Supplemental Table 2. Frequency of X^D in *D. recens* populations and locations of X^D and X^{ST} chromosomes used in polymorphism analyses

Population	Year sampled	X^D frequency among males	Polymorphism analyses	
			X^D	X^{ST}
Charlottetown, PEI	2003	0/4	-	-
Chebeague Island, ME	2004	2/37	2	-
Bethlehem, NH	2002	0/7	-	7
Rochester, NY	2002	0/9	4	9
Rochester, NY	1994	3/76	-	-
Big Moose Lake, NY	1994	2/42	-	-
Mattawa, ON	2004	2/78	5	-
Smoky Mountains, TN	2001	1/47	1	-
Munising, MI	2002	0/10	-	7
Bemidji, MN	2002	1/33	1	6
Minot, ND	2002	0/28	-	-
Winston Churchill, AB	2002	0/20	-	-
Edmonton, AB	2002	0/0	1	-
Total		11/391 (0.028)	14	29

Populations are listed approximately east to west. Data from New York populations from 1994 are from Jaenike (1). For the polymorphism analyses, the additional X^Ds from some populations were identified from the offspring of wild-caught females. Of the four X^Ds from Rochester included in the polymorphism analyses, one was collected in 2001, one in 2002, and two in 2004.

1. Jaenike J (1996) *Amer Nat* 148:237-254.

Supplemental Table 3. Gene conversion tracts detected with the Betran *et al.* (1) approach

Gene	Alignment length, bp	Source	Recipient	Recipient strain(s)	Tract length, bp	Region in alignment	ψ
cp36	744	X^D	X^{ST}	MI ST 7	157	440-596	0.0089
per	648	X^D	X^{ST}	MI ST 3	394	216-609	0.0174
per	648	X^D	X^{ST}	MI ST 4	120	67-189	0.0174
v	759	X^D	X^{ST}	MI ST 13, NY ST 14	2	481-482	0.0408
v	759	X^D	X^{ST}	MI ST 3	23	515-537	0.0408
y	1602	X^D	X^{ST}	MN ST 8	443	101-1,555	0.0254
y	1602	X^D	X^{ST}	NY ST 16	68	1,477-1,555	0.0254
Average					172		0.0231

Each gene was analyzed separately. No gene conversion events were observed within *elav* or *runt*.

1. Betran E, Rozas J, Navarro A, Barbadilla A (1997) *Genetics* 146:89-99.

Supplemental Table 4. Primers used in this study

Gene	Primer	Use	Source	Sequence (5' to 3')
cp36	cp36-F	A, S	1	TGCAACTYGGTCTCTGGTTTG
	cp36-R	A, S	1	TGAGGCTGGCTGTAGACG
elav	elav-R	A	This study	GAYACACAGGCRCAGCTAATG
	elav-F	A	This study	GGCYTTGTTGGTCTTGAAGC
	elav-intF6	S	This study	GGTGCAACAGGCGATATTG
	elav-intR5	S	This study	GTTGAGGGCGCGTATTGC
per	per-F	A, S	2	ACAAGGAGAAGTCCAGGAAGAAG
	per-R	A, S	2	GAACGTCAACCCCAGGCGGAAGG
runt	runt-F	A, S	This study	CTGACCATCACCATTGCCAC
	runt-R	A, S	3	AAGTAGTCCGCGTAGCCGTA
v	verm-F	A	1	TAYGGMGARTAYCTSATGCTGGAC
	verm-R	A	1	CGRAAGTCCATRAARTCVARMGG
	verm-intF10	S	This study	TTTGAATTCGACTCCATACG
	verm-intR10	S	This study	AAGAGGATCAGTGTGCGATC
y	Y-univ+iv	A, S	From E. Dyreson	TGGCTAATGACTTCGGAAATTG
	Y-univ-ex2	A, S	From E. Dyreson	CTGTGATCCATGTTGATGTAGG

