

For the trisomics, the second backcross or self of the progeny with the A-phenotype from the first backcross should separate $2n$ from $2n + 1$ individuals, and to separate the different trisomic genotypes. As shown in Table 87, the second backcross should furnish more reliable information on distances of genes from the centromeres; particularly for the trisomics if the trisomic segregates in the first backcross progeny are used. Trisomics should be preferable over triploids because of their higher fertility.

In tetrasomics, the second backcross should distinguish quadruplex (AAAA) from triplex (AAAa) individuals; also individuals that are pentasomic vs. trisomic for the locus. The latter genotypes are expected when three of the four chromosomes pass to one pole and one to the other.

Ratios in terms of double reduction frequency (α)

Fisher's and Mather's approach to the ratios expected in a polysomic is based on a consideration of what he terms the laws of gametic output. For a single locus in any trisomic or tetrasomic, only two modes of gamete formation are distinguishable, i. e. the two genes (alleles) in any $n + 1$ gamete may be derived from different chromosomes of the parent multivalent or from the same chromosome. In the latter case they are identical at that locus since they are from sister chromatids, and result from double reduction as described earlier. Fisher and Mather (1943) have derived special expressions for the frequencies of the different kinds of gametes in terms of α , the total frequency of double reduction. α is no longer used for the "index of separation". How these expressions are derived is shown below, based on an explanation furnished personally by Dr. Fisher. For a tetrasome in which the four homologous alleles at one locus are designated a, b, c, and d, the possible kinds of gametes in which the two alleles were derived from sister chromatids would be aa, bb, cc, dd; i. e. four possible combinations. For the other mode of gamete formation in which the two alleles are derived from different chromosomes, there are six possible combinations, ab, ac, ad, bc, bd and cd. The relative frequencies of these two groups is unknown, but if α is assumed to be the total frequency of the group in which the gametes carry alleles from sister chromatids, i. e. double reduction, then $1 - \alpha$ is the total frequency for the other group. Since the four double reductional combinations are expected to be equally frequent, the probability of each is $\frac{\alpha}{4}$. Likewise the frequency of each combination in the other group is $\frac{1 - \alpha}{6}$. These relations are summarized in Table 88, along with the genetic constitutions of the corresponding gametes expected from a tetrasomic which is triplex at the Aa locus, i. e. AAAa.

Table 88. Kinds of gametes from a tetrasomic classified as to the two modes of gamete formation and their frequencies in terms of α , which is the total frequency of double reduction. The second and fifth columns are the genetic genotypes for each combination from an AAAa tetrasomic.

Group 1 The two alleles from the same chromosome			Group 2 The two alleles from different chromosomes in the quadrivalent		
Combinations	Gene constitution from AAAa	Frequency	Combinations	Gene constitution from AAAa	Frequency
aa	AA	$\frac{\alpha}{4}$	ab	AA	$\frac{1 - \alpha}{6}$
bb	AA	"	ac	AA	"
cc	AA	"	ad	Aa	"
dd	aa	"	bc	AA	"
			bd	Aa	"
			cd	Aa	"
Total		α	Total		$1 - \alpha$

As shown in Table 88, in a tetrasomic whose genotype is AAAa, only one of the four combinations resulting from double reduction, i. e. aa, is recognizable. The frequencies of it and the other gametes may be derived from the table. The aa gamete is derived only by double reduction, and its frequency is $\alpha/4$. The Aa gametes are all derived from combinations between different chromosomes, group 2; three of the 6 combinations, or $\frac{3(1-\alpha)}{6}$. The AA gametes are derived from both groups, three from group 1 and three from group 2; or $\frac{3\alpha}{4} + \frac{3(1-\alpha)}{6}$. The frequencies of the three kinds of gametes reduce to:

$$\frac{2+\alpha}{4} AA + \frac{2-2\alpha}{4} Aa + \frac{\alpha}{4} aa.$$

Using the above scheme, the frequencies in terms of α of the various kinds of 2-chromatid gametes from any genotype heterozygous for a single locus in trisomics or tetrasomics may be derived. These are summarized in Table 89.

Table 89. Summary of expected gametic frequencies in terms of α for trisomics and tetrasomics heterozygous for a single locus.

