Gross Genetic Dissection and Interaction of the Chromosomal Region 95E;96F of *Drosophila melanogaster*

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Manuscript received February 4, 1989
Accepted for publication July 6, 1989

ABSTRACT

Making use of deficiencies, inversions and translocations, we have genetically dissected the region 95E to 96F of *Drosophila melanogaster*. We localized cytologically the loci *abnormal spindle* (*asp*: 3-85.2: 96A20-25;96B1-10) and $M(3)96C^2$ (96C1;96C5). We have also found several new phenotypes associated with lesions in the 95E to 97B region: (1) *Minute(3)96A* (*M(3)96A*) is a haplo-insufficient phenotype of thin and short bristles presented by individuals deficient for the region 95E6-8;96A1-5. (2) *abdominal-one reduced* (*aor*) shows two different phenotypes associated with the distal breakpoint of $In(3R)Ubx^{7L}$ (89E;96A1-7). One is the increase of the *Ubx* phenotype, but its effect requires the presence of lesions in *Ubx*. The other phenotype is a drastic reduction or disappearance of the first abdominal segment. Both phenotypes might be due to lesions in the same gene. (3) *metaphase arrest* (*mar*) is associated with the breakpoint of the *T(Y;3)B197* (96B1-10) and produces a phenotype typical of mitotic mutants with arrest of the cell cycle during prometaphase or metaphase. There is another region localized in 97B which interacts with *asp*: in a background homozygous for *asp*, three doses of this region enhance the *asp* phenotype.

THE cell division locus *abnormal spindle* (*asp*, RIPOLL *et al.* 1985) resides in one of the rather "unexplored" regions of the Drosophila genome where the available genetic information is scarce. The closest accurately positioned genetic markers are *M(3)95A* (95A1) and *Pr* (96F11). Besides *asp*, we only know of the following loci in the *M(3)95A-Pr* region: *M(3)96C*, located in 96A-C (LINDSLEY *et al.* 1972), *E(spl)*, in 96F11-14 (PREISS, HARTLEY and ARTAVANIS-TSAKONAS 1988), *boss*, in 96F8-11 (REINKE, and ZIPURSKY 1988), *tld* in 96A-C (JURGENS *et al.* 1984), and *crb* in 95E-96A (JURGENS *et al.* 1984). Through complementation analyses with two deficiencies described in this report two other loci have been located in this region: *ash-2* (SHEARN, HERSPERGER and HERSPERGER 1987; SHEARN 1989), and *bam* (A. SPRADLING and D. MCKEARIN, personal communication).

To better localize *asp* and characterize other loci in its vicinity we have roughly dissected the region surrounding *asp*, making use of deficiencies, inversions and translocations as genetic tools. In this report we describe the cytological localization of *M(3)96C* and *asp*, several new phenotypes associated with lesions in the 95E-96F interval, and some genetic interactions between lesions in the region and other genes outside of it. A summary of our results is presented in Figure 1.

MATERIALS AND METHODS

Flies were reared on standard Drosophila medium at 25°. Except when noted, all the mutations and chromosomes are described in LINDSLEY and GRELL (1968), CRAYMER (1984), and LINDSLEY and ZIMM (1985, 1986, 1987). The terminal duplications and interstitial deficiencies were generated following LINDSLEY *et al.* (1972). We have used the rearrangements shown in Table 1. All the translocations are described in LINDSLEY *et al.* (1972).

Tandem duplications were obtained following the method previously described by GRELL (1969). Briefly, X-irradiated *red* females were mated to $M(3)96C^2$ males, and the offspring of this cross was scored for suppression of the *Minute* phenotype. All the dominant suppressors except one were recovered from the sample of eggs laid by irradiated females during the first 72 hours after treatment. With the exception of $Dp(3;3)M96C^{2+}10$, in which the tandemly duplicated fragment is in inverted sequence, all other suppressors turned out to be regular tandem duplications. Their sizes ranged from 5 to *ca.* 450 bands.

