DOUBLE REDUCTION AND NUMERICAL NON-DISJUNCTION IN TETRAPLOID MAIZE

D. G. CATCHESIDE

Department of Genetics, University of Adelaide *

Received 11.xi.55

I. MODES OF SEGREGATION

GENETIC segregation in tetraploids is complicated by modes of gamete formation not found in diploids. For a single locus, there are three of these modes. Besides (1) the contribution to a gamete of one chromatid from each of two chromosomes out of four, there are the additional possibilities of (2) the contribution of two chromatids from one chromosome (Mather, 1936) and (3) the contribution of more or less than two chromatids to a gamete. The second mode is the result of what is usually known as chromatid segregation, in contrast to the normal chromosome segregation, the contribution of two sister chromatids and their genes to one gamete occurring through double reduction. For a particular locus, the frequency with which sister genes are present in the same gamete is measured by the parameter α (Fisher and Mather, 1943). The third mode is a consequence of numerical non-disjunction. This is a factor which cannot be neglected, at least in maize, because a substantial fraction of the progeny of a tetraploid have fewer or more than the tetraploid chromosome number. This has never been properly estimated for a particular chromosome, but is probably of the order of about 5 per cent. for each chromosome in maize.

In all previous genetic studies of tetraploids the effect of non-disjunction has not been separated from that of double reduction which it resembles superficially. This paper is an attempt to estimate the magnitude of these two factors for a particular locus in maize.

2. MATERIAL AND METHODS

The locus studied was sugary-1 (*su-1*) in chromosome 4. In the diploid, by the use of structural changes, this locus has been placed at not less than 9 map units from the centromere and in the shorter arm (Anderson and Randolph, 1945).

All the material was derived from a starchy tetraploid stock donated by Dr L. F. Randolph and a sugary tetraploid donated by Dr Janaki Ammal. Chromosome numbers have been determined in root tips, pretreated with 8-hydroxyquinoline usually in a refrigerator at 5° C. for three to four hours, and fixed in acetic alcohol. Squash preparations were made in acetic orcein after hydrolysis for five minutes in N HCl. The identification of the chromosomes in a somatic metaphase plate can be accomplished fairly reliably by taking into account the ratio of the long to the short arm for each chromosome and the ratio of the total length of the chromosome to the long arm of chromosome 6, which may always be identified by the trabant present on the short arm. The average statistics for identification are shown in fig. 1. It is unfortunate that chromosome IV is not a particularly easy one to identify, but it is believed that the data are reasonably reliable.

* Present address: Department of Microbiology, University of Birmingham.

3. CHROMOSOME NUMBERS OF PROGENY OF TETRAPLOIDS

The chromosome numbers of progeny of parent plants with 40 chromosomes have been determined. They are compared in table 1 with the pooled data collected by Kadam (1944). The two sets of data are consistent with one another.

FIG. 1.—Diagram showing the statistics of chromosomes I to V and VII to X, with respect to one another and to chromosome VI.

Taken as a whole the data show 259 chromosomes in excess or deficient in 503 plants. If we assume that only numerically balanced pollen grains function as gametes, there were 259 non-disjunctions amongst 503 eggs, a mean of 0.515 per gamete, or a chance of 0.0515

TABLE 1
Chromosome numbers of progeny of 40 chromosome tetraploid maize

Source	36	37	38	39	40	41	42	43	Totals	Means
(1)	4	1	36	43	261	73	8	3	429	39.92±0.046
(2)	...	...	3	8	50	13	...	...	74	39.99±0.078
Totals	4	1	39	51	311	86	8	3	503	39.93±0.041

(1) Kadam, 1944. (2) Present paper.

per chromosome per female gamete for non-disjunction. If we assume that numerically unbalanced pollen grains are not handicapped in functioning as gametes, the non-disjunctional chance per chromosome

per female gamete could be as low as 0.02575. Presumably the actual value is somewhere between these limits.

