Breeding and linkage

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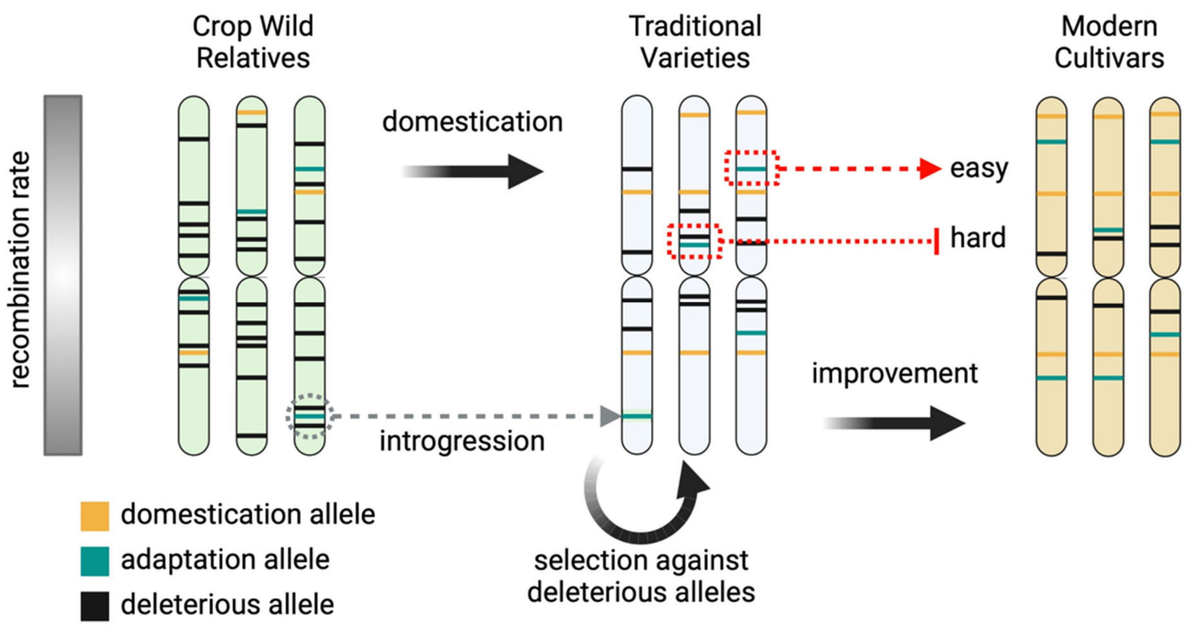
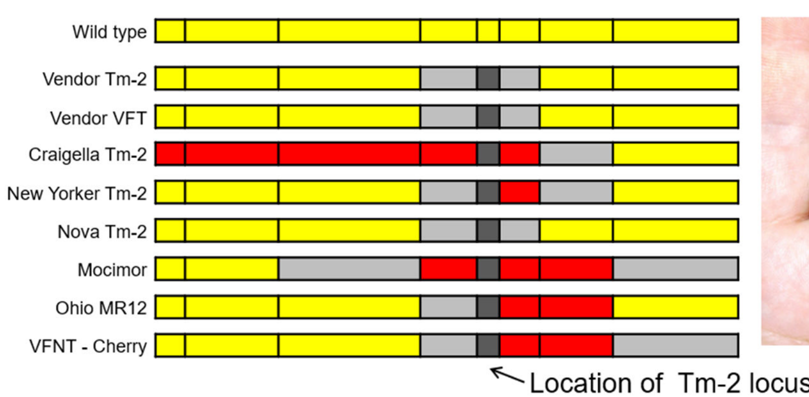


The need to break linkages

Young and Tanksley, 1989



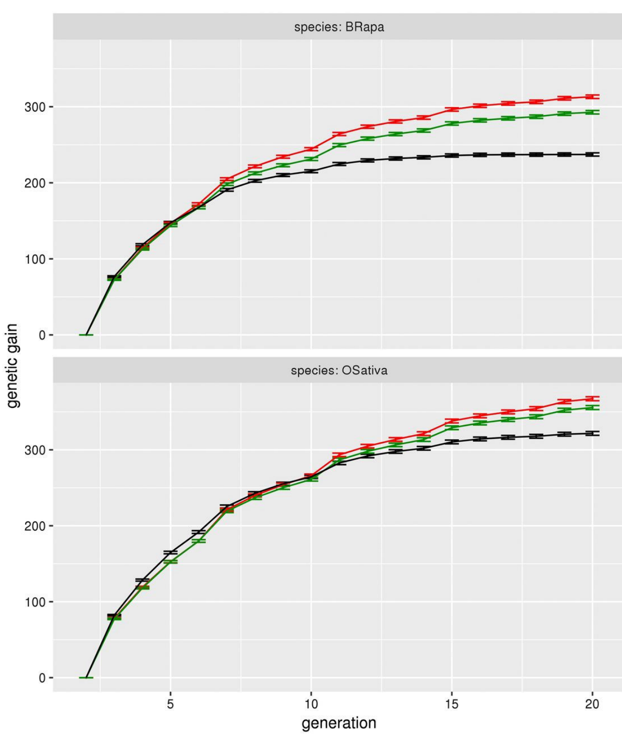
Red blocks illustrate DNA segments introgressed from the wild species donor of the Tm-2 locus. Lots of linkage drag is present.

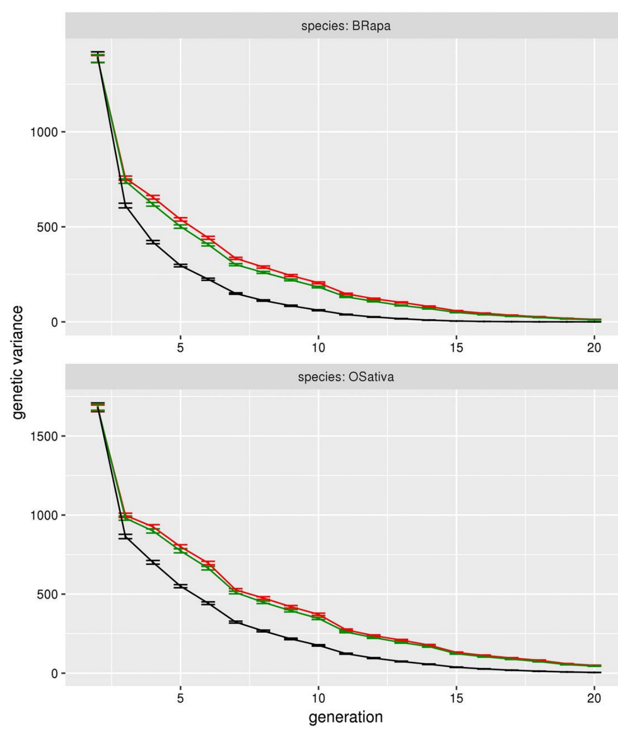
Significance of CO and its manipulation

Wijnker & de Jong, 2008

Increased recombination

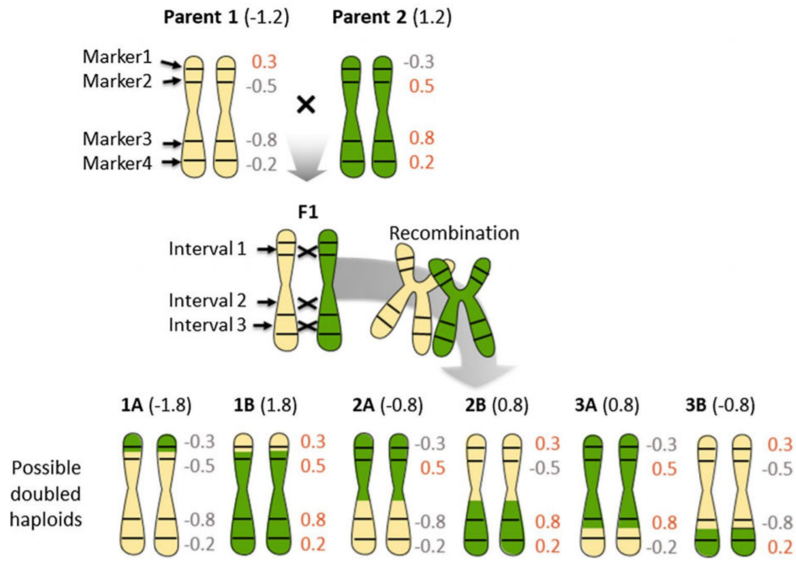
Tourette et al, 2019





Targeted recombination

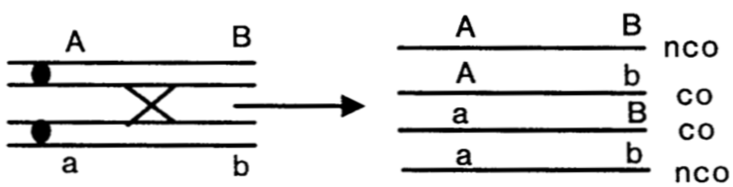
Ru & Bernardo, 2019



RG(x#1) =

RG(x#2) =

Recombination & genetic maps



Alfred Sturtevant

"In the latter part of 1911, in conversation with MORGAN . . ., I suddenly realized that the variations in strength of linkage, already attributed by MORGAN to differences in the spatial separation of the genes, offered the possibility of determining sequences in the linear dimension of a chromosome. I went home and spent most of the night (to the neglect of my undergraduate homework) in producing the first chromosome map."

Photo: Alfred Sturtevant, http://www.nap.edu/readingroom/books/biomems/asturtevant.html

3-point test cross

Data from **Hutchison, 1922**

Parents: C/C sh/sh Wx/Wx ✕ c/c Sh/Sh wx/wx
(purple; shrunken; starchy) (colorless; full; waxy)

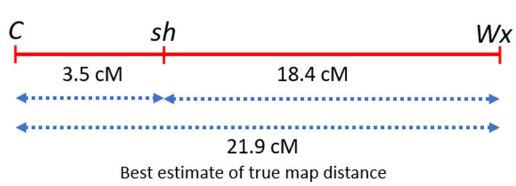
F_1 x tester: C/c sh/Sh Wx/wx ✕ c/c sh/sh wx/wx
(purple; full; starchy) (colorless; shrunken; waxy)

Figure 1. Nannas & Dawe, 2015

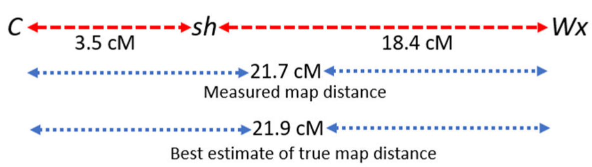
Measuring the distance

Kernel Phenotype	F_1 gamete genotype	Number
Purple, shrunken, starchy		2538
Purple, shrunken, waxy		601
Purple, full, waxy		116
Purple, full, starchy		4
colorless, full, waxy		2708
colorless, full, starchy		626
colorless, shrunken, starchy		113
colorless, shrunken, waxy		3
Total		**6708**

Kernel Phenotype	F_1 gamete genotype	Number
Purple, shrunken, starchy		2538
colorless, full, waxy		2708
Purple, shrunken, waxy		601
colorless, full, starchy		626
Purple, full, waxy		116
colorless, shrunken, starchy		113
Purple, full, starchy		4
colorless, shrunken, waxy		2
Total		**6708**

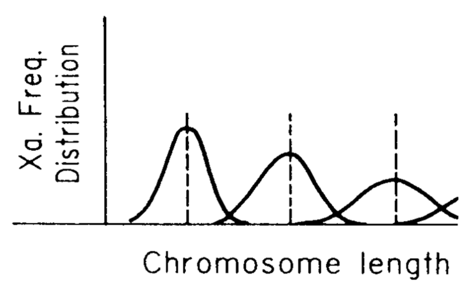


Accounting for unseen DCOs



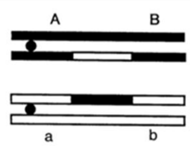
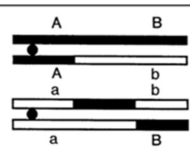
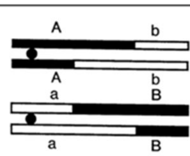
Interference

Muller, 1916; McPeek and Speed, 1995



2, 3, and 4-strand DCOs

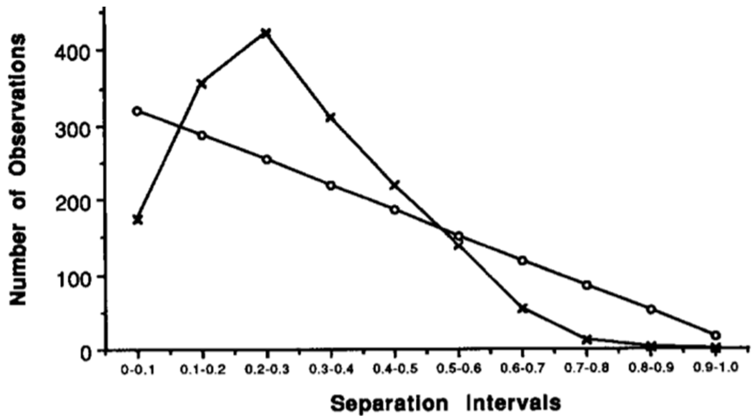
A 2-plane model used for clarity (not to endorse the 2 plane model!)