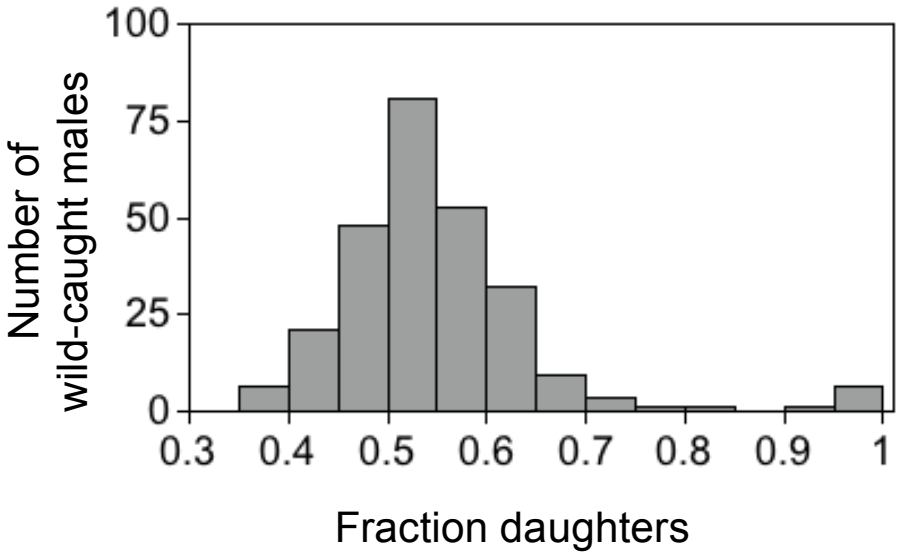
elav and *per* are all coding sequence; the primers of *cp36*, *runt*, and *v* are anchored in exons but amplify across an intron; the *y* primers are anchored in an intronic enhancer at the 5' end and an exon at the 3' end, yielding primarily noncoding sequence. Primers were used for amplification (A) and/or sequencing (S).

1. Dyer KA, Jaenike J (2004) *Genetics* 168:1443-1455.

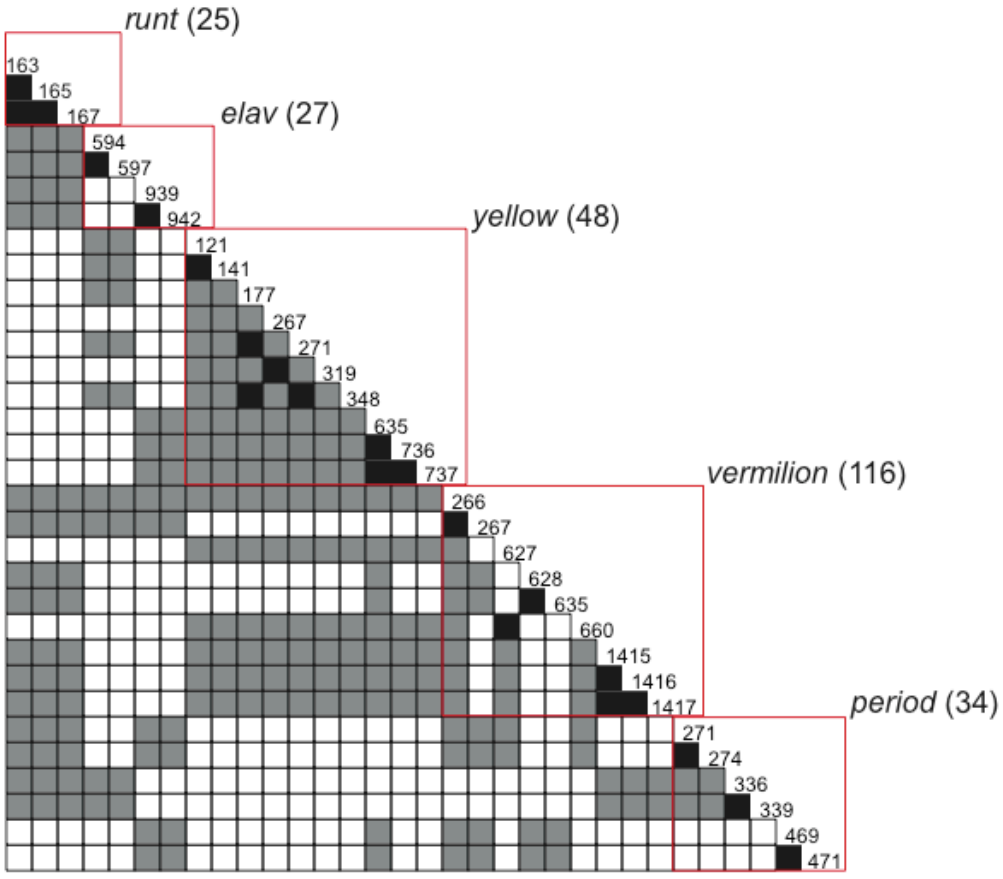
2. Shoemaker DD, Dyer KA, Ahrens M, McAbee K, Jaenike J. (2004) *Genetics* 168:2049-2058.

