes recovered*	Paternal gametes recovered				Ratio of
	$y^{u2}su$–w^aw^a; C	Y; O	y^2su–w^aw^a; O	Y; C	X; C↔Y to X↔Y; C
	(*a*) y^2su–w^aw^a/Y; C males (29 experiments)				
X; O	1637	—	—	937	1·748:1
	(1677)†	—	—	(721)	(2·326:1)
X; C	—	8453	7830	—	1·080:1
	—	(8650)	(7417)	—	(1·167:1)
	$X.Y$; C	Y; O	$X.Y$; O	Y; C	Ratio of $X.Y$; C↔Y to $X.Y$↔Y; C
	(*b*) $X.Y/Y$; C males (9 experiments)				
X; O	130	—	—	473	0·275:1
	(129)	—	—	(472)	(0·273:1)
X; C	—	500	1367	—	0·366:1
	—	(472)	(1375)	—	(0·343:1)

* Maternal X chromosomes were either both y^2su–w^aw^a or y^2su–w^aw^a and $X.Y$.

† In parentheses, number of progeny assuming all ambiguous progeny were due to maternal non-disjunction (see footnote to Table 2 and text).

Preferential segregation of the sex chromosomes from the compound was also observed in males carrying a free Y chromosome (Table 3). Progeny in which the Y chromosome segregated from the paternal compound were recovered more frequently than those in which the X chromosome segregates from the paternal compound (Table 3*a*). On the other hand, progeny in which the Y chromosome segregated from the paternal compound were recovered less frequently than those in which the $X.Y$ chromosome segregated from the paternal compound (Table 3*b*).

Table 4. *Segregation of the* X . Y *chromosome from the entire compound chromosome II in females heterozygous for a regular* X *chromosome and the* X . Y *chromosome, mated to* X . Y *; C males (24 experiments)*

Paternal gametes recovered	Maternal gametes recovered				Ratio of X; C ↔ X . Y to X ↔ X . Y; C
	y^2su–w^aw^a; C	X . Y; O	y^2su–w^aw^a; O	X . Y; C	
X . Y; O	4126	—	—	1001	1:0·243
	(4132)	—	—	(1001)	(1:0·242)
O; C	—	2538	1103	—	1:0·435
	—	(2540)	(1103)	—	(1:0·434)

In parentheses, progeny from either maternal non-disjunction or paternal X . Y; C ↔ O segregation were assumed to be due to the non-disjunction (see footnote to Table 2 and text).

Table 5. *Progeny of* X . Y/sn; *C, + females mated individually to* X . Y *; C,* b bw *males*

Paternal gametes recovered	Maternal gametes recovered				Ratio of X; C ↔ X . Y to X ↔ X . Y; C
	X; C	X . Y; O	X; O	X . Y; C	
	(*a*) *nonrecombinants between* y *and* sn				
X . Y; O	2586	—	—	636	1:0·246
O; C	—	838	680	—	1:0·811
	(*b*) *double recombinants between* y *and* sn				
X . Y; O	6	—	—	2	1:0·333
O; C	—	13	11	—	1:0·846

(*c*) *Sex chromosome segregation in females with compound II*

The correlated segregration of the compound autosome and the sex chromosomes in males raised the question whether a similar preferential segregation could also be observed in the meiosis of oocytes. Females with the compound and heterozygous for the X . Y and the y^2su–w^aw^a (or sometimes the unmarked) chromosome were mated to X . Y; C males and their progeny were scored (Table 4). In females too the X . Y chromosome segregates more often from the compound than it is going with it: X . Y ↔ X; C segregation is more frequent than X . Y; C ↔ X segregation.

Since the X . Y chromosome is inverted in relation to the y^2su–w^aw^a chromosome, most progeny would be those of non-exchange gametes. In order to follow segregation of recombinants between the X . Y and the regular X chromosome from the compound, individual X . Y/sn; C, + females were mated to X . Y; C, *b bw* males and their progeny scored (Table 5). Note that the X . Y chromosome carries a whole X-chromosome inversion besides the marker *y* for the distal end of the chromosome. *sn* is located 21.0 crossing over units more proximally than *y* but still some 40 crossing over units away from the centromere. Thus no single recombinant would be recovered but most double recombinants between the X . Y and *sn* chromosomes would be detected.

