

## Quadrant II – Transcript and Related Materials

**Programme:** Bachelor of Science (Third Year)

**Subject:** Botany

**Paper Code:** BOC 108

**Paper Title:** Cytogenetics and Plant Breeding

**Unit:** 04

**Module Name:** Recombination frequency, Two-point and Three-point test crosses

**Module No:** 22

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### Notes

In crossing over, non-sister chromatids of two homologous chromosomes pair up with each other and exchange different segments of genetic material to form recombinants.

**Recombination frequency** is the measure of genetic linkage. It is the frequency with which a single chromosomal cross will take place between two genes during meiosis.

To understand this concept, let us take one example.

Consider a cross between two organisms      AABB X aabb

The gametes produced are      AB   ab

Therefore the genotype of F1 generation will be      AaBb

We work out the recombination frequency to find out whether A and capital B or a and b are linked completely. We do a test cross, wherein the individual from F1 generation is crossed with the homozygous recessive individual.

AaBb   X   aabb

On crossing we get four types of phenotypes

|    | AB   | ab   | Ab   | aB   |
|----|------|------|------|------|
| ab | AaBb | aabb | Aabb | aaBb |
|    | 212  | 224  | 85   | 79   |

Notice that AaBb with value 212 and aabb with value 224 are parental. Whereas 'Aabb' with a value of 85 and 'aaBb' with a value of 79 are recombinants.

We calculate recombination frequency by the formula:

$$\text{Recombination frequency} = \frac{\text{Number of recombinants}}{\text{Total number of progeny}} \times 100$$

On substituting the values we get

$$\begin{aligned}\text{Recombination frequency} &= \frac{164}{500} \times 100 \\ &= 32.8\%\end{aligned}$$

It means that there is 32% chance for a recombination to take place.

When two genes are located on different chromosomes or when they are widely separated on the same chromosome, the recombination frequency is 50%

When two genes are close together on the same chromosome, they are said to be linked and have a recombination frequency of less than 50%.

The use of recombination frequencies has been used to develop linkage maps.

## Two-point and Three-point test crosses

### Two-point test crosses

The percentage of crossing over between two linked genes is calculated by test crosses in which a F1 dihybrid is crossed with a double recessive parent. Such crosses because involve crossing over at two points, so called two point test cross. For example, a dihybrid cross having the genotype AC/ac is test crossed with a double recessive parent (ac/ac), then among F2 test cross hybrids we may get 37% dominant genes at both gene loci (AC/ac), 37% recessive at both gene loci (ac/ac), 13% dominant gene at first gene locus and recessive at the second gene locus (AC/ac), and 13% recessive gene at first gene locus and dominant gene at second gene locus (Ac/AC). The last two groups (i.e.13% Ac/ac) and 13% Ac/ac) were produced by

crossover gametes (13+13) were of cross over types and the distance between loci A and C is estimated to be 26 centimorgans. Because double crossovers usually do not occur between genes less than 5 centimorgans apart, so for genes further apart, the three point test crosses are used.

### Three-point test crosses

A three point test cross or trihybrid test cross (involving three genes) gives us information regarding relative distances between these genes, and also shows us the linear order in which these genes should be present on the chromosome. Such a three point test cross may be carried out if three points or gene loci on a chromosome pair can be identified by marker genes. If in addition to genes A and C, a third marker gene B is located in fairly close proximity in the same linkage group, all three markers may be used together in conducting a more precise analysis of the map distance and the relative position of the three points. Suppose we test cross trihybrid individuals of genotypes ABC/abc and find in the progeny the following:

|                   |                                                   |                                                   |                      |
|-------------------|---------------------------------------------------|---------------------------------------------------|----------------------|
| 36%<br>ABC/abc    | 9%<br>Abc/abc                                     | 4% Abc/abc                                        | 1% AbC/abc           |
| 36% abc/abc       | 9%<br>Abc/abc                                     | 4% abC/abc                                        | 1% aBc/abc           |
| 72% Parental type | 18% Single cross overs between A and B (region I) | 8% Single cross overs between B and C (region II) | 2% Double crossovers |

To find the distance A-B we must count all cross overs (both singles and doubles) that occurred in region I =  $18\% + 2\% = 20\%$  or 20 map units between the loci A and B. to find the distance B-C we must again count all crossovers (both singles and doubles) that occurred in region II =  $8\% + 2\% = 10\%$  or 10 map units between loci B and C. the A- C distance is therefore, 30 map units when double crossovers are detected in a three point linkage experiment and 26 map units when double crossovers are undetected in the two-point linkage experiment.

For any three linked genes whose distances are known, the amount of detectable crossovers between the two outer markers A and C when the middle marker B is missing is

$(A-B \text{ cross over percentage}) + (B-C) \text{ crossover percentage} - (2 \times \text{double crossover percentage})$

### **Significance of two point test cross and three point test cross in chromosome mapping**

A two point test cross and a three point test cross is used to determine the loci of genes in an organism's genome.

Enables to map all of the tens of thousands of genes found on the chromosomes of plants and animals.

Chromosome mapping done with the help of two point or three point test cross data is very important in the treatment of certain diseases as it allows the experts to identify and locate the disease causing genes