Some indication of the relative likelihood with which non-disjunctive gametes function as eggs and as sperm is given by Punyasingh's (1947) data on the chromosome numbers of tetraploid $\times$ diploid and reciprocal crosses. These indicate rates of non-disjunction per chromosome per gamete of 0.0748 amongst eggs and 0.0524 amongst sperm, a ratio of 1.43. Apart from the fact that these values are higher than amongst the progeny of tetraploids, one cannot be sure that the relative performance of gametes in a diploid $\times$ tetraploid cross would be exactly similar to that in the tetraploid $\times$ tetraploid,

TABLE 2
*Representation of the ten different chromosomes (I to X) in
progeny of tetraploid maize*

(a) 74 unselected progeny of 40 chromosome plants

Chromo- somes per plant	I	II	III	IV	V	VI	VII	VIII	IX	X	Totals
3	...	1	4	2	...	3	1	2	...	...	13
4	73	71	69	70	73	71	73	71	70	73	714
5	1	2	1	2	1	...	...	1	4	1	13

(b) 27 *su-1* progeny from triplex $\times$ nulliplex.

Chromo- somes per plant	I	II	III	IV	V	VI	VII	VIII	IX	X	Totals omitting IV
3	...	1	1	9	1	2	2	...	3	1	11
4	27	25	26	18	26	25	25	25	23	25	227
5	...	1	...	...	...	...	...	2	1	1	5

nor that the relative viabilities of unbalanced near triploids would be similar to those of unbalanced near tetraploids. Nevertheless, it would appear reasonable to assume that amongst our material, the ratio of non-disjunction amongst female and male gametes is similar and that the non-disjunctive chance per chromosome per female gamete is about $0.0515 \times \frac{1.43}{2.43} = 0.0382$.

During the present observations, attempts were made to determine the numbers of each of the ten different chromosomes. These attempts were by no means exhaustive. Wherever the count departed from 40, the chromosomes were identified to discover which ones departed

from being present in quadruplicate. Where the count was 40, the chromosomes were identified to see that there were no departures from there being four of chromosome VI, 20 chromosomes longer than VI (numbers I to V) and 16 chromosomes shorter than VI (numbers VII to X). In two cases, the number of 40 was found to be unbalanced, being internally compensated through five of one chromosome making up the deficit due to three of another chromosome. No doubt other internal compensations escaped detection.

The results are given in table 2, separately for unselected plants and plants which, arising from the triplex backcross, showed the recessive sugary-1. The unselected plants show a mean of 0.0352 non-disjunctions per chromosome per plant; the selected ones, omitting chromosome 4, show a mean of 0.0684. The values are not significantly different, and the weighted mean is 0.0432. It will be noticed that a third of the selected plants have only three of chromosome IV (plate, fig. 2) and it is reasonable to conclude, as discussed in the next section, that each of these owed its recessive phenotype to non-disjunction in the formation of the female gamete, rather than to double reduction.

4. SEGREGATION RATIOS IN TETRAPLOIDS

In a tetraploid organism, there are three possible sorts of heterozygote for a given digenic locus, respectively with one, two or three doses of a recessive gene. These heterozygotes are usually known as triplex, duplex and simplex heterozygotes, from the presence of three, two and one dominant genes. The regular gametes formed contain two of each sort of chromosome and with regular chromosome segregation the simple gametic ratios given in the first section of table 3 are expected. In the case of a locus not situated at the centromere of the chromosome, a proportion of double reduction may be expected as a result of crossing-over between the locus and the centromere, followed by non-disjunction of the sister genes at both the first and second division of meiosis. If the coefficient of such double reduction is α , the gametic ratios shown in the second part of table 3 are expected.

Besides regular segregation in a tetraploid, numerical non-disjunction of the chromosomes may occur, resulting in gametes receiving either three or one of a particular kind of chromosome instead of the usual two. If numerical non-disjunction occurs in a proportion x of meiotic cells, so that a similar proportion of gametes shows the effects of such non-disjunction, the gametic ratios shown in the third part of table 3 may be expected if the complication of numerical non-disjunction is simply added to chromosome segregation. If chromatid segregation and numerical non-disjunction are both assumed to occur in a tetrasomic organism the gametic ratios shown in the fourth part of table 3 may be expected, if the two events do not occur together in a cell.