		Actual	Observed
2-strand DCO 		NCO	NCO
		DCO	
		DCO	
		NCO	
3-strand DCO 		NCO	50% CO
		SCO	
		DCO	
		SCO	
3-strand DCO 		SCO	50% CO
		DCO	
		SCO	
		NCO	
4-strand DCO 		SCO	100% CO
		SCO	
		SCO	
		SCO	

DCO graphics from http://www.mun.ca/biology/desmid/brian/BIOL2250/Week_Five/CXORF.jpg

Interference vs chromosome position

Sherman and Stack, 1995



Xmata vs recombination maps

Sherman and Stack, 1995

Chromosome	Length based on xma	Length based on classical map	Length based on molecular map
1	124.0	161	131.5
2	104.0	74	124.2
3	105.0	111	126.1
4	94.5	89	124.6
5	83.5	55	97.4
6	86.5	113	101.9
7	88.5	71	91.1
8	84.0	67	96.9
9	79.0	62	111.0
10	83.0	132	90.1
11	83.0	97	88.0
12	79.5	31	93.1
Total	1094.5	1063	1275.9

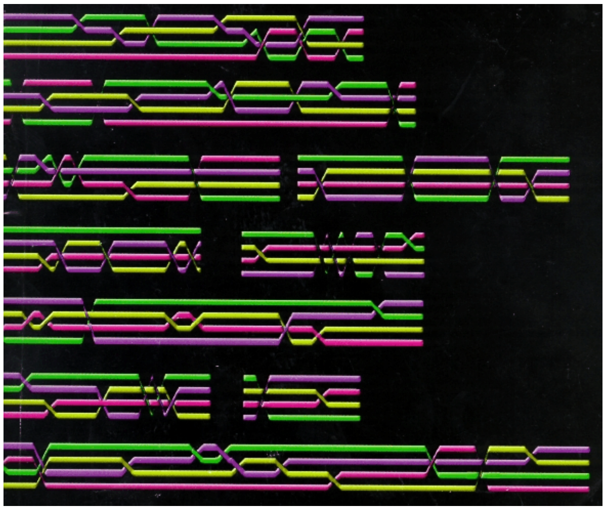
Nilsson et al., 1993

Chromosome of maize	Genetic length (cM) estimated from					
	Chiasma Counts	Linkage data				
		1934	1950	1976	1990 classical	1990 molecular
1	187	102	156	161	176	238
2	163	58	128	155	155	229
3	150	92	121	128	167	194
4	148	80	111	143	137	174
5	148	44	72	87	107	235
6	110	52	64	68	78	169
7	123	50	96	112	112	131
8	123	20	28	28	42	173
9	110	52	71	138	140	132
10	98	68	57	99	95	115

Why are molecular maps longer than chiasma maps?

Species	2n	Estimated CO/meiosis	
		xma counts	molecular maps
Brassica campestris	20	10.0-18.5	37.0
Brassica oleracea	18	12.8-14.8	22.2
Hordeum vulgare	14	13.5-15.6	22.7
Lactuca sativa	18	14.6-20.7	28.1
Lycopersicon esculentum	24	16.2-17.0	25.5
Oryza sativa	24	18.9-27.6	36.7
Pisum sativum	14	10.3-18.1	29.3
Solanum tuberosum	12/24	13.2-14.1	20.7
Zea mays	20	17.4-25.0	35.8

CO's assumed in mapping functions



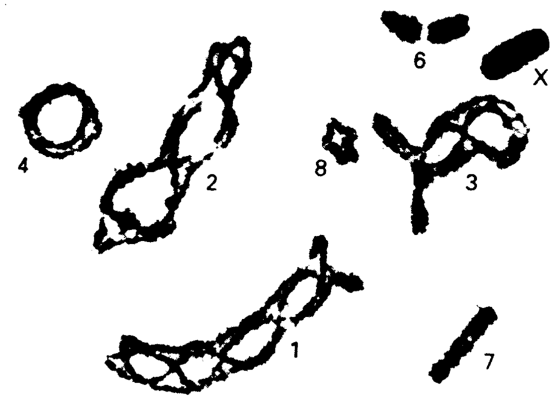
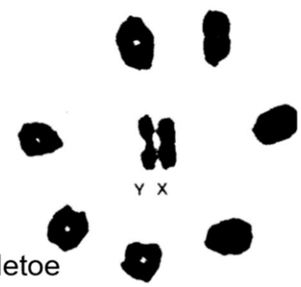


Figure 2. Diplotene in male Meadow grasshopper, Chorthippus parallelus

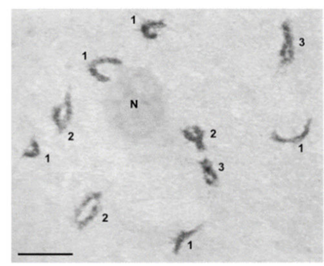
Figure 3. Crossovers in yeast, as determined by allele detection on a DNA chip

How common are DCOs in plants?

Barley



Mistletoe



Maize- Anderson et al. 2003

Tanksley et al., 1989

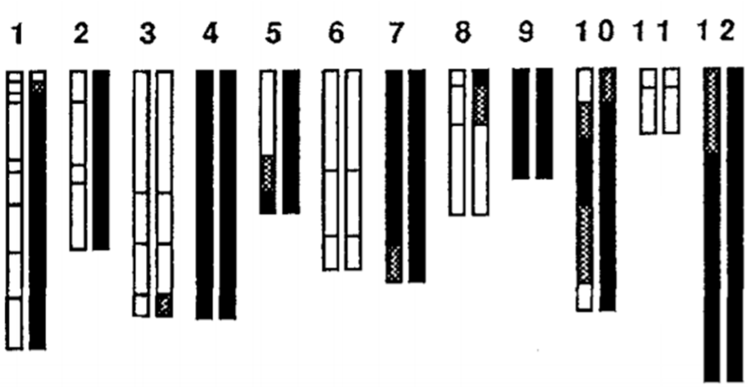
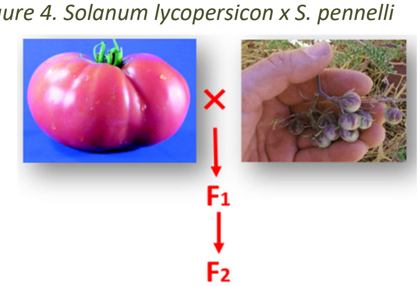


Figure 4. Solanum lycopersicon x S. pennelli

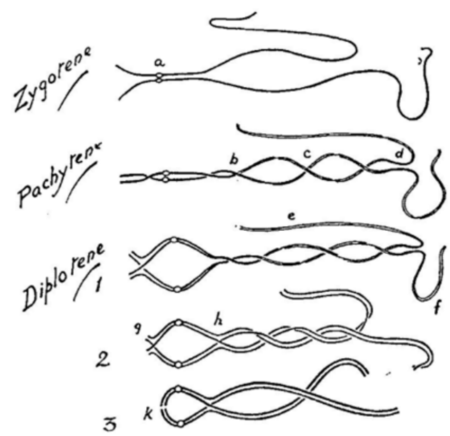


Sherman and Stack, 1995

- Counted xma in 5228 paired chromosomes of tomato

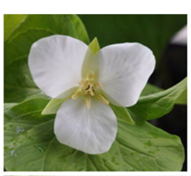
Terminalization

Darlington 1935



Localized Chiasmata


PMC of *Paeonia lutea*, 2n = 10 with distal chiasmata. John, 1990

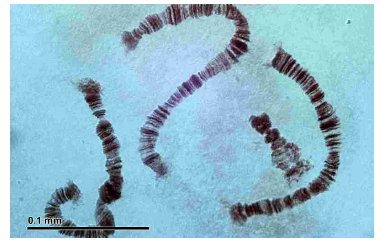



Species:	Chiasmata Site:			% Proximal Site
	Proximal	Interstitial	Distal	
Trillium kamtschaticum				
Clone 1	466	5	2	98
Clone 2	407	33	20	89
Allium fistulosum	2056	41	40	96

Recombination is variable

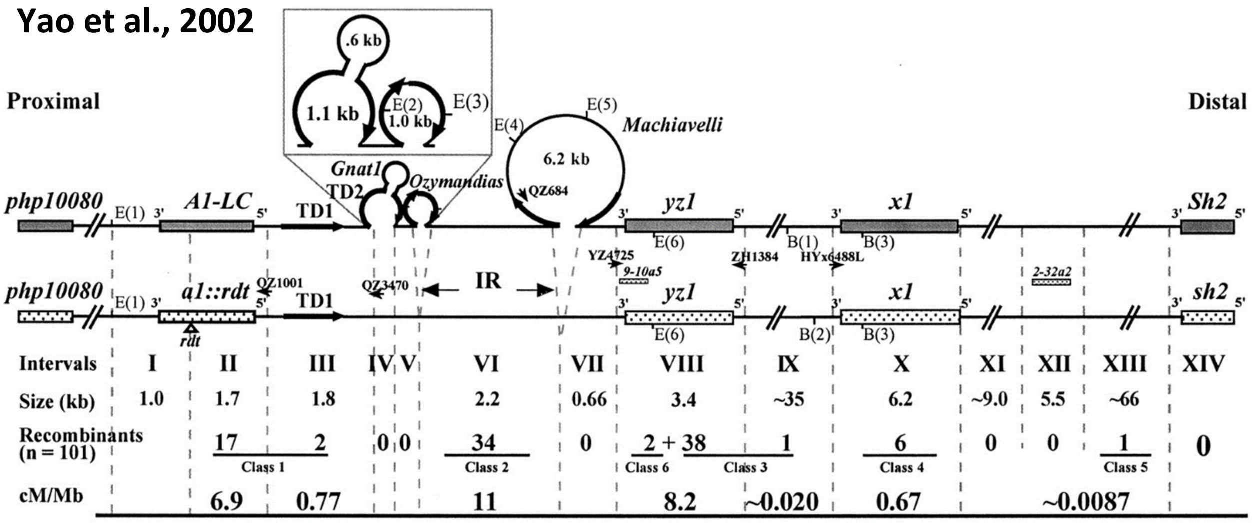
Data from Bridges, from the *Drosophila* sex chromosome
Order of genes = *y* - *pr* - *w* - *rst* - *fc* – centromere