Genotypes	AA	Aa	aa	Total
<u>2n + 1</u>				
AAa	$\frac{1+\alpha}{3}$	$\frac{2-2\alpha}{3}$	$\frac{\alpha}{3}$	1
Aaa	$\frac{\alpha}{3}$	$\frac{2-2\alpha}{3}$	$\frac{1+\alpha}{3}$	1
<u>2n + 2</u>				
AAAa	$\frac{2+\alpha}{4}$	$\frac{2-2\alpha}{4}$	$\frac{\alpha}{4}$	1
AAaa	$\frac{1+2\alpha}{6}$	$\frac{4-4\alpha}{6}$	$\frac{1+2\alpha}{6}$	1
Aaaa	$\frac{\alpha}{4}$	$\frac{2-2\alpha}{4}$	$\frac{2+\alpha}{4}$	1

Substitution for α of the value $1/6$ for maximum equational segregation gives the expected gametic frequencies determined earlier (Tables 83 and 84). For example, for the simplex Aaaa tetrasomic the values are $\frac{1}{24} : \frac{10}{24} : \frac{13}{24}$.

Maximum likelihood equations may be derived from the expressions for gametic frequencies given in Table 89. From these, α may be calculated from data in which the frequencies of the different kinds of gametes have been determined, e. g. from second backcross data.

Unless supplementary evidence is available on the frequency of quadrivalents and the behavior of chiasmata subsequent to crossing over, α values may be of use only in determining the relative distances of genes from the centromere. That is, low values indicate the gene is closer to the centromere than do higher values. The map distances corresponding to different values cannot be determined without information on the various factors affecting the frequency of double reduction. This might be obtained if values for a series of factors in one linkage group are determined together with measures of recombination between the genetic markers. Methods for doing this will be discussed in a later section.

Genetic data

TRISOMICS. Data from Datura in which the trisomics and disomics were classified separately are summarized in Table 90 (Blakeslee, Belling and Farnham 1923 a, b).

Table 90. Data from globe trisomics in Datura, heterozygous for three different gene markers sc = slender capsule, bd = Baltimore bad, pl = pale leaves.

										<u>Numbers in progeny</u>				<u>% rec. in 2n+1</u>	<u>progeny from 2n x 2n+1 het.</u>	
										<u>2n</u>		<u>2n + 1</u>				
										<u>+ recessive</u>		<u>+ recessive</u>				
Sc	Sc	sc	x	sc	sc	95	48	63*	11*	14.9	58	30				
Sc	sc	sc	x	sc	sc	61	121	9*	0*	0						
Bd	Bd	bd	x	bd	bd	988	111	310	9	2.8						
Bd	bd	bd	x	bd	bd	315	284	150	52	25.7						
P1	P1	p1	x	p1	p1	190	119	45	3	6.3	216	88				
P1	p1	p1	x	p1	p1	49	117	41	27	38.2						

* Only part of the 2n + 1 plants were recorded for Sc vs. sc.

In the progeny of plants duplex (Sc Sc sc etc.) for each of the three genes, there are 2n + 1 individuals that were recessive (nulliplex) (Table 90). Judging by their frequencies, sc must be farther away from the centromere than either bd or Pl.

In the data in Table 68 (page 163) trisomic 7 in barley with the genotype SSs had two homozygous recessive trisomic offspring in a total of 47 plants, indicating that crossing over occurs between this gene and the centromere.

In tomatoes, only one of the 9 genes tested in duplex genotypes produced nulliplex individuals (Rick and Barton 1954).

Only limited data are available from experiments in which second backcross progeny or selfs of progeny of the first backcross progeny were used to determine the genotypic makeup of the gametes from the original polysomic parent. In such a test in tomatoes for simplex Rrr plants backcrossed to rrr, there were 3 duplex, 45 simplex and 68 nulliplex plants (Sansome, reported by Mather 1936). The α value based on the first two classes is .118. The number in the nulliplex class appears to be too high.

AUTOTETRAPLOIDS. The only available data from tetrasomes are from autotetraploids. The ratios from tetrasomic (2n + 2) plants should be similar. It is possible, however, that there might be differences in physiology which might affect chromosome behavior.

The first published results from selfing and backcrossing heterozygous tetraploids were for Primula sinensis (Gregory 1914). They were interpreted as 15:1 and 3:1 ratios but Muller (1914) showed that the observed numbers gave better fits to 35:1 and 5:1 ratios.

The data obtained by Blakeslee, Belling and Farnham (1923) from tetraploid Datura for armed vs. inermis (A vs. a) capsules and for purple vs. white (P vs. p) flowers are given in Table 91.