TABLE 3

The effects of double reduction and numerical non-disjunction upon the proportion of gametes of different constitutions formed by tetrasomic heterozygotes

Type : Constitution :		TRIPLEX AAAA	DUPLEX AAaa	SIMPLEX Aaaa
Gametes formed by CHROMOSOME SEGREGATION	AA	$\frac{1}{2}$	$\frac{1}{6}$	...
	Aa	$\frac{1}{2}$	$\frac{2}{3}$	$\frac{1}{2}$
	aa	...	$\frac{1}{6}$	$\frac{1}{2}$
Gametes formed with CHROMATID SEGREGATION (Double Reduction)	AA	$\frac{2+\alpha}{4}$	$\frac{1+2\alpha}{6}$	$\frac{\alpha}{4}$
	Aa	$\frac{1-\alpha}{2}$	$\frac{2-2\alpha}{3}$	$\frac{1-\alpha}{2}$
	aa	$\frac{\alpha}{4}$	$\frac{1+2\alpha}{6}$	$\frac{2+\alpha}{4}$
Gametes formed with NUMERICAL NON-DISJUNCTION	AAA	$\frac{x}{8}$	...	...
	AAa	$\frac{3x}{8}$	$\frac{x}{4}$	...
	Aaa	...	$\frac{x}{4}$	$\frac{3x}{8}$
	aaa	...	...	$\frac{x}{8}$
	AA	$\frac{1-x}{2}$	$\frac{1-x}{6}$	...
	Aa	$\frac{1-x}{2}$	$\frac{4(1-x)}{6}$	$\frac{1-x}{2}$
	aa	...	$\frac{1-x}{6}$	$\frac{1-x}{2}$
	A	$\frac{3x}{8}$	$\frac{x}{4}$	$\frac{x}{8}$
	a	$\frac{x}{8}$	$\frac{x}{4}$	$\frac{3x}{8}$
Gametes formed with DOUBLE REDUCTION and NUMERICAL NON-DISJUNCTION	AAA	$\frac{x}{8}$	...	...
	AAa	$\frac{3x}{8}$	$\frac{x}{4}$	...
	Aaa	...	$\frac{x}{4}$	$\frac{3x}{8}$
	aaa	...	...	$\frac{x}{8}$
	AA	$\frac{(2+\alpha)(1-x)}{4}$	$\frac{(1+2\alpha)(1-x)}{6}$	$\frac{\alpha(1-x)}{4}$
	Aa	$\frac{(1-\alpha)(1-x)}{2}$	$\frac{(2-2\alpha)(1-x)}{3}$	$\frac{(1-\alpha)(1-x)}{2}$
	aa	$\frac{\alpha(1-x)}{4}$	$\frac{(1+2\alpha)(1-x)}{6}$	$\frac{(2+\alpha)(1-x)}{4}$
	A	$\frac{3x}{8}$	$\frac{x}{4}$	$\frac{x}{8}$
	a	$\frac{x}{8}$	$\frac{x}{4}$	$\frac{3x}{8}$

Strictly speaking, the expectations in the fourth part of table 3 need correcting in respect of non-disjunctional gametes in which double reduction has occurred. This will affect only those gametes which receive three allelic genes. The consequential corrections are as follows :

Gamete	Triplex	Duplex	Simplex
<i>AAA</i>	$\frac{x(1+\alpha)}{8}$	$\frac{\alpha x}{12}$	
<i>AAa</i>	$\frac{3x-2\alpha x}{8}$	$\frac{x(3-\alpha)}{12}$	$\frac{\alpha x}{8}$
<i>Aaa</i>	$\frac{\alpha x}{8}$	$\frac{x(3-\alpha)}{12}$	$\frac{x(3-2\alpha)}{8}$
<i>aaa</i>		$\frac{\alpha x}{12}$	$\frac{x(1+\alpha)}{8}$

It will be noticed that chromatid segregation and numerical non-disjunction each contribute to the presence of gametes bearing only recessive genes, for the locus in question, when none are expected (the triplex heterozygotes) and also to a greater frequency of such gametes than are expected on purely chromosome segregation (the duplex and simplex heterozygotes). The expectations of recessives are $\frac{x}{8} + \frac{\alpha(1-x)}{4}$ from a triplex heterozygote, $\frac{2+x}{12} + \frac{\alpha(1-x)}{3}$ from a duplex and $\frac{1}{2} + \frac{\alpha(1-x)}{4}$ from a simplex. In the case of the triplex

the expectation is approximately $\frac{x+2\alpha}{8}$, for $\frac{\alpha x}{4}$ is relatively negligible.