Mitotic phenotypes were observed in brains of third instar larvae. Larvae of the appropriate genotype were selected using sex, *yellow*, *red* and *Tubby* as larval markers. Brains were dissected in saline solution (0.7% NaCl) and fixed in 45% acetic acid. After this, they were stained with aceto-orcein, squashed, and observed under a Zeiss Universal Microscope. The mitotic index was quantified following GONZÁLEZ, CASAL and RIPOLL (1988) using the average number of mitotic figures per optical field as a measure. Adult cuticles were prepared by cutting the appropriate pieces under the dissecting microscope. The internal organs were digested with hot 10% KOH and the cuticle washed in alcohol and mounted in Euparal for examination.

RESULTS AND DISCUSSION

The *asp* locus was previously located by meiotic recombination to the right arm of chromosome *3* at

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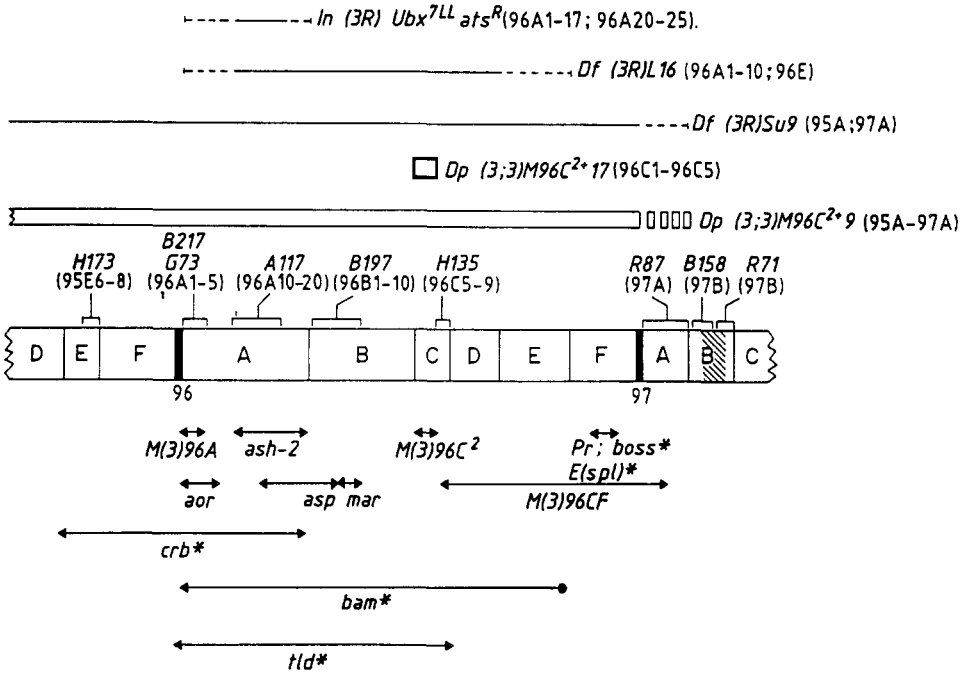

FIGURE 1.—Cytological and genetic analysis of the 95E;97B region with reference to Bridges' map. Deficiences (thin lines) and duplications (open boxes) are shown in the upper part of the scheme. The location of the different phenotypes studied in this work (exceptions are marked with asterisks) are shown below. Breakpoints of the *T(Y;3)*'s employed are represented with brackets on the map. The shadowed region between the B158 and R71 breakpoints is the region that when duplicated enhances the *asp* phenotype.

TABLE 1

Rearrangements used in this work

Rearrangements	Breakpoints	Ref.[a]
T(Y;3)L18	64C	a
T(Y;3)H173	95E6–8	a
T(Y;3)B217	96A1–5	a
T(Y;3)G73	96A1–5	a
T(Y;3)A117	96A10–20	a
T(Y;3)B197	96B1–10	a
T(Y;3)H135	96C5–9	a
T(Y;3)R87	97A	a
T(Y;3)B158	97B	a
T(Y;3)R71	97B	a
T(Y;3)R78	98E	a
T(Y;3)L129	100B–C	a
Df(3R)L16	96A1–10; 96E	e
Df(3R)Su9[b]	95A; 97A	e
Df(3R)D816	97A; 97D–E	b
In(3R)Ubx^{7L}	89E; 96A1–7	c
In(3R+3L)ats	63C; 72E; 84E; 88A; 89C; 96A20–25	d
In(3R)Ubx^{7LL}*ats*^{R}	Deficient for 96A1–7; 96A20–25	e
Df(3R)P9, Ubx^{-}	89E1; 89E4–5	c
Dp(3;1)P115, Ubx^{+}	89B7; 89E6–7	f
Dp(3;3)M96C^{2+}*9*	95B; 97A	e

