

RELATION OF ASSOCIATION AND ORIENTATION OF CHROMOSOMES IN INTERCHANGE HETEROZYGOTES OF RICE AND THEIR FERTILITY⁽¹⁾

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In the case of interchange heterozygotes obtained either spontaneously or artificially induced, the quadripartite group of chromosomes may be oriented at MI so that either alternate or adjacent chromosomes are directed toward the same pole. The alternate type of segregation would have the zigzag configuration. Whereas the adjacent type would have the open one. Theoretically, the adjacent segregation would have two different types. In plant material of some species, the alternate chromosomes of this interchange complex would pass to the same pole 70-90% of the time, while in others, alternate and adjacent segregation are about equally frequent. The former is called the directed segregation type with low ovule and pollen sterility. Examples are *Oenothera*, *Triticum monococcum*, *T. durum*. The other type is non-directed segregation with pollen and ovule abortion usually about 50%. Examples are *Zea mays*, *Pisum sativum*, etc. (Burnham 1956). This model had been quoted repeatedly in the textbooks of genetics and cytogenetics. In his extensive review of chromosomal interchanges in plants, Burnham (1956) also mentioned that:

1. Chains can be formed instead of rings if one long piece has exchanged with a very short one.

2. In species with very short chromosome, or with low crossover frequency, a reciprocal translocation may be present, but the association is two bivalents.

There may be other types of association present and the orientation of these different types of association would be different either genetically controlled or differentiated by mechanical structural difference. The gametes so produced would also vary. In view of the complexity of the problem and the problem and the paucity of experimental data to correlate the types of association and orientation of this interchange complex in relation to the fertility of the plant concerned, especially lacking being, the quantitative data, we try in

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this paper to present the data obtained concerning this problem.

Only very recently, Ahloowalia (1962) working with rye gave some quantitative data about the type of orientation and association of chromosomes in relation to pollen fertility. In all, there were five inbred strains studied, and 120 cells counted. Ahloowalia found: zigzag 54, ring 12, N-chain 40, U-chain 7, and saucer-pan 7. The pollen fertility was found to be 74.6% for these 5 inbred strains studied, and 88.1% for 5 normal strains. By our recalculation, this was 85.81% fertile as compared with the normal. If the types as zigzag, N-chain and saucer-pan can be counted to give rise to fertile pollen, then theoretically the percentage of pollen fertility was 84.17% which was very close to that of the observed.

In rice, since the chromosomes are small, therefore very little work has been done along this line. Katayama (1963), made some attempt to associate the orientation and association of chromosomes in translocation heterozygote with the fertility of rice plant, but the data presented were too meager to be of any significance.

Material and methods






Four varieties of rice, two japonica and two indica were treated by different doses of X-rays and thermal neutrons in the Brookhaven National Laboratory back in 1961. We were very grateful to this said institution. This project has been aimed at the production of induced mutations, particularly erectoides, early lines, and others for a large scale breeding program. As many as 800 panicles in X_1 were fixed and examined cytologically. Structural mutations such as reciprocal translocations ($\odot 4, 2\odot 4, \odot 6$ or $\odot 8$), telocentric and acentric fragments, inversions, pseudo-isochromosomes, monosomes, trisomes, and others were found (unpublished). In trying to reevaluate some of these structural mutants, about 200 X_3 lines were fixed and examined cytologically. It occurred to us then that we had many different lines of interchange heterozygotes and these lines differed greatly in the types of association and orientation for interchange complex. If quantitative data could be obtained, they would offer some explanation concerning either pollen or ovule fertility, or both.

Propiano-carmin was used exclusively in this study.