Gene pair:	# Bands:	Map distance:	%CO / band
y – pr	57	0.8	0.014
pr – w	18	0.7	0.038
w – rst	2	0.2	0.100
rst – fc	2	1.3	0.650

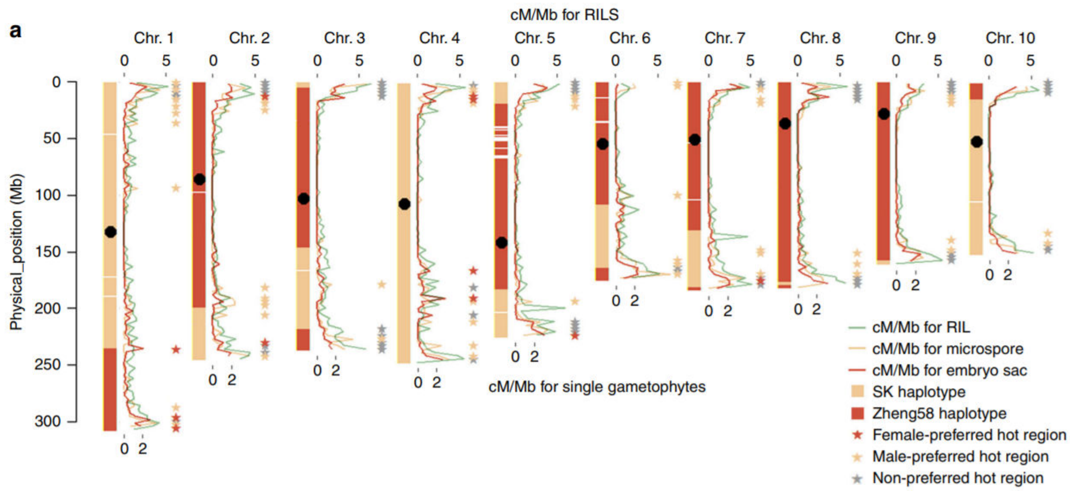


Recombination hotspots

Yao et al., 2002

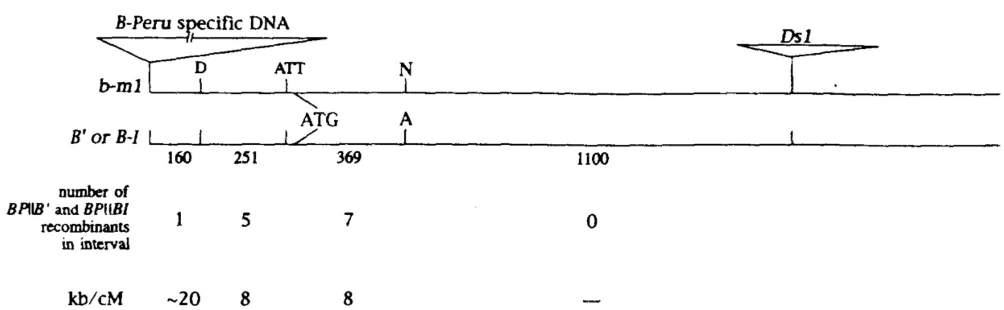


Luo et al., 2019

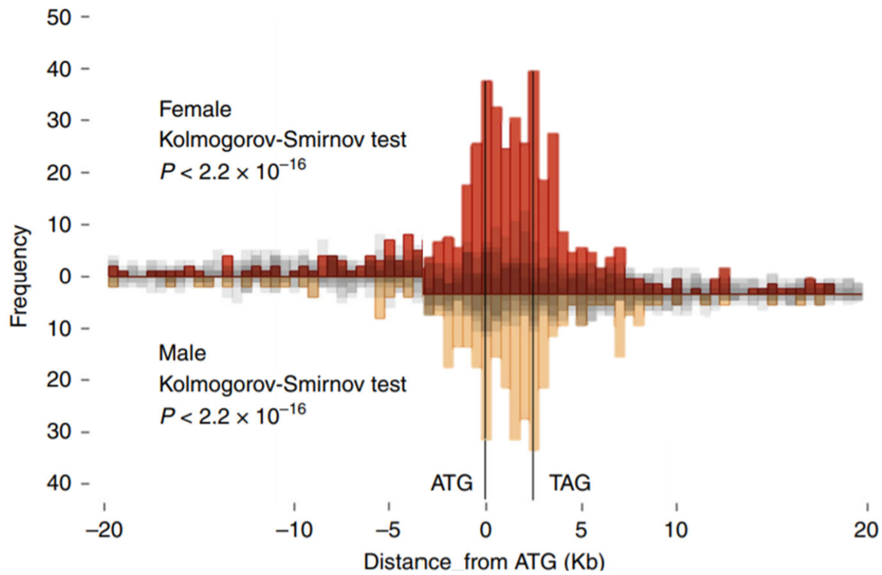


Recombination can vary within a gene, with hotspot near ATG

Patterson et al., 1995

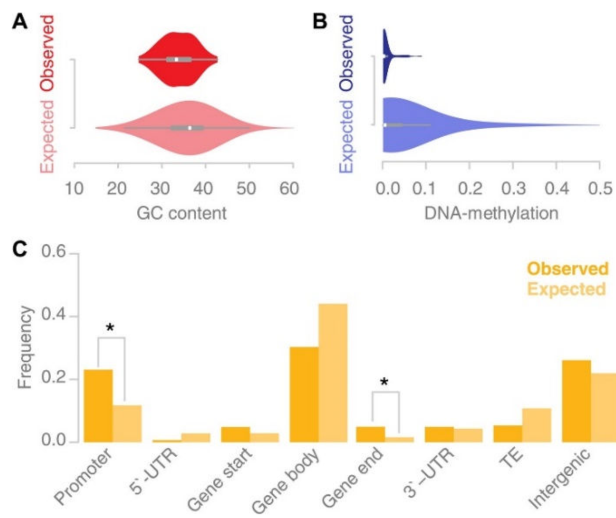


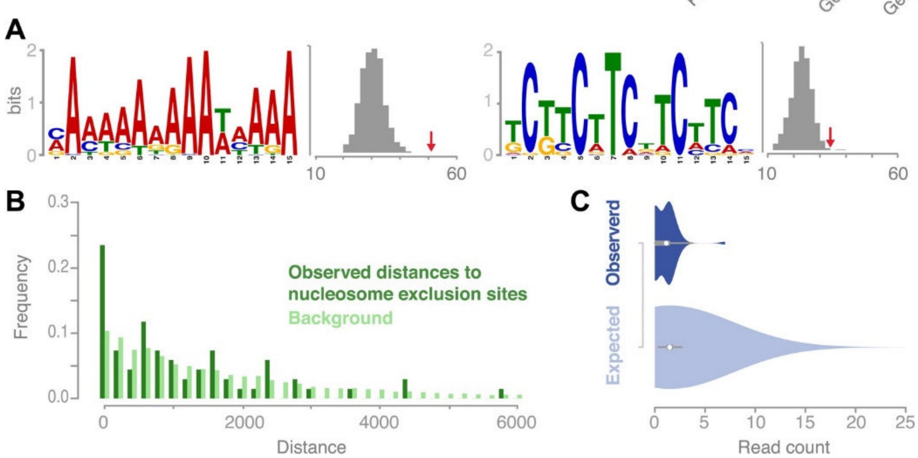
Luo et al., 2019



Preferred recombination spots

Wijnker et al., 2023

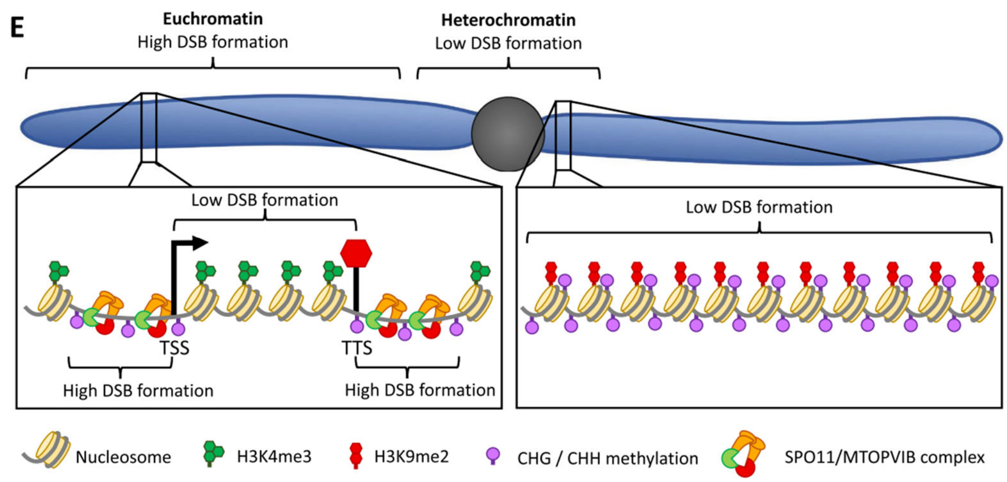




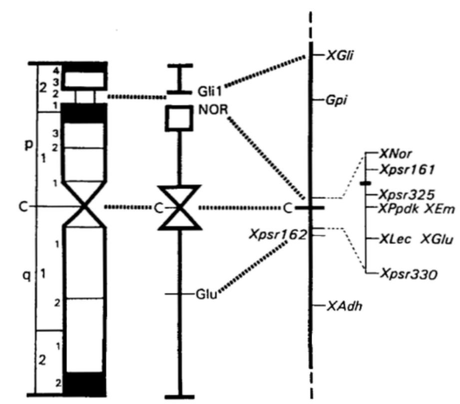
Nucleosome exclusion sites are defined as $(A)_{10}$ and $((GC)_3NN)_3$

Model of CO locations

Lloyd, 2022



Physical vs genetic maps



Heslop-Harrison, 1991

Crossover sites can be altered

Jones, 1967

Secale dighoricum x *S. turkestanicum*

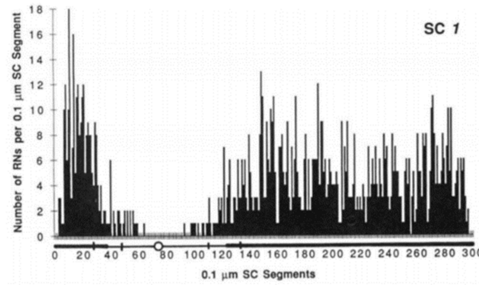
Chiasmata	Parental		F_2	
Distribution:	Observed	Expected	Observed	Expected
Distal	546	192	199	134
Interstitial	30	192	132	134
Proximal	0	192	71	134
Total	576		402	