The value of x in this case is not simply the proportion of non-disjunction, but includes a factor, not separately estimated, dependent upon the fact that an unbalanced gamete (and zygote), arising from non-disjunction, is less viable than a balanced one. A difference between the viability of gametes (and zygotes) with one chromosome short or in excess could be expected, but has been assumed absent in the present discussion.

Very small corrections to the duplex and simplex expectations should be applied if the formation of three gene double reductional gametes is taken into account. The exact expectations are $\frac{2+x}{12} + \frac{\alpha(4-x)}{12}$ for duplex and $\frac{1}{2} + \frac{\alpha(2-x)}{8}$ for simplex. The less exact expectations have been used in the following calculations, mainly because they are simpler to apply and display (table 5). However, the results of calculations using the more exact formulæ will be given.

The available information may be used in two ways to yield the parameters x and α . In the first place, we may consider jointly the

progeny of the three sorts of heterozygote, for which the expectations and observations, pooled from a large number of families, are given in table 4. The maximum values of the parameters, each on the assumption that the other is zero, may be estimated using Fisher's (1946) method of scores. This yields $\alpha = 0.0377 \pm 0.0059$ and $x = 0.0825 \pm 0.0136$. The value of x is well above what is plausible in view of the cytological data on rates of chromosome non-disjunction previously considered. We may therefore conclude that α does not take a zero value. On the other hand, there is no *a priori* expectation as to the maximum value of α for any particular locus, though it is

TABLE 4
*Observed segregations in backcross progeny of tetrasomics,
heterozygous for sugary-1, used as seed plants*

Heterozygote	Progeny	Observed	Expected proportions
TRIPLEX . . .	<i>su-1</i> ⁺	3326	$1 - \frac{x}{8} - \frac{\alpha(1-x)}{4}$
	<i>su-1</i>	33	$\frac{x}{8} + \frac{\alpha(1-x)}{4}$
DUPLEX . . .	<i>su-1</i> ⁺	5110	$\frac{5}{6} - \frac{x}{12} - \frac{\alpha(1-x)}{3}$
	<i>su-1</i>	1115	$\frac{1}{6} + \frac{x}{12} + \frac{\alpha(1-x)}{3}$
SIMPLEX . . .	<i>su-1</i> ⁺	3078	$\frac{1}{2} - \frac{\alpha(1-x)}{4}$
	<i>su-1</i>	3126	$\frac{1}{2} + \frac{\alpha(1-x)}{4}$

possible to make plausible guesses by the means discussed later, provided various other cytological and genetical parameters are known.

Maximum likelihood estimates of the parameters may be obtained jointly from the observed numbers. This is done by partial differentiation, for the two parameters in question, followed by the construction of scores and information per plant scored. The various items in the estimation and the values of the scores and information in a late round of iteration are given in table 5. It is clear that the estimates are not very precise, though the limits would not be symmetrically arranged about the estimates, because the probabilities of the events x and α are small.

After the parameters had been computed in this way, they were given a trial in the more exact formulæ, which take account of three gene, double reductional gametes. After several iterations this gave $x = 0.03380$, $V_x = 0.0019015$, $\alpha = 0.02382$, $V_\alpha = 0.00037295$.

These estimates of x and α may now be used to calculate, for each of the kinds of progeny, numerical expectations which may be compared with the observations. The comparison yields a χ^2 , for one

degree of freedom, of 0.235778, for which P is between 0.5 and 0.7. Thus the data are fitted very well by these estimates of the parameters.

The second method of estimation depends upon a joint genetical and cytological examination of the progeny of the triplex backcrosses. Amongst these there were 33 sugary seeds in the grand total of 3359.