3. Kovacevic M, Shaeffer SW (2000) Genetics 156:155-172.

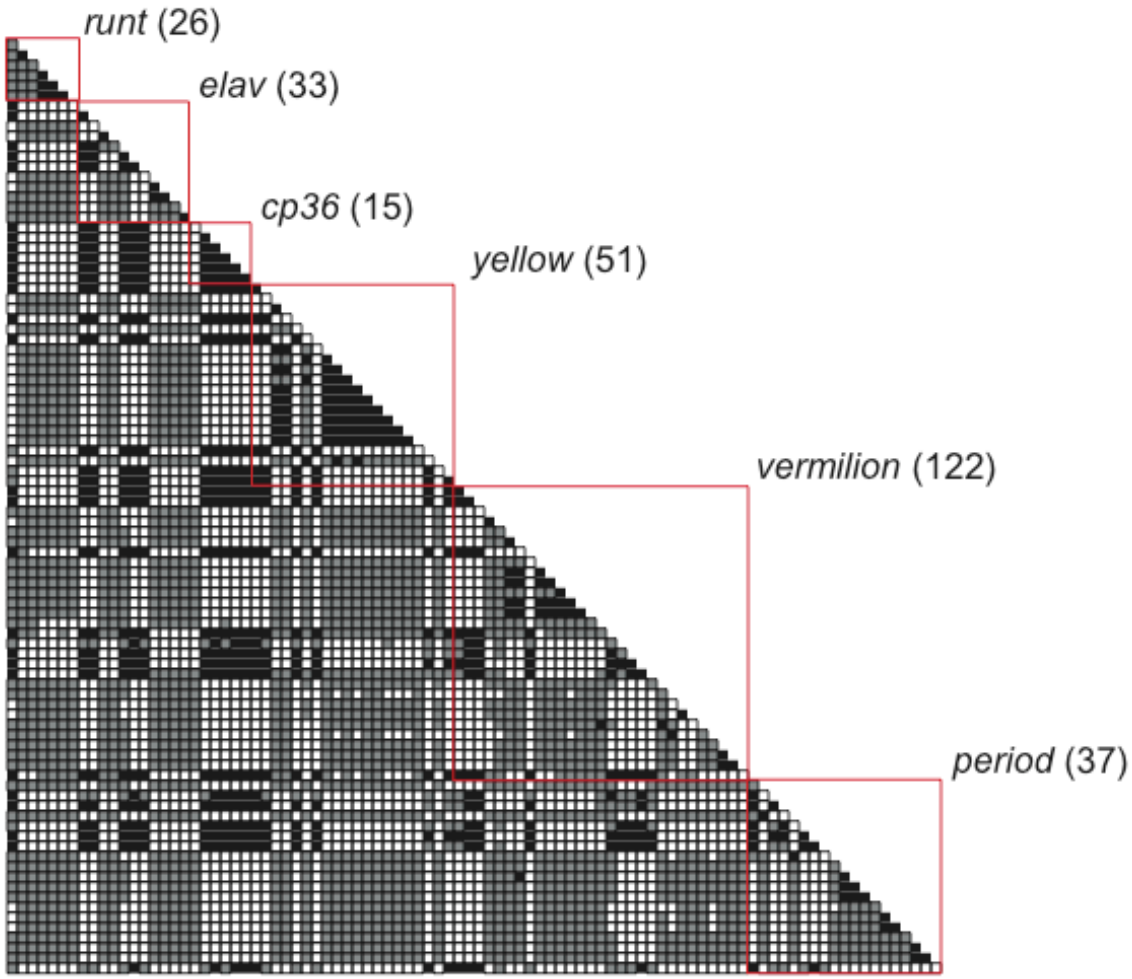
Supplemental Figure 5. Histogram of the offspring sex ratio of wild-caught males. Males were characterized as carrying an X^D if they produced ≥ 10 offspring, of which ≥ 90% were female; otherwise they were characterized as X^{ST}, as long as they sired at least 10 offspring. Only males that sired ≥ 10 offspring are included in the graph.