[a] a, LINDSLEY *et al.* (1972). LINDSLEY *et al.* defined the breakpoints to the level of lettered subdivision. In some instances we have defined the breakpoints more precisely; b, ANDERSON, JURGENS and NUSSLEIN-VOLHARD (1985); c, LEWIS (1978); d, SIERRA and COMENDADOR (1984); e, this work; f, SANCHEZ-HERRERO *et al.* (1985).

[b] HEMIZYGOUS lethal: kept in stock over *Dp(3;3)M96C*$^{2+}$*9*.

84 map units (RIPOLL *et al.* 1985), based on 25 recombinant chromosomes between *ebony* (*e*: 70.7) and *claret* (*ca*: 100.7). According to the meiotic location of *asp*, the closest genetic markers with cytological positions defined to single bands in the giant chromosomes of the salivary glands are *Minute(3)95A* (*M(3)95A*: 79.7, 95A1, BRODERICK and ROBERTS 1982) and *Prickly* (*Pr*:90.0,96F11, our observations). Thirty-three recombinant chromosomes recovered from *red asp/ M(3)95A Pr* females confirmed the location of *asp* within this interval. Other genes cytologically mapped into this region are *Minute(3)96C* (*M(3)96C*:84.5) and *Minute(3)96CF* (*M(3)96CF*:90.2) in 96A-C and 96C-97A, respectively (LINDSLEY *et al.* 1972). The recovery of wild-type chromosomes from *M(3)96C*^{2}*/red asp* females placed *asp* proximal to *M(3)96C*^{2}. A more accurate meiotic localization of *asp* using *bar of chromosome 3* (*bar-3*:79.1) and *taxi* (*tx*:91.0) as flanking markers allowed us to place the gene in position 85.2, based on 64 recombinants in this interval.

Localization of *M(3)96C*^{2}: *M(3)96C*^{2} was located between 96B1-10 and 96C5 using different terminal duplications. Individuals of genotype *M(3)96C*^{2}*/+/ Dp*, where *Dp* stands for the Y^P3^D element of translocations between the *Y* chromosome and the right arm of chromosome *3*, were scored for the bristle phenotype typical of *Minute* mutants. The smallest terminal duplication that rescued the *Minute* phenotype was generated from the translocation *B197* (96B1-10), and the largest terminal duplication that could not rescue the *Minute* phenotype was generated from the translocation *H135* (96C5). This location of *M(3)96C*^{2} was confirmed by: (1) the *Minute* phenotype of individuals heterozygous for the synthetic deficiency generated with the same translocations, and (2) the lethality of this deficiency in *trans* with *M(3)96C*^{2}. These data do

TABLE 2

Cytological breakpoints of tandem duplication covering *M(3)96C²*

Dp(3:3)M96C²⁺	Breakpoints
8	96A20–25; 96E
9	95B; 97A
10	96B; 96D
13	96B1–10; 96E
14	94C7–9; 98A
16	96A1–10; 100F
17	96C1; 96C5
18	96A20–25; 99F1–4
19	94A1–5; 96F
20	91B2–7; 97F
21	94C7–9; 97D

not exclude the possibility of other *Minute* loci in addition to *M(3)96C²* between 96B1-10 and 96C5.

We utilized the haplo-insufficiency of *Minute* phenotypes (SCHULTZ 1929) to select for a collection of duplications that include the wild-type allele of *M(3)96C²* following the procedure of GRELL (1969). The cytology of eleven lines thus generated is presented in Table 2. With the exception of *Dp(3;3)M96C²⁺10*, in which the tandemly duplicated fragment is in inverted sequence, all other suppressors were regular tandem duplications. Their sizes ranged from 5 to *ca.* 450 bands. The smallest duplication, *Dp(3;3)M96C²⁺17*, allowed a more precise localization of *M(3)96C²* to 96C1-5.

Localization of *asp*: Several deficiencies were used to define the cytological position of *asp*.

