

## Supplemental Data

### HIM-8 Binds to the X Chromosome

### Pairing Center and Mediates

### Chromosome-Specific Meiotic Synapsis

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**Table S1. Frequencies of Interhomolog Associations in Wild-Type and *him-8* Hermaphrodites**

<b><i>XL</i></b>	<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>
<b>wild-type (N2)</b>	18.5 (260)	65.7 (341)	96.8 (438)	97.9 (336)	96.4 (165)
<b><i>him-8 (mn253)</i></b>	16.2 (266)	17.3 (329)	18.5 (330)	20.0 (260)	21.6 (148)
<b><i>him-8 (me4)</i></b>	20.8 (207)	21.5 (311)	13.7 (315)	14.5 (255)	18.6 (129)

<b><i>XR</i></b>	<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>
<b>wild-type (N2)</b>	14.6 (260)	48.7 (341)	91.1 (438)	95.8 (336)	94.5 (165)
<b><i>him-8 (mn253)</i></b>	13.9 (266)	16.4 (329)	17.3 (330)	14.6 (260)	19.6 (148)
<b><i>him-8 (me4)</i></b>	18.4 (207)	20.3 (311)	12.1 (315)	12.5 (255)	17.1 (129)

<b><i>VR (5S rDNA)</i></b>	<b>1</b>	<b>2</b>	<b>3</b>	<b>4</b>	<b>5</b>
<b>wild-type (N2)</b>	8.5 (260)	50.1 (341)	93.8 (438)	97.3 (336)	94.5 (165)
<b><i>him-8 (mn253)</i></b>	7.5 (266)	33.1 (329)	92.1 (330)	99.6 (260)	98.6 (148)
<b><i>him-8 (me4)</i></b>	7.2 (207)	23.8 (311)	77.8 (315)	99.2 (255)	98.4 (129)

These data correspond to the graphs presented in Figure 2. FISH probes were derived from the left and right ends of the X chromosome (XL and XR) as well as the 5SrDNA locus on the right arm of Chromosome V (VR). The frequency at which each probe was observed as a single signal or two signals  $\leq 0.7 \mu\text{m}$  apart is scored in each of 5 zones (1-5). The total number of nuclei scored for each probe in each zone is indicated in parentheses.

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**Table S2. X-Chromosome Pairing Analysis in Wild-Type and *him-8(me4)* Hermaphrodites by Analysis of HIM-8-Staining Foci**

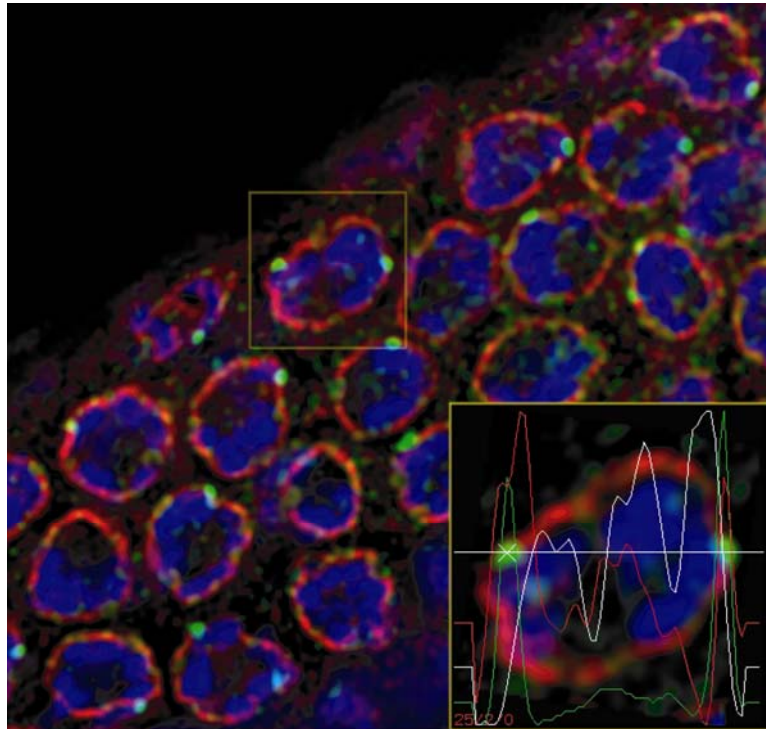
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	1	2	3	4	5
<b>wild type (N2)</b>	0.0 (39)	62.6 (99)	98.0 (102)	100.0 (85)	100.0 (80)
<b><i>him-8 (me4)</i></b>	0.0 (70)	0.9 (112)	2.3 (176)	4.7 (150)	2.1 (95)

