CHAPTER III

MATERIAL AND METHODS
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The chapter comprises of the methods employed and materials utilized during the course of investigation on tomatoes (*Lycopersicon esculentum* Mill.) undertaken at Agriculture Research Farm, College of Agriculture, Indore (M.P.) during 1983-84 and 1984-85. The investigations carried out are grouped under the following:

1. **Analysis of the genetical architecture of the yield components in** *Lycopersicon esculentum* **Mill.** based on estimation of gene effects through analysis of generation means following the method proposed by Hayman (1958).

2. **Estimation of components of heterosis and inbreeding depression in** $F_1$, $F_2$ and $F_3$ **generations.**

The material for investigation comprised six promising genotypes of *Lycopersicon esculentum* Mill. The genotypes were selected on the basis of diversity of origin, morphological differences and possible utility in breeding programme aimed at high yield, disease resistance (cracking/rotting) and nutritional quality. A list of these genotypes with brief description is as follows:
### Pedigree and Brief Description of Genotypes of *Lycopersicon esculentum* Mill. used in Genetic Analysis

<table>
<thead>
<tr>
<th>No.</th>
<th>Genotype</th>
<th>Pedigree</th>
<th>State in which selected/developed</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>ACC 99</td>
<td>Single plant selection of germplasm received from USA through Plant Introduction Division, I.A.R.I., New Delhi.</td>
<td>M.P.</td>
<td>Typical dwarf type with medium large fruits and early in ripening. It yields 450 q/ha but highly susceptible to fruit cracking/rotting.</td>
</tr>
<tr>
<td>2.</td>
<td>ACC 72</td>
<td>- do -</td>
<td>M.P.</td>
<td>Tall type with small medium sized fruits and late in ripening. It yields 400 q/ha, and moderately resistant to fruit cracking/fruit rot.</td>
</tr>
<tr>
<td>3.</td>
<td>ACC 5</td>
<td>- do -</td>
<td>M.P.</td>
<td>Tall type with large fruits (upto 400 g/fruit) and very late in ripening. Highly susceptible to fruit cracking and fruit rot.</td>
</tr>
<tr>
<td>4.</td>
<td>Pusa Ruby</td>
<td>Sioux and Improved Meerut</td>
<td>Delhi</td>
<td>Indeterminate tall type with medium sized fruits and early in ripening. It yields 300 q/ha, and moderately resistant to fruit cracking and fruit rot and excess rains.</td>
</tr>
<tr>
<td>5.</td>
<td>Sweet 72</td>
<td>Pusa Red Plum and Sioux</td>
<td>M.P.</td>
<td>Tall type with small sized fruits and late in ripening. It yields 250 q/ha and fairly resistant to fruit cracking and fruit rot.</td>
</tr>
<tr>
<td>6.</td>
<td>Marglobe</td>
<td>Marvel globe</td>
<td>America</td>
<td>Medium type with medium sized fruits and moderately resistant to fruit cracking and fruit rot.</td>
</tr>
</tbody>
</table>
Seed of these genotypes were made available by the Head, Department of Vegetable Crops, Jawaharlal Nehru Krishi Vishwa Vidyalaya, Jabalpur. The parent seed material was sown in pots in order to make sufficient F₁'s and later on F₂'s and F₅'s seeds.

**Field Plot Technique:**

During 1983-84 and 1984-85, the parents, F₁, F₂ and F₅'s were grown at Agricultural Research Farm, College of Agriculture, Indore. The material was planted in randomized block design with three replications. Each genetic group was considered as a single unit in the process of randomization. The components of each genetic group viz. P₁, P₂, F₁, F₂ and F₅ plots consisted of 1, 1, 1, 4 and 8 rows, respectively, each surrounded by border row from the same genetic material.

The experiment was conducted on black cotton soil and experimental plots received a basal dose of 30 kg N, 80 kg P₂O₅ and 40 kg K₂O/ha. The seeds were sown on 30th November, 1983 and 22nd December, 1984 during 1983-84 and 1984-85, respectively and 35 days old seedlings were used for transplanting. The seeds were treated with Diathane M-45 before sowing. The weeding and earthing operations were done to provide the better growth conditions,
timely. 70 kg N was applied as top dressing in two split
doses at 20 days and 45 days intervals after transplantation.
The plots were also irrigated at desired intervals.
Malathion (insecticides) and Diathane M-45 (fungicide) were
sprayed twice in the season to provide plant protection
umbrella against insects pests and diseases.