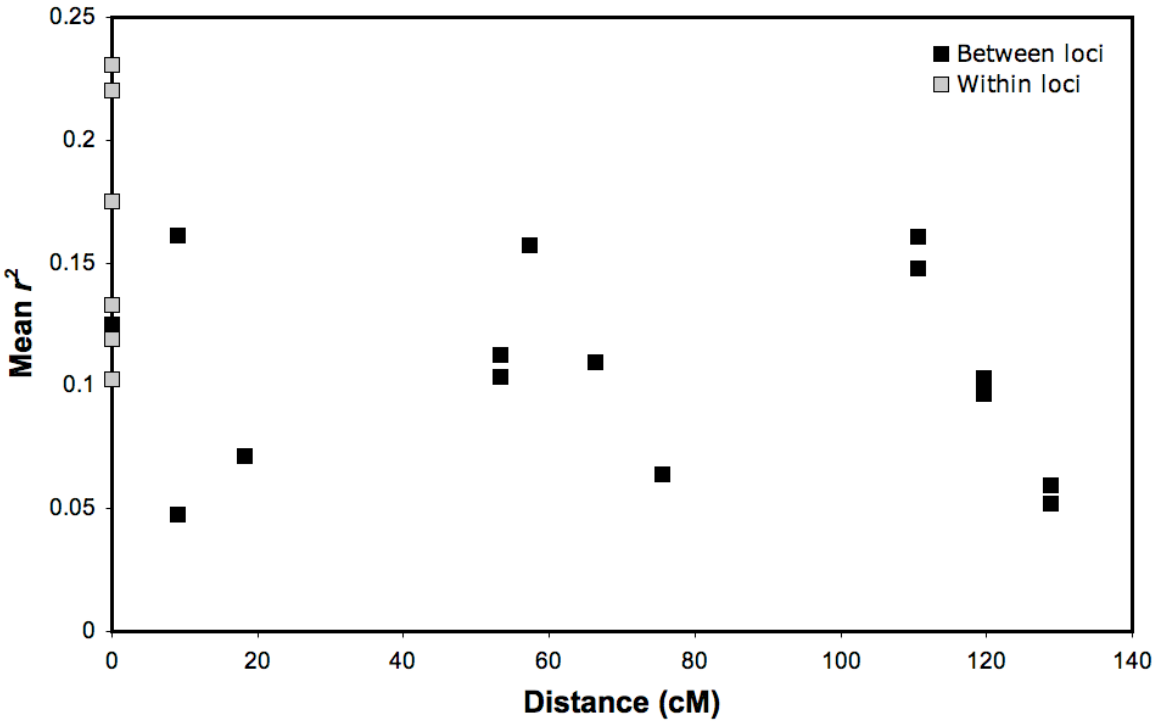
Supplemental Figure 6. Decay of linkage disequilibrium within X^{ST} chromosomes in *D. recens*. Using the concatenated dataset there were 259 informative segregating sites and 10,560 valid comparisons. After applying a sequential Bonferroni correction to account for multiple comparisons, 22 pairs of sites showed a significant association. All of these pairs of sites were within loci, indicating no signature of interlocus LD. The numbers in parentheses are the total numbers of informative segregating sites in each locus. Black squares indicate significant associations between pairs of segregating sites, and grey squares indicate valid comparisons that were not significant. Sites that did not show a significant association with any other site are not shown. *cp36* is not included in the figure because none of its 9 informative segregating sites showed significant association.



