The Meiotic Behavior of an Inversion in *Caenorhabditis elegans*

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ABSTRACT

The rearrangement *hln1(I)* was isolated as a crossover suppressor for the right end of linkage group (LG) I. By inducing genetic markers on this crossover suppressor and establishing the gene order in the homozygote, *hln1(I)* was demonstrated to be the first genetically proven inversion in *Caenorhabditis elegans*. *hln1(I)* extensively suppresses recombination in heterozygotes in the right arm of chromosome I from unc-75 to unc-54. This suppression is associated with enhancement of recombination in other regions of the chromosome. The enhancement observed maintains the normal distribution of events but does not extend to other chromosomes. The genetic distance of chromosome I in inversion heterozygotes approaches 50 map units (m.u.), approximately equal to one chiasma per meiosis. This value is maintained in *hln1(I)/szTI(I;X)* heterozygotes indicating that small homologous regions can pair and recombine efficiently. *hln1(I)/hT2(I;III)* heterozygotes show no unaltered homologous regions and segregate randomly, suggesting the importance of chiasma formation in proper segregation of chromosomes. The genetic distance of chromosome I in these heterozygotes is less than 1 m.u., indicating that crossing over can be suppressed along an entire chromosome. Since one of our goals was to develop an efficient balancer for the right end of LG I, the effectiveness of *hln1(I)* as a balancer was tested by isolating and maintaining lethal mutations. The meiotic behavior of *hln1(I)* is consistent with other genetic and cytogenetic data suggesting the meiotic chromosomes are monocentric. Rare recombinants bearing duplications and deficiencies of chromosome I were recovered from *hln1(I)* heterozygotes, leading to the proposal the inversion was paracentric.

CHROMOSOME rearrangements which result in crossover suppression are useful for a wide range of genetic experiments including the dissection of chromosomal features responsible for meiotic behaviour. An understanding of the mechanisms responsible for the elimination of meiotic events in the presence of the rearrangement can lead to the discovery and description of sites necessary for recognition and synopsis of homologs, meiotic exchange, and subsequent disjunction. For example, studies of translocations in *Caenorhabditis elegans* have led to the proposal that each chromosome contains a single region necessary for homolog recognition and pairing (ROSENBLUTH and BAILLIE 1981; McKIM, HOWELL and ROSE 1988; reviewed by ROSE and MCKIM 1992). Translocations are the major class of dominant crossover suppressors in *C. elegans* (HERMAN 1978; ROSENBLUTH and BAILLIE 1981; HERMAN, KARI and HARTMAN 1982; FERGUSON and HORVITZ 1985; CLARK et al. 1988; MCKIM, HOWELL and ROSE 1988), although intrachromosomal crossover suppressors have also been described (HERMAN 1978; ANDERSON and BRENNER 1984; ROSENBLUTH, JOHNSEN and BAILLIE 1990). For example, deletions of the chromosome V end that does not contain the region necessary for homolog recognition were found to suppress recombination for several map units beyond the breakpoint of the deletion (ROSENBLUTH, JOHNSEN and BAILLIE 1990). The authors proposed that the deletions eliminated sites required for meiotic synopsis which occurs after homolog recognition has taken place. Insertional duplications have a polar effect on recombination (HERMAN, ALBERTSON and BRENNER 1976; MCKIM 1992; reviewed by ROSE and MCKIM 1992). The intrachromosomal suppressor *mnC1(II)* has been used to balance a large region of chromosome II (HERMAN 1978). The meiotic properties of *mnC1(II)* are compatible with a transposition, an inversion, or a homozygous viable deletion. Although *mnC1(II)* is widely believed to be an inversion, no reversal of gene order has been demonstrated.

In this paper we report the first genetically proven inversion in *C. elegans*. Since this inversion, *hln1(I)*, suppresses crossing over in a region not previously balanced by translocations, it is representative of a new class of balancers for the genome. Furthermore, we have characterized the meiotic behavior of *hln1(I)* with respect to homolog recognition and the centromeric behavior of chromosome I.

MATERIALS AND METHODS

**General methods:** *C. elegans* populations consist largely of self-fertilizing hermaphrodites (5AA:XX). Males (5AA:YO) arise spontaneously as a result of X chromosome nondisjunction (HODGKIN, HORVITZ and BRENNER 1979).
and were maintained by mating to hermaphrodites. Wild-type and mutant strains were maintained and mated on petri plates containing nematode growth medium (NGM) and streaked with *Escherichia coli* strain OP50 (Brenner 1974). All strains were mutagenized carried out as outlined (McKim et al. 1988). The wild-type strain N2 and most mutant strains of *C. elegans* var. Bristol used in this study were obtained from D. L. BAILLIE at Simon Fraser University, British Columbia, or from the Caenorhabditis Genetics Center at the University of Missouri, Columbia. RW3072 was supplied by R. W. WATERSTON at Washington University School of Medicine, Missouri. The following genetic markers were used:

- **LG I:** bli-3(e379); unc-11(e47); dpy-5(e61); bli-4(e373); unc-29(e193); lin-1(n389); unc-75(e950); unc-73(h1041); unc-73(h1042); unc-101(m1); unc-39(e261); lev-11(a12); let-49(st44); unc-54(e190); unc-54(st60); unc-54(h1040); let-50(st33).
- **LG III:** dpy-18(e364); unc-36(e251).
- **LG V:** unc-42(e270); dpy-11(e224); him-5(e1467).
- **LG X:** lon-2(e678); unc-1(e719); dpy-3(e277).

The locations of some genes on chromosome I are shown in Figures 1 and 2 (see RESULTS). The following translocations were used in this study: *stT(I;X)* (Fodor and DEAK 1985; McKim, Howell and Rose 1988), and *hZnl(I;III)* (K. S. McKim, K. Peters and A. M. Rose, unpublished).

- **stT(I;X):** is inviable as a homozygote and is marked with the lon-2 mutation on *I* (K). *hZnl(I;III)* is viable as a homozygote and is marked with *bli-4* on *III*. C. elegans nomenclature of genes and alleles conforms to the system outlined by Horvitz et al. (1979). The nomenclature of rearrangements has undergone some modification since that time. Mutations on translocation chromosomes (T) are shown in square brackets (McKim, Howell and Rose 1988) and the formal name is used when discussing both components of the translocation (e.g., *stT(I;X)*). When discussing the individual component chromosomes of a translocation, the nomenclature describes the segregational properties of the new chromosomes. The translocation *hZnl(I;III)* is comprised of two chromosomes; *hZnl(I;III)* (of structure *I*), which segregates from chromosome I and *hZnl(I;III)* (of structure *II*), which segregates from chromosome III. Similarly, *stT(I;X)* is comprised of stT(I;X) (of structure *I*X*), which segregates from chromosome I and stT(I;XX) (of structure *I*X), which segregates from the X chromosome. Inversions in *C. elegans* are written In (Horvitz et al. 1979). Mutations on inversion chromosomes are shown in square brackets (e.g., *hln(I)[dpy-5 unc-54]*), similar to the system in use for translocations. The nomenclature does not necessarily provide information on gene order and does not implicate the marker in the rearrangement, indicating only that the mutations are linked to the inversion.