Only one pppp individual from the triplex PPPp genotypes was reported. This supposedly arose by double reduction. The triplex AAAa genotype produced a considerably

Table 91. Genetic data from autotetraploid *Datura stramonium* (Blakeslee, Belling and Farnham, 1923, data from Table 5, p. 348, and Table 8, p. 362, Bot. Gaz. 76, Univ. of Chicago Press)

Cross	Observed			Expected % recessives	
	Dom.	:	Rec.	% Rec.	Chromosome seg. Max. eq. seg.
*AAAA x AAAa	747	:	7	0.9	0.0 0.174
*AAAA x aaaa	257	:	6	2.3	0.0 4.2
AAAA x aaaa	518	:	137**	20.9**	16.7 22.2
AAAA x AAAa or self	3383	:	118**	3.4**	2.8 5.2
Aaaa x aaaa	144	:	144	50.0	50.0 50.2
*PPpp self	1280	:	0	0.0	0.0 0.174
*PPpp x pppp					
or reciprocal	160	:	1	0.6	0.0 4.2
PPpp x pppp	905	:	179	16.5	16.7 22.2
PPpp x PPpp or self	9199	:	225	2.4	2.8 5.2
Pppp x pppp and reciprocal	696	:	682	49.5	50.0 54.2
Pppp x Pppp or self	7547	:	2619	25.8	25.0 29.0

* Certain plants may have been AAAA or PPPP.

** Statistically significant deviations from the numbers expected for chromosome segregation and in the direction expected for maximum equational segregation.

higher number of nulliplex individuals. Hence this gene must be farther away from the centromere than P is in its chromosome.

As shown by these results, F_2 and backcross data are not very efficient in determining distances of genes from the centromeres.

A study of the frequency of double reduction for ten loci in autotetraploid maize has been made by El-Ghawas (1955). Summaries of the frequencies of the various genotypes in the progeny of individuals simplex for the loci are in Table 92.

Table 92. Frequencies of duplex and simplex genotypes from simplex Aaaa x nulliplex aaaa crosses in maize.

Chromosome	Gene	Numbers		total
		duplex	simplex	
1	bm ₂	8	14	22
2	lg ₁	3	25	28
3	A ₂	6	89	95*
4	su ₁	2	98	100*
9	C	0	62	62
"	wx	1	48	49*
"	I	0	31	31
10	g	11	47	58

* These figures are given by El-Ghawas (1955), the others were derived from his tables.

For su, c, wx and I the frequencies of duplex genotypes indicate that these genes are closer to the centromeres than are the other genes. For bm₂, lg, and g, the percentage of simplex is higher than the 1:10 expected for maximum equational segregation.

One difficulty in genetic studies of segregation in tetrasomes is that two chromosomes do not always segregate to each pole. Cytological studies of 100 plants made by El-Ghawas on the same crosses used in the genetic tests, showed a range from 36 to 47 chromosomes with 58 of the plants having 40 chromosomes and probably tetrasomic for all 10 chromosomes. The associations at diakinesis in autotetraploid plants ranged from disomes to hexasomes or even higher. The non-tetrasomic ratios observed by El-Ghawas in some plants could be explained on the basis of trisomic, and others by higher than tetrasomic inheritance. That these off-types may be important in determinations of frequencies of double reduction is indicated by results reported by Catchside (1956). Of 33 su seeds in a total of 3359 progeny obtained from the cross of Su Su Su su x su su su su, 27 were checked further. Nine were trisomic rather than tetrasomic for chromosome 4, and therefore had received only one su chromosome from 3-1 disjunction in the triplex parent, thus erroneously increasing the apparent frequency of double reduction.

RATIOS IN DI- AND TRI- HYBRIDS IN AUTOTETRAPLOIDS. The expected genetic ratios in di- and tri- hybrids in autopolyploids are of interest in relation to the breeding of polyploids. What are the theoretical F_2 ratios for an individual duplex for two independent loci, AAaa BBbb with no special interaction and complete dominance? Both loci may be close to a centromere, or one may be close, the other distal; or both may be distal. If maximum equational segregation is assumed when the locus is distal and chromosome segregation when the locus is close to the centromere, the expected F_2 ratios are those shown in Table 93.

Table 93. F_2 ratios expected for dihybrids of AAaa BBbb genotype as affected by proximity to the centromere.