TABLE 5
(a) Scores and information for joint estimation of x and α

Class	Progeny	x scores	α scores	i_{xx}	$i_{x\alpha}$	$i_{\alpha\alpha}$
TRIPLEX	$su-I^+$	$-\frac{1-2\alpha}{8-t}$	$-\frac{2(1-x)}{8-t}$	$\left. \begin{array}{l} \frac{(1-2\alpha)^2}{t(8-t)} \\ \frac{2(1-2\alpha)(1-x)}{t(8-t)} \end{array} \right\}$	$\frac{2(1-2\alpha)(1-x)}{t(8-t)}$	$\frac{4(1-x)^2}{t(8-t)}$
	$su-I$	$\frac{1-2\alpha}{t}$	$\frac{2(1-x)}{t}$			
DUPLEX	$su-I^+$	$-\frac{1-4\alpha}{10-u}$	$-\frac{4(1-x)}{10-u}$	$\left. \begin{array}{l} \frac{(1-4\alpha)^2}{(2+u)(10-u)} \\ \frac{4(1-4\alpha)(1-x)}{(2+u)(10-u)} \end{array} \right\}$	$\frac{4(1-4\alpha)(1-x)}{(2+u)(10-u)}$	$\frac{16(1-x)^2}{(2+u)(10-u)}$
	$su-I$	$\frac{1-4\alpha}{2+u}$	$\frac{4(1-x)}{2+u}$			
SIMPLEX	$su-I^+$	$\frac{\alpha}{2-v}$	$-\frac{1-x}{2-v}$	$\left. \begin{array}{l} \frac{\alpha^2}{(2-v)(2+v)} \\ -\frac{\alpha(1-x)}{(2-v)(2+v)} \end{array} \right\}$	$-\frac{\alpha(1-x)}{(2-v)(2+v)}$	$\frac{(1-x)^2}{(2-v)(2+v)}$
	$su-I$	$-\frac{\alpha}{2+v}$	$\frac{1-x}{2+v}$			

$$t = x + 2\alpha - 2\alpha x; u = x + 4\alpha - 4\alpha x; v = \alpha - \alpha x.$$

(b) Computations for $x = 0.03213$, $\alpha = 0.02463$

Class	Progeny	x scores	α scores	I_{xx}	$I_{x\alpha}$	$I_{\alpha\alpha}$
TRIPLEX	$su-I^+$	-399.2532	-812.8935	$\left. \begin{array}{l} 4803.3095 \\ 9779.7050 \end{array} \right\}$	9779.7050	19911.8255
	$su-I$	393.1138	800.3936			
DUPLEX	$su-I^+$	-466.6050	-2003.8737	$\left. \begin{array}{l} 240.8554 \\ 0.9410 \end{array} \right\}$	1034.3734	4422.2002
	$su-I$	472.4582	2029.0108			
SIMPLEX	$su-I^+$	38.3629	-1507.5215	$\left. \begin{array}{l} 0.9410 \\ -36.9795 \end{array} \right\}$	-36.9795	1453.1404
	$su-I$	-38.0432	1494.9608			
Totals		0.0335	0.0765	5045.1059	10777.0989	25807.1661

$$x + \delta x = 0.03213(2837) \\ \alpha + \delta \alpha = 0.02463(1759)$$

$$V_x = 0.00183628 \\ V_\alpha = 0.00035897$$

Efforts were made to determine the numbers of chromosome IV present in each of the sugary progeny. The results (table 2b) for 27 which were successfully analysed showed 9 which each had only three of chromosome IV in their somatic complements. This is altogether a very much higher frequency of non-disjunction than has been encountered for any other chromosome and it is very probable

that the egg from which each of these trisomic tetraploids was formed had only one chromosome IV, and that this single chromosome carried the *su-1* gene. The alternative explanation would be that the pollen parent introduced a single chromosome IV into the zygote. In this case the zygote must owe its sugary phenotype to double reduction in the formation of the egg. This would imply an unreasonably high frequency of functioning of male gametes deficient for one chromosome IV unless there were preferential fertilisation by them of eggs showing double reduction. In what follows it will be assumed that the 9, each with only three of chromosome IV, owe their origin to non-disjunction in egg formation and that the 18, each with four of chromosome IV, owe their origin to double reduction in egg formation.