### General recombination mapping

**Recombination frequency in the hermaphrodite were measured by scoring the number of recombinant progeny of a cis-heterozygote under the conditions described by Rose and BAILLIE (1979).** The recombination frequency (p) between two genetic markers was calculated using the formula

\[ p = 1 - \left(1 - R \right)^n \]

where *R* was the number of visible recombinant individuals divided by the total progeny number (Brenner 1974). Since the double homozygote class was not scored because of its reduced viability, the total progeny number was estimated by \(n\) (number of wild-type progeny + one recombinant class). In some intervals, recovery of only one recombinant class was possible. In these instances, \(R = 2 \times (n)\) recombinant class divided by the total progeny number. In experiments using *bli-3* as a marker, *Bli-3* recombinants were scored as wild types and later subtracted because Bli-3 penetrance is low. The 95% confidence intervals were estimated using the statistics of Crow and Gardner (1959). In the event that the number of recombinants exceeded 300, confidence intervals were approximated using the equation

\[ n = R/2R \]

where *x* is the number of recombinants, *n* divided by the number of wild types plus recombinants, and *y* is equal to 1 − *x*.

### Isolation of hln(I)

**N2 males were treated with 1500 rads of \(\gamma\)-radiation (Rosenthal, Cuddeford and BAILLIE 1985) and mated to unc-101(m1) unc-54(e190) homozygotes.** unc-101 unc-54/+ + hermaphrodites resulting from this mating were individually plated and their progeny screened for the absence of Unc-101 recombinants. In total, 900 chromosomes were screened and one isolate recovered that suppressed crossing over.

### Egg hatching frequency

Hermaphrodites of the genotype unc-101 lev-11/hln(I)[+/+], hln(I)/hln(I), hDp131/unc-101 leu-11 or hDp131/unc-101 unc-54 were individually plated and allowed to lay eggs for two 10–12 hr periods. The hermaphrodites were then transferred and the eggs remaining on the plate counted. All resulting progeny were counted 3 days later.

### Induction of genetic markers on hln(I)

**hln(I) homozygous males were treated with 25 mm ethyl methanesulfonate (EMS) (Rosenthal, Cuddeford and BAILLIE 1985) using the procedure described by Brenner (1974).** The mutantized males were then mated to unc-75(e950) unc-101(m1) homzygotes for 24 hr. These hermaphrodites were individually plated and their progeny screened for the presence of Unc-75 individuals. Since the unc-75 unc-101 interval is located in the crossover suppressed region of hln(I) (heterozygotes), any Unc-75 individuals recovered were expected to be the result of an induction of a new mutation on the hln(I) chromosome. A total of 18,700 chromosomes were screened and two mutations were recovered; unc-75(h1041), and unc-75(h1042). Both new unc-75 alleles were lethal as homozygotes and were maintained as heterozygotes. Both new alleles produced the Unc-75 visible phenotype when crossed to males heterozygous for unc-75(e950). To induce an unc-54 mutation on the hln(I) chromosome, hln(I) males mutagenized in the procedure described above were mated to lev-11 let-49 +/+ + unc-54 let-50 (RW3072) hermaphrodites for 24 hr. These hermaphrodites were then individually plated, and their progeny screened for Unc-54 individuals. A total of 5200 chromosomes were screened, of which one-half are heterozygous with the unc-54 let-50 chromosome. Some Unc-54 isolates could arise from recombination between unc-54 and let-50 in the parental strain. To test if the new unc-54 mutations were linked to hln(I), the progeny of putative hln(I)[+/+ unc-54]/unc-101 leu-11 + hermaphrodites were screened for the presence of Unc-101 and Leu-11 recombinants. One strain, KR2151 exhibited complete recombination suppression indicating the new mutation, unc-54(h1040), was linked to hln(I).

### Recombination in hln(I)

**Hermaphrodites were examined in hln(I) homzygotes in three intervals; dpy-5 unc-75, dpy-5 unc-29, and dpy-5 unc-54. To examine crossing over in dpy-5 unc-75, Dpy-5 recombinants were picked from amongst the progeny of hln(I)[+/+ unc-75 +/+ hln(I)]/dpy-5, and dpy-5 recombinants and mated to unc-75(h950)/+ males to confirm the presence of unc-75(h950) or h1042). Unc-75 progeny resulting from this cross were then mated to dpy-5 unc-75(e950)/+ + males to ensure the dpy-5 mutation was still present. The resulting hln(I)[+/+ dpy-5 unc-75]/dpy-5 unc-75(e950) progeny were examined in hln(I)[+/+ unc-54]/+ + males and a fraction of the wild-type individuals resulting from this cross were of the desired genotype hln(I)[+/+ unc-54]/hln(I)[+/+ unc-54 +].**
experiment also confirmed the gene order in \( hZnl(I) \) was 
\( dpy-5 \) unc-54 unc-75 (see RESULTS). Knowing the map distance 
between \( dpy-5 \) and unc-54 in \( hln(I) \) homozygotes (see 
Table 3), recombination was measured in the unc-54 unc-75 
interval in the same heterozygotes used in the three-factor 
experiment using the formula:

\[
p = 9 - \frac{(81 - 20(2D + W)(9D - W))/(D + W)^2}{10(2D + W)/(D + W)}
\]

where \( D \) is the number of \( Dpy-5 \) recombinants and \( W \) is the 
number of wild-type progeny. Since \( dpy-5 \) and unc-29 are 
outside the boundary of \( hln(I) \) crossover suppression, the 
mutations can be crossed onto the inversion chromosome. 
Since \( hln(I) \) is viable as a homozygote, recombination was 
measured in \( hln(I)[+ +]/hln(I)[dpy-5 \ unc-29] \) and 
\( hln(I)[+ +]/hln(I)[dpy-5 \ unc-54] \) heterozygotes using the 
general mapping methods described above.

