

Plant Breeding

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A backcross is a cross between a F_1 and one of its parents.

There are single backcrosses and repeated backcrosses.

A F_2 population, born from a segregation of a F_1 holds 3ⁿ (n=number of different loci) genotypes. The same F_1 backcrossed to one of its homozygous parents holds 2ⁿ genotypes. Hence a backcross decreases drastically the number of genotypes, theoretically facilitating the discovery or detection of the wanted genotype.



The offspring of a backcross statistically holds 75% of the genes of the backcross parent $([1-(1/2)^m]; m \text{ is number of backcross generations}).$

Is this true for each individual in this offspring?

Repeated backcrosses to the same *recurrent parent* (P_r) expulse gradually the genes of the *donor parent* (P_d).



Critical success factors in backcrossing are :

- One needs an excellent P_r: an elite line, elite inbred line, elite variety.
- The genotype of P_r should be recovered after a reasonable number of backcrosses.
- The traits of interest in P_d should have a high heritability. A monogenic trait with a low heritability is difficult to work with; on the other hand a quantitative trait may be easy to introgress if its heritability is high, even if the number of genes concerned is high or not known.







- The degree of homozygosity in a certain generation equals (2^m-1)/2^m, m = number of backcrosses.
- The number of homozygous individuals equals [(2^m-1)/2^m]^x; x being the number of heterozygous loci in F₁.
- Convince yourself that the number of homozygous individuals becomes quite high after 3-4 backcrosses <u>in</u> <u>case the number of segregating loci is limited.</u>



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			r (Number of Plants to Be Recovered)										
p *	q^{\dagger}	1	2	3	4	5	6	8	10	15			
0.95	1/2	5	8	11	13	16	18	23	28	40			
••••	- 1 3	8	13	17	21	25	29	37	44	62			
	1	11	18	23	29	34	40	50	60	84			
	1	23	37	49	60	71	82	103	123	172			
	$\frac{1}{16}$	47	75	99	122	144	166	208	248	347			
	$\frac{1}{32}$	95	150	200	246	291	334	418	500	697			
	$\frac{1}{64}$	191	302	401	494	584	671	839	1002	1397			
0.99	$\frac{1}{2}$	7	11	14	17	19	22	27	32	45			
	13	12	17	22	27	31	35	44	52	71			
	14	17	24	31	37	43	49	60	70	96			
	1	35	51	64	77	89	101	124	146	198			
	$\frac{1}{16}$	72	104	132	158	182	206	252	296	402			
	$\frac{1}{39}$	146	210	266	218	268	316	508	597	809			
	$\frac{1}{64}$	293	423	535	640	739	835	1020	1198	1623			

 Table 28-1
 Total Number of Plants Needed to Obtain Required Number

 with Desired Genes
 Plants Needed to Obtain Required Number

*p = probability of recovering r plants with the desired genes.

 $\dagger q$ = frequency of plants with desired genes.

Source: Sedcole, 1977.





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Advantages of BC

- Provides breeder a high degree of control
- It is repeatable
- Extensive field trials are not required
- Few notes (record keeping)



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Disadvantages of BC

- Improved variety is same as recurrent parent except for trait transferred
- Minimal recombination
- Linkages to deleterious genes



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Marker-assisted backcrossing (MABC)

one does not have to wait for phenotypic expression of traits



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It has the potential to improve the reliability of the selection process On top of an improvement of the reliability, the breeding goal might be achieved with less backcrosses, i.e. faster.

 P_d : aabbCCddEEFF..... x AABBccddEEff..... P_r

gametes : abCdEF ABcdEf

F1 :AaBbCcddEEFf.....



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Gametes made from F₁ **individuals**: among many different gametes, $AB\underline{C}dEf$ may occur: it is nearly isogenic to the P_r gametes - with the exception of the wanted allele *C*.

From B_1F_1 (=BC1) on, one is looking for plants with a genotype as close as possible to the genotype of the recurrent parent, with the exception of the donor gene(s).

This means : <u>one is looking for plants with</u> <u>the highest possible degree of homozygosity</u> <u>of the recurrent genome, with the exception of the donor</u> gene(s).





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Bt-maize

Ragot et al. 1995



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Ragot et al., 1995



SELECTED BC1

Figure 1-a: Genetic maps of the backcross-derived individuals selected in the first four generations of a marker-assisted backcross program. The locus to be introgressed (Bt) is located on chromosome 1.