Factors Affecting Recombination

Landmarks/Chromosome Structure

a. The centromere

Sherman and Stack, 1995

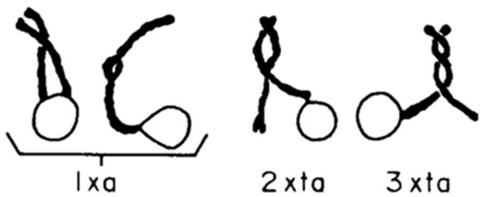


Location of COs in chromosome 1 of tomato

b. Heterochromatin

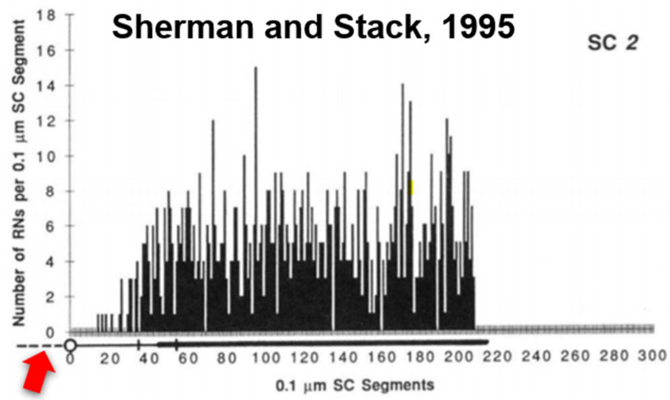
		Euchromatin			Heterochromatin		
C-some	Length (µm)	Length (µm)	Ave no. RNs / II	RNs / µm	Length (µm)	Ave no. RNs / II	RNs / µm
1	30.0	22.5	2.44	0.112	7.5	0.04	0.005
2	21.3	17.1	2.05	0.122	4.2	0.03	0.007
3	23.1	16.0	2.07	0.131	7.1	0.03	0.004
4	20.8	13.7	1.87	0.138	7.1	0.02	0.003
5	16.2	9.5	1.63	0.168	6.7	0.04	0.006
6	18.5	12.9	1.69	0.132	5.6	0.04	0.007
7	18.5	11.6	1.75	0.155	6.9	0.02	0.003
8	18.5	11.9	1.66	0.143	6.6	0.02	0.003
9	16.2	10.0	1.58	0.160	6.2	0.004	0.0006
10	16.2	10.0	1.64	0.160	6.2	0.02	0.003
11	16.2	9.7	1.63	0.165	6.5	0.03	0.005
12	14.0	8.2	1.54	0.183	5.8	0.05	0.009

c. The NOR



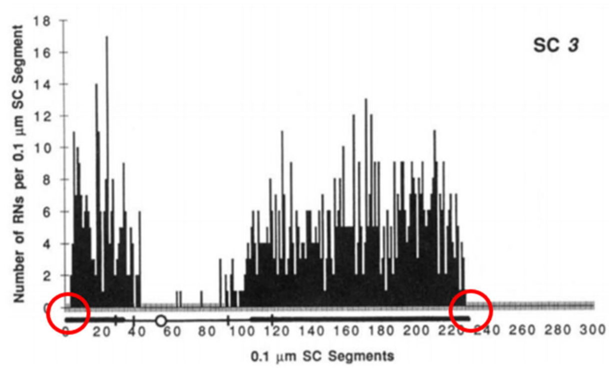
COs in *Eremurus spectabilis*

Upcott, 1936 via **John, 1990**



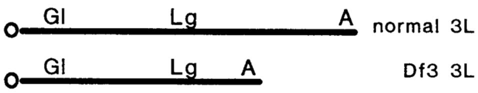
d. Telomeres

Sherman and Stack, 1995; Barton et al, 2008



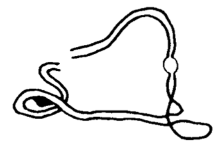
e. Interchromosomal effects

Rhoades, 1958

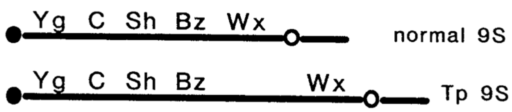


Cross	*Gl – lg*	*Lg – A*
N3L / N3L	28.0	30.6
N3L / N3L*	29.0	28.2
N3L / D3L	35.1	12.8

Marcus Rhoades
1903 - 1991



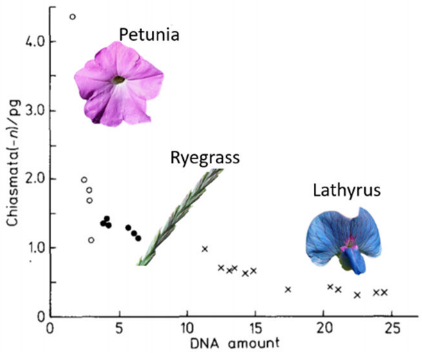
An indel during pairing



Cross	*Yg – Sh*	*Sh – Wx*
N9 / N9	21.0	17.0
Tp9 / N9	2.0	2.0
Tp9 / Tp9	29.0	18.0

f. Chromosome length

Rees & Durant, 1986; Sherman & Stack, 1995; Anderson et al., 2003



Rees & Durrant, 1986

Table from John, 1990: Species	2C DNA (pg)	DNA content (pg) of II with means of:		
		1.5 xta	2 xta	3 xta
Lathyrus clymenum	13.43	1.0	1.4	2.2
L. cicero	14.64	1.3	1.8	2.8
L. sativus	16.78	2.1	2.7	3.8
L. tinitanus	22.08	2.4	3.1	4.4
Lolium perenne	4.16	0.36	0.8	--
L. temulentum	6.23	0.51	1.2	--

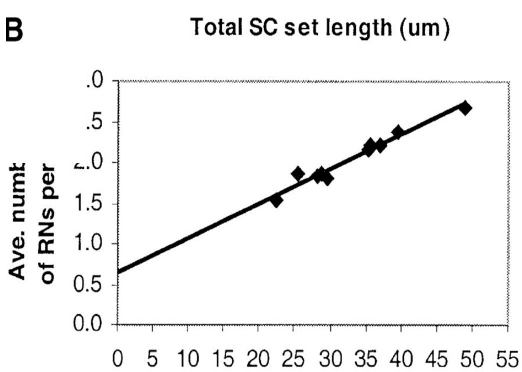


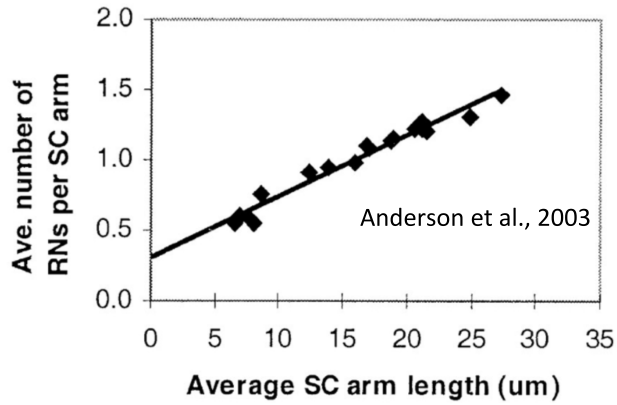
Figure 5. Anderson et al, 2003

- The following data are from tomato (***Sherman and Stack, 1995***):

Chromosome	Mean Length	CO/Chromosome	Unit Length per CO
1	30.0	2.48	12.10
2	21.3	2.08	10.24
3	23.1	2.10	11.00
4	20.8	1.89	11.00
5	16.2	1.67	9.70
6	18.5	1.73	10.69
7	18.5	1.77	10.45
8	18.5	1.68	11.01
9	16.2	1.58	10.25
10	16.2	1.66	9.75
11	16.2	1.66	9.76
12	14.0	1.59	8.80

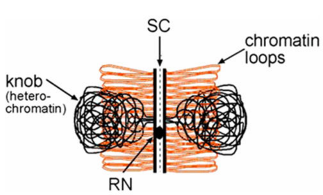
g. Arm length

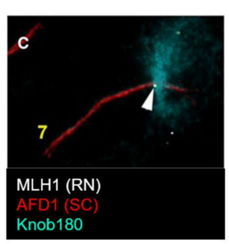
Sherman and Stack, 1995



h. Knobs

Naranjo and Lacadena, 1980; Stack et al., 2017

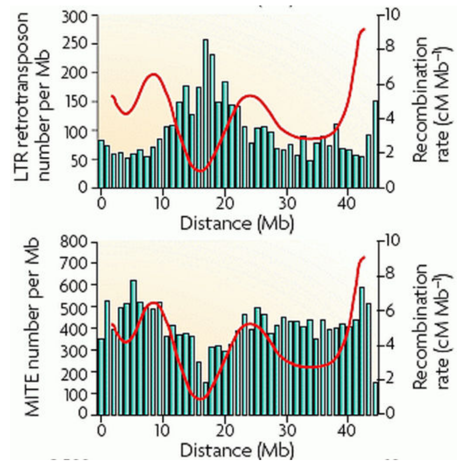




SC	SCs obs	No. MLH1 foci			No. of MLH1 foci per µm of SC length		
		SC	Near knob	In knob	SC	Near knob	In knob
5	89	139	7	4	0.042	0.039	0.026
6	72	96	8	3	0.043	0.056	0.052
7	94	132	10	9	0.046	0.053	0.056
Total	255	367	25	16			