**Analysis of recombinants from \( hln(I) \) heterozygotes:**
Four rare recombinants falling into two classes, duplications 
and deficiencies, were recovered from \( hln(I) \) heterozygotes. 
The deficiencies \( hDf11 \) and \( hDf12 \) were recovered from 
\( hln(I)[+ +]/unc-101 \ unc-54 \) and \( hln(I)[+ +]/unc-75 \ unc-101 
\) heterozygotes respectively. The duplications \( hDp113 \) and 
\( hDp132 \) were both recovered from \( hln(I)[+ +]/unc-101 \ lev-11 
\) heterozygotes, based on the Lev-11 visible phenotype. 
These duplications, were mapped with respect to 
visible markers. For example, markers inside the region of 
\( hln(I) \) crossover suppression were tested by mating males 
of the genotype \( unc-75 \ unc-29 \) to \( hDp113/unc-75 \ unc-101 \) to 
\( hDp113/unc-101 \ lev-11 \) hermaphrodites. A fraction of the wild-type progeny 
from the cross were of the desired genotype \( hDp11/unc-75 \ unc-101 \) 
\(+ +/unc-101 \ lev-11 \). Upon examining the 
progeny of such individuals, Unc-75 individuals were 
observed if the duplication did not carry unc-75(+). In 
the event the duplication did carry unc-75(+), no Unc-75 
individuals were observed. A similar procedure was followed 
for markers outside the region of crossover suppression with 
the exception that males heterozygous for a wild-type rather 
than a \( hln(I) \) chromosome were used. To determine if the 
duplications also carried unc-54(+), \( hln(I)[+ +]/unc-75 \ unc-54 
\) males were mated to \( hDp11/unc-75 \ unc-101 \) heterozygotes. 
A number of the progeny resulting from this mating 
were of the genotype \( hDp11/unc-75 \ unc-101 \) unc-75 \ unc-54 
\(+ +/unc-101 \ unc-75 \ unc-54 \). Since Unc-75 Unc-54 
independent individuals are similar in phenotype to Unc-54 individuals, several wild type progeny 
from the latter heterozygote were plated and their progeny 
examined. If the duplication carried unc-54(+), a fraction 
of these individuals would be of the genotype \( hDp11/unc-75 \ unc-54 
\). The deficiencies \( hDf11 \) and \( hDf12 \) were complementation 
tested with several visible markers by mating \( hDf11/unc-101 \ unc-54 \) 
and \( hDf12/unc-75 \ unc-101 \) hermaphrodites to either 
\( unc-x+ \) or \( lev-11+/+ \) males. The \( F1 \) progeny resulting from 
this mating were screened for both males and 
hermaphrodites Unc-x or Lev-11 in phenotype, the presence of which 
indicated the deficiency did not carry either \( unc-x+ \) or \( lev-11+/+ \).

**Interaction of \( hln(I) \) with \( stT1(I;X) \):** Recombination 
between the boundary of \( hln(I) \) crossover suppression and 
the \( stT1(I;X) \) breakpoint was measured by scoring the 
Unc-101 progeny from the segregating phenotype \( hln(I) \) 
\(+ +/stT1I(X)/unc-101; lon-2 \). To measure crossing over 
between \( stT1(I;X) \) and chromosome \( I \), Unc-101 
hermaphrodite progeny were scored from \(+/stT1I(X)/unc-101; 
lon-2 \) hermaphrodites. In both cases, the crossover frequency 
(\( p \)) between the \( stT1(I;X) \) breakpoint and unc-101 
(or the \( hln(I) \) boundary of crossover suppression) is defined 
by the following formula:

\[
p = \frac{4 - (16 - 60U/(U + W))^2}{6}
\]

where \( U \) is the number of Unc-101 recombinants and \( W \) is the 
number of wild-type progeny.

**Interaction of \( hln(I) \) with \( kT2(II;III) \):** To examine 
the interaction of \( hln(I) \) with the translocation \( kT2(II;III) \), 
recombination was measured in \(+ +/,+ +kT2(II;III)/bl-4 \ dpy-5 
unc-54 \) and \( hln(I)[+ +]/+,+ +kT2(II;III)/bl-4 \ dpy-5 \ unc-54 
\) heterozygotes using the formula:

\[
p = \frac{1 - [1 - 20D(3D + W)/(4D + 2W)]^2}{(3D + W)/(2D + W)}
\]

where \( D \) is the number of \( Dpy-5 \) recombinants and \( W \) the 
number of wild types. The segregation of \( hln(I) \) and 
\( kT2(II;III) \) was examined by scoring the \( Dpy-5 \ Unc-29 
progeny of a \( hln(I)[+ +]/+,+ +kT2(II;III)/bl-4 \ dpy-5 \ unc-29 \) 
hermaphrodite and \(+ +/+kT2(II;III)/bl-4 \ dpy-5 \ unc-29 \) control.

**Lethal screen using \( hln(I) \) as a balancer:** Hermaphrodites 
of the genotype \( hln(I)/unc-54/unc-101 \ lev-11 + \) were 
treated with 17 mm EMS (ROSENBLUTH, CUDDEFORD 
and BAILLIE 1985) using the procedure described by BRENNER 
(1974). Wild-type \( F1 \) progeny from these hermaphrodites were 
individually plated and their progeny screened for the 
absence of Unc-101 Lev-11 individuals.

**RESULTS**

Isolation of a crossover suppressor for the right 
end of LG I: \( hln(I) \) was identified in a screen for 
gamma mutations that suppressed crossing over 
between \( unc-101 \) and \( unc-54 \), a 14-map unit (m.u.) interval 
located at the right end of LG I. This map distance 
was reduced to 0.04 m.u. in \( hln(I)[+ +]/unc-101 \ unc-54 
\) heterozygotes (see Table 1). Since recombination 
in this interval was measured using the Unc-101 
recombinant class (Unc-54 recombinants are 
distinguishable from the double mutant), the possibility that 
\( hln(I) \) was a suppressor of the Unc-101 phenotype 
remained. For this reason, crossing over was examined in 
\( unc-101 \ lev-11 \) heterozygotes from which both 
recombinant classes were recovered. This interval 
was 9.0 m.u. in \( unc-101 \ lev-11+/+ \) heterozygotes and 0.07 
m.u. in \( hln(I)[+ +]/unc-101 \ lev-11 \) heterozygotes, 
demonstrating extensive crossover suppression of the 
right arm of LG I in \( hln(I) \) heterozygotes. Individuals 
nonhomogous for \( hln(I) \) were fertile and wild type in 
appearance. Since most crossover suppressors identified 
in \( C. elegans \) are translocations, the segregation of 
\( hln(I) \) from a normal homolog marked with an \( unc-101 
\) mutation was examined. The predicted segregation 
pattern of wild-type and Unc progeny for a 
translocation heterozygote is 5:1 (HERMAN 1978; 
ROSENBLUTH and BAILLIE 1981). \( hln(I) \) heterozygotes 
segretated wild-type and Unc progeny in a 3:1 ratio 
(2060 wild types: 672 Unc-101 individuals); a segregation 
pattern characteristic of an intrachromosomal 
rearrangement.
TABLE 1