We have then that the proportion of the triplex recessive gametes that arises through non-disjunction is one-third. The limit values,

TABLE 6

Rates of non-disjunction (x), double reduction (α) and $y = \alpha(1-x)$ for the sugary locus in tetraploid maize, using combined genetical and cytological analysis of triplex heterozygotes

Parameter	Experimental value	Upper limit $p = 0.05$	Lower limit $p = 0.05$
x	0.0262	0.0424	0.0130
y	0.0262	0.0328	0.0181
α	0.0269	0.0332	0.0189

probable at the 0.05 level, may be calculated with the aid of Steven's tables (1942). Using these numbers multiplied by a factor $\frac{33}{27}$, to take account of the fact that not all the sugary progeny were analysed cytologically, and by factors of 8 and 4 respectively, the upper and lower limits of x and y , $= \alpha(1-x)$, and thence α may be calculated. The results are given in table 6. The results of this method agree very well with those given by the first, purely genetic, method.

5. ESTIMATION OF THE PROPORTION (r) OF RECESSIVE GAMETES PRODUCED BY TRIPLEX HETEROZYGOTES

A triplex heterozygote may often be distinguished from a quadruplex (dominant homozygote) by its production of one or more recessive (nulliplex) offspring when pollinated by a nulliplex and always by the presence of simplex offspring in this same progeny. If no recessive offspring appear, leaving it uncertain whether the parent was quadruplex or triplex, the first backcross progeny must be tested by a second backcross to detect the presence of simplex plants or establish their absence with a given reasonable level of

probability to conclude that the original test plant was quadruplex rather than triplex. This second backcross test is laborious and time-consuming and has not been performed upon all the plants from which the data included here have been collected.

Some of the triplex plants used appeared in the progeny of selfed duplex plants. In this case, those plants which might have been quadruplex or triplex were tested by a second backcross. However, most of the triplex plants appeared in crosses between quadruplex and simplex plants, in which the majority of the offspring would be triplex or duplex, in equal proportion, and only a minority quadruplex through the occurrence of double reduction in the simplex parent. It has been assumed in this case that all families showing no recessives were in fact from triplex plants.

There were 33 *su* seeds among a total of 3359, or just under 1 per cent. The distribution of these recessives amongst the 24 families of triplex progenies is given in table 7.

TABLE 7
*Progenies from tetraploid maize, triplex for su-1⁺,
pollinated by nulliplex*

Number <i>su</i>	Sizes of progenies
0	184, 39, 105, 165, 170, 46, 65
1	211, 206, 76, 74, 18
2	259, 84, 65, 219, 86, 188, 131, 173, 181
3	251, 140
4	223

Mr G. N. Wilkinson (C.S.I.R.O. Division of Mathematical Statistics) has shown me that it is possible to obtain satisfactory estimates of the frequency of the recessives, omitting those families which lack any recessive. When dealing with truncated binomial probabilities of this description the expectation of a given family, among those considered (which must have at least one recessive) is

$$\frac{{}_n C_r p^r (1-p)^{n-r}}{1 - (1-p)^n}.$$

Maximum likelihood estimates of the proportion of recessives, and of their variance may be derived. Their computation involves an elegant mode of iteration, by means of reciprocals, that Mr Wilkinson will be describing elsewhere.

From the present point of view, it is interesting to note that the estimate of the size of the population sample containing the 33 *su* seeds is 3401.7, and leads to a parameter not significantly different from the value (33/3359) obtained on the assumption that all the zero class families in table 7 should be included in the estimate.

6. SEGREGATION RATIOS IN PLANTS TRISOMIC OR PENTASOMIC FOR A CHROMOSOME

Progeny which are trisomic or pentasomic may be expected to occur as a result of numerical non-disjunction in a tetrasomic plant. They may be detected, in some instances, by the special segregations shown when heterozygous for suitable markers. Two kinds of heterozygote, AAa and Aaa , are possible in the trisomic, and four kinds, $AAAAa$, $AAAaa$, $AAaaa$ and $Aaaaa$, in the pentasomic.