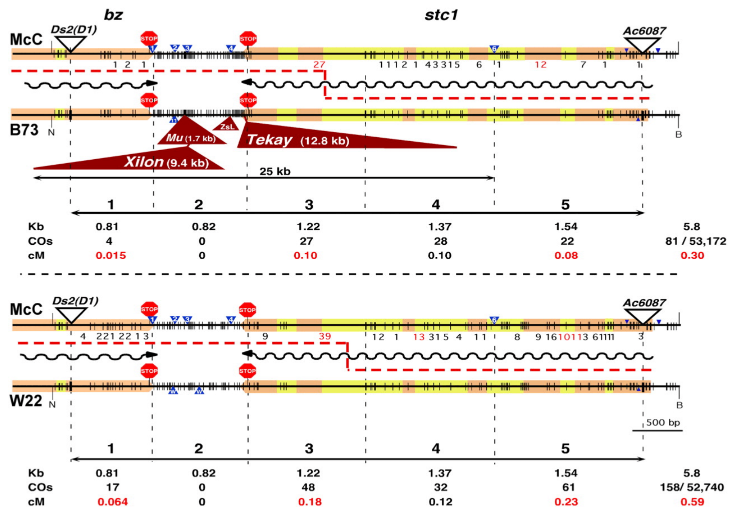
i. MITEs

Gaut et al., 2007



j. Retrotransposons

Dooner & He, 2008



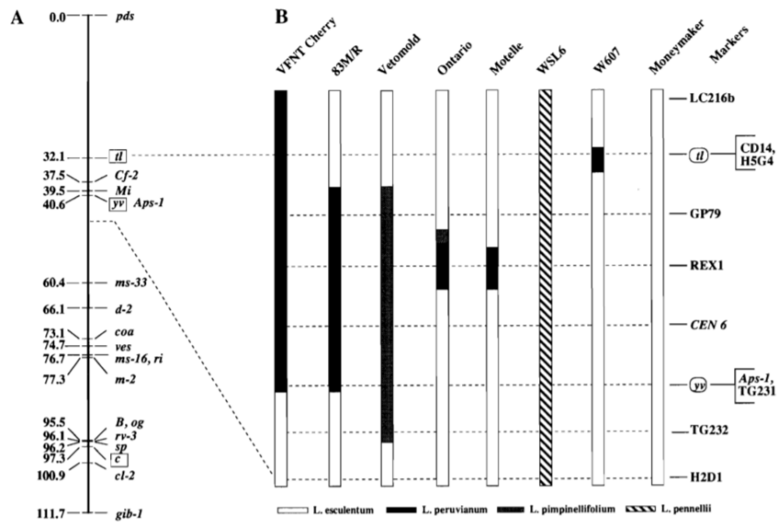
k. Gene density

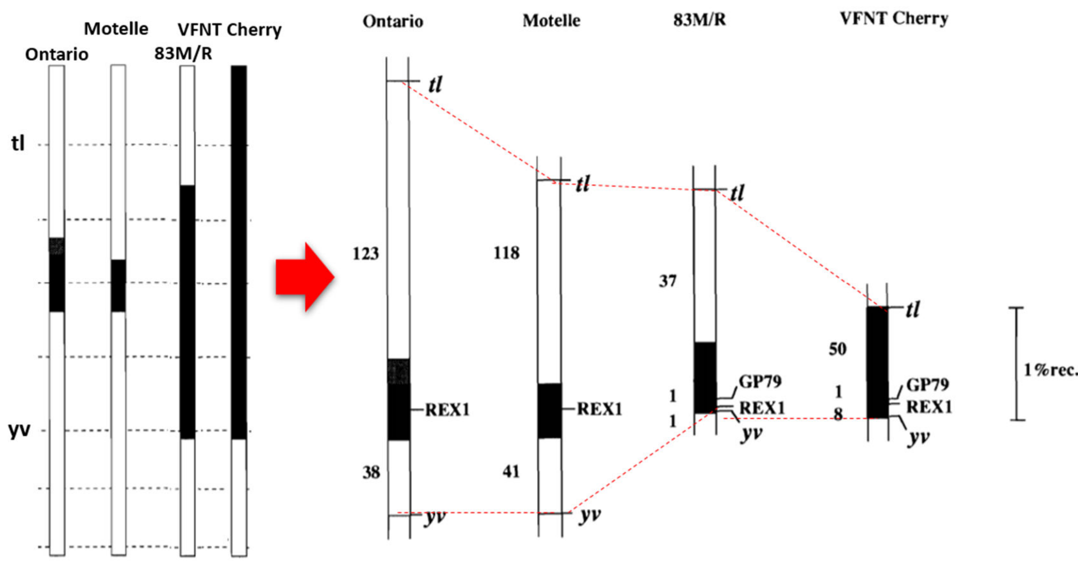
Fengler et al., 2007

Maize Chromosome	Genic sequences	Genetic length (cM)	Correlation between genes and recombination
1	3357	1137.9	0.96
2	2619	725.3	0.95
3	2478	829.9	0.90
4	2286	750.2	0.83
5	2618	676.7	0.93
6	1721	548.7	0.92
7	1757	618.4	0.85
8	1970	632.0	0.77
9	1608	638.7	0.93
10	1432	533.7	0.78
total/Ave	21,846	7090.0	0.87

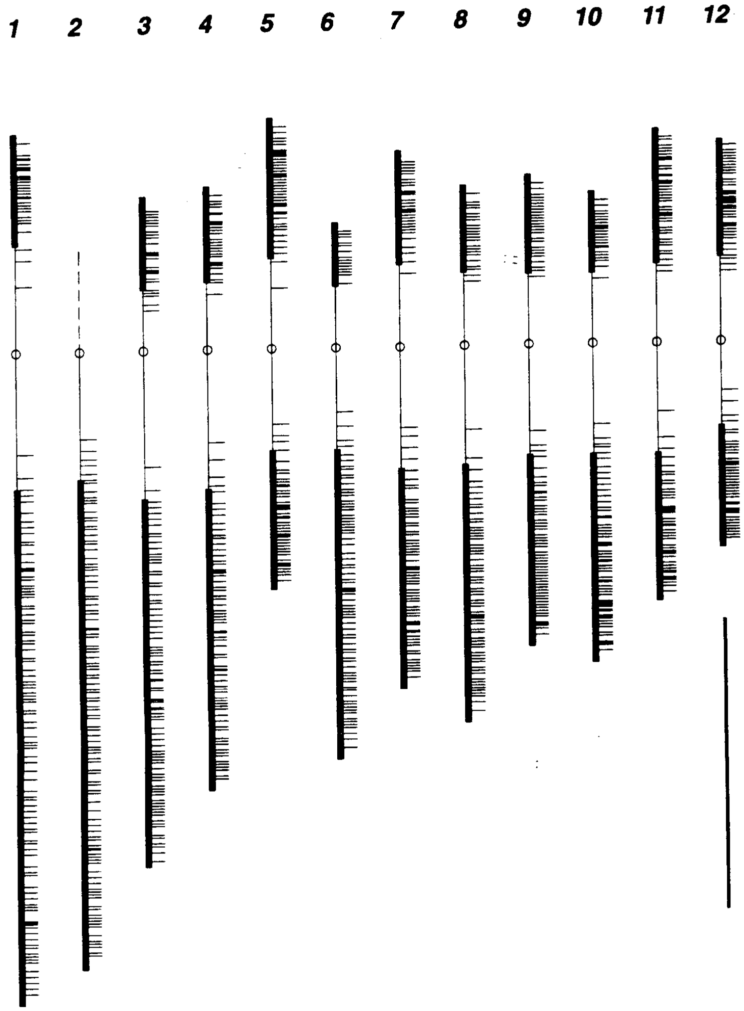
I. Alien introgressions

Liharska et al., 1996





Effect of CO variability on linkage maps



Sherman & Stack, 1995

Demarly, 1979

Factors Affecting Recombination

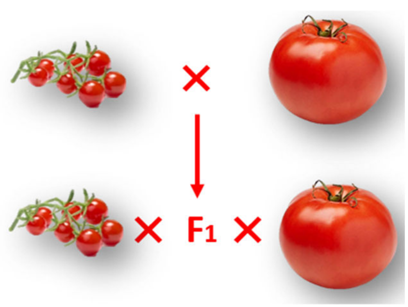
Genetics

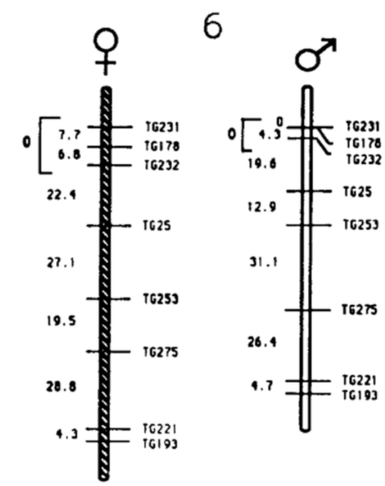
A. Gender - Heterochiasmy

John 1991

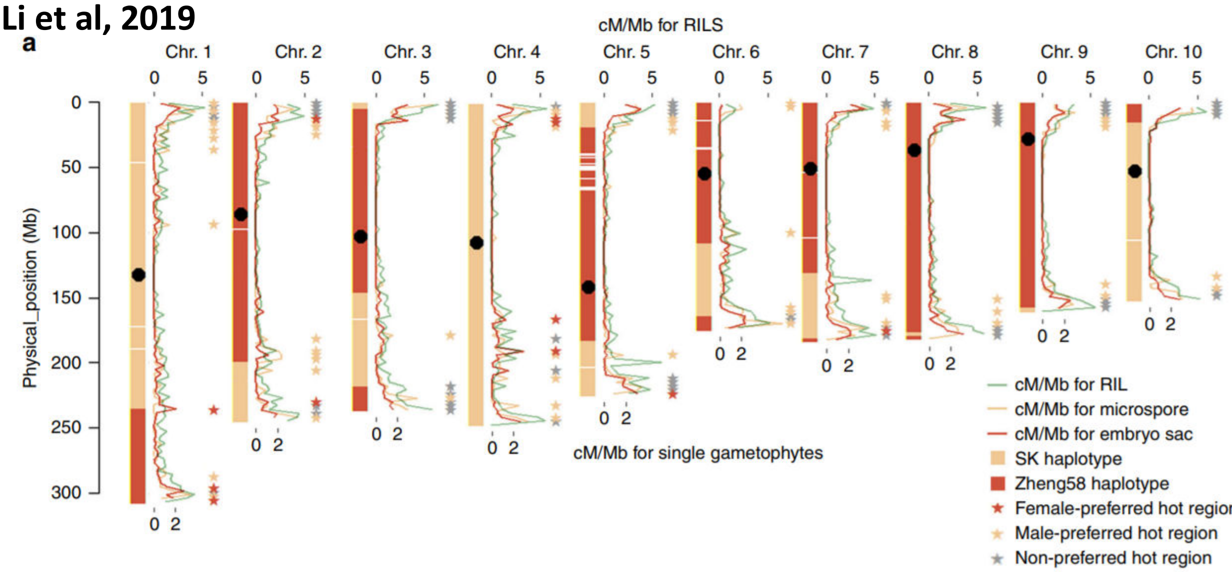
	Xma freq.		Dist'n	
	♀	♂	♀	♂
Fritillaria martagon	41.0	36.3		
F. meleagris	37.8	24.8		
F. longiflorum	31.4	27.3		
Allium nigrum	16.9	21.9	R*	R
A. consanguineum	17.5	21.9	R	R
A. cepa	17.9	22.4	R	R
A. kachrooi	15.0	12.9	P*	R

de Vicente & Tanksley, 1991



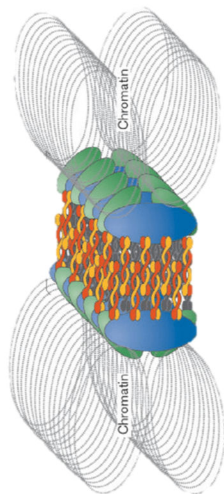


Li et al, 2019

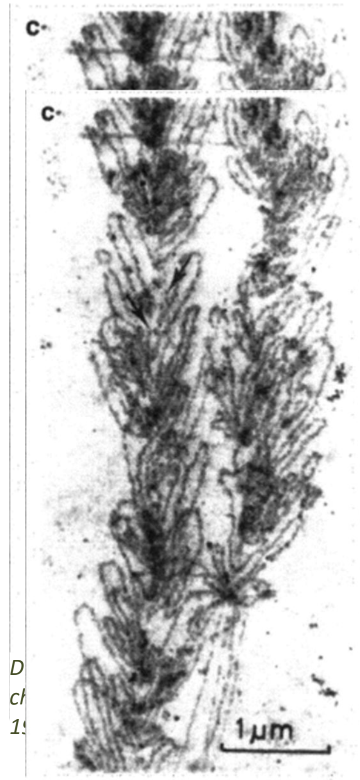


Heterochiasmy

Zickler & Kleckner, 1999; Koul & Nagel, 2002; Giraut et al, 2011; Lloyd, 2022



Popa. 2011



DNA loops on bumblebee pachytene chromosomes. Zickler & Kleckner, 1999

Koul & Nagel, 2002;

Säll & Nilsson, 1994

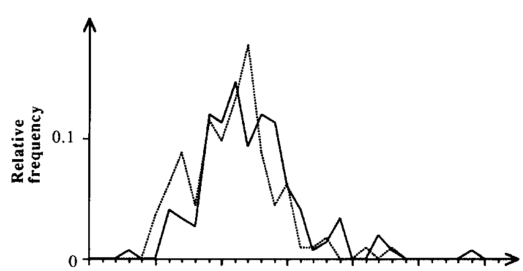


Figure 6. Comparison of CO in ♂ (dotted) and ♀ (solid) meiosis of barley

B. B chromosomes

John, 1990

Tp9/Tp9	*Yg-C*	*C-Wx*
+ 0 B	28.8	17.7
+ 1 B	12-13	37.0
+ 2 B	12-13	40.4
+ 3 B	12-13	42.0