Df(3R)Su9 (95A;97A) was recovered as a spontaneous event in a *red Dp(3;3)M96C²⁺9/red M(3)96C²* stock (Figure 2). Individuals *red Df(3R)Su9/red asp* or *red Df(3R)Su9/TM6B* do not survive, showing an effective lethal phase close to the end of larval development. Brain neuroblasts from third instar larvae of genotype *red Df(3R)Su9/red asp* show a phenotype similar to that found in *red asp* homozygous larval brains: a high mitotic index although lower than of *asp* homozygous, and extremely condensed X-shaped chromosomes. Contrary to homozygous *asp* larval brains, aneuploid and polyploid cells are rare, and no anaphases are found in *red Df(3R)Su9/red asp*. *Df(3R)Su9/TM6B* larvae have mitotically normal brains.

Df(3R)L16 (96A1-10;96E) was induced with X-ray treatment of a *red* strain, the same strain from which the original *asp* allele had been previously isolated (RIPOLL *et al.* 1985). Individuals *Df(3R)L16/+* or *Df(3R)L16/asp* have a *Minute* phenotype. This deficiency is practically lethal in *trans* with *asp*: relative to their *Df(3R)L16/TM6B* sibs, the viability of *Df(3R)L16 asp* individuals is reduced to less than 1%, the rest dying as third instar larvae. The few individuals of this genotype that reach adulthood have an increase in the cuticular defects (rough reduced eyes, nicked wings, etc.) similar to that shown by homozygous *asp* flies, and male and female sterility. The mitotic phenotype shown by late larval brains is identical to that shown by *Df(3R)Su9/asp* individuals. *Df(3R)L16/TM6B* larval brains present normal mitotic figures.

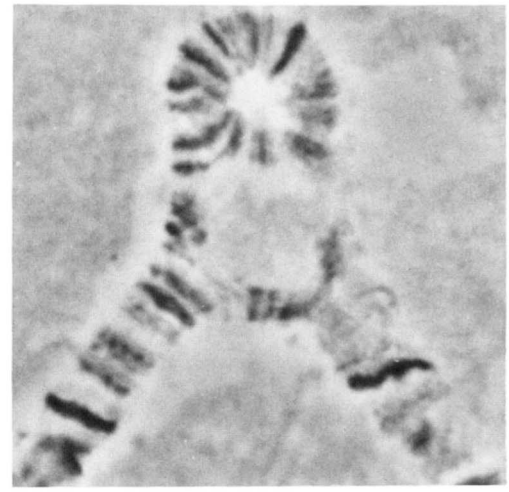

FIGURE 2.—Polytenic chromosomes of a *Df(3R)Su9/Dp(3;3)M96C²⁺9* larva.

At first sight the phenotype of individuals *asp* over a deficiency for the region seems to be less extreme than the homozygous phenotype. However, based on the following considerations, we believe that the hemizygous phenotype is closer to the one expected from total lack of function of the gene, indicating that *asp* is a hypomorphic allele. In the absence of the function of a gene needed for spindle function, cells should be able to divide until the wild-type gene product provided to the developing embryo by the maternal genome is exhausted. One might expect that cell division would be arrested once these maternally derived products are consumed or degraded, resulting in a population of diploid cells arrested at the stage of mitosis when the gene product is first needed. In the presence of a hypomorphic allele a fraction of the cells might be able to continue dividing while making mistakes in chromosome segregation, resulting in high frequencies of aneuploid and polyploid cells, as observed in homozygous *asp* larval brains. Additional experimental evidence supporting this interpretation, based on the analysis of several *asp* alleles will be published elsewhere.

In(3R)Ubx⁷ᴸᴸats^R was obtained by recombination between *In(3R)Ubx⁷ᴸ* (89E;96A1-7) and the inversion in 3R (89C;96A20-25) carried by *In(3L + 3R)ats*. The recombinant chromosome is duplicated for 89C;89E and deficient for the region 96A1-7;96A20-25. It is heterozygous viable, homozygous lethal, and fully complements all the phenotypic traits shown by *asp* homozygous individuals, thus narrowing down the

proximal limit of the location of *asp* to 96A20-25.