Effects of hlnl(Z) on crossing over on linkage group I

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Wild types</th>
<th>Recombinants</th>
<th>p × 100 (C.I.)a</th>
</tr>
</thead>
<tbody>
<tr>
<td>unc-101 unc-54/hln1</td>
<td>1584</td>
<td>1 Unc-101</td>
<td>0.04 (0.002–0.25)</td>
</tr>
<tr>
<td>unc-101 lev-11/++</td>
<td>1492</td>
<td>99 Unc 82 Lev</td>
<td>9.0 (7.8–10.5)</td>
</tr>
<tr>
<td>unc-101 lev-11/hln1</td>
<td>2062</td>
<td>1 Lev</td>
<td>0.07 (0.004–0.39)</td>
</tr>
<tr>
<td>unc-75 unc-101/++b</td>
<td>3192</td>
<td>68 Unc-101</td>
<td>3.2 (2.7–3.8)</td>
</tr>
<tr>
<td>unc-75 unc-101/hln1</td>
<td>2211</td>
<td>1 Unc-75</td>
<td>0.07 (0.003–0.30)</td>
</tr>
<tr>
<td>dpy-5 unc-101/++</td>
<td>889</td>
<td>79 Dpy 66 Unc</td>
<td>12.0 (10.1–14.0)</td>
</tr>
<tr>
<td>dpy-5 unc-101/hln1</td>
<td>1975</td>
<td>165 Dpy 148 Unc</td>
<td>11.7 (11.2–12.2)</td>
</tr>
<tr>
<td>unc-29 lin-11/+/+; him-5/+</td>
<td>1514</td>
<td>14 Unc-29</td>
<td>1.4 (0.8–2.3)</td>
</tr>
<tr>
<td>unc-29 lin-11/hln1; him-5/+</td>
<td>1381</td>
<td>44 Unc-29</td>
<td>4.7 (3.4–6.3)</td>
</tr>
<tr>
<td>dpy-5 unc-29 unc-75/++/++b</td>
<td>1598</td>
<td>36 Unc-29 Unc-75c</td>
<td>3.4 (2.6–4.2)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2 Unc-29d</td>
<td>6.0 (4.7–7.6)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>63 Unc-75d</td>
<td>5.5 (4.6–6.5)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>81 Unc-75d</td>
<td>7.5 (5.9–9.5)</td>
</tr>
<tr>
<td>unc-11 dpy-5/++b</td>
<td>3786</td>
<td>58 Dpy 61 Unc</td>
<td>2.3 (2.0–2.8)</td>
</tr>
<tr>
<td>unc-11 dpy-5/hln1</td>
<td>1345</td>
<td>46 Dpy 41 Unc</td>
<td>4.8 (3.9–5.9)</td>
</tr>
<tr>
<td>bli-3 unc-11/++b</td>
<td>1686</td>
<td>170 Unc</td>
<td>14.8 (12.4–17.4)</td>
</tr>
<tr>
<td>bli-3 unc-11/hln1</td>
<td>1232</td>
<td>191 Unc</td>
<td>22.7 (19.4–26.0)</td>
</tr>
</tbody>
</table>

a C.I. = 95% confidence interval.

b Data from ZETKA and ROSE (1990).
c dpy-5 unc-29 interval.
d unc-29 unc-75 interval.

dpy-5 unc-29 unc-75/hln1

TABLE 2

Egg hatching frequencies of hlnl(I) heterozygotes and recombinants

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Egg-hatching frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>hlnl(I)/hlnl(I)</td>
<td>0.985 (465/472)</td>
</tr>
<tr>
<td>unc-101 lev-11/hlnl(I)</td>
<td>0.983 (567/577)</td>
</tr>
<tr>
<td>unc-101 unc-54/hdf11</td>
<td>0.556 (73/205)</td>
</tr>
<tr>
<td>unc-101 lev-11/hdp131</td>
<td>0.942 (243/258)</td>
</tr>
</tbody>
</table>

hlnl(I) heterozygotes have wild-type zygote viability: To further confirm that hlnl(I) was an intrachromosomal rearrangement, the egg-hatching frequency of individuals homozygous and heterozygous for the mutation was determined and is shown in Table 2. The egg-hatching frequencies for heterozygotes is not statistically different than for homozygotes, both of which are high, suggesting that few or no aneuploid gametes are being produced in the former. The egg-hatching frequencies of two recombinants derived from hlnl(I) heterozygotes is also shown in Table 2 for comparison. The egg-hatching frequency of hdf11/unc-101 unc-54 and hdp131/unc-101 lev-11 heterozygotes was 36% and 94%, respectively.

Gene order is inverted in hlnl(I): STURTEVANT (1921) established that three loci in Drosophila melanogaster and Drosophila simulans were not in the same sequence on the genetic maps of the two species, thus defining the first inversion. To determine if hlnl(I) was an inversion, the order of genes was examined by the induction of three mutations on the rearranged chromosome: unc-54(h1040), unc-75(h1041) and unc-75(h1042). The unc-75 mutations were recessive lethals that produced an Unc-75 phenotype when heterozygous with unc-75(e950), and both were used in the following experiments. Dpy-5 and Unc-54 recombinant progeny from a hlnl(I)[dpy-5 unc-54]/+ [hlnl(I)[+] unc-75] hermaphrodite were individually mated to unc-75(e950)/+ males to determine if the recombinant chromosome carried one of the lethal unc-75 mutations. The normal order of these genes is dpy-5 unc-75 unc-54 (EDGLEY and RIDDLE 1990). If the order of unc-75 and unc-54 were reversed, all Dpy-5 recombinants should fail to complement unc-75(e950), whereas the Unc-54 recombinants should complement unc-75. Of 17 Dpy progeny examined with h1042 and 10 with h1041, all 27 failed to complement unc-75. Of 12 Unc-54 progeny examined with h1042 and 6 with h1041, all 18 complemented unc-75. This demonstrated that either the gene order in hlnl(I) is dpy-5 unc-54 unc-75, or that the order is unchanged but unc-54 is now tightly linked to unc-75. To distinguish between these two possibilities, Dpy-5 progeny from a hlnl(I)[dpy-5 + unc-75(h1042)]/hlnl(I)[+ unc-54 +] hermaphrodite were individually plated and their progeny examined for Dpy-5 Unc-54.
different in the control (1 1.7 and 12.0 m.u., respectively), thereby reduced to 0.07 m.u. in The raising two possibilities; recombination to the left of intervals to the left of and two spanning the boundary. The results are determine if crossing over occurred in homozygotes, a 1.5-fold increase in recombination was measured in the control. The total genetic length of chromosome Z is 44 m.u in controls and 41 m.u. in unc-75 was normal or the interval contained a region of recombination enhancement with an associated region of recombination suppression. To distinguish between these alternatives, recombination was examined in dpy-5 unc-29 unc-75 heterozygotes. Recombination in the dpy-5 unc-75 interval was 13.0 m.u. in hInl(I) heterozygotes and 9.4 m.u. in the control. Crossing over in the unc-29 unc-75 region was not significantly affected by the presence of hInl(I), whereas the dpy-5 unc-29 interval showed a 1.6-fold increase in recombination in hInl(I)[+/+]/dpy-5 unc-29 heterozygotes. To further map the boundary of crossover suppression, recombination was measured between unc-29 and lin-11. This interval was 4.7 m.u. in heterozygotes and 1.4 m.u. in controls (3-fold enhancement), thus localizing the hInl(I) boundary of crossover suppression between lin-11 and unc-75. To determine if the recombination enhancement observed in the dpy-5 unc-29 interval extended to the left arm of LG I, recombination was examined in the bli-3 unc-11 and unc-11 dpy-5 regions. In hInl(I) heterozygotes, a 1.5-fold increase in recombination was observed in the bli-3 unc-11, and a 2-fold increase was observed in the unc-11 dpy-5 interval when compared to controls. The total genetic length of chromosome I is 44 m.u. in controls and 41 m.u. in hInl(I) heterozygotes (see Figure 1).