Ragot et al., 1995

SELECTED BC2

1	2	3	4	5	6	7	8	9	10
CG100 CG161 CG259 CG363 CG363 CG320 BT CG415 CG263 CG378 CG264 CG264 CG264 CG264 CG324 CG312 CG267 CG143 CG246	CG270 CG305 CG350 CG350 CG465 CG346 CG262 CG460 CG354 CG354 CG451 CG153	 CG128 CG141 CG426 CG361 CG283 CG263 CG483 CG341 CG134 CG295 CG134 CG411 	CG653 CG125 CG094 CG327 CG095 CG095 CG335 CG406	CG323 CG548 CG450 CG069 CG077 CG279 CG102 CG315 CG107 CG265 CG110 CG349 CG185	CG382 CG286 CG386 CG397 CG463 CG196 CG095 CG095 CG177 CG079	CG608 CG262 CG176 CG228 CG316 CG329 CG237 CG167 CG186 CG573	CG483 CG224 CG485 CG536 CG382 CG128 CG292 CG168 CG122 CG302 CG302 CG302 CG302 CG492	CG214 CG437 CG321 CG042 CG123 CG451 CG363 CG080 CG170 CG337	CG326 CG454 CG334 CG204 CG317 CG305
U									

Figure 1-b: Genetic maps of the backcross-derived individuals selected in the first four generations of a marker-assisted backcross program. The locus to be introgressed (Bt) is located on chromosome 1.



generation	% phosphinolhricin resistant plants	RFLP genotyping			nb plants analyzed *	% homozygous recurrent parent genotype				nb heterozygous chromosome segments ***			
		nb plants	nb Ioci	nb datapoints		mean	std dev	5-best mean **	selected plant	mean	std dev	5-best mean **	selected plant
BC1 BC2 BC3 BC4	49.05 44.65 46.32	96 61 72 26	61 22 10 3	5856 1342 720 78	87 30 71 26	48.72 83.42 93.83 98.23	10.35 5.64 1.85 0.49	68.31 91.98 96.82 99.09	70.45 90.84 98.03 99.36	11.01 5.03 2.20 1.00	2.17 1.54 0.71 0.00	7.75 3.20 1.60 1.00	6 3 1 1

Table 1: Proportion and characteristics of plants carrying the genes of interest, in the first four generations of a marker-assisted backcross program.

· Plants for which two or more adjacent markers had missing values were not included in the analyses

** Mean value of the five individuals having the five highest percentages of homozygous recurrent parent genotype.

... Including the segment carrying the transgene construct.



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Repeated backcrossing to introgress Cladosporium fulvum resistance (dominant) from Lycopersicon pimpinellifolium (small tomatoes) in Lycopersicon esculentum (large tomatoes). Kuckuck, 1979.

Help of markers

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Repeated backcrossing to bring nuclear male sterility (recessive) in an elite line. Kuckuck, 1979.



I	II	Ш	IV	
ВХА	CXA	DXA	ΕXΑ	
AB X A	AC X A	AD X A	AEXA	
$A_2 B X A$	$A_2C \times A$	$A_2 D X A$	$A_2 E \times A$	
			· · ·	
		•		
$A_5B \times A$	$A_5C \times A$	$A_5 D \times A$	$A_5 E \times A$	
Selbstung, Spaltung- Auslese	wie I	wie I	wie I	
A' _(Res.B)	X A'(Res.C)	A' _(Res.D) X	· A' _(Res.E)	
	A'(Res.B) →	< A'(Res.C) X	$\left[A'_{(\text{Res.D})} \times A ight]$	^′(Res,E)] ◀ _]
		A'	Res.B+Res.C+R	es.D+Res.E)

Stepwise backcrossing to bring different traits in one elite parent (today we call this stacked traits): the backcrossings start at the same moment. Hoffmann et al., 1971.

Help of markers !!!!!





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BC 21

Backcross inbred lines (BIL's)

- Lycopersicon esculentum x L pennellii (small, green, sweet fruits) + a number of BC's with L. esculentum, followed by self-fertilization: result: BIL's
- Ideally : a number of homozygous lines, each containing a different piece of *pennellii* DNA on different chromosomal sections.
- Correlation between phenotypical performances genotype.
- Identification of some specific regions of *pennelli* DNA responsible for specific performances





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Different transgenic events stacked in elite plants

Crossing scheme



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