Phenotype	Chrom. seg. for both		Max. eq. for both		Chrom. seg. for A		Max. eq. for A, Max. eq. for B		Chrom. seg. for B	
	number	%	number	%	number	%	number	%	number	%
AB	1225	94.5	5929	89.4	2695	92.4	2695	92.4		
Ab	35	2.7	308	4.6	140	4.8	77	2.6		
aB	35	2.7	308	4.6	77	2.6	140	4.8		
ab	1	0.1	86	1.3	4	0.1	4	0.1		
Total	1296		6631		2916		2916			

In diploids the Ab and aB phenotypes are expected to be equal in frequency. Note that in an autotetraploid if one locus is close to the centromere, the other not, the Ab and aB phenotypes are unequal, as shown in Table 93. Data in which one gene is known to be close to a centromere might be helpful in determining if a second independent gene is close or far away from the centromere in its chromosome.

Another point to note is the low frequency of the multiple recessive class in F_2 ; lowest of course if both loci are close to the centromere. The expected F_2 ratio for a trihybrid in which each of the three loci is close to a centromere is: 42, 876:1, 225:1, 225:1, 225:35:35:35:1. Here there is only one multiple recessive in 46, 656. Hence F_2 's are not very efficient if one is selecting for new multiple recombinants.

Blakeslee et al. (1923) have reported the numbers obtained in F_2 for a di-hybrid in Datura of the PPpp AAaa genotype for flower color and the armed vs. inermis capsule characters:

	<u>PA</u>	<u>Pa</u>	<u>pA</u>	<u>pa</u>
Observed	1696	58	63	1
Calculated	1718.4	49.1	49.1	1.4
(chromosome seg.)				

Here, the Pa and pA classes are about equal in frequency. The ratio for P vs. p is 1754:64 or 3.53% p; that for A vs. a is 1759:59 or 3.25% a. The expected percentages for chromosome segregation and for maximum equational segregation are 2.8 and 5.19 respectively. The data show the two loci are similarly placed and relatively close to the centromeres.

POLYPLOID RATIOS IN FUNGI. Genetic markers may be used in yeasts and in *Neurospora* to permit genetic tests for polyploidy. For examples of polyploid segregations in yeast see Leupold and Hottinguer (1954), and Leupold (1956).

Rates of approach to homozygosis in tetraploids vs. diploids.

Haldane (1930), and Bartlett and Haldane (1934) have considered the effects of different types of mating on rates of decrease of heterozygosis in autopolyploids. The proportions of single factor homozygotes expected from selfing two autotetraploid genotypes, as calculated by Greenleaf (1938) for two types of segregation are shown in Table 94. The values for maximum equational segregation (double reduction = 1/6) have been added, since this is the theoretical maximum rather than 1/7 (see pages 186 and 188).

Table 94. Effect of autotetraploidy on percent of homozygosis at a single locus in successive generations of selfing (Greenleaf, 1938, from Table 5, page 463, Jour. Heredity 29, except columns 5 and 8).

Generations selfed	Diploid Aa	Percent of homozygosis					
		Tetraploid Aaaa			Tetraploid AAaa		
		Chromo-some seg.	Random Chromatid seg.	Max. equat. seg.*	Chromo-some seg.	Random Chromatid seg.	Max. equat. seg.*
1	.5	.25	.2883	.2951	.05	.0918	.0988
2	.75	.38	.4498	.4602	.194	.2714	.2848
3	.875	.4931	.5688	.5811	.3256	.4257	.4420
4	.9375	.5579	.6614	.6743	.4375	.5486	.5658
5	.96875	.6483	.7339	.7467	.5312	.6453	.6623

* Calculated by Neal Tuleen, checked by M. L. Wright.

As shown in Table 94, the rate of approach to homozygosity is expected to be much slower in autopolyploids than in diploids and slower with chromosome than with maximum equational segregation.

Parsons (1959), in considering the effect of different values of α on the rate of approach to homozygosity, has not included the value of 1/6. Geiringer (1949) has presented a mathematical treatment of the expectations from random mating and random chromatid segregation in autopolyploids.

Linkage in autopolyploids (Fisher 1947, 1948, Gates 1957)

Kinds of genotypes. As stated by Fisher linkage in autopolyploids as compared with diploids is much more complex because of the greater number of possible "gametic geno-