The trisomics will usually form gametes with one or two of these particular chromosomes and it is probable that those gametes with but one of these chromosomes and resulting zygotes with three (instead of four) would be less viable than those with balanced numbers.

TABLE 8

Segregation ratios in trisomic and pentasomic plants, with the viabilities of trisomic and pentasomic progeny respectively u and v

Trisomics	Gametes					Divisor	Ratios in backcross progeny $A : a$
	AA	Aa	aa	A	a		
AAa	1	2	...	$2u$	u	6	$3+2u : u$
Aaa	...	2	1	u	$2u$	6	$2+u : 1+2u$
Pentasomics	Gametes						Ratios in backcross progeny $A : a$
	AAA	AAa	Aaa	aaa	AA	Aa	
$AAAAa$	$2v$	$3v$	...	...	3	2	10
$AAAaa$	v	$6v$	$3v$	...	3	6	20
$AAaaa$	...	$3v$	$6v$	v	1	6	20
$Aaaaa$	...	...	$3v$	$2v$	...	2	10
							$1 : 0$ $9+10v : 1$ $7+9v : 3+v$ $2+3v : 3+2v$

Similarly, it is probable that the unbalanced gametes and zygotes with excess numbers, as in the pentasomic and its pentasomic progeny, would also be less viable than the balanced types. If the viability of the trisomic is u and of the pentasomic is v , the expectations with chromosome segregation are those shown in the two parts of table 8. With the great uncertainty introduced by the viability factor, it is hardly worth while considering any correction due to double reduction. The ratio of A to a in backcross progeny would vary with the value of u and v , according to the ways indicated in fig. 3. It will be noticed that Aaa and $Aaaaa$ may be confused with $Aaaa$ and AAa with $AAaa$ if u has a high value; $AAAAa$ will be confused with $AAAa$ under all circumstances, while $AAAaa$ and usually $AAaaa$ seem recognisable whatever value v might take.

One backcross family consisted of 160 su^+ and 57 su , a very bad agreement with the 5 : 1 ratio expected in a duplex backcross ($\chi^2 = 14.401$). The plant which gave this family was assumed to be a duplex pentasomic, namely $++su su su$. The viability, v , of the pentasomic, may be estimated as 0.229, and we should expect a fraction $\frac{9v}{7+9v}$, or 0.22746, of the dominant heterozygotes to be pentasomic, one-third of them duplex and two-thirds simplex. A

FIG. 3.—Relation between the expected gametic ratios of trisomic and pentasomic tetraploids and the viabilities of their trisomic and pentasomic progeny.

number of the dominant heterozygotes were planted. The chromosome number was counted in 39 plants and backcross progenies were obtained from every one of these. Two plants each had five chromosomes IV and their backcross progenies showed 45+ : 50 su and 94+ : 106 su , respectively. Evidently these two plants were $+su su su su$. If indeed the original parent were pentasomic we should expect the 39 plants tested to consist of 4.3 $++su su$, 25.82 $+su su su$, 2.96 $++su su su$ and 5.92 $+su su su su$. The observed numbers were 7, 30, 0 and 2. The χ^2 for three degrees of freedom is 7.9255

and P is 0.02-0.05. If the original plant had been tetrasomic, the duplex and simplex progeny would have been expected in the ratio of 1 : 4. While the results do not permit a perfectly definite conclusion, they are distinctly unfavourable to the assumption that the original plant was pentasomic. The two progeny that were pentasomic for chromosome IV could conceivably be fresh cases of numerical non-disjunction.

7. DISCUSSION

There can be no doubt that non-disjunction makes an appreciable contribution to gamete formation and that the effects of it have previously been confused with the effects of double reduction. This means that the rates of double reduction have been overestimated, particularly for loci close to the centromere. No other data suitable for analysis in the ways investigated above appear to be present in the literature. Frequently, one of the types of mating is missing from the data. In other cases, the numbers of progeny that have been enumerated are too small. Finally, some sets of tetrasomic segregation data show evidence of differential viability, which tend to swamp the effects due to non-disjunction and double reduction. In no other case has it been possible to differentiate, amongst the progeny of triplex heterozygotes, those with three or four of the chromosome concerned.