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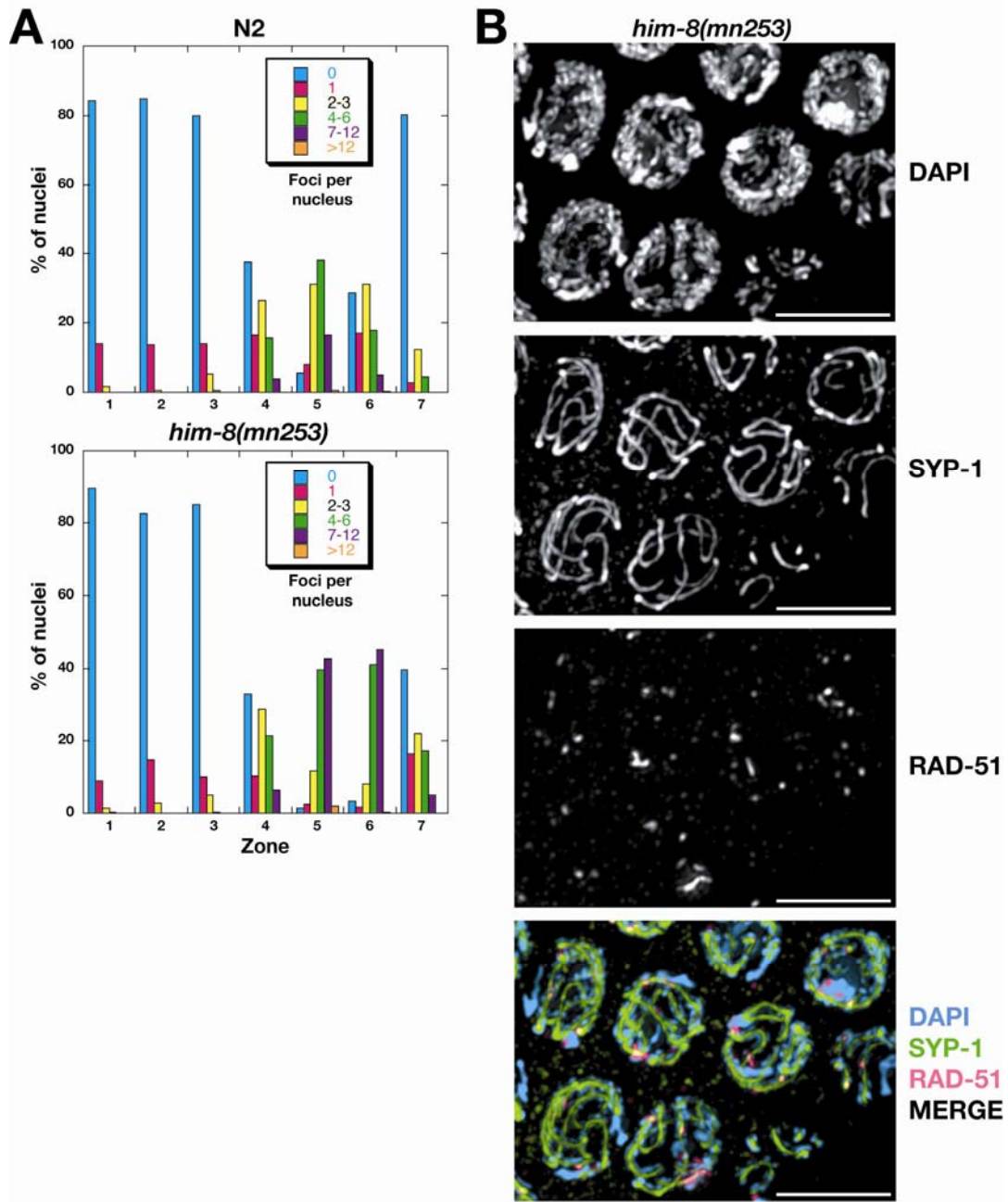
These data correspond to the graphs presented in Figure 5E. Immunofluorescence with the HIM-8 antibody was performed on wild-type and *him-8(me4)* hermaphrodites. The fraction of paired foci was scored in each of 5 zones of the gonad, which were defined in the same way as for the FISH analysis (Figure 2). The number of nuclei scored for each zone is indicated in parentheses.