	Mean chiasmata/cell									
Species:	0B	1B	2B	3B	4B	5B	6B	7B	8B	10B
Crepis capillaris	↑ 4.08	3.90	4.65	4.68	5.33					
Puschkinia libanotica	↑ 9.07	10.82	11.70	11.62	13.80					
Lolium perenne	↓ 11.93	11.21	10.00							
Festuca mairei	↑ 18.87	20.95	22.85	26.25	28.00					
Zea mays	↑↓ 18.50		19.50	19.70	19.80		22.53		23.80	19.80
Secale cereale -inbred	↓ 14.85	13.43	13.22	13.29	12.64	12.86	13.20	12.64	13.03	
- wild	↑ 13.20	15.12	16.57	17.78	18.40					
Listeria ovata - PMC	↑ 26.9	28.9	28.2	30.3	29.1					
- EMC	↑ 30.3	32.6	31.3	32.5	32.7					

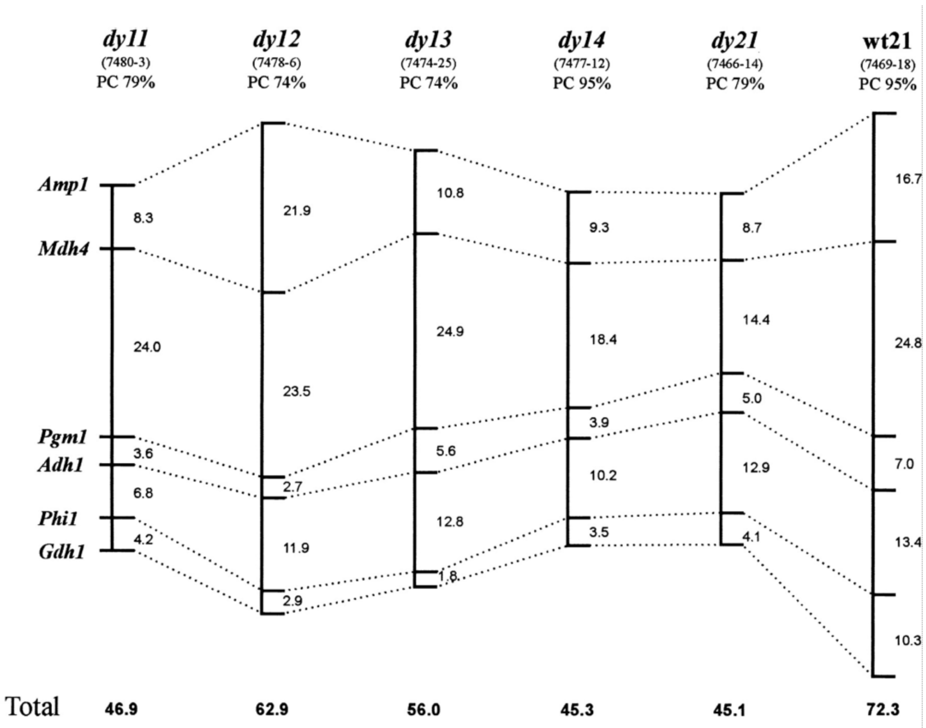
C. Genes

Robert, Farcy, & Cornu, 1991

	rm1/rm1	*Rm1/rm1*	*Rm1/Rm1*
Hf1-Lg1	15.3%	26.9%	
An2-Rt	0-0.5%	6%	26%

D. Desynaptic mutants

Ji et al., 1999



E. Species

Price et al., 1993

Interval	Distance (cM) Tomato	Pepper
CT268 - TG273	23.0	5.7
TG197 - TG158	52.2	34.6
r45S - TG31	2.8	5.0
TG48 - CD30A	15.6	6.0
CT128A - CT166	28.9	20.3
TG366 - TG244	86.3	75.3
TG264 - TG574	15.5	15.4
TG363 - CD64	18.3	23.2
TG623 - TG379	49.0	0.0
TG232 - TG253	46.3	51.9
TG20 - TG499	10.6	9.2
TG201 - TG496	10.6	9.2
TG47 - TG400	6.4	13.7
TG618 - TG296	39.9	0.0
TOTAL:	441.2	282.0

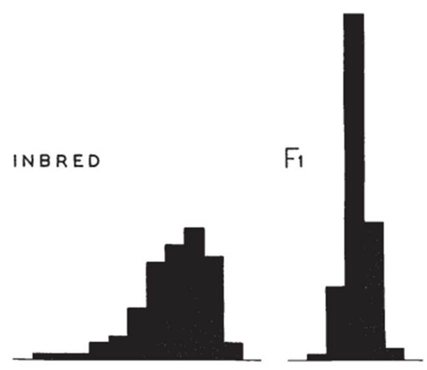
F. Zygosity

Robbins et al., 1995

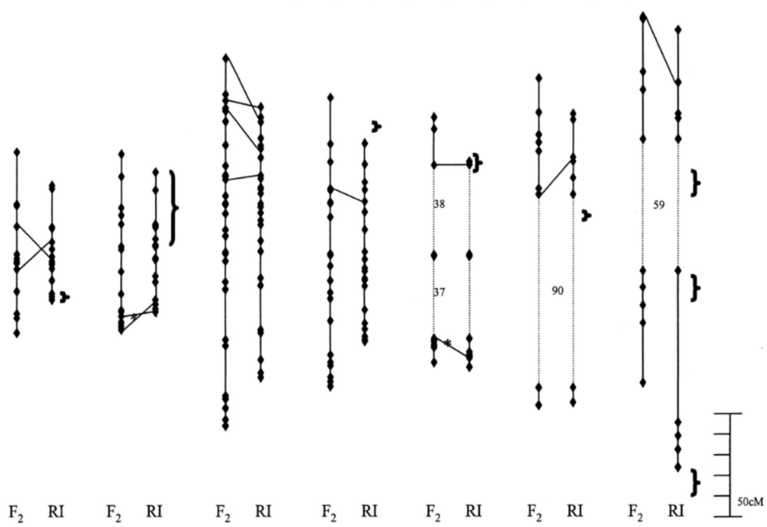
Case I: homozygosity increases CO

Rees & Thompson, 1956

Case II: homozygosity decreases CO

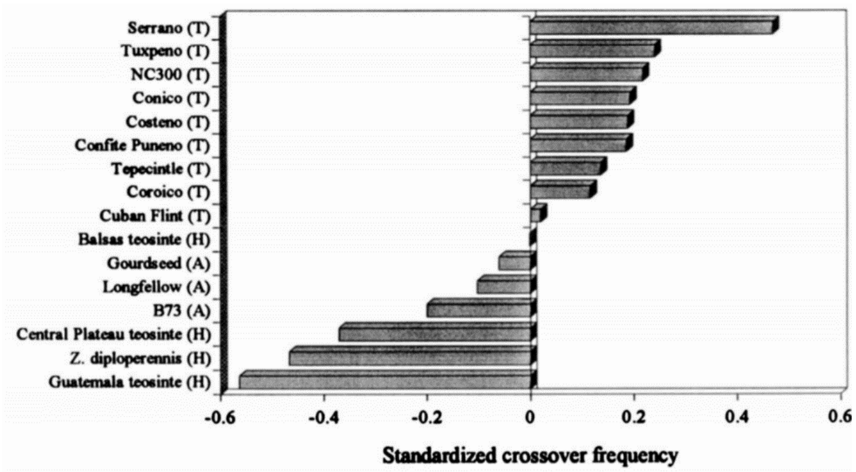


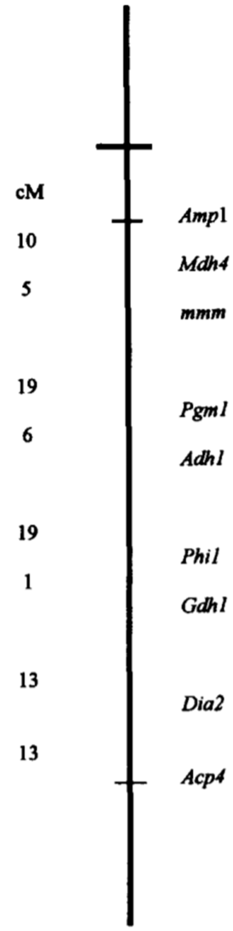
Knox & Ellis, 2002



G. Genotype

Williams, Goodman, and Stuber, 1995

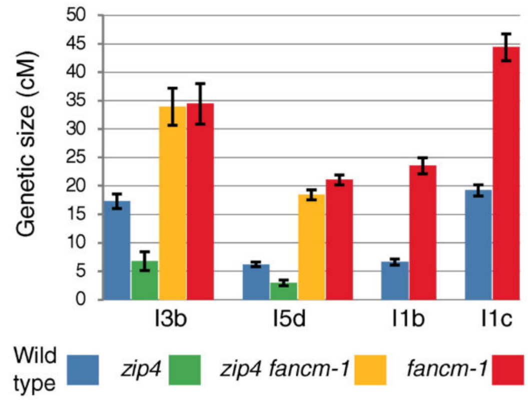




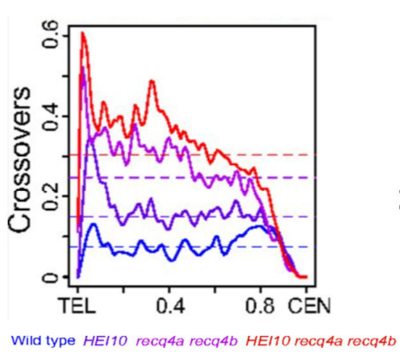
MAP	amp1 - mdh4	mdh4 - pgm1	pgm1 - phi1	phi1 - dia2	dia2 - acp4	Total cM ×1000
Wendel (1989)	**15.0**	**19.0**	**25.0**	**14.0**	**13.0**	**86.0**
Composite	**9.7**	**15.9**	**17.4**	**14.4**	**12.7**	**70.2**
Composite US genotypes	**12.4**	**19.3**	**15.0**	**9.3**	**10.5**	**66.5**
B73	8.5	16.2	11.7	11.6	9.4	57.4
Gourdseed	6.9	17.5	19.2	11.1	12.3	66.9
Composite Exotic genotypes	**11.8**	**19.1**	**17.6**	**15.7**	**12.9**	**77.2**
NC300	12.1	17.4	18.9	14.7	15.0	78.1
Serrano	13.0	25.1	20.6	22.8	13.0	94.5
Tepecintle	11.0	17.9	12.6	20.9	12.3	74.7
Tuxpeño	12.5	21.2	21.6	8.0	15.1	78.3
Confite Puneño	16.7	23.0	18.7	7.1	11.5	76.9
Cónico	6.3	17.7	11.1	19.1	20.4	74.6
Coroico	15.4	13.2	15.6	19.9	10.8	74.9
Costeño	10.4	24.0	22.2	15.0	9.0	80.6
Cuban Flint	8.8	13.9	17.0	17.8	12.3	69.6
Composite High	**12.5**	**21.2**	**20.3**	**14.9**	**14.3**	**83.2**
Composite maize × teosinte	**4.8**	**6.3**	**18.6**	**12.7**	**13.5**	**56.0**
Balsas teosinte	14.0	9.3	24.5	12.2	9.8	69.8
Zea diploperennis	0	3.9	11.8	15.2	18.1	49.0
Central Plateau teosinte	0	5.6	20.7	10.6	12.7	49.7