It is interesting to consider how far the estimate of double reduction determined above agrees with the value expected from the interaction of the various contributory factors. The statistics for the cytological factors are largely based on data in a Cambridge Ph.D. thesis written by Dr Jillela Venkateswarlu, to whom I am indebted for permission to use the relevant material.

The coefficient of double reduction, α , is dependent upon the following coefficients: (1) the amount of crossing-over (c) between the centromere and the locus; (2) the frequency (q) of quadrivalent formation; (3) the frequency (p) of adjacent (or parallel) disjunction of quadrivalents; and (4) the frequency (d) with which adjacent disjunction of the quadrivalent results in non-disjunction of the genes in paired chromosome arms. These parameters are related by the formula $\alpha = cqpd$, since the half chance of having the necessary disjunctional arrangement at division II of meiosis is offset by the double chance of the necessary cross-over in each cell. Dr Venkateswarlu's data indicate that the value of q , averaged for all chromosomes (I to X), is 0.769 and the value of p is 0.373. The value of d may be taken as 0.5, in the absence of any evidence of how the difference in lengths of chromosome arms affects the chances of the two parallel arrangements. Thus in maize the average relation would be $\alpha = 0.769 \times 0.373 \times 0.5 \times c = 0.1434c$. For the sugary-1 locus, c is not less than 0.09 (Anderson and Randolph, 1945) and this would indicate 0.0129 as a minimum value for α . Alternatively, from our measurement of α , we could argue that the value of c is closer to 0.19.

It would be useful to obtain information of this nature for a series of loci at different positions with respect to the centromere. We should expect that the contribution due to non-disjunction would be constant for a given chromosome, and that α would rise from a zero value at the centromere to a maximum of $\frac{1}{2}$, except that this might be exceeded if certain conditions of interference were fulfilled. The maximum value of one-seventh could be achieved only if q were unity and p were one-half.

8. SUMMARY

1. There are three components of variation in tetraploid segregation, *viz.* chromosome and chromatid segregation, and numerical non-disjunction. The two latter components have not been discriminated from one another in previous studies, but it has been possible to separate all three in an experiment with the sugary-1 locus in chromosome 4 of tetraploid maize. The ten different chromosomes can be distinguished at mitosis, and unbalanced complements have been demonstrated in many plants.

2. The coefficient of double reduction (α) at the sugary-1 locus is approximately 2.7 per cent. The coefficient of non-disjunction (x) is about 2.6 per cent., when estimated from triplex backcross progeny, amongst which the recessives are analysed cytologically. Estimates made from triplex, duplex and simplex data treated together give maximum likelihood estimates of $\alpha = 2.4$ per cent. and $x = 3.4$ per cent.

9. REFERENCES

- ANDERSON, E. G., AND RANDOLPH, L. F. 1945. Location of the centromeres on the linkage maps of maize. *Genetics*, 30, 518-526.
- FISHER, R. A. 1946. A system of scoring linkage data, with special reference to the pied factors in mice. *Amer. Nat.*, 80, 568-578.
- FISHER, R. A., AND MATHER, K. 1943. The inheritance of style length in *Lythrum salicaria*. *Ann. Eugenics*, 12, 1-23.
- KADAM, B. S. 1944. Chromosome studies in relation to fertility and vigour in inbred and open-pollinated strains of autotetraploid maize. I. Behaviour of 40-chromosome plants. *Indian J. Genet. and Plt. Brdg.*, 4, 8-22.
- MATHER, K. 1936. Segregation and linkage in autotetraploids. *J. Genet.*, 32, 287-314.
- PUNYASINGH, K. 1947. Chromosome numbers in crosses of diploid, triploid and tetraploid maize. *Genetics*, 32, 541-554.
- STEVENS, W. L. 1940. Accuracy of mutation rates. *J. Genet.*, 43, 301-307.

FIG. 2.—The somatic chromosome complement of a 39 chromosome plant having only three of chromosome IV.