To localize *asp* more precisely we used translocations between chromosomes *Y* and *3* with breakpoints in the region (Table 1) to construct individuals of genotype *Df/red asp* (where *Df* is defined by the combination of the $3^{P}Y^{D}$ element of the *T(Y;3)* with the more proximal breakpoint and the $Y^{P}3^{D}$ element of the *T(Y;3)* with the more distal breakpoint; see LINDSLEY *et al.* 1972). Analysis of these individuals made it possible to correlate the larval brain, cuticular and testicular phenotypes of *asp* with the region spanning the interval from 96A10-20—the breakpoint of *T(Y;3)A117*—to 96B1-10, the breakpoint of *T(Y;3)B197*. The mitotic phenotype of brains of genotype $Df(3R)A117^{P}B197^{D}/red\ asp$ is identical to that of *Df(3R)Su9/red asp* or *Df(3R)L16/red asp*. All other interstitial deficiencies fully complemented all traits associated with the *asp* phenotype.

We confirmed the location of *asp* by phenotypic analysis of homozygous *asp* individuals bearing terminal duplications. We constructed larvae of genotype *red asp/red asp/Dp*. While the phenotype of *red asp/red asp/A117*^{D} larval brains is wild type, that of *red asp/red asp/B197*^{D} larvae is mutant, indicating that *asp* lies in the 96A20-25;96B1-10 interval. Surprisingly, the phenotype of *red asp/red asp/B197*^{D} is not the same as the phenotype of *red asp/red asp* individuals. Instead, the larval brain phenotype of *red asp/red asp/B197*^{D} individuals resemble that of *red Dfasp*$^{-}$*/red asp* larvae: a high mitotic index, anaphases are rare and the frequency of aneuploid or polyploid cells is reduced. Most metaphases show the X-shaped chromosomal phenotype typical of mitotic arrest. The enhancement of the *asp* phenotype is accompanied by a decrease in the relative frequency of aneuploid cells. The duplication derived from *B197*^{D} appears to act as an enhancer of *asp*.

To identify the locus responsible for the enhancement of the *asp* phenotype we have constructed larvae of genotype *red asp/red asp/Dp* using a series of translocations with breakpoints distal to 96B1-10. The mitotic index of larvae of this genotype was measured and compared with the mitotic index found in their sibs of genotype *red asp/red asp*. Data from these experiments are presented in Table 3. As can be seen, duplications proximal to *T(Y;3)B158* (97B) enhance the *asp* phenotype, while duplications broken distally to *T(Y;3)R71* (97B) do not, indicating that the locus responsible for the enhancement resides within 97B (Figure 1). These results suggest that the breakpoint of *T(Y;3)B158* must be proximal to that of *T(Y;3)R71*. That the enhancement is not due to nonspecific effects of duplications is shown by adding a duplication in the other chromosome arm of size equivalent to that of *T(Y;3)B197*, the largest duplication used not covering *asp* (see *T(Y;3)L18* in Table 3).

TABLE 3

Mitotic index and degree of aneuploidy in male larvae of genotype *red asp/red asp/Y*^{P}*3*^{D} (genotype 1) and their sisters of genotype *red asp/red asp* (genotype 2)

		Mitotic index			Percent aneuploidy		
$Y^{P}3^{D}$	Breakpoint	1 *asp/asp/Dp*	2 *asp/asp*	Ratio, ½	1 *asp/asp/Dp*	2 *asp/asp*	Ratio, ½
B197	96B	1.65	2.91	0.57	5.9	11.7	0.50
H135	96C	1.77	3.00	0.59	9.2	15.0	0.62
B158	97B	1.98	2.94	0.67	6.5	16.4	0.40
R71	97B	2.69	2.15	1.25	12.8	12.9	0.99
R78	98E	3.36	3.31	1.01	12.3	13.1	0.95
L129	100BC	2.79	2.55	1.09	8.6	8.1	1.06
L18	64C	2.30	2.00	1.15	9.3	10.6	0.90

Other loci in the region: We have found several new phenotypes associated with lesions in the 95E to 97B region.

