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Segregation of centric *Y*-autosome translocations in *Drosophila melanogaster*

I. Segregation determinants in males

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SUMMARY

A special screening procedure for the detection of induced Y-autosome translocations with centric breakpoints was applied. A series of Experimental stocks was constructed, each containing a different half of one of the induced T(Y; 2)'s (T element). The three other elements that were involved in the segregation experiments in each stock were a sex chromosome (X element), an inverted chromosome 2 (A element), and a free arm of chromosome 2 (F element). It is not feasible to determine the relative frequencies of all the 16 possible gamete types by mating an Experimental stock to one tester, nor to different testers that have each at least one class of progeny of the same genotype. Each Experimental stock was mated to four different Tester stocks and the data were calibrated so that a coherent segregation pattern could be obtained.

Segregation patterns in meiosis of males from 15 Experimental stocks, each with a different T element were studied. In most Experimental stocks the T element was of the left autosomal arm, while the F element was of the right autosomal arm. In four Experimental stocks the X element segregated independently of the A, F and T elements. In these Group 1 stocks, both the F and the T elements disjoined regularly from the A element. It was concluded that the T element of these stocks had no sex-chromosome disjunction determinants ('S-determinants') to interact with the determinants on the X element. Both the T elements and the F elements carried autosomal disjunction determinants ('H-determinants') that secured the segregation of the autosomal elements. The H-determinants of the left autosomal arm were qualitatively different from those of the right arm.

In the remaining 11 Group-2 Experimental stocks the X and T elements disjoined regularly, indicating the presence of S-determinants on the T elements of these stocks. The segregation of the T and the A elements in these stocks varied from nearly complete dependence to complete independence. It was concluded that this gradation reflected differences in the quantity of H-determinants present on the T elements of these Experimental stocks. It was impossible to discriminate between a model of continuous H determinants activity and one of a finite discrete number of determinants. The results do not agree with the claim that there are no autosomal disjunction determinants in the proximal heterochromatin of chromosome 2.

The S-determinants on the $B^{S}Yy^{+}$ chromosome were located both adjacent to the centromere and distally on the long arm. The latter were probably translocated to the Y chromosome together with the B^{S} marker.

1. INTRODUCTION

The mechanisms that control regular disjunction* of homologous chromosomes during meiosis are still among the most attractive, yet most enigmatic ones in genetic and cytogenetic research, in spite of the intensive attention that the subject has received for over six decades.

For regular disjunction of homologous chromosomes during the first meiotic division, conjunction (whether this is of the nature of physical pairing or not) between homologues is necessary. There is little doubt that usually exchange pairing plays a major role in determination of chromosome disjunction. Yet in *Drosophila melanogaster* males, and in many other species (see, for example, Gassner, 1969; Welch, 1973; Noda, 1975; Debus, 1978; Serrano, 1981) regular chromosome disjunction occurs without exchange or even exchange pairing. Furthermore, even in females of Drosophila, exchange is not a necessary precondition for regular chromosome disjunction. A perennial problem of cytogenetic research is thus the existence and the nature of specific segregation determinants on the chromosomes.

Mutants of the first meiotic division of Drosophila are limited in their effects either to females or to males, while those affecting the second meiotic division may do so in both sexes. Thus it was concluded that while the second meiotic division is under similar control in both sexes, the regulation of the first meiotic division in males differs from that in females (Sandler *et al.* 1968; Baker & Carpenter, 1972).

The long-time accepted major role that the basal heterochromatic segments, proximal to the centromeres of the chromosomes, play in the determination of disjunction of the chromosomes of Drosophila has been repeatedly challenged in recent years. It has been suggested that in females chromosome disjunction is determined either by exchange pairing that is essentially limited to the euchromatic segments of the chromosomes, or by a pairing process, that depended on the total length of the chromosomes involved, rather than on their contents and organization (Grell, 1962, 1964). For Drosophila males there seems to be general agreement that specific sites in the proximal heterochromatin of the *X* chromosome determine *X–Y* chromosome pairing and disjunction (Cooper, 1964, 1965; Yamamoto & Miklos, 1977; Applels &Hilliker, 1982; Ault, Lin & Church, 1982). But it has been claimed that in the autosomes neither the basal heterochromatin as such, nor any specific pairing sites in these heterochromatic segments, play any role in male meiotic pairing processes (Yamamoto, 1979; Hilliker, Holm & Appels, 1982). Furthermore, studies of non-exchange chromosomes in Drosophila indicated that forces that governed chromosome disjunction in females did not act in males (Grell, 1976;

* Throughout the series of these papers an attempt has been made to reserve *disjunction* (and *non-disjunction*) for events between homologues. *Segregation* is used for the relation between (at least some) non-homologous chromosomes. Thus, when four elements of a translocation heterozygote interact with each other, segregation may occur. If, however, the four elements interact like two pairs of homologues, the result may be disjunction.

Carpenter, 1972). In view of this, a claim for the existence of segregation determinants in the basal heterochromatin needed further experimental support.

Many efforts have been directed in recent years to the study of mutations in genes that effect meiotic processes (see Baker *et al.* 1976), and very recently also to the isolation of the DNA segments that might incorporate sequences involved in specific functions of the meiotic mechanism (centromeres, see, for example, Clarke & Carbon, 1983). Still, the study of the segregation of chromosomes in heterozygotes for rearrangements, especially translocations, continues to be one of the most efficient approaches to the problem. Because the disjunction of chromosomes during the anaphase of the first meiotic division is considered to reflect interaction of the homologues, or some sites along them, during early stages of meiosis, shuffling the putative disjunction determinants through chromosomal rearrangements should affect in a specific manner the segregation pattern of the chromosomes involved. Thus one way to find out whether segregation determinants exist and whether they are common to meiosis in females and in males would be to compare segregation patterns of given chromosome rearrangements in males with those in females in which exchange pairing had been eliminated or at least greatly reduced. For this purpose a series of induced Y-autosomal translocations, T(Y; 2)'s, with centric breakpoints was recovered (Falk & Baker, 1984) and the segregation of translocated elements was studied in Experimental stocks constructed from them.

It is important for the successful performance of experiments with such translocation heterozygotes that it should be feasible to identify (*a*) each of the chromosomal *elements* involved in the interaction of the rearranged complex, and (*b*) every possible segregational *configuration* of the elements involved. Unfortunately, it is rare that these premises can be met completely in cytological observations of meiosis as well as in genetic analyses of the progeny recovered from the gametes of such meiotic events. In genetic analyses, for euploid viable progeny to be recovered the genetic contents of the gametes produced by one parent must complement that produced by the other parent. Even when all elements involved in a translocation are marked differently, it is usually not possible to design a single experimental mating that would allow recognition of all the segregational configurations (see, for example, Lindsley *et al.* (1972); both their 3:1 and their 1:3 segregations contain products of two meiotic configurations each; cf. Fig. 2). Furthermore, for the quantitative analysis of the relative frequencies of the segregational configurations in one (tested) parent, a previous acquaintance with the relative frequency of the segregational configurations of the other (tester) parent is essential. Finally, matings are often arranged so that a progeny gets either both or no translocated element of a given reciprocal translocation from the same parent. In such cases, on comparing results from different translocations, one actually considers simultaneously effects accrued by two variables and their possible interactions.

In our study, only one of the chromosomal elements of a T(Y; 2) (that is, either the left or the right half translocation) was present in a given Experimental stock. The other half of the T(Y; 2) was replaced by a 'standard' free chromosome arm, i.e. a chromosome 2 from which one arm was deleted. Thus, the segregation pattern of four chromosomal elements, three of them being identical in different

Experimental stocks, and one varying from one stock to the other, could be followed.

As noted, it is impossible to detect all eight meiotic configurations that may be obtained from the four chromosomal elements by mating an Experimental stock to one, universal tester stock. To detect all eight meiotic configurations, the Experimental stocks were mated to four different Tester stocks. The results were then normalized, so that the results obtained with the different Tester stocks could be assembled into estimates of the frequency of each meiotic configuration. Although possible systematic errors between crosses to Tester stocks left a component of uncertainty in our analysis, we consider that at least first order factors that determine segregation in Experimental stocks could be well established.

We conclude (1) that the proximal segments of both autosomal arms carry disjunction determinants, (2) that the determinants on the left autosomal arm were different from those on the right arm, although some interactions between the determinants of both arms took place, (3) that there were several determinants on each autosomal arm, or that the determination function was spread over some length of the arms, and (4) that it was improbable that all these determinants were located exclusively in the euchromatic segments distal to the basal heterochromatin of the autosome. We could also confirm the presence of sex-chromosome disjunction determinants on translocated *Y* chromosomes. However, it appears that at least some sex chromosome determinants on the translocated (modified) *Y* chromosomes were of *X* chromosome origin.