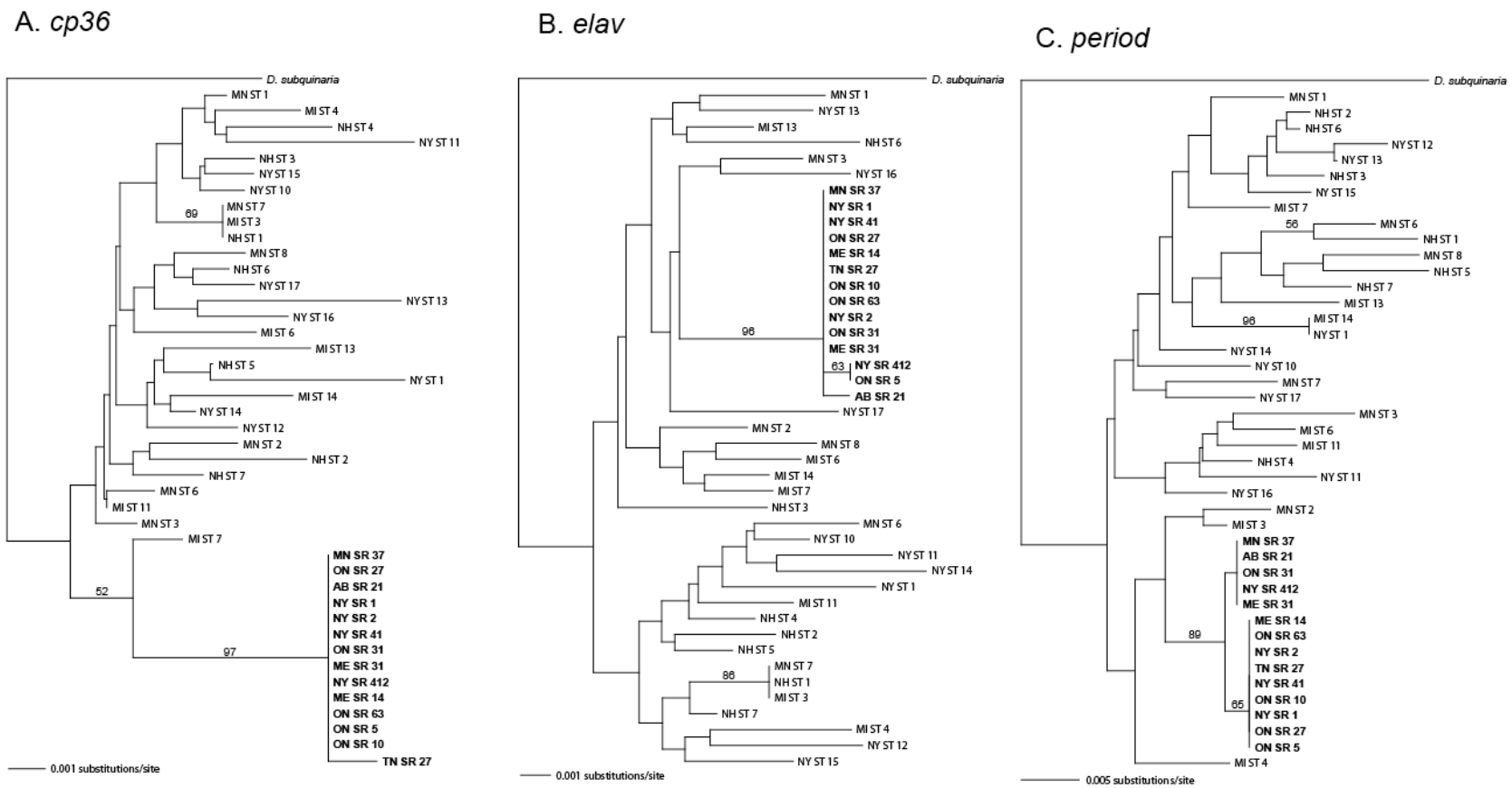
Supplemental Figure 7. Widespread linkage disequilibrium between X^{ST} and X^{D} chromosomes in *D. recens*. Using the concatenated dataset there were 284 informative segregating sites and 14,290 valid comparisons. After applying a sequential Bonferroni correction, 561 pairs of sites showed a significant association; 325 (58%) of these pairs are between loci, indicating significant LD between the X^{ST} and X^{D} chromosome types. The numbers in parentheses are the total numbers of informative segregating sites in each locus. Black squares indicate significant associations between pairs of segregating sites, and grey squares indicate valid comparisons that were not significant. Sites that did not show a significant association with any other site are not shown.



Supplemental Figure 8. Due to the complex set of inversions that differentiate the X^D and X^{ST} chromosomes, we do not know the gene order of the loci on the X^D chromosome; thus it is not entirely appropriate to determine how LD between types declines with distance. However, if we use the cM between loci on the X^{ST} as a proxy for distance, the mean r^2 (which measures LD between sites) for interlocus comparisons between X^D and X^{ST} does not significantly decline as distance between loci increases (Spearman's rho = -0.30, $P = 0.2769$). Furthermore, the fraction of pairs of sites that are in significant LD also does not decline significantly with this measure of distance (Spearman's rho -0.22, $P = 0.42$; graph not shown).



Supplemental Figure 9. Phylogenetic tree of each locus. A. *cp36*, B. *elav*, C. *per*, D. *runt*, E. *v*, F. *y*. The bold samples are those characterized as X^D. Each tree is a neighbor-joining tree with support values from 10,000 bootstrap samples. Sites with gaps were excluded from the analyses. Note that the X^{ST} samples most closely related to the X^D differ among genes, indicating that recombination has had time to shuffle diversity within the X^{ST} to alter the sequence of the ancestral X^{ST} haplotype.



Supplemental Figure 9 (cont).