Minute(3)96A (*M(3)96A*): This is a haplo-insufficient phenotype shown by individuals heterozygous for the deficiency synthesized by combining *T(Y;3)H173* (95E6-8) and *T(Y;3)G73* (96A1-5). These flies have thin bristles with full penetrance and expressivity. Since LINDSLEY *et al.* (1972) did not find this phenotype using the synthetic deficiency between the breakpoints of *T(Y;3)H173* and *T(Y;3)B217* (96A1-5), we suggest that *T(Y;3)B217* is broken proximal to *T(Y;3)G73*, and that *M(3)96A* is located between the breakpoints of these translocations, although this possibility has not been confirmed. We have named this locus as *Minute* following the criterium used by LINDSLEY *et al.* (1972): haploinsufficiency of the thin bristles phenotype. However, this locus does not have some of the more general phenotypes of *Minutes*, namely decreased viability and developmental delay.

abdominal-one reduced (*aor*): Adult flies heterozygous for $In(3R)Ubx^{7L}$ and a deficiency for 96A (genotypes 2 and 3 in Table 4) show two separable phenotypes in different combinations, both apparently related to the function(s) of the bithorax complex (LEWIS 1978). One consists of a drastic reduction or disappearance of the first abdominal segment of the imago (Figure 3), a phenotype resembling that of *bithoraxoid* (*bxd*: 58.8) adult individuals. This seems to be a recessive loss of function phenotype. As in *bxd* homozygous individuals, third instar larvae of *aor* genotype have normal histoblasts in A1. The other phenotype is an enhancement of the *Ubx* transformation of haltere into wing produced by the inverted chromosome (Figure 3). Genetically, it behaves as a dominant loss of function phenotype. This enhancement does not require for its expression the presence of $In(3R)Ubx^{7L}$ in the genome: it results from the interaction of lesions in *Ubx* with the haplo-insufficiency of the region 96A (genotypes 5, 7, 8 and 10 in Table 4). Haploidy for this region alone, in the pres-

TABLE 4

Phenotypic variations affecting the first abdominal segment (Ab1) and the haltere of different genetic combination

	Genotype	Ab1	Haltere
1	*In(3R)Ubx^{7L}/+*	Wild type	*Ubx*
2	*In(3R)Ubx^{7L}/Df(3R)G73^{P}A117^{D}*	Absent	Strong *Ubx*
3	*In(3R)Ubx^{7L}/Df(3R)L16*	Absent	Strong *Ubx*
4	*Df(3R)G73^{P}*A117^{D}/+	Wild type	Wild type
5	*Df(3R)G73^{P}*A117^{D}/TM2	Wild type	Strong *Ubx*
6	*Df(3R)L16/+*	Wild type	Wild type
7	*Df(3R)L16/TM2*	Wild type	Strong *Ubx*
8	*Df(3R)L16/Df(3R)P9, Ubx^{-}*	Wild type	Strong *Ubx*
9	*In(3R)Ubx^{7LL}ats^{R}/+*	Wild type	Wild type
10	*In(3R)Ubx^{7LL}ats^{R}/TM2*	Wild type	Strong *Ubx*
11	*Dp(3;1)P115, Ubx^{+}/In(3R)Ubx^{7L}/Df(3R)L16*	Absent	Wild type
12	*In(3R)Ubx^{7L}/Dp(3;3)M96C^{2+}9*	Wild type	*Ubx*[a]
13	*Dp(3;3)M96C^{2+}9/TM2*	Wild type	*Ubx*

[a] This phenotype is slightly weaker than what is found in *In(3R)Ubx^{7L}/+* or *Dp(3;3)M96C^{2+}9/TM2*. Individuals with *Ubx* phenotype are those with phenotypes as strong or weaker than those of genotype 1 (Figure 3a). Strong *Ubx* phenotypes refer to individuals with haltere into wing transformations at least as strong as the one shown by individuals of genotype 3 (Figure 3b).