2. MATERIALS

Flies were grown on standard cornmeal–yeast–agar medium at 25 °C. All experimental matings were performed with about 20 males and a few more females per culture bottle. Flies were transferred twice, after 2 days and again after another 2 days, to fresh culture bottles. All experimental matings with a given Experimental stock were performed within as short a time as possible of each other, and not exceeding 2 weeks.

Progeny were counted every other day from the 10th to the 18th day after a culture was established.

Most stocks and their markers are described in the text; because the experimental procedure depended to a large extent on the structure of a few stocks, they will be described here in some detail. For further details on markers and chromosomes, see Lindsley & Grell (1968).

(i) *C(2L)SH1/F(2R)VH2*, bw

The left arms of chromosome 2 are present as a compound metacentric chromosome. The chromosome carries also 'at least half of the 2R heterochromatin' (Hilliker, Holm & Appels, 1982). The right arms of chromosome 2 are present as two 'free' right arms, from which the left arm euchromatin and most of the heterochromatin was deleted.

(ii) *F(2L)*, dp/*C(2R)RM*, cn

The left arms of chromosome 2 are present as two 'free' chromosome arms, from which the right arm euchromatin was deleted. In microscopic slides of larval brain mitoses, it could be seen that this free arm had an extensive heterochromatic 'right' arm. Its origin is unknown, but as will be seen from the later results, there are some indications that this is of autosomal origin. The right arms of chromosome 2 are present as a compound reverse metacentric chromosome. This is in all probability not the same compound described by Gethmann (1976), by Hilliker *et al.* (1982) and by Yamamoto (1979), which carried some 2L proximal euchromatin, and possibly also some 2L heterochromatin, because its segregation from a C(2L)RM chromosome in C(2L)RM, *b*/(2R)RM, *cn* males is random.

These two stocks were kindly put at our disposal by E. Novitski, Eugene, Oregon. In later stages we introduced into both stocks an *X*-chromosome marked with *y*.

A third stock that was intensively used throughout these experiments is the entire compound chromosome 2, C(2)EN. It is described in detail elsewhere (Novitski, 1976; Novitski, Grace & Strommen, 1981; Falk, 1983).

3. PROCEDURES AND METHODS

(i) *Recovery of Experimental stocks*

T(Y; 2)'s with centric breakpoints were recovered as follows (see Fig. 1). Males with a *Y* chromosome marked with B^S on the long arm with y^+ on the short arm were irradiated. The irradiated males were mated to females with attached-*X* chromosomes (C(1)DX, *y f*) and an inverted, dominantly marked chromosome 2, In(2LR)CyO, *Cy* dp^{lvI} *pr* cn^2. All daughters with both the *Y*-chromosome markers and the dominant autosomal marker were mated *en masse*, either to C(2L)SH1/F(2R)VH2, *bw* males or to F(2L), *dp*/C(2R)RM, *cn* males. No progeny were expected from the non-translocated daughters, except for some due to rare autosomal non-disjunction. In practice, only F_1 daughters that carried centric autosomal translocations with either the *Y* chromosome or chromosome 4 could produce euploid viable zygotes with the gametes provided by the paternal stocks. Using this procedure, 17 T(Y; 2)'s with centric breakpoint were detected (Falk & Baker, 1984). This amounts to a frequency of 0·8 % centric T(Y; 2)'s obtained after irradiating the post-meiotic cells of the males with an X-ray dose of 3000 R. Lindsley *et al.* (1972) reported that 43 of their T(Y; 2)'s were broken in the centric heterochromatin, after irradiating the males with an X-ray dose of 4000 R. This amounts to an induction frequency of approximately 0·5 %.

Some progeny of F_1 daughters carried only the left half of the T(Y; 2), others only the right half. Some half translocations were recovered together with the dominantly marked autosome and a complementary free arm, others with the complementary compound and a homologous free arm. All these could be recognized with the help of the genetic markers (see Fig. 1). Attempts were made to establish a stock from each half translocation by mating the appropriate brothers and sisters. Sometimes it was necessary to repeat the backcrosses to flies of the parental genotypes.

In order to transform these stocks to Experimental stocks it was necessary (1) to eliminate the free *Y* chromosome, (2) to replace the females' C(1)DX, which caused lethality unless an appropriate segment of a *Y* chromosome was present,

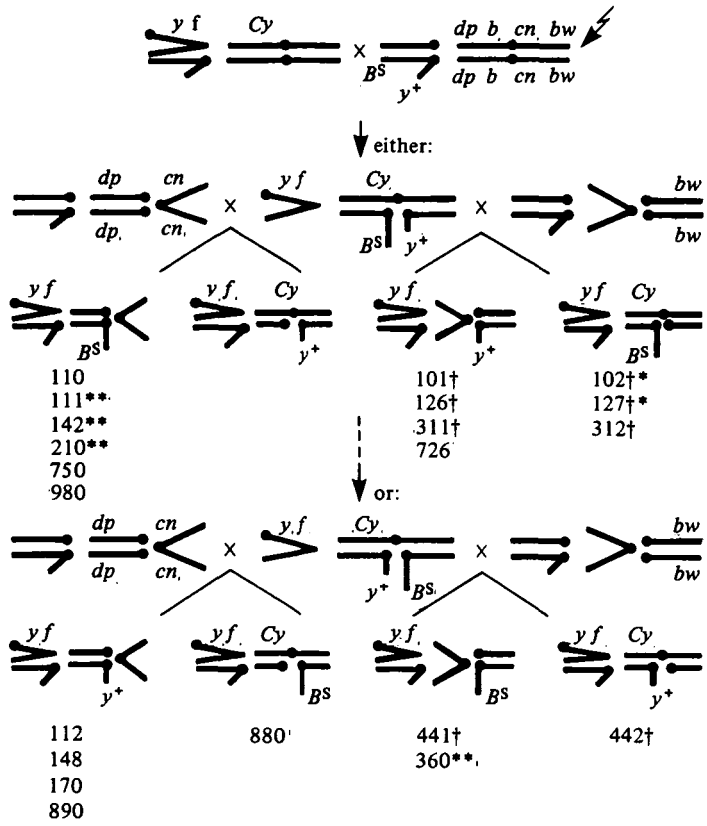

Fig. 1. Mating scheme used for the production of T(Y; 2) translocations, and the half-translocation stocks obtained, with their Experimental numbers. * Sterile; ** no Experimental stock could be established; † both left and right halves of the T(Y; 2) were recovered. Although only F_1 daughters of founder T(Y; 2) females are shown, both daughters and sons were obtained. Stocks were established by mating these daughters and sons. When not enough of the appropriate progeny were available, or when they were too infertile, flies of the appropriate genotypes were backcrossed to those of the paternal genotype.

with C(1)RM, reversed metacentric *X* chromosomes, (3) to replace the *X* chromosome of the males with an attached-*XY* chromosome ($Y^S X . Y^L$). In addition, all half-translocations recovered as stocks with the half translocation complemented by a compound and a homologous free arm (see Fig. 1) were transferred into stocks with the In(2LR)CyO and the complementary free arm. Most half translocations were finally transferred to Experimental stock genotypes; 14 of these were tested.

(ii) *Classification of chromosome segregation patterns*

Let us denote the chromosomal elements involved in an Experimental stock as follows:

X Attached-X, C(1)RM, y^2 *su*(w^a) w^a in females *and* attached-XY, $Y^SX \cdot Y^L$, In(1)EN, *y B* or $X^SX \cdot Y^L$, *y cv v* . y^+ in males.

A The multiply inverted, dominantly marked chromosome 2, In(2LR)CyO, *Cy* dp^{lvI} *pr* cn^2.

F (May sometimes be specified as F_L or F_R.) The left or the right free arm of chromosome 2, F(2L), *dp* and F(2R), *bw*, respectively.

T (May sometimes be specified as T_L or T_R.) The half-translocation element of the various Experimental stocks, which may carry either 2L or 2R.

Thus we may denote the Experimental stocks XATF (or XAT_LF_R) and XAFT (or XAF_LT_R), according to whether the T element carries 2L or 2R, respectively.

It is important to remember that the procedure for the recovery of the T(Y ; 2)'s was such that the T element could be deficient for only a short segment of the euchromatin of the chromosomal arm present. This means that the chromosomal break was either in the basal heterochromatin or in a nearby site in the proximal euchromatin, otherwise the cumulative effect of the missing genes would cause dominant lethality. Thus it can be only rarely that direct cytological observation would indicate whether the autosomal breakpoint was to the left or to the right of the centromere, i.e. whether the T element was provided with a *Y* chromosome centromere or with a chromosome-2 centromere.

Hardly any homozygotes for the T and F elements (i.e. non-*Cy* flies) survived in any of the Experimental stocks. This cannot be taken as evidence for the location of the autosomal breakpoints in the euchromatin rather than in the heterochromatin. Sites whose deletion caused recessive lethality have been identified within the basal heterochromatin of both the left and the right arms of chromosome-2 (Hilliker & Holm, 1975; Hilliker, Appels & Schalet, 1980). The possibility must also be considered that the segments of the *Y* chromosome, translocated to the autosomal arm suppressed essential autosomal genes by position effect variegation.