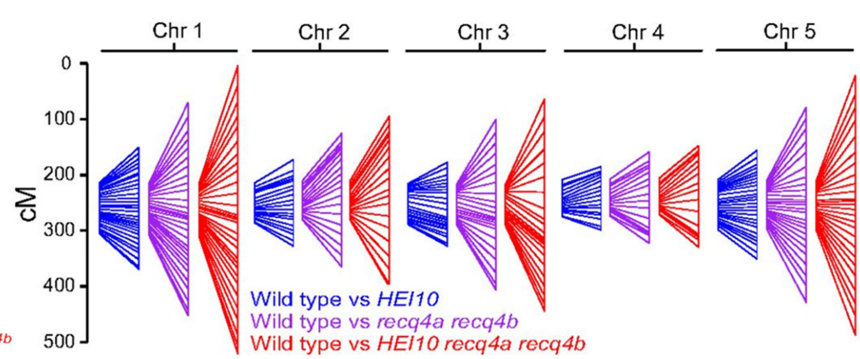
I. Helicases FANC & Req4

Crismani et al., 2012

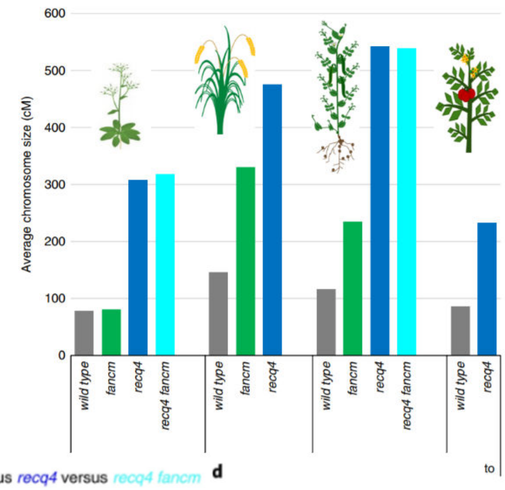


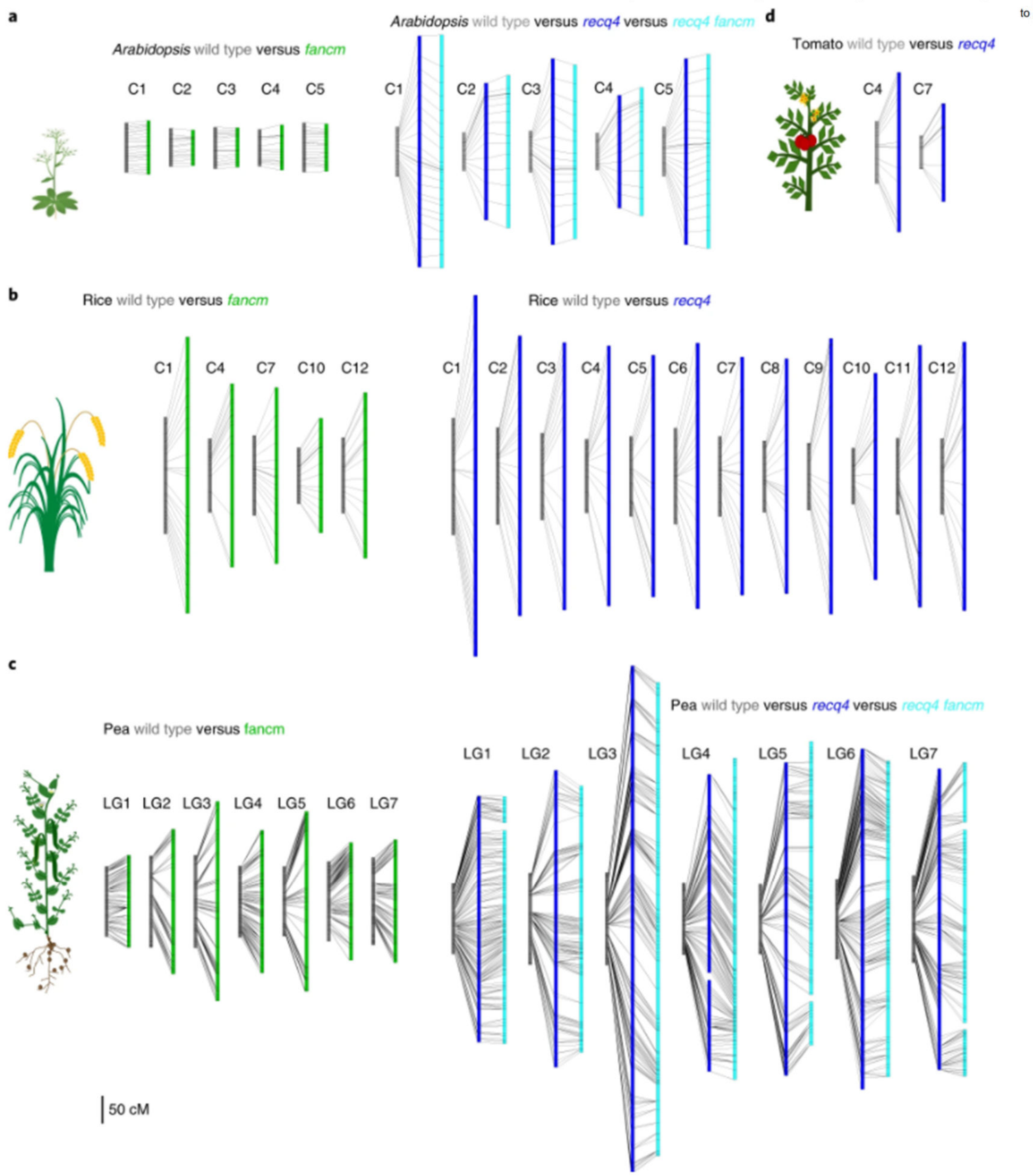
Serra et al, 2018



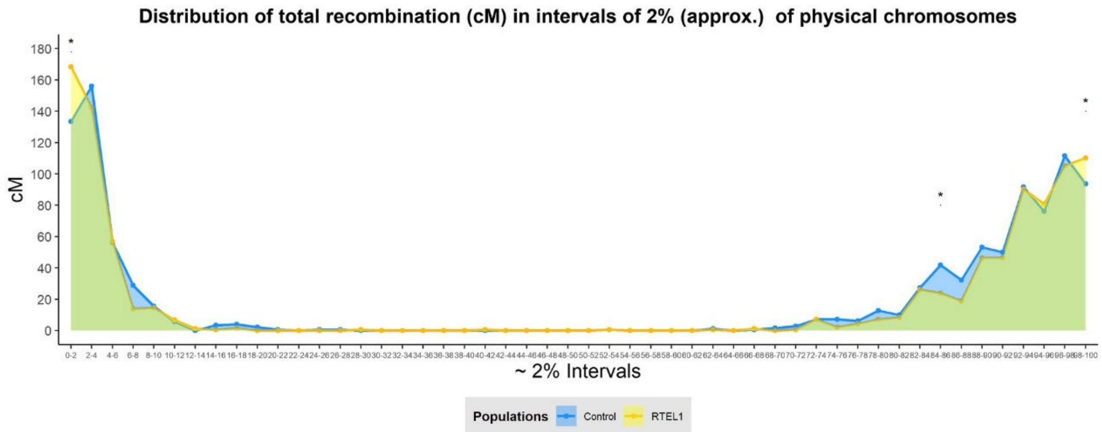


Mieulet et al., 2018





Barakate et al, 2021

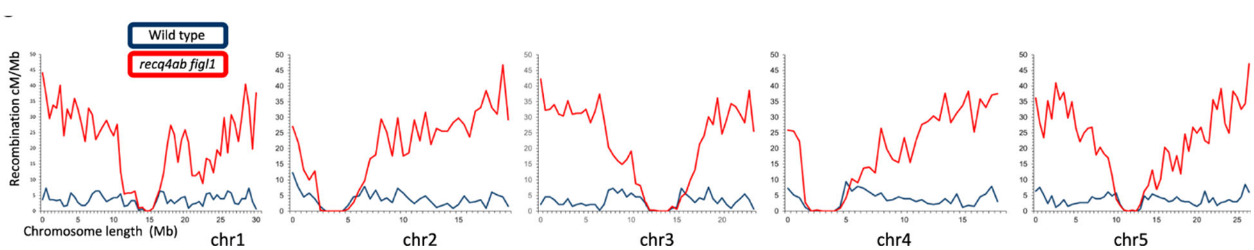


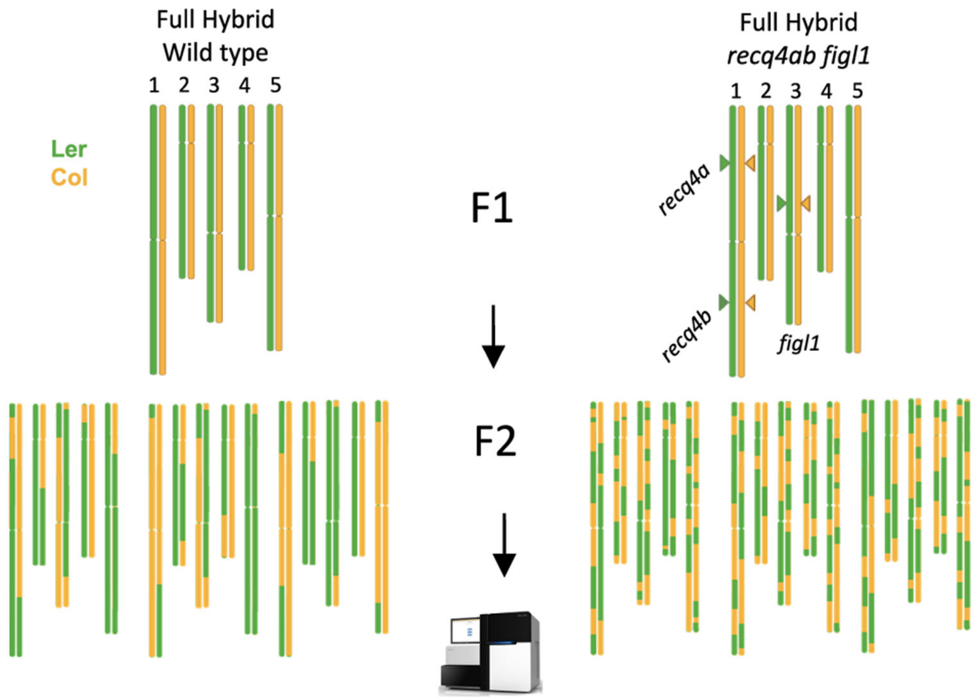
Capilla-Pérez et al, 2024

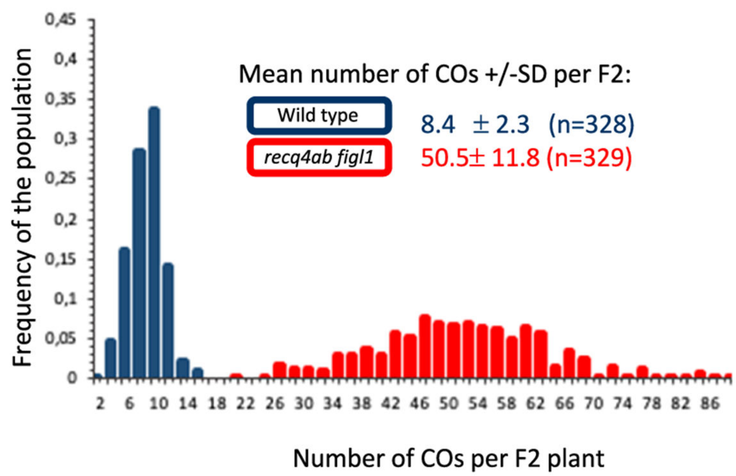
There are two types of crossovers

- Class I
  - The most common
  - Subject to interference
  - Limited by dosage of
    - HEI10
    - Phosphatase X1
    - Synaptonemal proteins ZYP1/SCEP1/SCEP2

- Class II
  - Limited by 3 protein complexes
    - TOP3/RECQ4AB/RMI1
    - FANCM
    - FIGL1/FLIP