Although the number of double recombinants was rather low, the preferential

Table 6. *Frequencies of maternal nondisjunction in* $y^2su\text{–}w^aw^a$; *C*, + *and* b bw *females*

Maternal non-disjunction gametes recovered		Frequencies of non-disjunction
X/X; O	36/1827 (173/5307)	0·0197 (0·0326)
O; C	201/5127 (368/11130)	0·0392 (0·0331)
X/X; C	8/807	0·0099
O; O	1/435 (2/467)	0·0023 (0·0043)

In parentheses, maximum frequencies assuming all ambiguous progeny were due to maternal nondisjunction (see footnote to Table 2 and text).

segregation of the X . Y from the compound over that of the regular X from the compound was found also among the double recombinants. The significance of the preferential segregation among the double recombinants is underlined by the results for (single) recombinants between w^a and *sn* in the $y^2su\text{-}w^aw^a/sn$ sisters of the experimental females: one recombinant X chromosome segregated from the compound in 22 cases and the complementary recombinant X chromosome in 23 cases.

(*d*) *Sex chromosome non-disjunction in flies with compound II*

Non-disjunction of the sex chromosomes was studied in females with compound autosomes and a pair of regular isosequential $y^2su\text{–}w^aw^a$ chromosomes. The frequency of primary non-disjunction of the X chromosomes in compound II females was 6 % or even higher (5·92 % if twice the frequency of maternal X/X gametes is considered, or 7·1 % if the sum of the frequencies of maternal X/X and no-X gametes are considered). This is as high as the frequency of secondary non-disjunction in XXY (no compound autosome) females. Furthermore, the segregation of the two X chromosomes is not independent of that of the compound II, both X chromosomes segregate from the compound much more frequently than they segregate with it (Table 6).

If the test females were mated to X/Y; C males the same phenotypes would have been obtained in most matings from maternal X/X; C ↔ O segregation as from paternal X/Y ↔ C segregation and from maternal X/X ↔ C segregation as from paternal X/Y; C ↔ O segregation. To avoid this ambiguity between maternal and paternal non-disjunction we preferred to mate the females to X . Y; C males. However, such males produce only few of the gametes necessary for the recovery of maternal X/X; C ↔ O non-disjunction gametes. To recover the latter gametes, the experimental female were mated in separate experiments to X . Y/Y; C males.

Table 7. *Gametes recovered from entire compound chromosome II males,* $y^2su–w^aw^a/y^+Y$; *C, in seven experiments*

	Paternal gametes recovered		
	$y^2su–w^aw^a$; O	y^+Y; O	$y^2su–w^aw^a/y^+$Y; O
Number	3285	3130	406
Frequency	0·482	0·459	0·060
	$y^2su–w^aw^a$; C	y^+Y; C	O; C
Number	26	23	99
frequency	0·176	0·155	0·669

The fertility of such males is low, so that the data for X/X; C↔O non-disjunction are not as extensive as those for the other non-disjunction type.

Also note that in some matings it was impossible to distinguish phenotypically between progeny from maternal non-disjunction and progeny from the rare paternal $X.Y$; C↔O segregation mode. Therefore, whenever the distinction was impossible, the maximum non-disjunction frequencies, assuming that all ambiguous progeny were due to non-disjunction rather than to the rare paternal segregation, were given in parentheses.

When the Y chromosome was marked by a y^+ allele of X chromosome origin, non-disjunction could be followed directly in males (Table 7). As for females, in males too both sex chromosomes may segregate with the compound or from it. In matings to $y^2su–w^aw^a/X.Y$; C females it was shown that the X/Y; C↔O non-disjunction pattern in $y^2su–w^aw^a/y^+Y$; C males was extremely rare. It was not included in Table 7 and all ambiguous progeny that could be due to paternal X/Y; C↔O segregation or to maternal X/X↔C non-disjunction (in matings to $y^2su–w^aw^a$; C females) were considered to be due to the latter.

The very unequal recovery of progeny with the maternal compound and the paternal compound (6821 : 148) was unexpected even in this type of experiments with compound autosomes. The possibility must be kept in mind that the extremely high frequency of X/Y↔C non-disjunction (0·669) was related to the presence of the marked y^+Y chromosome instead of the usual unmarked Y chromosome (the y^+Y chromosome may contain other heterochromatic segments from the X chromosome, besides the euchromatic segment containing the y^+ allele).