The classical nomenclature of the meiotic distribution of chromosomes in translocation heterozygotes, is based on the disjunction of centromeres. This is inappropriate in our experiments, because we do not know the source of the centromere of the T element. Of the four elements involved in our Experimental stocks, either two or three of the centromeres may be of the autosomal origin, while two or one may be of *Y*-chromosome origin.

Because there are eight possible meiotic configurations that may give 16 different types of gametes, we adopted a neutral, numerical nomenclature (Fig. 2). It may be noted that when the T element is provided with a *Y* chromosome centromere, i.e. when there are two autosomal and two *Y* chromosome centromeres, configuration I would be the classic 'alternate' disjunction and configuration III 'adjacent-1' disjunction, while configuration V would be 'adjacent-2' disjunction.

Meiotic configuration	Gametes		Schematic representation XATF	Schematic representation XAFT	Lindsley *et al.* (1972) Gametes	Nomenclature
I	XA	XA	X — •	X — •	*XX* Bal D P	alt
	TF	FT	— • —	— • —		
II	XTF	XFT	X — • —	X — • —	*XX* D P Bal	3 : 1
	A	A	— •	— •		
III	XF	XF	X • —	X — •	*XX* P D Bal	ad1
	AT	AT	= •	= •	or: *XX* D Bal P	or: ad2
IV	XAT	XAT	X = •	X = •	*XX* D Bal P	3 : 1
	F	F	•	•	or: *XX* Bal P D	or: 1 : 3
V	XT	XT	X — •	X • —	*XX* D Bal P	ad2
	AF	AF	= •	= •	or: *XX* P D Bal	or: ad1
VI	XAF	XAF	X — =	X = —	*XX* Bal P D	1 : 3
	T	T	— •	• —	or: *XX* D Bal P	or: 3 : 1
VII	X	X	X	X	*XX* D Bal P	1 : 3
	ATF	AFT	= • —	= • —		
VIII	XATF	XAFT	X = —	X = —	*XX* D Bal P	4 : 0
	O	O	O	O		

Fig. 2. Segregation pattern of XATF and XAFT Experimental stocks. X: attached-X in females, attached-XY in males; A, multiply inverted *Cy* chromosome 2; F, 'Free' left, or 'free' right arm of chromosome 2; T, half-translocation element. The corresponding gametes and nomenclature of Lindsley *et al.* (1972) is given for comparison; XX, attached X; Bal, multiply inverted autosome; D, half-translocation element with the distal part of chromosome Y; P, half-translocation element with the proximal part of chromosome Y.

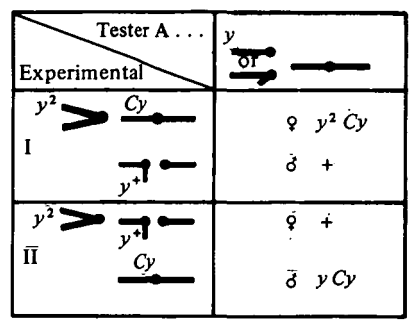

Fig. 3. Schematic representation of the gamete configurations of Experimental stock females that give viable progeny with Tester stock A males, and the phenotypes obtained.

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(iii) *Detecting meiotic configurations with Tester stocks*

Four Tester stocks were used to determine the relative frequencies of the eight meiotic configurations that may be obtained in XATF and XAFT Experimental stocks.

(*a*) *Tester stock A*

This stock carries normally arranged chromosomes, in which the *X* chromosome was marked with *y*. It produces essentially only X;2 gametes in females, and an equal frequency of *X*;2 and *Y*;2 in males.

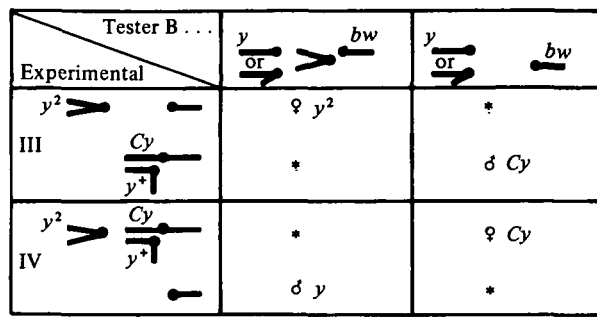

Fig. 4. Schematic representation of the gamete configurations of Experimental stock females that may give viable progeny with Tester stock B males, and the phenotypes obtained. * Non-viable.

This stock is adequate for the detection of gametes from configurations of type I and type II (Fig. 3). Rare autosomal non-disjunction gametes (our unpublished results: $0{\cdot}82 \times 10^{-3}$; see also Frost, 1961, and Baker & Hall, 1976) may produce euploid zygotes with gametes of configurations VII and VIII. Because the recovery of such zygotes depends on the very limited availability of the appropriate gametes in the Tester stock, it cannot be determined to what extent the few progeny obtained from such zygotes reflect the limited supply of Experimental stock or of Tester stock gametes. The progeny produced from such combinations of gametes were not included in the calculations.

(*b*) *Tester stock B*

This stock carries the compound left arm of chromosome 2 and two free right arms, *y*; C(2L)SH1/F(2R)VH2, *bw*. Both males and females of this stock produce predominantly gametes with the compound left arms and one free right arm C(2L)/F(2R), and gametes with one free right arm F(2R), with equal frequencies (Yamamoto, 1979). These gametes should complement gametes of configurations III and IV of the XATF and gametes of configurations V and VI of the XAFT Experimental stocks (see Fig. 2). However, only half of the gametes of the Tester stock would produce viable euploid zygotes with the appropriate gametes of the Experimental stocks (Fig. 4).

Culture bottles of Tester stock B produce a couple of hundred progeny each. In

order to find out how frequently these Tester stock flies produce other than the prevailing types of gametes, they were mated to two different complementary compound arms stocks, C(2L)RM, *b*/C(2R)RM, *cn* and C(2L)RM, *j*/C(2R)RM, *px*. These stocks produce four gamete types, C(2L), C(2R), C(2L)/C(2R) and O, with equal frequencies in males and predominantly the first two types in females (Yamamoto, 1979; Hilliker, Holm & Appels, 1982). Of these gametes the first two

Table 1. *Mean number of progeny per culture bottle obtained from gametes produced by Tester stock B females and males that were mated to tester* (*a*) *C(2L)RM*, b/*C(2R)*, cn *and to tester* (*b*) *C(2L)RM*, j/*C(2R)RM*, px *males and females, respectively*

(Results were calibrated according to the rules described in section iv below, and were pooled for testers (*a*) and (*b*). The number of progeny obtained from sex-chromosome non-disjunction of Tester stock B were doubled to compensate for the non-viability of half of non-disjunction zygotes as compared with the corresponding disjunction zygotes.)

Tester stock B:	*X*/*X*;C/F/F		*X*/*Y*;C/F/F	
Gametes:	*X*; 0/F/F	7·2	*X*; 0/F/F and *X*/*Y*; 0/F/F	0·75
	X; C/0	5·6	*Y*; C/0 and 0; C/0	0
	X; 0	0	*X*; 0 and *X*/*Y*; 0	0·25
	X; C/F/F	0	*Y*; C/F/F and 0; C/F/F	0
	X/*X*; C/0	0	*X*; C/0 and *X*/*Y*; C/0	1·5
	0; 0/F/F	0·4	*Y*; 0/F/F and 0; 0/F/F	0·25
	X/*X*; C/F/F	0·2	*X*; C/F/F and *X*/*Y*; C/F/F	0·25
	0; 0	5·8	*Y*; 0 and 0; 0	0·25
	X/*X*; 0	0		
	0; C/F/F	0		
	X/*X*; 0/F/F	0		
	0; C/0	0		
No. of cultures:				
Tester (*a*)	3		3	
Tester (*b*)	2		1	

would detect F(2R)/F(2R) gametes, and C(2L) gametes of the Tester stock, respectively (these complement none of the gametes of the Experimental stocks). The last two would detect nullo-2 gametes or C(2L); F(2R)/F(2R) elements of the Tester stock (which complement gametes of configurations VIII and VII, respectively, of the Experimental stocks).

Only few progeny were obtained upon these test-matings (Table 1). Thus we may consider Tester stock B to produce an excess of C(2L)/F(2R) and F(2R) gametes with only a negligible number of other types of gametes.

To further exclude the possibility that the few C(2L)/F(2R)/F(2R) and O gametes recovered from Tester stock B males were not the result of the limited supply of complementary gametes in females of its tester, Tester stock B males were mated to females with the entire compound chromosome 2. These produce only gametes complementary to the C(2L)/F(2R)/F(2R) and O gametes of the Tester stock. The very few progeny obtained from this mating (3 progeny in 5 culture bottles) confirm that only a negligible number of such gametes are produced by Tester stock B males.