Factors Affecting Recombination

Environment

1. Age

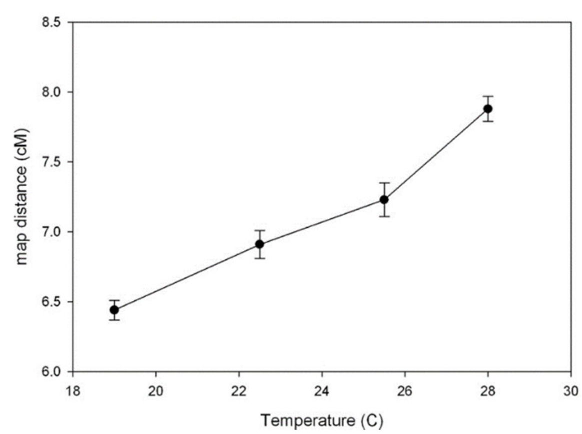
Bridges, 1915

2. Temperature

Dowrick, 1957

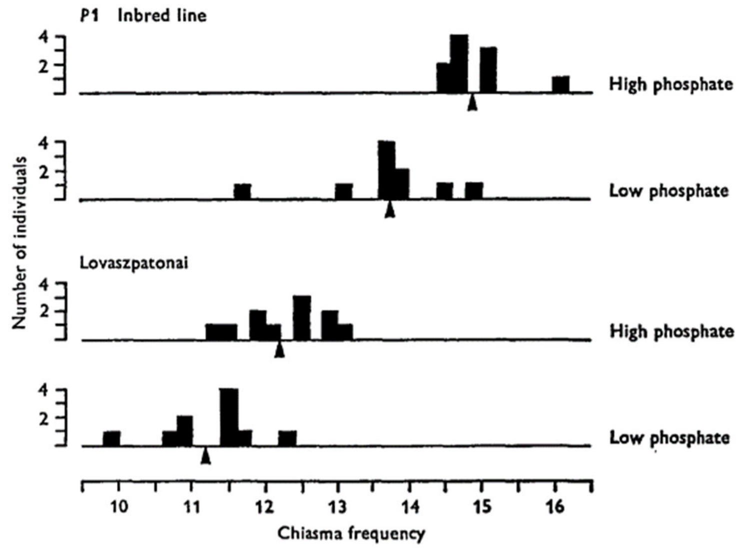
°C	Time (h)	% interstitial xmata	Total xmata (100 PMC)	°C	Time	% interstitial xmata	Total xmata (100 PMC)
control	--	2.3	743	control	--	2.2	757
20	12	4.8	773	34	12	8.8	832
	24	6.2	836		18	18.9	984
	48	11.9	847		24	21.6	1006
	96	15.3	842		48	12.1	860
	192	13.7	883		96	0.7	431
control	--	2.0	707	control	--	2.3	743
27	12	7.4	757	39	18	16.8	841
	24	15.8	845		24	41.2	1152
	48	17.4	849		27	29.4	961
	144	17.3	851		30	19.3	787
					48	0	72

Francis et al., 2007

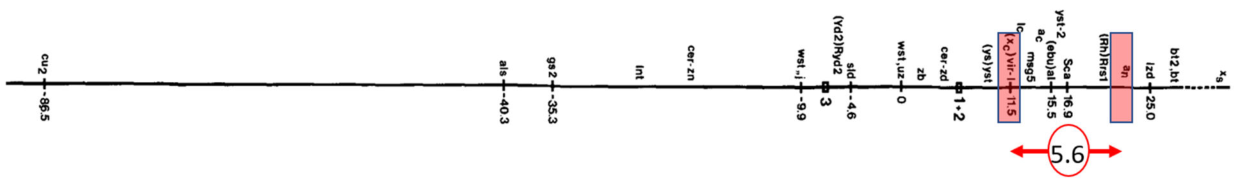


3. Stress

Bennett & Rees, 1970



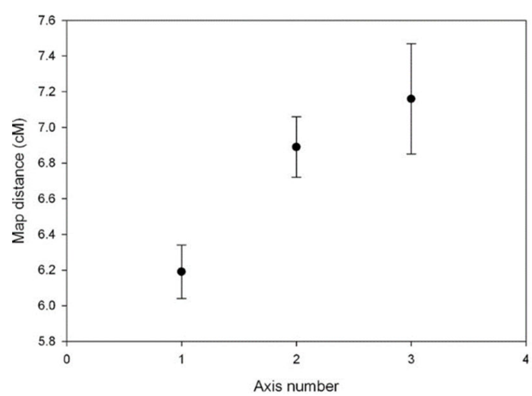
Sinha & Helgason, 1969



- Lethal xc (xantha) and an (albino)
  - Actinomycin D
  - Diepoxybutane
- Increased recombination distance to

4. Position on flower head

Francis et al., 2007



<u>A NOTE OF CAUTION</u>

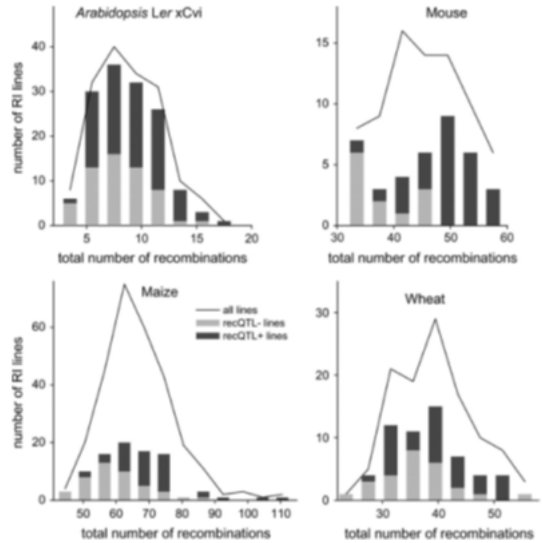
Barth et al, 2000

Manipulating Crossovers

Yanagira et al., 1992

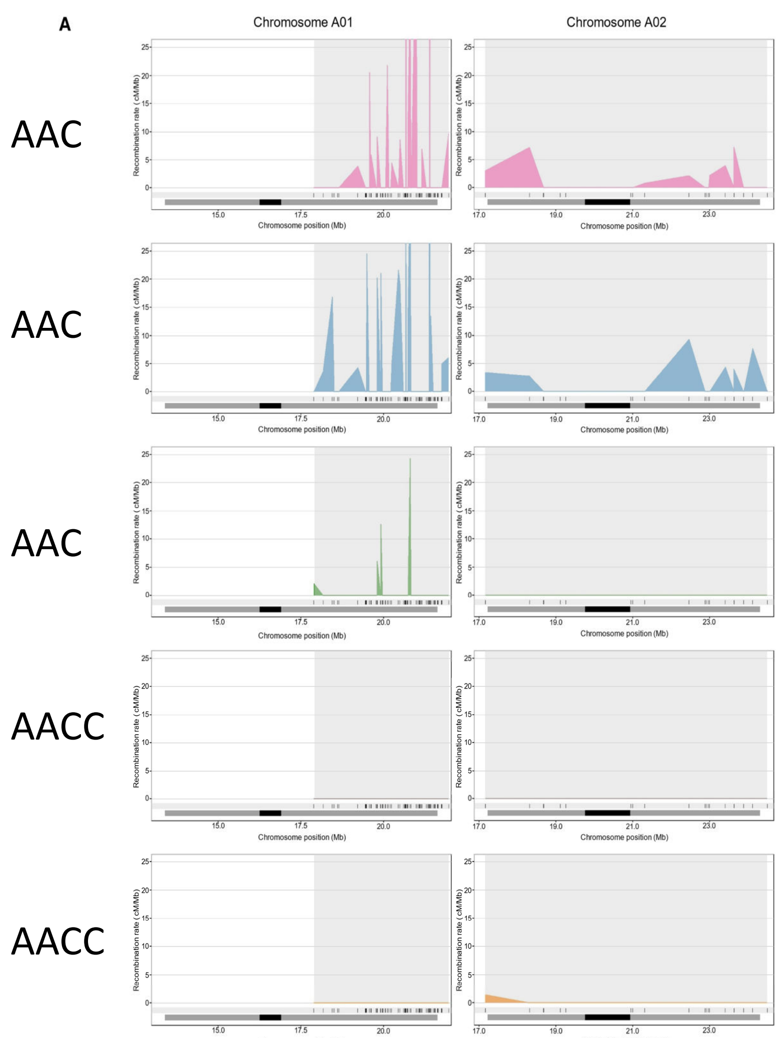
Gene order: *Wx* – *C* – *S-5* on chromosome 6

Esch et al., 2007



Boideau et al, 2024

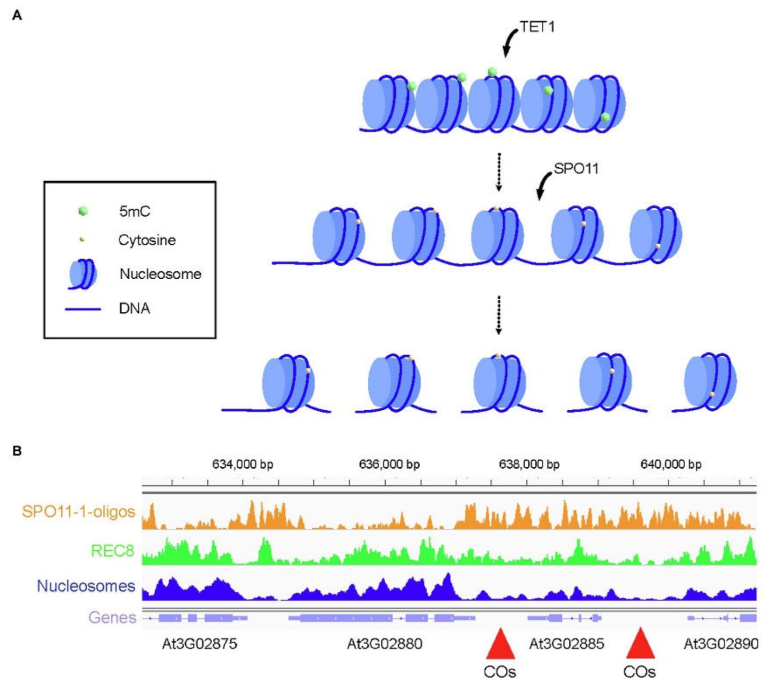
Allotriploidy



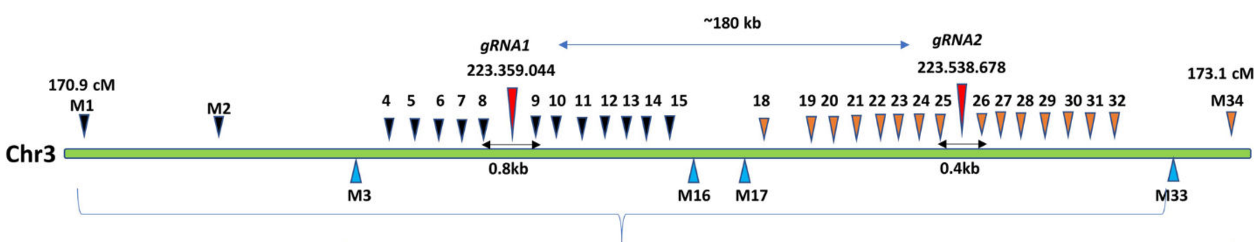
- Gray box = heterozygous area
- X axis = chromosome
- Black = centromere
- Dark gray = pericentric heterochromatin
- Vertical bars = genetic markers

Engineering recombination hotspots

Kuo et al, 2021



Kouranov et al, 2022



Limitations

Taagen et al., 202