(*e*) *Evidence for homology between the* X *chromosomes and the compound II*

One possible explanation for the correlated segregation of the sex chromosomes and the compound autosome is that during the construction of the compound a segment of the sex chromosome, significant for regular chromosome disjunction, was translocated into it.

The sc^4sc^8 chromosome is devoid of most of the proximal heterochromatin of the sex chromosome; sc^4sc^8/O males do not survive. The sex chromosomes of females

Table 8. *Segregation of the* sc^4sc^8 *chromosome from the compound in* $sc^4sc^8/y^2su-w^aw^a$; *C*, + *and* b bw *females mated to* X . Y; *C*, b bw *and* + *males*

Maternal gametes recovered	Paternal gametes recovered	
	X . *Y*; O	O; C
sc^4sc^8; C	1555	—
y^2su–w^aw^a; O	—	1662
sc^4sc^8; O	–	38
y^2su–w^aw^a; C	1531	—
sc^4sc^8/y^2su–w^aw^a; O	—	837
O; C	561	—

Frequency of $X/X \leftrightarrow$ C non-disjunction 561/3646 – 0·154.
Relative viablity of sc^4sc^8/O; C males 38/1662 – 0·023.

heterozygous for this chromosome and a regular *X* chromosome (with free autosomes) disjoin regularly, but in males primary non-disjunction of sc^4sc^8 and *Y* is grossly elevated.

Males of the genotype sc^4sc^8/Y; C proved to be sterile. When sc^4sc^8/y^2su–w^aw^a; C, + or *b bw* females were mated to *X* . *Y*; C, *b bw* or + males, a low but consistent frequency of sc^4sc^8/O; C sons were recovered (Table 8). This proves that at least some genes missing in the sc^4sc^8 chromosome are intercalated in the compound II. Note also that maternal nondisjunction in females heterozygous for the deleted *X* chromosome is at least four times higher than in females with the compound and non deleted *X* chromosomes.

4. CONCLUSIONS AND DISCUSSION

The findings of this study may be summarized as follows:

(1) A segment from the proximal heterochromatin of the sex chromosomes had been intercalated in the entire compound chromosome II during its construction. The low viability of sc^4sc^8/O; C males indicates that only a fraction of the bb^+ genes, deleted in the sc^4sc^8 chromosome, is intercalated in the compound (Table 8). This conclusion has been substantiated by Dr E. Lifschytz of the Technion in Haifa, who showed that plasmids carrying specific sex chromosome heterochromatic DNA fragments ('insertion-1 elements') hybridize *in situ* with the compound (personal communication).

(2) The compound autosome segregates non-randomly from the sex chromosomes in both male meiosis (Tables 2 and 3) and female meiosis (Tables 4 and 5).

(3) Sex chromosomes differ in their affinity to the compound autosome. When only one sex chromosome is present, as in *X* . *Y*/O; C males, this chromosome segregates regularly from the compound (Table 2). In the presence of two sex chromosomes the *X* . *Y* segregates more often from the compound than both the *Y* chromosome (in males, Table 3) and the regular *X* chromosome (in females,

Table 4). On the other hand, the *Y* chromosome segregates more often from the compound than does the regular *X* chromosome (in males, Table 3). This leads to the conclusion that in both the females and the males specific determinants direct the disjunction of the sex chromosomes and the compound. At least some of these determinants were included in a segment intercalated into the compound, and some were lost in the segment deleted from the sc^4sc^8 chromosome. More determinants were present in the *X*. *Y* chromosome than in either the regular *X* or the *Y* chromosomes alone.

The existence of sex chromosome disjunction determinants in Drosophila has been inferred many years ago. Cooper (1959) showed that *X*–*Y* disjunction in males depended on elements, which he called 'collochores', in the proximal heterochromatin of the *X* chromosome. These elements have been shown recently also in electron micrographs of prometaphase–metaphase I in males (Ault, Lin & Church, 1981). Gershenson (1940), who studied secondary non-disjunction induced by free deleted *X* chromosomes of various lengths, suggested that there were four pairing sites in the proximal heterochromatin: two that function both in males and in females, one that functions in females only, and one that functions in males only. Lindsley & Sandler (1958), who repeated Gershenson's experiments and extended them, confirmed his conclusions (see also Peacock & Miklos, 1973). Although the importance of such sites for the disjunction of sex chromosomes in Drosophila males has been widely accepted, their significance for sex chromosome disjunction in females has been ignored in favour of models based on less specific mechanisms, such as that of the distributive pairing (Grell, 1962*b*).