Note the high frequency of sex chromosome non-disjunction (or loss) in Tester stock B females. A similarly high frequency of sex chromosome non-disjunction was found by Hager and Holm (1980) in C(3L)/C(3R) stocks. The sex chromosome non-disjunction was easily detected in our experimental matings, and the frequencies of the recovered gametes were corrected accordingly.

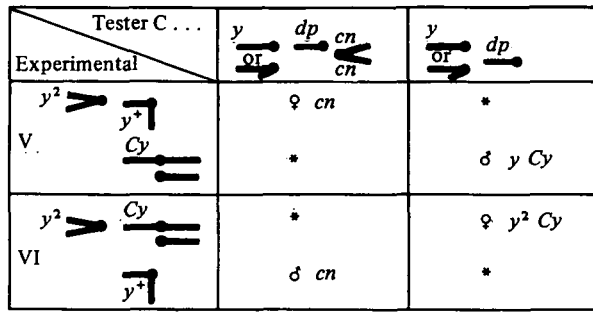

Fig. 5. Schematic representation of the gamete configurations of Experimental stock females that may give viable progeny with Tester stock C males, and the phenotypes obtained. * Non-viable.

(*c*) *Tester stock C*

This stock carries two free left arms of chromosome 2 and a compound right arm: *y*; F(2L), *dp*/C(2R)RM, *cn*. Both males and females of this stock produce predominantly gametes with one free left arm and the compound right arms F(2L)/C(2R) and gametes with one free left arm only F(2L), with equal frequencies. These gametes should complement gametes of configurations V and VI of the XATF and gametes of the configurations III and IV of the XAFT Experimental stocks (see Fig. 2). Only half of these gametes of the Tester stock would produce viable euploid zygotes with the appropriate gametes of the Experimental stocks (Fig. 5).

The scarcity of other types of gametes that could be produced by Tester stock C females and males has been verified by a procedure similar to that shown for Tester stock B, and the argument will not be repeated here.

(*d*) *Tester stock D1*

This stock carries an entire compound chromosome 2: y^2 *su*(w^a) w^a; C(2)EN, +. These flies produce only gametes with either the entire chromosome 2 or no chromosome 2 element. They served to recover gametes of configurations VII and VIII of the Experimental stocks. Only half of the gametes of the Tester stock produce viable euploid zygotes with the appropriate gametes of the Experimental stocks (Fig. 6, left half). Sex chromosome non-disjunction is frequent in this stock (Falk, 1983), but it is easily detected, and the results were corrected accordingly.

(*e*) *Tester stock D2*

Another possibility to recover configurations VII and VIII of the Experimental stocks was to use the C(2L)/C(2R) stocks as Tester stocks. As noted, half of the

paternal gametes of these stocks and a variable frequency of maternal gametes may complement Experimental stocks' gametes. Of these only half would produce viable euploid zygotes with the appropriate gametes of the Experimental stocks (Fig. 6, right half).

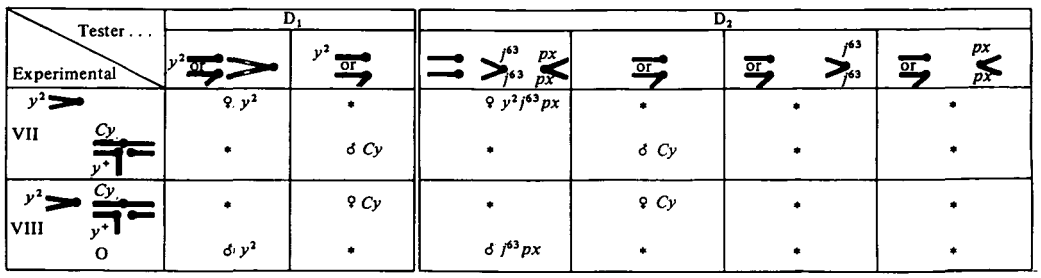

Fig. 6. Schematic representation of the gamete configurations of Experimental stock females that may give viable progeny with Tester stock D_1 males and with Tester stock D_2 males, and the phenotypes obtained. * Non-viable.

(iv) *Calibration of the frequencies of gametes obtained with different Tester stocks*

It is common practice to calculate the relative frequencies of various gametes produced by flies of a given genotype from the relative numbers of progeny produced from appropriate matings. Often the data must be corrected for deviations due to factors such as meiotic drive, or differences in the viability of the various types of progeny. In the present study another factor of error was introduced by comparing the numbers of progeny produced in *different* matings. It would have been desirable to have at least one identical genotype among the progeny of two different matings, in order to allow corrections for systematic differences between matings. Unfortunately, this was not possible.

An additional factor must be considered. The relative frequencies of progeny should, of course, properly reflect the relative frequencies of the gamete types produced by an Experimental stock, rather than those produced by the Tester stock. Therefore, only progeny produced from those Tester stock gametes that were available in excess were considered (see previous section). Many of the abundant gametes of the Tester stocks may encounter gametes of the Experimental stocks that would not complement them to give euploid zygotes. However, if pairing of gametes is random, and the relative frequency among the abundant gametes of the Tester stock that would fail to produce viable progeny with gametes of the Experimental stocks is known, a correction factor for this loss of progeny may be introduced.

When Experimental stocks were mated to Tester stock A, practically all tester gametes should complement experimental gametes of configurations I and II (Fig. 3). When, however, Experimental stocks were mated to Tester stocks B, C and D1, only half of the abundant tester gametes would complement the gametes of configurations III and IV, V and VI, and VII and VIII, respectively (Figs. 4–6). Therefore, the number of progeny obtained per culture bottle from these matings should be doubled in relating them to the number of progeny per culture obtained

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with Tester stock A. For similar reasons the numbers obtained from Experimental stock females mated to Tester stock D2 males were multiplied by a factor of four, in relating them to those obtained with Tester stock A flies (Fig. 6). Note that no Tester stock D2 females were used, because their segregation pattern was not adequately known.

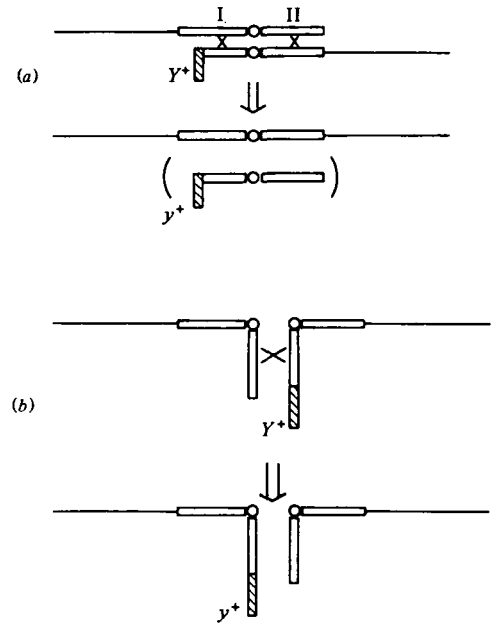

Fig. 7. Two possibilities for exchange between the T and F elements of XAFT stocks. (*a*) Upper figure, exchange between the left (I) or the right (II) arms of F and T. (*b*) Lower figure, exchange between the left arm of T and the right arm of F. (Only the T and F elements are presented.) Simple line, euchromatic arms of chromosome 2; empty boxes, basal heterochromatic segments of chromosome 2; hatched box, translocated *Y*-chromosome arm. Circle, chromosome 2 centromere. Only a single exchange is considered.

(v) *The Experimental stocks*

Of the 17 T(Y; 2)'s with centric breakpoints, in only four were both half-translocations recovered in separate progeny, while in the remaining 13 only one half-translocation was recovered. Of the former, two left half-translocations caused sterility (102 and 127). We then attempted to transfer the remaining half-translocations to Experimental stocks. In five (111, 142, 210, 360 and 441) the flies were so inviable or infertile that either no Experimental stocks could be established, or we were unable to test them properly. For the other 14 Experimental stocks were maintained and tested (see Fig. 1).

Nine of the Experimental stocks were XATFs (110, 112, 148, 170, 312, 442, 750, 890, and 980), while the remaining five were XAFTs (101, 126, 311, 726, and 880). The structure of all stocks was confirmed by cytological examinations of mitoses of larval brain cells.

In three of the XAFT Experimental stocks (126, 311, and 726) a spontaneous

process of transfer of the y^+ marker of the T element (carrying 2R) to the F(2L) element took place. By the time we performed the experimental matings, the y^+ marker of all flies of Experimental stock 311 and most flies of Experimental stocks 126 and 726 had already transferred to the F(2L) element. When the latter two stocks were retested a couple of months later, all flies were marked on the left arm rather than on the right arm. As noted before, the F(2L) element had a long heterochromatic right arm.