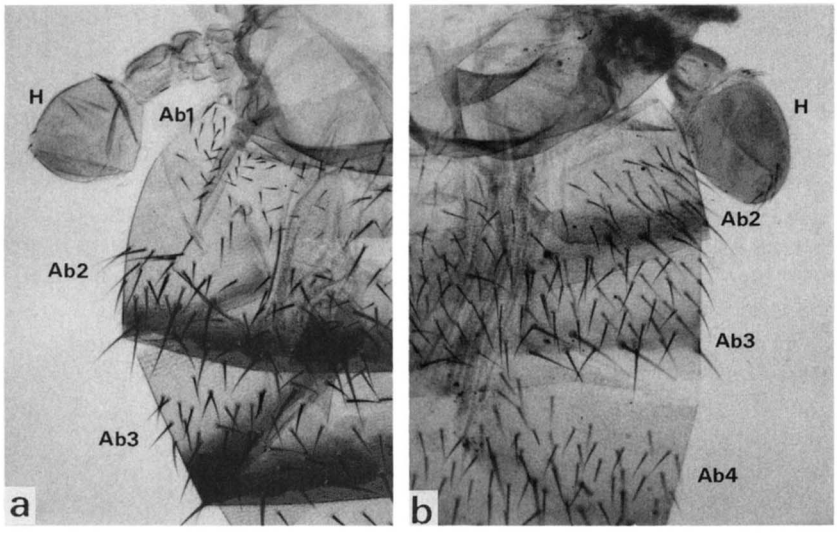

FIGURE 3.—and b) Phenotypical characteristics of *In(3R)Ubx^{7L}/+*, and *In(3R)Ubx^{7L}/Df(3R)-L16* individuals. *a*, *In(3R)Ubx^{7L}/+*; the haltere (H) presents a typical *Ubx* transformations showing several wing-type bristles; the first abdominal segment (Ab1) is characterized by shorter bristles (compare with Ab2 and Ab3). b, *In(3R)Ubx^{7L}/Df(3R)L16* shows the two *aor* phenotypes; the *Ubx* transformation of the haltere is enhanced; the haltere is larger, and has more wing-type bristles and hairs than *In(3R)Ubx^{7L}/+*; the first abdominal segment is absent. Note that the bristles are thinner than in *In(3R)Ubx^{7L}/+* because of the haploinsufficiency of *M(3)96C^{2}* uncovered by *Df(3R)L16*.

ence of two wild-type doses of the bithorax complex, is unable to give rise to the enhancement of the *Ubx* transformation (genotypes 4, 6, 9 and 11 in Table 4). In contrast, the reduction of the first abdominal segment is a recessive phenotypic trait independent of mutations in *Ubx*. The two phenotypes (reduction of the first abdominal segment, and enhancement of the *Ubx* transformation) could result from lesions in the same gene. If this is the case, the *Ubx* transformation produced by heterozygosity of the inverted chromosome should be due to the enhancement of a weaker *Ubx* phenotype caused by the proximal breakpoint in 89E. That is to say, in the presence of an extra dose of the wild type region covering 96A (*In(3R)Ubx^{7L}/Dp(3;3)M96C^{2+}9*) the *Ubx* phenotype should be weaker than when there is only one wild-type dose of 96A (*In(3R)Ubx^{7L}/+*). This is in fact what happens in genotype 12 in Table 4: addition of an extra dose of region 96A does partially suppress the *Ubx* phenotype shown by *In(3R)Ubx^{7L}/+* individuals, indicating that the inverted chromosome is also defective for the function that when mutated enhances the *Ubx* phenotype.

There is another locus in the region 96A, *ash-2* (SHEARN, HERSPERGER and HERSPERGER 1987), that gives rise to homeotic transformations when mutated. This locus is uncovered by *Df(3R)L16* (96A1-10; 96E) (SHEARN 1989), and by *In(3R)Ubx^{7LL}ats^{R}* (deficient from 96A1-7 to 96A20-25). *aor* and *ash-2* do not appear to be allelic. While *In(3R)Ubx^{7L}/ash-2* individuals do not show the *ash-2* phenotype, in contrast *In(3R)Ubx^{7LL}ats^{R}/ash-2* are lethal. These results localize *ash-2* distal to *aor*.