TABLE 3
Recombination in hInl(1) homozygotes

<table>
<thead>
<tr>
<th>Genotype</th>
<th>Wild types</th>
<th>Recombinants</th>
<th>p × 100 (C.I.)a</th>
</tr>
</thead>
<tbody>
<tr>
<td>hInl[dpy-5 unc-29]/hInl[+ +]</td>
<td>1500</td>
<td>35 Dpy 46 Unc</td>
<td>3.7 (2.9-4.6)</td>
</tr>
<tr>
<td>dpy-5 unc-75/hInl[+ +]</td>
<td>1464</td>
<td>151 Dpy</td>
<td>15.2 (12.7-17.7)</td>
</tr>
<tr>
<td>hInl[dpy-5 unc-75]/++</td>
<td>1285</td>
<td>136 Dpy</td>
<td>15.6 (13.2-18.2)</td>
</tr>
<tr>
<td>hInl[dpy-5 unc-75]/hInl[+ +]</td>
<td>1312</td>
<td>87 Dpy</td>
<td>9.8 (7.9-11.9)</td>
</tr>
<tr>
<td>hInl[dpy-5 unc-75(h1042)/hInl[+unc-54+]</td>
<td>1155</td>
<td>243 Dpy</td>
<td>9.0 (6.6-11.4)</td>
</tr>
</tbody>
</table>

a C.I. = 95% confidence interval.
b unc-54 unc-75 distance measured in trans. see MATERIALS AND METHODS.
Rare recombinants from hln1(I) heterozygotes contain duplications and deficiencies: Single crossovers within a classical inversion heterozygote produce chromosomes that contain duplications and deficiencies. Four rare (~1/2500) recombinants were recovered from hln1(I) heterozygotes. Three of these originated from mapping experiments (see Table 1), while the fourth was isolated independently from a hln1(I)[+]/unc-101 lev-11 hermaphrodite on the basis of its visible Lev-11 phenotype. To determine if the individuals homozygous for the chromosome of interest were viable, all four recombinants were crossed to N2 males. The progeny of wild-type hermaphrodites resulting from this cross were screened for the presence of individuals with the original recombinant phenotype. Two of the four recombinants proved to be homozygous lethal, and both failed to complement unc-59 and lev-11 establishing them as deficiencies, later designated as hDf11 and hDf12. hDf11 was known to complement unc-54 because of the original phenotype of the recombinant (Unc-101 when heterozygous with an unc-101 unc-54 chromosome). hDf11 complemented unc-75, indicating the left deficiency breakpoint is to the right of this gene. It is not known whether hDf11 includes unc-101 or if the deficiency bearing chromosome carries the original unc-101 mutation. hDf12 also complemented unc-54 and was known to complement unc-101 based on the original recombinant phenotype (Unc-75 when heterozygous with an unc-75 unc-101 chromosome). Thus the left breakpoint of hDf12 is to the right of unc-101. The extent of these deficiencies is shown diagramatically in Figure 2. The remaining two recombinants, hDp131 and hDp132, were both Lev-11 in phenotype when heterozygous with an unc-101 lev-11 chromosome. When crossed to N2 males, however, all resulting wild-type progeny segregated Unc-101 Lev-11 individuals, suggesting the recombinants were diploid for unc-101 lev-11 chromosome and carried a duplication of unc-101. These two duplications were mapped to visible markers and segregated from chromosome I as though unlinked. Both hDp131 and hDp132 have breakpoints between unc-29 and unc-75, and carry unc-75(+), unc-101(±), unc-59(+), and unc-54(±). That the duplications are Lev-11 in phenotype when heterozygous with unc-101 lev-11 chromosomes suggests that they are linked to the original lev-11 mutation. The extent of the duplications and their known breakpoints is shown in Figure 2.

hln1(I) has no effect on crossing over on other chromosomes: In D. melanogaster, inversion heterozygosity produces interchromosomal effects; an increase in crossing over in regions surrounding the centric heterochromatin and the distal tips of chromosome arms on the other pairs of chromosomes (Schultz and Redfield 1951; Ramel 1962; reviewed by Lucchesi 1976). To determine if hln1(I) produces a similar effect in C. elegans, recombination was measured on other chromosomes in hln1(I) heterozygotes. The results are shown in Table 4. Two regions located on autosomes and one located on the X chromosome were examined. In all three cases, the presence of hln1(I) did not significantly affect recombination in heterozygotes.

hln1(I) recombines with szTl(I;X): The meiotic behaviour of the translocation szTl(I;X) has been extensively characterized (Fodor and Deak 1985; McKim, Howell and Rose 1988). The breakpoint of the translocation on LG I is close to the left of unc-29, and translocation homozygotes are inviable. The extent of crossover suppression was determined; recombination was suppressed to the left of the breakpoint and enhanced to the right (McKim, Howell and Rose 1988). Since crossing over is suppressed in the unc-75 unc-54 interval in hln1(I) heterozygotes, it was of interest to determine if pairing was possible between the two rearrangements. Crossing over between the szTl(I;X) breakpoint and the hln1(I) boundary of crossover suppression was measured in hermaphrodites of the genotype hln1(I)[+]/szTl(I;X); unc-10 lon-2]. The map distance between the breakpoint of szTl(I;X) and the boundary of hln1(I) crossover suppression (between lin-11 and unc-75) was 45 m.u. The recombination frequency between the szTl breakpoint and unc-101 was measured in +/+ szTl(I;X)/unc-10 lon-2] controls and was 25 m.u., approximately 2-fold lower. The data for these experiments are shown in Table 5.

hln1(I)/hT2(I;III) heterozygotes suppress crossing over on LG I: The translocation hT2(I;III) is comprised of two chromosomes; hT2(I;III) segregates from chromosome I and, hT2(I;III) segregates from chromosome III. In heterozygotes, recombination on LG I is suppressed to the left of unc-101 and enhanced to the right of this marker. Since hln1(I) suppresses recombination from unc-75 to unc-54, it was of interest to determine whether recombination could be completely suppressed on LG I. Crossing over between dpy-5 and unc-54 was measured in hln1(I)[+/+] hT2(I;III)[dpy-5 bli-4 unc-54] heterozygotes and +/+ hT2(I;III)[dpy-5 bli-4 unc-54] controls (results shown in Table 5). The map distance of chromosome I was reduced to 0.8 m.u. in hln1(I) heterozygotes, compared to 32.1 m.u. in controls, thus demonstrating recombination could be effectively suppressed along the entire length of the chromosome.