It appears that some exchange process took place between the F element and the T element of these Experimental stocks. It could not have been due to recombination between homologous autosomal sites (heterochromatic or euchromatic), because such events would result in a two-armed chromosome 2 (Fig. 7*a*), which was not the case. The recombination had to occur between the right arm of F(2L) and the left arm of the T(2R) (Fig. 7*b*). Thus it must have been an exchange between the heterochromatic arms. It is unknown why the original arrangement was selected against. At any rate, in the three Experimental stocks 126, 311 and 726, the T element is actually composed of the left arm of the F element, with at least some of the Y-chromatin from the original T element attached to it. The F element of these stocks, on the other hand, contains actually the right arm of the original T element. Thus, although Experimental stocks 126, 311 and 726 were originally recovered as XAFTs they were tested as XATFs.

No such transfer of markers was observed in the remaining two XAFT Experimental stocks (101 and 880).

An additional Experimental stock, originally from the *Y*-autosomal translocation collection of Lindsley *et al.* (1972), was obtained from the Umeå Drosophila Stock Centre. This stock, B190, is a T(*Y*; 2) broken in section 60 of the polytene chromosomes. T(*Y*; 2) B190 (like many of the other stocks of this collection) had lost the B^S marker. We showed y^+ to be located on the right half of the translocation. Because this stock carried both complementary half-translocation elements, it will be labelled as an XATT stock.

4. RESULTS

As noted before, the frequencies of different meiotic configurations measured by crosses to different Tester stocks are subject to greater systematic errors than the frequencies of different meiotic configurations measured by crosses to the same Tester stock. Several tests were performed to obtain an estimate of this source of error.

The coefficient of variance – that is, the standard deviation between the number of progeny of a given genotype (in the five culture bottles of the same mating), expressed as the proportion of the mean number of these progeny per culture bottle – was calculated. This allows a comparison of the variations in the numbers of progeny of *different* genotypes in *different* matings. Because pooling such data would make the values meaningless, only a sample of the coefficients of variance for progeny obtained in two test matings for five Experimental stocks is given in Table 2. The coefficient of variance usually did not exceed 30% for those genotypes that were represented by many progeny (more than 100). When the

number of progeny was less than ten or up to a couple of dozens the coefficient of variance often amounted to 50–80 % of the mean number of progeny. When only single progeny were obtained per culture, fluctuations were too extensive to make the values reliable. Note also that pooling the figures for the two types of progeny produced from gametes of the same configuration, gave considerably reduced coefficients of variance.

Table 2. *Mean number of progeny per culture-bottle* ($\bar{x}$) *and the coefficient of variance* (*c.v.*) *of number of progeny in five culture-bottles obtained with different Experimental stock males that were mated to Tester stocks B and C females*

Experimental stock	Females		Males		Pooled		Females		Males		Pooled	
	$\bar{x}$	c.v.	$\bar{x}$	c.v.	$\bar{x}$	c.v.	$\bar{x}$	c.v.	$\bar{x}$	c.v.	$\bar{x}$	c.v.
	TESTER STOCK B											
	Configuration III						Configuration IV					
101	117·0	0·35	60·0	0·78	177·0	0·48	1	0·71	1·4	0·81	2·4	0·70
110	186·1	0·29	199·1	0·28	385·2	0·28	8·2	0·79	3·6	0·42	11·8	0·61
112	122·2	0·19	174·6	0·20	296·8	0·20	0	—	0·4	1·37	0·4	1·37
890	233·8	0·20	136·4	0·24	370·2	0·21	0·2	—	2·2	1·99	2·4	2·01
980	93·4	0·29	149·8	0·30	243·2	0·28	0·2	—	0	—	0·2	—
	TESTER STOCK C											
	Configuration V						Configuration VI					
101	7·6	0·30	6·6	0·50	14·2	0·29	14·4	0·33	7·8	0·52	22·2	0·37
110	17·6	0·46	23·0	0·09	40·6	0·23	26·6	0·19	31·4	0·16	58·0	0·16
112	13·0	0·49	18·2	0·51	31·2	0·46	9·6	0·85	28·4	0·35	38·0	0·44
890	6·2	0·84	5·6	0·54	11·8	0·68	3·0	0·62	12·6	0·56	15·6	0·43
980	9·0	0·28	4·4	0·52	13·4	0·27	2·2	0·75	8·2	0·56	10·4	0·59

Some Experimental stocks were tested more than once, at intervals of several months. Although there were sometimes considerable differences in the mean number of progeny per culture bottle between repeats of the same mating (e.g. 617·5 and 110·6 for Experimental stock 126, or 330 and 990 for Experimental stock 726), there was good agreement between the proportion of the different configurations (see Table 5).

A control experiment was performed with flies with normal (non-attached) sex-chromosomes, a free left and a free right arm and the inverted *Cy*-marked chromosome 2. These flies, denoted X/X; $A/F_L/F_R$, were mated to Tester stocks A, B, and C (Table 3). Note that in females of this stock configurations I and II reduce to a single configuration, as do configurations III and IV, configurations V and VI, and configurations VII and VIII. In males there was good agreement between pairs of configurations recovered in the same mating: I with II, III with IV, and V with VI, as expected when the *X* and *Y* chromosomes disjoined independently of the autosomal elements. The frequency of configurations III and IV was similar to that of configurations V and VI, in both females and males, suggesting that both left and right free arms fail to disjoin equally frequently from the *Cy* chromosome. On the other hand, the number of progeny from complementary genotypes of the same configuration varied sometimes by as much as a factor of

two, which presumably reflects differences in viability of the genotypes. These findings indicate that the variability between data obtained from different matings need not exceed variability in data obtained from the same mating. Note also that matings to Tester stocks B and C males, which carry chromosomal rearrangements, produce as many (or even more) offspring per culture bottle as do matings to Tester stock A males whose chromosomes are free of major chromosomal rearrangements.

Table 3. *Numbers of progeny and calculated relative frequencies of gametes obtained on mating females and males of a control stock with non-attached sex-chromosomes, an inverted* Cy*-marked chromosome 2, a free left-arm chromosome and a free right-arm chromosome to Tester stock A, B and C*

Tester stock...				A		B		C		Corrected mean/ culture	Proportion
				♀♀	♂♂	♀♀	♂♂	♀♀	♂♂		
				(*a*) X/X; $A/F_L/F_R$ gametes							
I and II	X	A		178	264	—	—	—	—	435	1
	X	F_L	F_R	217	211	—	—	—	—		
III and IV	X		F_R	—	—	193	205	—	—	660·7	1·52
	X	A	F_L	—	—	291	302	—	—		
V and VI	X	F_L		—	—	—	—	152	184	654	1·50
	X	A	F_R	—	—	—	—	322	323		
No. of cultures				2		3		3			
				(*b*) X/Y $A/F_L/F_R$ gametes							
I	X	A		249	—	—	—	—	—	477	1
	Y	F_L	F_R	—	228	—	—	—	—		
II	X	F_L	F_R	211	—	—	—	—	—	423	0·89
	Y	A		—	212	—	—	—	—		
III	X	F_R		—	—	38	—	—	—	46·7	0·10
	Y	A	F_L	—	—	—	35	—	—		
IV	X	A	F_L	—	—	29	—	—	—	42·7	0·09
	Y	F_R		—	—	—	35	—	—		
V	X	F_L		—	—	—	—	39	—	57·3	0·12
	Y	A	F_R	—	—	—	—	—	51		
VI	X	A	F_R	—	—	—	—	38	—	71·3	0·15
	Y	F_L		—	—	—	—	—	65		
No. of cultures				1		3		3			
Correction factor				1		2		2			

Therefore, it can be safely claimed that the observed differences in relative frequencies of the configurations between females and males of the control stock indicate the sensitivity of the procedure to inherent differences in segregation patterns. We conclude that our procedure is valid for detecting at least first-order differences in the segregation pattern of Experimental stocks.

The raw data for all 15 Experimental stocks are given in Table 4. The number of culture bottles on which the results for each mating with Tester stocks are based are given at the bottom of each column. The relative frequencies of the eight meiotic configurations for all 15 Experimental stocks are given in Table 5. Included

Table 4. *Number of progeny recovered for the different types of gametes produced by the Experimental stocks*

(The number of culture bottles of each Tester stock upon which the counts were based are given at the bottom of each column. In few cases a culture was not transferred to a new bottle after 2 days (and was considered 1/3) or was transferred only once to a new bottle (and was considered 2/3).)