(4) The presence of a compound induces primary non-disjunction in both females and males (Tables 6, 7). Among these, segregation of both sex chromosomes *from* the compound is signficantly more frequent than *with* it.

(5) Non-disjunction in females heterozygous for the deleted sc^4sc^8 chromosome in the presence of the compound autosome is nearly as high as in sc^4sc^8/Y males with free autosomes (Table 8).

The excess segregation of both sex chromosomes from the compound over that of both chromosomes with the compound is expected when each arm of the compound may interact with a sex chromosome, probably forming trivalents. On the other hand, an *X*-chromosome like sc^4sc^8, from which pairing sites are missing, would segregate at random in relation to the regular segregation of the compound and the other *X* chromosome.

(6) The preferential segregation of one sex chromosome from the compound over another (*X*. *Y* over regular *X*) is maintained for recombinant chromosomes as well as for the nonrecombinants (Table 5).

Yamamoto & Miklos (1977) showed that in females homozygous for chromosomes that lacked most of the proximal heterochromatin and all the postulated pairing sites, disjunction was very near to normal. Obviously, when conditions for pairing in the euchromatic segments were favourable 'most of the basal heterochromatin does not play a major and indispensable role in pairing and subsequent segregation of the chromosome in the female'. Yet, exchange-determined disjunction is not enough to secure regular disjunction even in Drosophila females, where non-

exchange tetrads are quite frequent. The experiments presented in this study support the notion that it is the presence of specific sites in the proximal heterochromatin of the sex chromosomes that ensures regular disjunction of non-exchange chromosomes in both sexes. But the observation that sex chromosomes that exchanged were determined to segregate from the compound just as non-exchange chromosomes did, indicates that the participation of an *X* chromosome in one kind of disjunction determination need not exclude the involvement of the same chromosomes in the other determination, or interfere with it, i.e. the chromosomes do not necessarily belong to two separate disjunction pools.

In her experiments on recombination and non-disjunction in Drosophila females Grell (1962*b*) proposed that the fact that the frequency of recombination between the *X* chromosomes was increased in the presence of an additional *Y* chromosome in the nucleus, rather than decreased, proved the independence of the distributive pool of disjunction from that of the exchange pool. Grell observed, however, an intrabrachial effect on crossing over both in females heterozygous for *X* chromosome inversions and in those with an additional *Y* chromosome. An inversion in one segment of the chromosome caused an increase in crossing over in the same arm further away from the inversion. In *XXY* females the frequency of crossing over among the regular offspring was decreased near the heterochromatin (to 99·4 % of that in *XX* females in the *f–car* interval and to 95·3 % in the *wy–f* interval, see fig. 3 of Greel, 1962*b*), while it was increased in the segments further away (to 104·8 % in the *cv–v* interval and to 119·4 % in the y^2–*cv* interval). In *XXY* females that were also heterozygous for inversions both intrabrachial effects appear to operate next to each other: crossing over in In(1)sc^7/+ females was only 23·2 % of the normal in the *cv–v* interval, next to the inversion, but 144·0 % of the normal in the *f–car* interval, furthest away from the inversion. In In(1)sc^7/+/Y females, on the other hand, crossing over was still 23·8 % in the *cv–v* interval, and only 94·4 % in the *f–car* interval. Grell believes that the two effects of the *Y* chromosome (on recombination and on non-disjunction) 'do not arise from a single cause' and that 'the *Y* must affect exchange prior to distributive pairing'.

I would like to suggest that increased crossing over frequencies in regular offspring of *XXY* females, which have been observed since the classical work of Bridges (1916), and secondary non-disjunction both arise from a single cause, namely the competition between the three sex chromosomes for the specific heterochromatic pairing sites. This competition leads to the formation of both *X–Y–X* trivalents as suggested by Cooper (1948) and *X–X–Y* trivalents (besides the two types of bivalents that may be formed by leaving one element unpaired). *X–X–Y* trivalents would be formed by pairing between one *X* and the *Y* at the specific sites proximally, and by exchange pairing between the two *X*'s distally. Since the *X* and the *Y* chromosomes do not recombine proximally, crossing over between the *X* chromosomes would be enhanced distally (the intrabrachial effect). That such *X–X–Y* trivalents are formed in *XXY* females is suggested by the formation of *X* chromosome trivalents in triploid females (Bridges & Anderson, 1925). In such females the *X* chromosome that exchanged with one of the other two *X* chromosomes proximally, often exchanged with the third *X* chromosome

distally. Since X–X–Y trivalents contribute only to the regular offspring of XXY females (and X–Y–X trivalents only to the exceptional offspring), they could go undetected, but for the increase in the distal crossing over in XXY females. Thus the intrabrachial effect indicates that determination of disjunction of nonexchange sex chromosomes occurs prior to, or simultaneously with, that of the exchange chromosomes.