hln1(I) and hT2(I;III) segregate randomly: While examining recombination in hln1(I)[+ +]; +/+ hT2(I;III)[dpy-5 bli-4 unc-54] heterozygotes, an unusually small number of Dpy-5 Unc-54 progeny, representing the viable translocation homozygote, were recovered. To investigate the possibility that hln1(I)
An Inversion in *C. elegans*

**Figure 2.**—Position of breakpoints of recombinant chromosomes derived from *hlnl(I)* heterozygotes. *hDf11* complements *unc-75* but it is not known if the deletion includes *unc-101*. The right breakpoints of *hDp131* and *hDp132* are not known but both the duplications cover *unc-54*.

<table>
<thead>
<tr>
<th>Table 4</th>
<th>Effect of <em>hlnl(I)</em> on crossing over on other chromosomes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype</td>
<td>Wild types</td>
</tr>
<tr>
<td><em>dpy-18 unc-36/+</em></td>
<td>1561</td>
</tr>
<tr>
<td><em>dpy-18 unc-36/+;hlnl/+</em></td>
<td>1881</td>
</tr>
<tr>
<td><em>unc-1 dpy-3/+</em></td>
<td>1375</td>
</tr>
<tr>
<td><em>unc-1 dpy-3/+;hlnl/+</em></td>
<td>2147</td>
</tr>
<tr>
<td><em>unc-42 dpy-11/+</em></td>
<td>1357</td>
</tr>
<tr>
<td><em>unc-42 dpy-11/+;hlnl/+</em></td>
<td>1324</td>
</tr>
</tbody>
</table>

* C.I. = 95% confidence interval.

<table>
<thead>
<tr>
<th>Table 5</th>
<th>Effect of <em>hlnl(I)</em> on crossing over with LG I translocations</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotype</td>
<td>Wild types</td>
</tr>
<tr>
<td><em>+/st1(I;X)[unc-10 lon-2]</em></td>
<td>1110</td>
</tr>
<tr>
<td><em>hlnl(I;+;st1(I;X)[unc-10 lon-2]</em></td>
<td>1775</td>
</tr>
<tr>
<td><em>++;st2(I;III)[dpy-5 bli-4 unc-54;+]</em></td>
<td>441</td>
</tr>
<tr>
<td><em>hlnl(I)/st2(I;III)[dpy-5 bli-4 unc-54]</em></td>
<td>591</td>
</tr>
</tbody>
</table>

* C.I. = 95% confidence interval.

Both Dpy individuals were fertile and gave Dpy progeny.

and *hT2(I;III)* were segregating abnormally, segregation was examined in *hlnl(I)+;+/hT2(I;III)[dpy-5 bli-4 unc-29]* heterozygotes and *++;+/hT2(I;III)[dpy-5 bli-4 unc-29]* controls. Since both *dpy-5* and *unc-29* map in the crossover suppressed arm of *hT2(I;III)*, the recovery of the double mutant, representing the viable translocation homozygote class, is dependent upon the proper segregation of the translocation from the normal homologs. In the control, the predicted ratio (5:1) of wild types to Dpy-5 Unc-29 progeny was observed (771 Wild types: 164 Dpy Unc). The frequency observed in *hlnl(I)* heterozygotes however, was 13.7:1 (411 Wild types: 30 Dpy Unc), close to the predicted ratio of 11:1 if *hT2(I;III)* and *hlnl(I)* were segregating randomly, resulting in aneuploid gametes (shown in Figure 3). Both the recovery of rare recombinants in the previous experiment and the difference between the predicted and observed segregation ratios may be explained by a low frequency of pairing between the two rearrangements.

**hlnl(I) effectively balances lethal mutations:** One objective in isolating a crossover suppressor for a region associated with the homolog recognition region was the demonstration that such rearrangements, presumably intrachromosomal, would be effective balancers. The efficiency of *hlnl(I)* was tested by screening for recessive lethal mutations in the region of crossover suppression. In total, 1412 mutagenized chromosomes were screened and 54 mutations, including those resulting in adult sterility, were recovered. Strains representing the recovered mutations were effectively balanced in *hlnl(I)* heterozygotes for at least 20 generations (before being frozen) without breakdown of the balancer being observed.

**Discussion**

In this study, we have presented evidence for an inversion in *C. elegans*, *hlnl(I)*, that inverts a region of chromosome I, including the genes *unc-75* and *unc-54*. The meiotic properties of *hlnl(I)* were similar to
and chromosome, has been well characterized. Zn(l)dl-49, 
satory increases in recombination are large in 
pairing by forming loops in mitotic cells 
bing efficiently with the translocation 
gation 
are duplicated for (see RESULTS). Empty boxes represent presumed lethal zygotes 
synthesis, but that physical constraints inside the inver-
reasons. Firstly, in Drosophila, crossing over inside 
reduction in crossing over in heterozygotes despite cy-
tological evidence that this inversion was capable of 
resulting from severe aneuploidy. Viable classes are indicated by 
exchange  events are rare seems likely for three 
that the two rearrangements also synapse 
and any aneuploidy they might carry. hDp134 progeny 
phenotype and any aneuploidy they might carry. 
are capable of homolog recognition and 
synapsis, but that physical constraints inside the inver-
some and is even smaller than 
linked with the chromosome to which it is attached. 
es that reduce the frequency of chiasma formation 
such inversions when heterozygous. By analogy, 
hln l(I), which is also located at the end of a chromo-
and is even smaller than ln(l)dl-49, would also 
expected to experience constraints in pairing for 
recombination in heterozygotes. Secondly, compensatory increases in recombination are large in hln l(I) heterozygotes, as would be expected if exchanges within the inversion were rare (i.e., in hln l(I) heterozygotes, the map distance from bci-3 to lin-11, normally 22 m.u., approaches 50 m.u.). The fact that hln l(I) heterozygotes efficiently recombine in other regions of the chromosome indicates that the ability of the homologs to recognize one another is intact, and that recombination suppression on the right arm is limited to the inverted segment. Thirdly, reciprocal recombinant events were isolated from hln l(I) heterozygotes, suggesting that all meiotic products can be recovered, but that their frequency is low.