Experimental stock...	126	311	442	726	880	750	312	110	890	170	112	980	101	148	B190
Gametes															
I X A	978	419	488	1037	1986	1430	4116	1211	2147	1735	1221	497	1783	1057	1240
T F	1361	618	930	1603	1955	1144	3086	886	2445	2289	1962	616	2668	1784	1799
II X T F	1128	558	746	1419	2	47	179	43	71	37	53	10	10	29	1
A	1068	434	708	1110	13	52	266	30	88	78	47	13	29	43	13
III X F	138	121	156	396	240	531	1227	605	949	1173	742	1212	1279	1105	1474
A T	178	269	162	453	159	279	983	314	995	1291	987	734	1922	1272	259
IV X A T	122	168	148	368	7	3	22	5	41	37	0	1	5	3	0
F	130	112	95	265	1	3	28	7	8	12	2	14	3	10	143
V X T	426	289	44	592	4	38	92	38	88	102	69	31	101	115	90
A F	514	189	63	676	5	33	201	33	146	127	91	28	144	149	209
VI X A F	343	137	43	489	1	71	233	72	134	80	48	15	158	41	29
T	357	246	62	470	1	46	268	39	157	237	142	63	138	217	44
VII X	0	0	0	—	2	7	1	16	18	20	27	46	15	0	98
A T F	0	0	2	—	11	5	4	38	9	6	25	19	6	56	92
VIII X A T F	0	0	0	—	0	0	0	12	0	0	0	1	1	6	5
0	0	1	1	—	0	0	0	1	0	0	0	0	2	0	1
							No. of culture bottles								
A	3	2	4	4	2	2	$6^{2/3}$	2	2	3	2	1	4	3	3
B	8	5	7	13	5	5	12	5	5	10	$5^{1/3}$	5	16	5	5
C	10	5	4	11	5	5	8	5	5	$10^{2/3}$	5	5	9	5	5
D_1	4	3	6	0	5	5	7	6	5	5	5	5	4	5	5

Table 5. *Corrected and calibrated relative frequencies of gametes from different meiotic configurations (configuration I set as 1)*

Numbers in parentheses, corrected for viability differences. Repeats of the same Experimental stocks are presented separately.)

(*a*) Group 1 Experimental stocks

Configurations	Control X/Y; A/F/F	126 XATF y^+		311 XATF y^+	442 XATF y^+	726 XATF y^+	
		1st	2nd			1st	2nd
I X A T F	1	1	1	1	1	1	1
II X T F A	0·89	0·93	0·95	0·96	1·03	0·93	0·97
III X F A T	0·10	0·10	0·10	0·30	0·26	0·32	0·19
IV X A T F	0·09	0·09	0·07	0·22	0·20	0·27	0·18
V X T A F	0·12	0·20	0·23	0·37	0·15	0·37	0·32
VI X A F T	0·15	0·13	0·18	0·30	0·15	0·28	0·25
VII X A T F	—	0	—	0	0·002	—	—
VIII X A T F 0	—	0	—	0·001	0·001	—	—

(*b*) Group 2 Experimental stocks

Configuration	880 XAFT B^s	750 XATF B^s	312 XATF B^s		110 XATF B^s	890 XATF y^+	170 XATF y^+		112 XATF y^+	101 XAFT y^+			980 XATF B^s	148 XATF y^+	B190 XATT $(B^s)y^+$
			1st	2nd			1st	2nd		1st	2nd	3rd			
I X A T F	1	1	1	1	1	1	1	1	1	1	1	1	1	1	1
II X T F A	0·004	0·04	0·06	0·06	0·04	0·03	0·02	0·03	0·03	0·003	0·01	0·01	0·02	0·02	0·005
III X F A T	0·08	0·25	0·32	0·27	0·33	0·34	0·53	0·23	0·41	0·57	0·67	0·71	0·70	1·01	(1·16) 0·68
IV X A T F	0·002	0·002	0·004	0·004	0·005	0·01	0·01	0·002	0·001	0·01	0·003	0·004	0·005	0·01	(0·11) 0·06
V X T A F	0	0·02	0·05	0·07	0·03	0·04	0·04	0·03	0·04	0·05	0·04	0·03	0·02	0·11	(0·12) 0·09
VI X A F T	0·002	0·04	0·07	0·15	0·04	0·05	0·04	0·04	0·05	0·04	0·03	0·01	0·03	0·11	0·03
VII X A T F	0·003	0·004	0·001	—	0·02	0·005	0·005	—	0·01	0·01	—	—	0·02	0·02	0·08
VIII X A T F 0	0	0	0	—	0·004	0	0	—	0	0	—	—	0	0·003	0·002

are also the results of the control stock X/Y; A/F/F. All frequencies are normalized to configuration I. The arm of the T elements involved, as well as the Y chromosome marker attached to each T element are given under the serial number of each Experimental stock in Table 5.

Where Experimental stocks were tested repeatedly the corrected relative frequencies for each repeat are given separately. No corrections were introduced in the numerical results for what appeared to be viability differences between the complementary genotypes of a given meiotic configuration, except in the case of Experimental stock B190 where the number of the larger class in configurations III, IV and V was doubled instead of taking the sum of the two complementary genotypes (The uncorrected numbers were given in parentheses.)

Non-disjunction of the sex chromosomes of the Tester stocks B, C and D1 was often observed. Such progeny were included with those of the appropriate paternal configuration.

The results presented in Table 5 reveal that in the segregation pattern of four Experimental stocks, 126, 311, 442 and 726 (Group 1) configurations I and II were more or less equally frequent, and all other configurations were rare. In the remaining Experimental stocks 101, 110, 112, 148, 170, 312, 750, 880, 890, 980 and B190 (Group 2) configuration II is much rarer than configuration I. The next most common one is configuration III.

(i) *Group 1 Experimental stocks*

In all four Experimental stocks the relative frequencies within the pairs of configurations I and II, III and IV, V and VI, and VII and VIII were quite similar. In both configurations of each such pair the segregation pattern for the A, T and F elements is the same; they differ only in the segregation of the X element. The similarity in the frequencies of the pairs of segregation classes indicates that the X segregates independently of the other three elements in these four stocks. Thus, if there were determinants for disjunction of the sex chromosomes ('S-determinants') on the X element, they had no matching counterparts to interact with on the T elements of these Experimental stocks.

Because in Experimental stocks 126, 311 and 726 the y^+ marker and an unknown amount of Y and second chromosome heterochromatin have been transferred from the T element to the F element, it could have been argued that the sex-chromosome determinants remained on the new F elements, rather than on the new T elements. The results do not indicate this: both the F elements and the T elements segregate independently of the X elements. Rather, we conclude that in these four Experimental stocks no S-determinants were present, either on the T elements or on the F element.

In all four Experimental stocks the T and F elements segregate regularly from the A element (configurations I and II), although non-disjunction of either the T element (configurations III and IV) or the F element (configurations V and VI) from the A element, is quite common (10–30 %). The regular segregation of the T and F element suggests that all three carry some kind of autosomal disjunction determinants ('H-determinants'). Because configurations I and II, in which the

T and F did not segregate from each other, were more frequent than configurations III to VI, where T segregated from F, we conclude that there was no major disjunctional interaction between these two elements. If there are H-determinants on the autosomal arms, those of the left arm are different from those of the right arm. The observed frequency of non-disjunction of both the T and F elements (configurations VII and VIII) was, however, at least one order of magnitude less frequent than that expected on the basis of independence. This suggests that even though determinants of the left arm do not disjoin from those of the right arm, there is some interaction between the arms. This kind of interaction between arms is also suggested by the frequency of non-disjunction of the uninterrupted, whole chromosome 2, which is at least one order of magnitude below that expected from the independent non-disjunction of the T and F elements (Frost 1961; Falk, unpublished).

The segregation pattern of the control X/Y; A/F/F stock is similar to that of the four group I stocks, as would be expected of this stock with no T(Y;2) element.

It is impossible to tell whether the differences in the relative frequencies of non-disjunction between the four stocks (and between them and the control stock) reflect differences in the strength or number of H-determinants, or whether these are just experimental fluctuations.

(ii) *Group 2 Experimental stocks*

Contrary to what was observed for Group 1 Experimental stocks, here the frequency of configuration II is much rarer than that of configuration I, and also the frequency of configuration IV is rarer than that of configuration III. (It is difficult to say whether configuration V is rarer than VI and whether configuration VIII was rarer than VII, because the error component of these frequencies was considerable.) This means that in these Experimental stocks the X element tends to segregate regularly from the T element. We conclude that the T element of Group 2 Experimental stocks carries S-determinants that interact with S-determinants present on the X element, so as to induce the two to disjoin from each other.

Another feature of the Experimental stocks of Group 2 is that they may be arranged in a series of increasing relative frequencies of configuration III (Table 6).

This series reflects a decreasing dependence of the disjunction of the T element on the A element. Whereas in Experimental stock 880 the T and A elements disjoin more or less regularly from each other (with about 8% non-disjunction), in Experimental stock 148 the T and A elements segregate completely independently of each other. Thus, there remains little doubt that the T element may lose its capacity to disjoin from the A element. In other words, there are H-determinants on chromosome 2 responsible for its disjunction, and these determinants can be lost. Whether these determinants are discrete localizable sites or the effect is continuous and cumulative over a whole section of the chromosome arm, cannot be decided at this stage of the experiments.