Results

There were altogether 52 lines studied finally. Our original plan was aimed at least 100 lines or more. But since only one panicle was fixed from each plant, it was rather difficult to make good preparations with only few spikelets available. Furthermore, in all the lines of one variety of the four dealt with, the chromosomes were apt to clump together, so that the interchange complexes

Table 1. Types of interchange complex association and orientation

Pedigree	Open ring	Zigzag	Chain	U-chain	N-chain
					
1. T25-33-17	6			4	1
2. T25-24-6	38	1	9		1
3. C4-4-2	49		4		2
4. I25-7-12	3		7	12	15
5. C4-5-7	46	3		3	
6. I25-5-2	20		3		
7. C15-19-2	25	12	1		
8. C5-9-20	17		1	3	4
9. I25-3-7	30			10	8
10. C5-6-7	26	4	10	1	6
11. I20-5-6	5	8		1	8
12. I25-1-17	4			17	7
13. I25-2-13	17	5		3	2
14. C20-22-10	9	2	1	1	1
15. I20-5-9	13	2		5	4
16. T25-25-4	34	4	3		1
17. T25-25-3	24		1	1	
18. T15-13-15	20	3	1		2
19. T25-18-3	36	6			
20. T25-30-6	10	1			
21. T25-20-1	10	3	1		2
22. T25-15-10	25				
23. T25-8-12	12	7	2	2	6
24. T25-8-17	26	1	1	1	4
25. T25-9-20	28		2	1	
26. T25-18-91	26	11	1		
27. T15-29-10	13	4			2
28. T15-18-3	6	1			1
29. T15-29-2	10	15	2		
30. T20-21-6	22	4	1	2	2
31. C20-12-9	18	5	1		8
32. C20-5-16			3	20	18
33. W15-3-6	26	4			
34. W25-5-9	24		2		4
35. T52-26-1	17	10		1	1
36. W21-4-1	18		4		11
37. C20-6-13	19	8			
38. T15-7-17	32				
39. T15-7-14	36				1
40. T15-14-10	28		1		1
41. T15-7-1	29	1			2
42. C5-3-20	6	6	5		
43. C5-4-14	8	1	4		5
44. C4-5-19	19	6	3		1
45. T15-7-7	29	5	1		
46. T25-15-12	14	4			
47. C4-4-3	33	5		1	2
48. C3-4-4	32	1	2		3
49. C3-5-20	22	23			3
50. T25-12-19	11	9	1		
51. T25-7-9	12	5	1	2	2
52. T25-16-13	8	6			2

in different plants together with pollen and seed fertility

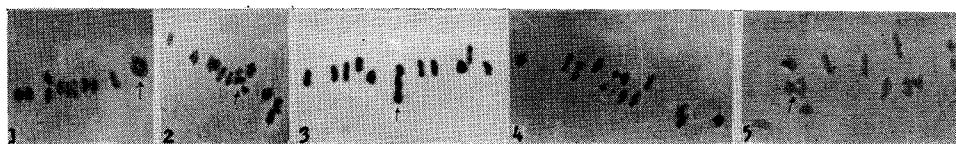
Saucerpan	Bivalent	Total	Theoretical expectancy %	Pollen Fertility %	Seed Fertility %
	() ()				
	12	23	28.00	18.49	17.73
	14	63	12.69	43.55	8.63
	13	68	12.65	6.25	10.16
	7	44	42.00	66.13	33.89
	10		17.70	30.38	13.95
	8		19.20	42.24	31.09
	5	43	33.70	43.10	17.31
	20	45	31.11	22.06	23.51
	5	53	19.39	31.40	22.08
1	22	70	30.14	38.39	15.67
1	8	31	67.74	57.82	31.45
2	6	36	33.33	30.90	6.97
	35	62	39.51	56.52	21.01
1	6	21	28.57	34.61	44.64
4	10	38	39.73	38.80	28.73
	10	52	19.23	36.12	8.79
	3	29	5.17	28.61	18.24
2	28	56	37.50	39.09	26.04
	14	56	23.21	36.16	23.25
11	22	44	52.27	47.06	19.23
	13	29	39.65	57.42	23.01
	8	33	12.12	38.02	17.93
	16	45	46.66	58.54	20.63
2	13	48	28.12	56.23	34.37
	19	50	19.00	37.11	26.04
	4	42	30.95	30.00	14.13
	13	32	39.06	51.36	22.02
	17	25	42.00	42.26	23.25
1	6	34	55.88	62.58	25.46
1	15	47	30.85	46.33	26.81
	5	37	47.29	61.54	32.56
	6	47	44.67	53.85	53.24
	20	50	28.00	48.50	23.58
6	4	40	30.00	41.29	3.87
	8	37	40.54	50.66	15.50
5	6	44	43.18	44.24	38.86
	14	41	37.07	40.20	16.83
	9	41	10.97	36.00	24.03
	5	42	8.33	24.66	5.41
1	9	40	16.25	18.00	18.52
	5	37	14.86	36.54	22.60
14	20	51	58.82	66.34	47.46
11	33	62	45.16	67.32	24.68
	23	52	33.86	63.84	37.39
1	6	42	21.43	35.03	18.59
1	14	33	36.36	38.05	18.44
	13	54	25.00	29.97	9.87
1	17	56	22.32	22.22	29.17
1	15	64	53.84	43.25	30.12
	11	32	45.31	46.63	30.44
	9	31	37.09	50.64	35.91
5	23	44	55.68	46.74	24.55