metaphase arrest (*mar*): Individuals of genotype *T(Y;3)B197/Df(3R)L16* die as late third instar larvae with few escapers. The third instar larval brains have a phenotype typical of mitotic mutants leading to arrest of the cell cycle during prometaphase or metaphase. In the absence of any treatment, such as culturing in colchicine or hypotonic shock, arrested cells have

highly condensed X-shaped chromosomes, the mitotic index is twice that of wild type, and there is a decrease in the number of cells in anaphase (30-fold reduction of the ratio of anaphases to metaphases). Culturing these brains in colchicine for 90 min prior to fixation does not affect the phenotype either qualitatively or quantitatively, with the expected exception of the disappearance of the few anaphases found without treatment. Practically all the cells are diploid, with only about 1% of the cells being tetraploid. Although the *mar* phenotype closely resembles the phenotype shown by hemizygous *asp* larval brains, it does not result from either a breakpoint in *asp* or to variegation of *asp* due to the *Y*-heterochromatin attached to the autosomal breakpoint of *T(Y;3)B197*, because *T(Y;3)B197/red asp* individuals are viable and the brains of larvae of this genotype are completely normal. Therefore *mar* is a recessive lesion in a gene needed for normal mitosis, associated with the breakpoint of *T(Y;3)B197* (96B1-10).

The similarity in mitotic defects between *asp* and *mar* suggests that *mar* might be involved in spindle function as *asp* is (RIPOLL *et al.* 1985). Another microtubule-related function (*ms(3)nc32*) is likely to reside in the same region based on its meiotic location between *bar-3* and *Pr* (FULLER 1986). *ms(3)nc32* is thought to be defective in some microtubular component since it interacts with mutations in *B2t*, the gene coding for β_2-tubulin (RAFF and FULLER 1984; FULLER 1986). Mutations in *ms(3)nc32* fail to complement *B2t* alleles for male sterility. Neither *asp* nor *T(Y;3)B197* show this interaction with *B2t* alleles (our unpublished observations). The proximity of *asp* and *mar*, plus the possibility that *ms(3)nc32* being in their vincinity, provides tantalizing grounds to speculate about the possible existence of a cluster of cell-division-related genes in the region.

T(Y;3)B197 has a curious maternal effect: females of *T(Y;3)A117/TM6* genotype were crossed to *T(Y;3)B197/TM6B* males. The viability of the resulting interstitial deficiency-bearing individuals was 53% that of those carrying the complementary duplication (89 *vs.* 169). In the reciprocal cross, this ratio was 19% (74 *vs.* 383). Since the viability of *T(Y;3)B197/TM6B* (or *T(Y;3)B197/TM6*) individuals does not change depending on the genotype of the mother, the decrease in viability described above must be due to the interaction of the maternal effect of the breakpoint of *T(Y;3)B197* (defining *mar*) with the zygotic effect of some locus, possibly *asp*, included in the interstitial deficiency. The viability of *T(Y;3)B197/red asp* individuals decreases a maximum of 15% when the translocation is introduced via the female, relative to the viability when the translocation is introduced via the male. Since *asp* is a hypomorphical allele, this decrease in viability suggests, but does not prove, the involvement of *asp* as the zygotic component of the interaction. The maternal effect shown by *T(Y;3)B197* is not unusual among mutants altering cell division, and the presence of maternally provided products has been used to explain why many mitotic mutants are able to survive until late stages of larval development (GATTI *et al.* 1983; RIPOLL, CASAL and GONZALEZ 1987).

The phenotype associated with other translocation breakpoints was studied by constructing individuals heterozygous for each translocation and *Df(3R)L16*. In one case—*T(Y;3)A117* (96A10-20)—the individuals died as late larvae and their brains did not show any mitotic abnormalities. Individuals of genotype *T(Y;3)H135/Df(3R)L16* are viable and morphologically normal. This is not surprising because it is known that not all breakpoints affect essential functions (LEFEVRE 1981).

Finally, we know of at least one locus in the region uncovered by *Df(3R)L16* that is essential for male and female fertility but not for viability (*bag of marbles*; *bam*, A. SPRADLING and D. MCKEARIN, personal communication).

We are indebted to J. CASANOVA, and J. BOTAS for their help in part of this work. We thank E. LEWIS, L. SIERRA, M. COMENDADOR, A. SHEARN, and Bowling Green Stock Center for providing us with some fly stocks, and M. CARMENA, A. BUSTURIA and M. SCOTT for comments on the manuscript. We are also obliged to M. FULLER for her editorial efforts, and the two anonymous reviewers for very useful comments. This work was supported by grants from Dirección General de Investigación Cientifica y Técnica (PR84-0178-C05-04), and by institutional grant from Fondo de Investigaciones Sanitarias and Ramón Areces Foundation.

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Communicating editor: M. T. FULLER