We have examined recombination between hln l(I) and two translocations; szTl(I;X) and hT2(I;II). The pairing portion of szTl(I;X) and hln l(I) share sequences not included in either rearrangement, whereas the pairing portion of hT2(I;III) and hln l(I) have no common unarranged sequences. The crossover frequency between hln l(I) and szTl(I;X) was 0.45, demonstrating that synapsis and recombination are being efficiently conducted between the two in spite of the small size of homologously paired DNA. In contrast, the frequency of recombination between hln l(I) and hT2(I;III) was less than 0.01. These results agree with the conclusion that exchanges within the inversion are rare, since the only DNA available for pairing is within the inverted segment.

DARLINGTON (1937) suggested that the formation of a chiasma between homologs during meiosis facilitates their proper disjunction (reviewed by HAWLEY 1988). One consequence of the crossover suppression observed in hln l(I)/hT2(I;III) heterozygotes was the random segregation of hln l(I) and hT2(I;III). This suggests that in C. elegans, as in Drosophila, the formation of a chiasma between two homologs is important in ensuring proper disjunction during meiosis.

In Drosophila it has been observed that inversions can effect increases in recombination frequency on the rearranged chromosome and on the other major chromosomes (STURTEVANT 1919; 1931; DOBZHANSKY 1933; reviewed by LUCHESSI 1976). We have found the total genetic length of chromosome I to be 41 m.u. in hln l(I) heterozygotes and 44 m.u. in controls. These values are similar to the recombination frequency reported for the pairing portion of chromosome I in individuals heterozygous for four translocations involving chromosome I: hTl(I;V), szTl(I;X) (MCKIM, HOWELL and ROSE 1988), hT3(I;X), and hT2(I;III) (MCKIM 1990). These results indicated that while compensatory increases can occur on both arms of LG I, the amount of exchange is limited to approximately one crossover event per meiosis. Unlike inversions in Drosophila, hln l(I) mediated recombination enhancement in heterozygotes did not extend to other linkage groups. No increase in crossing over was observed in the three intervals examined in the presence

<table>
<thead>
<tr>
<th></th>
<th>µ1 ln l(I; hZn(Z))</th>
<th>µ1 ln l(I; hZnZ)</th>
<th>µ1 ln l(I; hT2(I;ZZZ))</th>
<th>µ1 ln l(I; hT3(I;V))</th>
<th>µ1 ln l(I; szT1(Z;X))</th>
</tr>
</thead>
<tbody>
<tr>
<td>µ1 ln l(I; hZn(Z))</td>
<td>µ1 ln l(I; hZnZ)</td>
<td>µ1 ln l(I; hT2(I;ZZZ))</td>
<td>µ1 ln l(I; hT3(I;V))</td>
<td>µ1 ln l(I; szT1(Z;X))</td>
<td></td>
</tr>
<tr>
<td>µ1 ln l(I; hZnZ)</td>
<td>µ1 ln l(I; hT2(I;ZZZ))</td>
<td>µ1 ln l(I; hT3(I;V))</td>
<td>µ1 ln l(I; szT1(Z;X))</td>
<td></td>
<td></td>
</tr>
<tr>
<td>µ1 ln l(I; hT2(I;ZZZ))</td>
<td>µ1 ln l(I; hT3(I;V))</td>
<td>µ1 ln l(I; szT1(Z;X))</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

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Figure 3.—Punnett square diagramming the predicted segregation of a hln l(I; + +); +/hT2(I;III)dp-5 unc-29; +] heterozygote (see RESULTS). Empty boxes represent presumed lethal zygotes resulting from severe aneuploidy. Viable classes are indicated by phenotype and any aneuploidy they might carry. hDp134 progeny are duplicated for µIII and are viable (K. S. MCKIM and A. M. ROSE, unpublished results).
of *hln1(I)*, regardless of their location on the autosomes (small interval inside the cluster or large interval spanning the cluster) or on the *X* chromosome. Interchromosomal effects have been observed in *C. elegans* with mutations that result in *X* chromosome nondisjunction (Hodgkin, Horvitz and Brenner 1979; Herman and Kari 1989). Thus in *C. elegans*, as in *D. melanogaster*, the mechanism that regulates the number of crossovers per meiosis may involve compensatory increases of events on other chromosomes in the event crossing over is suppressed or reduced along an entire chromosome. The failure to observe interchromosomal effects in *hln1(I)* heterozygotes may have been expected since recombination was not reduced on chromosome *I* as a whole.

Exchange events resulting from an intrachromosomal effect are not distributed randomly along the chromosome. In *Drosophila* for example, such increases occur in regions sufficiently removed from the inversion breakpoint (Grell 1962), and near the centromeric heterochromatin and distal tips of other chromosomes, regions of low intrinsic exchange (Schultz and Redfield 1951; RAMEL 1962). Each of the autosomes in *C. elegans* are marked by a region where genes cluster on the meiotic map resulting from a reduction in recombination per base pair compared to the genomic average (Brenner 1974; Greenwald et al. 1987; Kim and Rose 1987; Prasad and Baillie 1989; Starr et al. 1989). In *hln1(I)* heterozygotes, recombination frequency was enhanced in intervals both inside (1.5 fold in *dpy-5 unc-29*) and outside (1.5-fold in *bli-3 unc-11*) the chromosome *I* gene cluster. This suggests that the regulatory mechanism responsible for establishing the distribution of crossing over is independent of the mechanism determining the number of exchanges. The meiotic pattern specific to chromosome *I* is retained; the enhancement observed is not greater in the cluster than it is at the left end. We have found the frequency and distribution of exchange events to be normal in *hln1(I)* inversion homozygotes. This suggested that the pairing difficulties experienced by heterozygotes were removed in homozygotes, exchange within the inversion maintained the distribution observed in wild types. Chromosomal sites that are necessary for normal levels of meiotic exchange have been mapped in *Drosophila* (Hawley 1980; Szauter 1984). A similar mechanism may exist in *C. elegans* since recombination frequency is enhanced in the region adjacent to the szTl(I;X) breakpoint on LG 1, suggesting the break may have disrupted the mechanism responsible for the regional distribution of exchange (McKim, Howell and Rose 1988). If this mechanism is mediated by chromosomal elements, the level of crossing over in *hln1(I)* homozygotes suggests that such elements can operate normally in either orientation.