of the preparations of the plants of this variety could be studied only with great difficulty. So finally, we had to give up the study of this variety altogether.

There were altogether 5 plants fixed from one line. Theoretically, as far as interchange complexes were concerned, the genetical constitution of one plant would be more or less the same as the plant with the same line. However, with rye interchanges, Rees (1961) found that in case of two independent interchanges, A and B studied in F_3 to F_7 in inbred lines, the heterozygotes of A were favored by selection compared with homozygotes, those of B were not. These interchanges had been studied before by Thompson (1956) who suggested that the disjunction of these interchange complexes was under genotypic control and that it was possible to select the zigzag segregation (from F_3 - F_6) in the interchange heterozygote. Since the material used in our study consisted of F_3 plants, there would be some interplant variation in the same line. Therefore, if it could be avoided, only one plant from a single line was used in this study.

As the chromosomes in rice were rather small. Counts were done only from preparation having well spread metaphase I figures. The association and orientation of the interchange complex encountered were open ring, zigzag, chain, U-chain, N-chain, saucer-pan, and 12 bivalents. Those with bivalents only but having partial sterility were not studied in detail and were not included.

These associations and orientations were shown in figures 1-5 inclusive. In our earlier plan we aimed to make counts of 50 cells or more from each plant but we could attain our goal only partially.



Figs. 1-5. The association and orientation of the interchange complexes at metaphase I. The arrow showing: Fig. 1, a ring, Fig. 2, N-chain, Fig. 3, a chain, Fig. 4, a saucer-pan, Fig. 5, a zigzag.

The theoretical expectancy was calculated by assuming that those zigzag, N-chain, and saucer-pan orientations would give all fertile gametes; bivalents 50%; and those open ring, chain, and U-chain would produce complete sterile gametes. Assumption was based on the supposition that the deficiency-duplication gametes would be completely sterile, and there were no extra chromosome gametes. In fact, Catcheside (1963) found in *Oenothera* that the progenies of the interchange heterozygote to be mostly of the balanced type (Table 2) and the frequency of duplication gametes as well as that of extra chromosome gametes was very small. Therefore, they could be discounted in

the calculation if rice would have comparable situation as that of *Oenothera*.