It should be noted that in Group 2 as in Group 1 H-determinants that are present on the T element and those present on the F element do not interact

regularly, or if they interact, this does not compete with the interaction of each with the A element. This is most easily seen on comparing configurations I, III and VI, in all of which the X element disjoins from the T element: The combined frequencies of configurations III and VI is less than that of I, i.e. the T and F elements segregate to different poles less frequently than they go to the same pole. Even when T segregates completely independently of the A element, as in Experimental stock 148, configuration VI is still much rarer than configuration I.

Table 6. *Relative frequency of configuration III, in increasing order, of Experimental stocks of group 2*

Experimental stock...	880	750	312	110	890	170	112	101	980	148	B190
Relative frequency of III	0·081	0·25	0·29	0·33	0·34	0·37	0·41	0·64	0·70	1·01	1·16

Table 7. *The ratio of configurations II/I in four Experimental stocks, with and without a free Y chromosome present*

XATF	112	170	312	750
No free-Y	0·031	0·025	0·060	0·038
With free-Y	0·045	0·034	0·071	0·043

In flies of four Experimental stocks of group 2 (112, 170, 312, and 750) a free *Y* chromosome was added to the four X, A, T and F elements. Such flies were mated to Tester stock A females. The more efficiently the free Y competes with the X and T elements for interaction with the S-determinants, the more should the ratio of configuration I to II change towards 3:1. In all four Experimental stocks the proportion of configurations II to I was increased only slightly in the presence of a free *Y* chromosome (Table 7). It must be concluded that X and T elements interacted much more efficiently with each other than any of these interacted with the free *Y* chromosome.

As noted, Experimental stock B190 is an XAT_LT_R stock, originally described by Lindsley *et al.* (1972). The segregation pattern of this Experimental stock suggests that two pairs of homologues disjoin independently of each other: the *X* from the T_L element and the A from the T_R element (because this is an XATT stock the notation of configurations III and IV versus configurations V and VI respectively, is arbitrary). In accordance with the previous considerations we concluded that its T_L element carries S-determinants with practically no H-determinants, and its T_R element carries H-determinants, with no S-determinants. However, the possibility must be kept in mind that S-determinants may be present on both arms of the T element and that these interact.

5. DISCUSSION

In their paper on segmental aneuploidy and the methods for its detection, Lindsley *et al.* (1972) note: 'The recovery of the various zygotic classes is influenced not only by viability but also by the relative frequencies of the various types of

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segregation. Our *impression* is that *roughly* alt > ad1 > 3 : 1 ⩾ ad2 > 1 : 3 > 4 : 0. . . .' The need for a more direct and complete experimental analysis of the segregation patterns in translocation heterozygotes (emphasized by our italicizing) is borne out by the same authors even more emphatically, when they discuss translocations whose breakpoints are in the vicinity of the centromere. They overcome the difficulties by 'assuming that adjacent II, but not adjacent I, disjunction is extremely rare in males but not in females – an assumption that appears to be borne out for the majority of T(*Y*; 2)'s with heterochromatic second chromosome breakpoints (Gethmann, unpublished)'. However, the short note of Gethmann (1974) on the segregational behaviour of Y-2 translocations with breaks near the centromere, does not clarify why this assumption was borne out.

The experimental procedure presented in this paper avoids the need for any *a priori* assumptions on the segregation pattern in the translocation heterozygotes, and allows the detection of gametes produced by all eight meiotic configurations. The price paid for this method in comparison to those that have been applied before is that the relative frequencies are compiled from progeny obtained in different matings. We believe that we have shown that by taking care of reasonable standardization procedures, and by correcting for known differences in segregation patterns of Tester stocks, we obtained results that are repeatable on the one hand and sensitive to differences in segregation patterns on the other. The uncontrolled fluctuations, whether systematic or random, appear to be not more than those usually tolerated in experiments of genetic analysis.

Already fifty years ago Dobzhansky observed that inverted chromosome sections produced qualitatively, though not quantitatively, the same effect on disjunction of translocation chromosomes in males as they did in females. He reached the conclusion that 'the occurrence of crossing over does not determine directly the course taken by the chromosomes at disjunction. Both crossing over and disjunction are determined by a third factor, which is, apparently, *the pairing of the chromosomes at the stages preceding the occurrence of crossing over*' (Dobzhansky, 1933).

Forty-five years later, Sandler & Szauter found that although meiotic mutants may relieve the constraints on crossing over in the euchromatic segment of the fourth chromosome of Drosophila females, they still bring about increase in chromosome non-disjunction of this chromosome. Thus, they conclude that either 'recombination-defective meiotic mutants affect only exchange', and disjunction is affected secondarily, or the mutants 'affect *some single property of meiosis* that is neither recombination nor disjunction, but *that secondarily affects both*' (Sandler & Szauter, 1978).

Novitski & Braver (1954) concluded that they could rationalize their results on crossing over within inversions in Drosophila females with various chromosomal arrangements if they 'simply assumed that the synaptic tendency along the euchromatic length of the chromosomes is very weak and ordinarily becomes manifest after homologous regions have been brought into proximity by stronger *pairing centres located in the heterochromatic regions* adjacent to the centromeres'.

The purpose of this series of studies was (*a*) to examine whether such pairing centers do exist in the basal heterochromatic regions of both the sex chromosomes

and the autosomes of Drosophila, (*b*) to examine whether such regions operate in chromosome disjunction in both females and males, and if they do, whether the same determinants operate in both sexes, and (*c*) to characterize the presumptive disjunction determinants as to their location and function. In the present study we deal with the first and last questions. The remaining question will be dealt with in the next paper of this series (Falk, Rahat & Baker, 1985).

The experimental data presented here support the conclusions that in Drosophila males: (1) H-determinants are present on both the left and right arms of chromosome 2, (2) the determinants on one autosomal arm are qualitatively different from those on the other arm, and (3) there are differences between the potency of disjunction determinants even within a given arm.

It should, however, be kept in mind that for purposes of comparison with segregation patterns in females the autosome in this study (the A element) was multiply inverted. As noted by Dobzhansky (1933), inversions affect disjunction of translocation chromosomes also in males. It cannot be excluded that segregation determinants play a different (and probably a more important) role in segregation of non-inverted autosomes

A reasonable assumption would be that, among the T elements of a given arm, those having less potent H-determinants were broken more distally from the autosomal centromere than those having more H-determinants on that arm. Hence, the breakpoints of the T elements recovered in Experimental stocks B190, 148, 980, 112, and 170 of Group 2, would be the most distal ones from the centromere on the left arm of chromosome 2 (Fig. 8). The breakpoints of the T elements of Experimental stocks 890, 110, 312, and 750 of Group 2 would accordingly be located either still more proximally on the left arm or on the proximal parts of the right arm, as would the breakpoint of Experimental stock 442 of Group 1. Similarly, on this argument the T element of Experimental stock 101 was broken relatively distally on the right arm, while the breakpoint of the T element of Experimental stock 880 was proximally either on the right arm (Fig. 8) or on the left arm. This in turn leads to the conclusion that the T elements of Experimental stocks B190, 148, 980, 170, 112, and 101 have *Y* chromosome centromeres, i.e. the autosomal arms were captured by the *Y* chromosomal segments. For the remaining Experimental stocks it is impossible to conclude whether they have an autosome or a *Y* chromosome centromere, without making further assumptions.

Four of the captured T elements induced by us are marked with y^+ (148, 170, 112, and 101), which means that the break in the *Y* chromosome occurred in the long arm. In only one the break was in the short arm of the *Y* chromosome (the T element of 980 is marked with B^S). Let us assume that in the remaining Experimental stocks too, the majority of the *Y*-chromosome breaks were induced in the long arm rather than in the short arm of the *Y* chromosome. Accordingly, most y^+-marked T elements would have *Y* chromosome centromeres (i.e. the *Y* chromosomes captured the autosomal arm), while most B^S marked T elements would have autosomal centromeres (i.e. the *Y* chromosome capped the autosomal arm). Such an assumption would, however, lead to contradictions with the results

of the segregation experiments. For example, according to this model the T elements of Experimental stocks 312 and 750 were broken in the right arm while the T element of Experimental stock 442 was probably broken in the left arm. Thus, Experimental stock 442 should have at most as many H-determinants as Experimental stocks 312 and 750. But the segregation experiments show unequivocally 442 to have *more* determinants than 312 or 750. Note also that originally, the T element of Experimental stock 311 was the complement of the T element of Experimental stock 312 in the same induced T(Y; 2). They had consequently the same breakpoint. If the left-armed T element of 312 was broken to the right of the centromere, then it should carry at least as many H-determinants as its original complementary right armed T element of Experimental stock 311. The segregation pattern indicates, however, that it carries *fewer* (the original right arm of 311 is now the F element of this Experimental stock). These contradictions led us to reject the model constructed on the assumption that most recovered breaks were induced in the long rather than in the short arm of the *Y* chromosome.