I wish to thank E. Novitski for his constant support and encouragement throughout this study. C. Auerbach's editorial comments were most valuable. Parts of this work were supported by grant GM18678 from the National Institutes of Health to E. Novitski.

REFERENCES

AULT, J. G., LIN, H.-P. P. & CHURCH, K. (1981). Sex chromosome morphology at prometaphase-metaphase I of meiosis in *Drosophila melanogaster* males. *Genetics* **97**, s4.

BRIDGES, C. B. (1916). Non-disjunction as proof of the chromosome theory of heredity. *Genetics* **1**, 1–52, 107–163.

BRIDGES, C. B. & ANDERSON, E. G. (1925). Crossing over in the X-chromosome of triploid females of *Drosophila melanogaster*. *Genetics* **10**, 418–441.

COOPER, K. W. (1948). A new theory of secondary non-disjunction in female *Drosophila melanogaster*. *Proceedings of the National Academy of Science U.S.A.* **34**, 179–187.

COOPER, K. W. (1959). Cytogenetic analysis of major heterochromatic elements (especially X*h* and Y) in *Drosophila melanogaster*, and the thoery of heterochromatin. *Chromosoma* **10**, 535–588.

GERSHENSON, S. M. (1940). The nature of so-called genetically inert parts of chromosomes. *Videnskap. Akad. Nauk, URSS* Kiev (in Ukrainian), pp. 3–116.

GRELL, R. F. (1962*a*). A new hypothesis on the nature and sequence of meiotic events in the female of *Drosophila melanogaster*. *Proceedings of the National Academy of Science U.S.A.* **48**, 165–172.

GRELL, R. F. (1962*b*). A new model for secondary nondisjunction. The role of distributive pairing. *Genetics* **47**, 1737–1754.

GRELL, R. F. (1976). Distributive pairing. In *The Genetics and Biology of Drosophila*, vol. 1*a* (ed. M. Ashburner and E. Novitski), pp. 435–486. Academic Press.

LINDSLEY, D. L. & GRELL, E. H. (1968). *Genetic Variations of Drosophila melanogaster*. Carnegie Institute of Washington Publication, no. 627.

LINDSLEY, D. L. & SANDLER, L. (1958). The meiotic behavior of grossly deleted X chromosomes of *Drosophila melanogaster*. *Genetics* **43**, 547–563.

NOVITSKI, E. (1964). An alternative to the distributive pairing hypothesis in Drosophila. *Genetics* **50**, 1449–1451.

NOVITSKI, E. (1975). Evidence for the single phase pairing theory of meiosis. *Genetics* **79**, 63–71.

NOVITSKI, E. (1976). The construction of an entire compound two chromosome. In *The Genetics and Biology of Drosophila* vol. 1*b* (ed. M. Ashburner and E. Novitski), pp. 562–568. Academic Press.

NOVITSKI, E. (1978). The relation of exchange to nondisjunction in heterologous chromosome pairing in the Drosophila female. *Genetics* **88**, 499–503.

NOVITSKI, E., GRACE, D. & STROMMEN, C. (1981). The entire compound autosomes of *Drosophila melanogaster*. *Genetics* **98**, 257–273.

PEACOCK, W. J. & MIKLOS, G. L. G. (1973). Meiotic drive in *Drosophila*: New interpretations of the segregation distorter and sex chromosome systems. *Advances in Genetics* **17**, 361–409.

PORTIN, P. (1975). Non-homologous chromosome pairing in female Drosophila. Before or after exchange? *Hereditas* **80**, 59–68.

YAMAMOTO, M. & MIKLOS, G. L. G. (1977). Genetic dissection of heterochromatin in *Drosophila*: The role of basal X heterochromatin in meiotic sex chromosome behaviour. *Chromosoma* **60**, 283–296.