Pollen fertility or stainability was obtained by the examination of no less than 300 pollen grains. Seed fertility was obtained by counting from table 1, it can be seen that the lines differed greatly in the kind of association as well as the orientation of the chromosomes in the interchange complex. Most of the lines would have predominantly the open ring orientation. Few would have the bivalents as the predominant type of association. In only three lines did we find the zigzag orientation to be the predominant type. The calculated theoretical expectancies based on these observations in MI varied from as low as 5.17 to as high as 67.70% fertility. The correlation coefficient calculated between the theoretical expectancy and pollen fertility $r=0.6424$, $P=0.01$, and $r=0.3512$, $P=0.025$ for theoretical and seed fertility. Both were significant statistically. (correlation was assumed to be zero)

Discussion

Association and orientation on the metaphase plate of the chromosomes of the interchange complex may be governed by:

1. Length of chromosome involved,
2. Position of the breaks,
3. Number and position of chiasmata, and
4. Degree of terminalization of chiasmata.

(Swanson 1957, P. 176)

These would make this problem more complex which was originally found in *Zea mays* (Burnham 1956).

With rice, having smaller chromosomes, there are other types of chromosome association and orientation on the metaphase plate found besides the zigzag and open ring ones. Some plants have all of these associations. Others would have a few, predominantly of one or two types. Since these plants differ greatly in the fertility (pollen and seed) from one another, and the correlation coefficients calculated from the data are statistically significant. Therefore conclusion can be drawn that fertility of the rice plant is highly correlated with the association and orientation of the chromosomes of the interchange complex in interchange heterozyotes.

However, the fit is not too ideal either between the expectation and pollen fertility or that and seed fertility. The discrepancy may be explained as follows:

1. To start with, the number of lines studied is not big enough, nor is the number of cells examined large enough to be representative for each plant, especially for those when only about 30 samples are taken.
2. As being mentioned earlier, the chromosomes of rice are rather small.

Some of the configurations may be wrongly classified, particularly the zigzag configuration is easily confused with the open ring type.

3. Pollen stainability of each plant observed is higher than the expected. Perhaps if both interchanged pieces are short, the pollen may be normal or nearly normal in appearance. Shortness of the deficiency is undoubtedly not the only determining criterion, rather it is the physiological effect when the genes are missing (Burnham 1956, P. 436). At least, this factor may offer to explain part of the discrepancy.

4. In general, seed fertility is lower than the expectation. Environmental factors would creep in to enhance the effect which is already being offered by the association and orientation of the interchange complex.

Summary

In all, 52 plants representing almost as many lines of interchange heterozygotes of rice plant in the X_3 generation involving three varieties of rice after the seeds were irradiated by X-ray and thermal neutrons.

About 50 cells were examined from each plant. According to kinds of association and types of orientation, the quadripartite chromosomes of the interchange complex were classified as open-ring, zigzag, chain, U-chain, N-chain, saucer-pan and bivalents, and their frequency recorded. From these the theoretical expectancy of fertility of each plant was calculated.

Correlation coefficients were calculated between the expected fertility with pollen fertility and seed fertility, with $r=0.6424$ and 0.3512 respectively, and they were found to be statistically significant.

It seemed that either pollen or seed fertility would depend primarily upon the type of association of the chromosomes of interchange complex and their orientation in meiosis.

水稻異質互換 (Interchange Heterozygotes) 的 染色體配對和排列方向與其可孕率的關係

李先聞 楊桂淑 李淑媛 何閨綺

本文所用的材料是三個水稻品種經放射線 (X光或熱中子) 處理種子的第三代 (X_3) 植株。52株可以代表52個異質轉座 (translocation heterozygote) 系。

每一植株儘可能觀察50個細胞按染色體配合的種類和排列方式, 由四條染色體組成的互換複合體 (interchange complex) 可以為環形, 鋸齒形, 鏈狀, U形鏈狀, N形鏈狀, 平底鍋形或12個二價體。由其發生的頻度計算每一植株的理論的期望可孕率。

期望可孕率和充實花粉率間以及期望的可孕率與結實率間的相關係數依次為 $r=0.6424$

和 0.3512。顯著性測驗的結果其相關顯著。

因此，花粉率和結實率主要是互換複合體在減數分裂時染色體的配合情形和排列方向來決定的。

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