Alternatively the frequencies of T–A disjunction in the segregation experiments reflect the relative position of the breakpoints (i.e. the more regular the disjunction of two elements, the more H-determinants are possessed by the T elements). The distribution of breakpoints obtained on this assumption is schematically presented in Fig. 8. The apparent inconsistency that occurs now, namely that there were fewer (or as many) breakpoints on the long arm of the *Y* chromosome than on the short arm of this chromosome, will be dealt with below.

In this presentation the relative frequencies of configurations I and III were accepted at face value. The presented results do not allow, however, to discriminate between this continuous model and one of a small number of discrete H-determinant sites on the chromosome arms.

(*a*) Only three of the originally five XAFT Experimental stocks were transformed into XATF stocks. One explanation to this could be that the other two Experimental stocks (101 and 880) had too short a left arm to exchange efficiently with the right arm of F(2L). Indeed, the model shows Experimental stocks 126, 311 and 726 to have relatively long left arms, while Experimental stock 101 was concluded to have only the Y^S as its left arm. If it is assumed that the T element of Experimental stock 880 was broken to the left of the autosomal centromere, rather than to the right (and thus carries only a small left arm) it too would be consistent with this interpretation. Alternatively, it could be that only Experimental stocks 126, 311 and 726, but not Experimental stocks 880 and 101, had on their left arms segments that were homologous to those on the right arm of F(2L). That is, the right arm of F(2L), as well as the left arm of these three stocks are mainly of autosomal origin rather than merely of *Y*-chromosome origin (see Fig. 8). If the 'other arms' of the T and F elements of Experimental stocks 126, 311, and 726 of Group 1 had enough in common to interchange, they could also have enough of the same arm's H-determinants in common to interact at disjunction. As could be expected on this assumption, there is more T–F disjunction (more A–T and A–F non-disjunction) in these stocks than in Experimental stocks of Group 2 (compare the relative frequencies of configurations IV–VI of Experimental stocks of Group

1, with those of Group 2 in Table 5). Experimental stock 442 was broken either proximally enough on the left arm or on the right arm, to have similar characteristics.

If this last interpretation is accepted, it would be a strong indication for the presence of H-determinants in the most proximal segments of the chromosome.

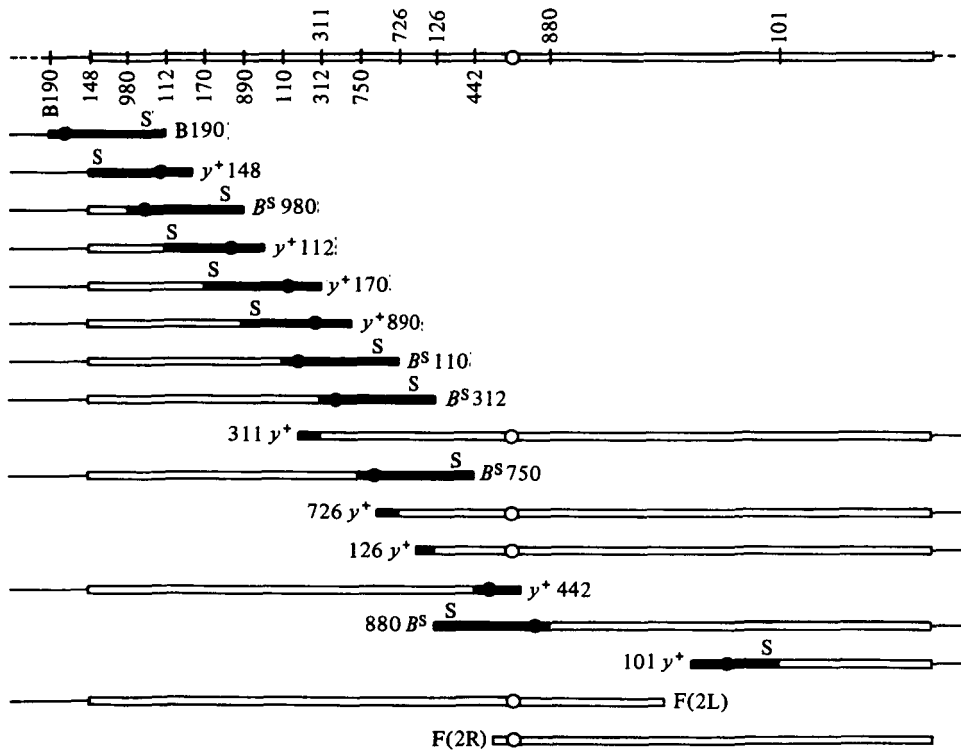

Fig. 8. Schematic representation (not to scale) of the relative positions of breakpoints of the T(*Y*; 2)'s in the basal segments of chromosome 2, as deduced from the segregation patterns of the Experimental stocks (upper line). The numbers of the recovered right arm T elements are given above the line, those of the recovered left arm T elements below the line. For each tested T element the presumed basal autosomal segment with the presumed *Y*-chromosome segment (in black) attached to it, are shown. S denotes the presence of 'S-determinants' on the *Y*-chromosome segment. B^S and y^+ denote the *X*-chromosome markers present on the translocated *Y*-chromosome segment. The T elements of Experimental stocks 126, 311, and 726 are shown before the exchange between the 'other arms' took place.

But even if one ignores it, it would be difficult to 'squeeze' all the disjunction determinants into the proximal euchromatic segments, as demanded by Yamamoto (1979) and Hilliker *et al.* (1982). Let us accept for the argument's sake a more extreme interpretation, namely that the 14 Experimental stocks belong to only two classes: Those with H-determinants in the proximal euchromatin (Experimental stocks 110, 112, 170, 312, 442, 750, and 890 on the left arm, and 126, 311, 726, and 880 on the right arm) and those with no disjunction determinants (Experimental stocks 148, and 980 on the left arm and 101 on the right arm). This

means that 3 of the 13 recovered breakpoints were in the proximal euchromatin and 10 in the basal heterochromatin (Experimental stocks 311 and 312 have one and the same breakpoint). The basal heterochromatin of chromosome 2 represents about 25 % of its physical length, but only 0·1 % of the genetic length of the entire chromosome (Hilliker & Holm, 1975). Lindsley *et al.* (1972) estimated that a deletion of 'three percent of the genome is the upper limit that the fly can tolerate'. Because chromosome 2 comprises about 40 % of the genome, a deletion of 7·5 % of its euchromatin would be the upper limit that a fly can tolerate. This would put the maximum expected ratio of recovered proximal euchromatic breakpoints among the induced T(Y; 2)'s at about 18 %, while even under the extreme interpretation made here it is still about 23 %. From these considerations we conclude that Yamamoto's (1979) and Hilliker *et al.*'s (1982) assertion that there are no autosomal disjunction determinants in the basal heterochromatin of chromosome 2 would be difficult to maintain. At least some H-determinants must be located in the proximal heterochromatin. It is appropriate to mention here that because of the scarcity of genetic markers, and the high redundancy of the DNA in the heterochromatin, it may be difficult to determine directly how many H-determinants are present on the chromosome arm and where exactly they are located. Both Yamamoto (1979) and Hilliker *et al.* (1982), had to resort, like us, to indirect reasoning.

Finally, the sex-chromosome determinants must be considered. Group 2 stocks all carry S-determinants on their T element. With the exception of Experimental stock 880, whose *Y* chromosome could cap as well as capture the autosomal fragment, all are assumed to have captured the autosomal segment. Our model has led us to conclude that there were as many, or even more breakpoints in the short arm of the *Y* chromosome, as in the the long arm, even though the short arm is only about a third as long as the long arm. Hence the segment distal to the S-determinants on the long arm is at most as long as that distal to the S-determinants on the short arm. Or, at least some S-determinants are located on the distal third of the long arm of the $B^{S}Yy^{+}$ chromosome.

Of the four Experimental stocks of Group 1, the T elements of which do not carry significant S-determinants, three breakpoints were apparently located on the short arm of the *Y* chromosome, distal to any possible S-determinants (Experimental stock 126, 311 and 726). The *Y* chromosome of Experimental stock 442 was broken either in the short arm distal to the S-determinants, or on the long arm proximal to the S-determinants. This scarcity of breakpoints proximal to the S-determinants on the long arm, suggests that S-determinants are located also quite proximally on the long arm or on the short arm of the *Y* chromosome.

Taken together, we conclude that there were both proximal S-determinants and distal S-determinants on the long arm of the $B^{S}Yy^{+}$ chromosome. This agrees with the conclusion arrived at earlier: the minute effect that the addition of an unmarked free *Y* chromosome had on the segregation pattern of the Experimental stocks indicated that at least part of the S-determinant activity of the T elements rested within the *X*-chromosome-segment (marked with B^{S}) that had been translocated on to our *Y* chromosome.

In conclusion, our results confirm the presence of S-determinants on the sex

chromosomes, and strongly indicate that at least some autosomal H-determinants present in the proximal heterochromatin of chromosome 2 are involved in the regulation of chromosome segregation in meiosis of Drosophila males.

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