Inversions are classically defined by their exclusion (paracentric) or inclusion (pericentric) of the centromere (Muller 1938; reviewed by Roberts 1976). Cytogenetic analysis of inversion heterozygotes demonstrated that inverted homologous segments were capable of pairing by forming a loop in salivary glands (Painter 1933). A single exchange in a paracentric inversion loop led to the formation of acenric and dicentric fragments. The formation of these structures had been observed cytogenetically during meiosis in *Zea mays* (McClintock 1933). Single exchanges within paracentric inversions were not observed in *Drosophila* until single crossover products were recovered from individuals heterozygous for a long paracentric inversion on the *X* using an attached chromosome (Sidorov, Sokolov and Trofimov 1935). These results demonstrated that single crossovers do occur but that single crossover recombinants are not recovered. Nevertheless, information transfer in the form of gene conversion occurred in undiminished frequency in inversion heterozygotes, except near the breakpoints where effective homologous pairing may not be possible (Chovnick 1973). Unexpectedly, no concomitant loss of zygote viability was observed in heterozygotes despite the formation of aberrant chromosomes (Sturtevant and Beadle 1936; Novitski 1952). To explain this, Sturtevant and Beadle (1939) proposed that chromatids involved in single exchanges were excluded from a functional nucleus, a theory later corroborated by genetic and cytological evidence (Sturtevant and Beadle 1939; Carson 1946; Hinton and Luchessi 1960). In contrast, single crossovers in pericentric inversion heterozygotes produced chromosomes with terminal duplications and deficiencies that were segregated into gametes and resulted in reduced fertility (Roberts 1967). The frequency at which single exchanges occurred was dependent on the size and location of the inversion; a reduced frequency of such events was observed with both small inversions, and inversions located at the ends of chromosome arms (Sturtevant and Beadle 1936; Novitski and Braver 1954). Individuals heterozygous for *hln1(I)* showed no reduction in egg-hatching frequencies, compatible with the behaviour of a paracentric inversion for which the products of single exchanges are either excluded from functional nuclei or for which single exchanges in the inverted segment are rare.

The recovery of a chromosomal rearrangement in *C. elegans* that behaves like a paracentric inversion may seem surprising given that the mitotic chromosomes are holokinetic (Albertson and Thomson 1982) and evidence for holocentric meiotic chromosomes has been reviewed (Herman 1988). Recent cytological studies, however, suggest that the ends of *C. elegans* chromosomes adopt centromeric functions...
for meiotic disjunction; one end holds the bivalent together and the other probably provides a site for the attachment of microtubules. These roles do not appear to be specific to one end of the chromosome and either end can be the inner or outer end of the bivalent (D. G. Albertson, unpublished results). This meiotic behaviour is similar to that observed in other mitotically holokinetic species; the nematode *Parascaris univalens* (Goday, Ciolfi Luzzatto and Pimpinelli 1985; Pimpinelli and Goday 1989; Goday and Pimpinelli 1989), the insects *Euchistus servus* (Hughes-Schrader and Schrader 1961) and *Myrmus miriformis* (Nokkala 1985), where the mitotic chromosomes are holocentric but during meiosis centromeric activity is restricted to a limited chromosome region, often at chromosome ends.

The results of genetic analyses in *C. elegans* have consistently been compatible with the predicted behaviour of monocentric chromosomes. For example, the segregation ratios of aneuploid and viable progeny observed from translocation heterozygotes were compatible with the presence of a single centromere (Herman 1978; Rosenbluth and Bailie 1981; McKim, Howell and Rose 1988; this paper). In the case of the translocation *etl(III;V)*, recombination is suppressed to one side of the translocation breakpoint while the other recombines and segregates from the chromosome with which it had paired (Rosenbluth and Bailie 1981). Thus, in any one meiosis, only one meiotic segregator (centromere) was functional. Our data strongly suggest that the meiotic chromosomes are monocentric, a suggestion compatible with both genetic and cytogenetic observations.

The isolation of four recombinants from *hlnI(1)* heterozygotes raised the possibility that their genotypes would provide information on the location of the meiotic centromere. Two apparently free duplications, *h*dp131 and *h*dp132 were recovered following a single exchange event inside the inversion loop. The structure of these duplications is consistent with the products formed by the events illustrated in Figure 4. In this model, *h*dp131 and *h*dp132 are represented by the acentric fragment that results from a single exchange within a paracentric inversion where the centromere is to the left of *unc-75*. The facts that the duplications were isolated independently, and that the left endpoints were nonrandom and coincided with the boundary of *hlnI(1)* crossover suppression (between *unc-29* and *unc-75*) support this interpretation.

In *Drosophila*, the acentric fragment generated by a single exchange within a paracentric inversion loop is not recovered under ordinary circumstances. In *C. elegans* however, free duplications are readily recovered (Herman, Albertson and Brenner 1976). According to the model shown in Figure 4, the reciprocal product is a duplication of the sequences to the left of the inversion including the centromeric sequences. This structure is analogous to the dicentric chromosomes generated by single exchanges within paracentric inversions in *Drosophila*. Although this reciprocal product was not recovered intact in our experiments, *hdf11* and *hdf12* may have resulted from its breakage. Dicentric chromosomes in other organisms have been observed to form chromatid bridges at anaphase I and as a result are meiotically unstable and subject to chromosome breakage (McClintock 1933, 1941; Carson 1946; Haber, Thorburn and Rogers 1984). The two deficiencies recovered, *hdf11* and *hdf12* could have resulted from a similar event followed by the broken end of one
An Inversion in C. elegans

product being capped by sequences on the right end (including unc-54(+)), presumably derived from its normal homolog to which it is still attached. To stabilize broken ends of chromosomes, double-stranded breaks can be repaired by recombining with homologous sequences (HABER and THORBURN 1984), or fusing with other chromosomes (McCLINTOCK 1941, 1942). Both deficiencies recovered have breakpoints independent of the site of the original exchange event. Thus, the structures of the recombinant chromosomes recovered are compatible with the interpretation that hln l(l) is a paracentric inversion with the centromere to its left.

The recombinant products recovered are not consistent with the predicted behaviour of a paracentric inversion with centromeric sequences to the right of the inversion. The recovery of hdp131 and hdp132 is not compatible with dicentric products (see below), and no acentric fragment of the predicted structure was recovered. The possibility that hln l(l) is a pericentric inversion cannot formally be ruled out, however it is unlikely for the following reasons. No significant reduction in egg-hatching frequency was observed in hln l(l) heterozygotes as would have been expected if the inversion were pericentric however, it is possible that the frequency of recombination within the inversion was so low that no reduction was observed. Most importantly, however, if hln l(l) included the centromere, hdp131 and hdp132 would be centric and would have segregated from their homolog at meiosis II. To recover these duplications, a nondisjunction event would be needed to generate a viable zygote and the probability of recovering two such rare events is very low. The reciprocal recombinant product would also have possessed one centromere and would have been meiotically stable. Size is unlikely to be a consideration because large, rearranged chromosomes exist in C. elegans and are meiotically stable (HERMAN, KARI and HARTMAN 1982; SUGURDSON et al. 1986; MCKIM 1990; D. G. ALBERTSON, unpublished results). We feel the simplest interpretation of our data is that this inversion is paracentric in nature.

In conclusion, we have characterized the meiotic behaviour of a C. elegans inversion hln l(l), that suppresses crossing over in a previously unbalanced region. We feel the simplest interpretation of our data is that hln l(l) is paracentric in nature. hln l(l) was used to successfully balance lethal mutations in a region previously impenetrable to extensive essential gene analysis, demonstrating its value as a new class of balancer for the genome of C. elegans.

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