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**Figure S1. HIM-8 Localizes near the Nuclear Envelope**

The larger image displays a cross-section through a field from the pachytene region of a *him-8(me4)* hermaphrodite stained with anti-LMN-1 (lamin) and anti-HIM-8 antibodies. DNA is stained with DAPI. Because the close association of HIM-8 foci with the nuclear lamina is not always apparent in 3D projections such as those shown in Figure 6, this image shows a single optical section from a deconvolved 3D data stack. Consequently, not all HIM-8 foci are visible in this plane. The DAPI (blue), anti-HIM-8 (green), and anti-LMN-1 (red) images are displayed in separate RGB channels rather than the mixed pseudocolors used in Figure 6 so that intensity values in each channel could be analyzed separately. The inset shows a line-scan of intensities through one nucleus (outlined in the larger image), which was selected for display here because both of the unpaired HIM-8 foci could be detected in the same optical section. A horizontal line that traverses the peaks of both HIM-8 foci (shown in white) was scanned in each wavelength. Intensity traces are shown in white (DAPI), red (lamin), and green (HIM-8). Note that the DAPI intensity is bounded by and internal to the major peaks of lamin intensity, but the HIM-8 foci correspond closely in their peak positions to the outer lamin peaks.

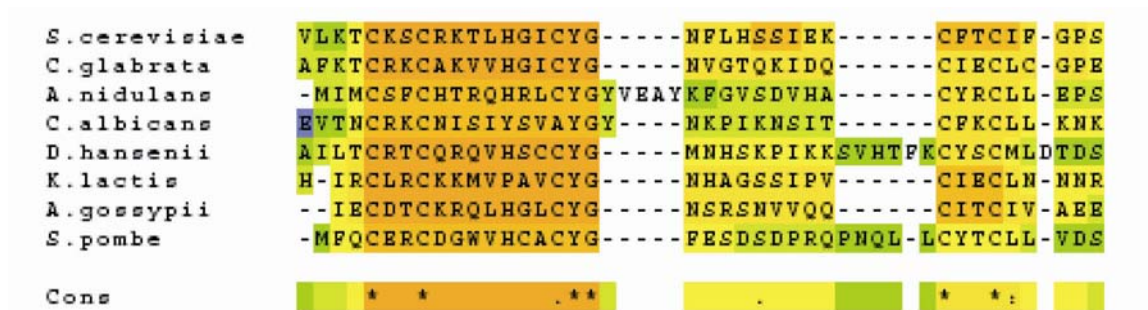


**Figure S2. Unsynapsed X Chromosomes Initiate Meiotic Recombination in *him-8* Mutants**

(A) Quantification of RAD-51 foci in individual nuclei throughout meiotic prophase reveals that *him-8* mutations cause a delay in the disappearance of recombination intermediates. This analysis was performed as in MacQueen et al. (2005), and the data for N2 worms are identical. The numbers of nuclei scored for each genotype were as follows

(in order by zone): N2: 344, 453, 471, 373, 327, 276, 138; total = 2382; *him-8(mn253)*: 312, 415, 436, 438 365, 243, 146; total = 2355.

(B) In nuclei in late pachytene, RAD-51 foci are frequently detected on the unsynapsed X chromosomes, which can be recognized as brightly DAPI-stained areas lacking extensive SYP-1 segments. In this projection, RAD-51 foci are most clearly seen on X chromosomes in the nucleus at the top right corner. Scale bars represent 5  $\mu$ m.



**Figure S3. The Zinc Finger of Hop1 Is Conserved among Homologs from Diverse Fungi**

Homologs of *S. cerevisiae* Hop1 were identified among GenBank entries by PSI-BLAST analysis. Protein sequences that share the zinc finger region were aligned using T-COFFEE. The portion of the alignment containing the zinc finger region is shown here. The conservation of this domain in *S. pombe* and *A. nidulans* is particularly relevant to this Discussion, since these fungi do not polymerize a central element of the SC, indicating that the zinc finger is likely to play a synapsis-independent role.