Details of Experimental Layout:

Dimensions of the experiment:

<table>
<thead>
<tr>
<th>Description</th>
<th>Dimensions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gross Area</td>
<td>47.0 M x 40 M</td>
</tr>
<tr>
<td>Net Area</td>
<td>27.0 M x 30 M</td>
</tr>
<tr>
<td>Planting distance</td>
<td>60 cm x 60 cm</td>
</tr>
<tr>
<td>No. of rows in each</td>
<td>125</td>
</tr>
<tr>
<td>replication</td>
<td></td>
</tr>
<tr>
<td>No. of plants for</td>
<td>10 plants/row</td>
</tr>
<tr>
<td>observation</td>
<td></td>
</tr>
</tbody>
</table>

Number of Parents and Crosses:

1. ACC 99                  Parent
2. ACC 72                  Parent
3. ACC 5                   Parent
4. Pusa Ruby               Parent
5. Sweet 72                Parent
6. Marglobe                Parent
7. ACC 99 x Pusa Ruby      F₁
8. ACC 99 x Sweet 72  
9. Pusa Ruby x ACC 5  
10. ACC 72 x ACC 99  
11. ACC 72 x Marglobe  
12. ACC 99 x Pusa Ruby  
13. ACC 99 x Sweet 72  
14. Pusa Ruby x ACC 5  
15. ACC 72 x ACC 99  
16. ACC 72 x Marglobe  
17. ACC 99 x Pusa Ruby  
18. ACC 99 x Sweet 72  
19. Pusa Ruby x ACC 5  
20. ACC 72 x ACC 99  
21. ACC 72 x Marglobe  

Crop and Climatic Conditions:

The meteorological data during the crop period were obtained from meteorological observatory, College of Agriculture, Indore and presented in Appendix-I. The climatic conditions were favourable for crop growth during both the years of experimentation.

Recording of Experimental Data:

The data were recorded on 10 randomly selected plants in each row. The number of plants in parents, $F_1$, $F_2$ and
$F_3$ were 10, 10, 10, 40 and 80, respectively. All the characters were measured on individual plant basis.

I. **Phenological Characters:**

1. **Days to 1st flowering:** The number of days taken from sowing to the date of opening of first flower in the generation plot.

2. **Days to 50% flowering:** The number of days taken from sowing to the date of opening of first flower in 50% plants.

3. **Days to 50% maturity:** The number of days taken from sowing to the date of maturity of fruits in 50% plants.

II. **Growth Characters:**

1. **Plant height:** The mean height of the largest branch from the base of plant to the base of the highest leaf was measured at the time of maturity.

2. **Number of branches per plant:** Total number of primary branches were recorded at maturity.

3. **Stem girth:** The mean girth of the stem was measured at the base of the main stem by caliper.
III. Yield Components:

1. **Weight per fruit:** Ten matured fruits were randomly selected from individual plant. Weight of these fruits was recorded and average weight per fruit was calculated.

2. **Size of fruit:** Ten matured fruits were selected randomly from each individual plant and length and breadth of fruit was measured by caliper. The fruit size was calculated by multiplying length and breadth.

3. **Yield per plant:** All the fruits harvested during the various picking from selected plants were weighed and average yield per plant was calculated.

4. **Number of locules per fruit:** The matured fruits were randomly selected from each individual plant and fruits were cut into two halves from middle of the fruit by sharp knife and number of locules per fruit were recorded.

**Cracked/Rotted Fruits per Plant:**

The fruits damaged due to cracking or fruit rot, which were not suitable for marketing were also separated and weighed per plant. The diseased fruits were also
examined for association of causal organism.

**Ascorbic acid and reducing sugar determination:**

Ascorbic acid was estimated as per the standard method of Colorimetric (Ruck, 1963) and reducing sugar was determined by using Hane’s Ferricyanide method described by Browne and Zerban (1952).

**Statistical techniques:**

The estimate of various mean effects of the gene and non-allelic interaction components based on the generation means following the analysis suggested by Hayman (1958) and reported by Singh and Choudhary (1979) was followed. Ignoring the linkage and taking the $F_2$ means as reference value $m$ to estimate:

$$d' = (d - i) \text{ additive minus additive x dominance effect.}$$

$$h = \text{dominance effect}$$

$$i = \text{additive x additive effect}$$

$$l = \text{dominance x dominance effect}$$

$$m = F_2$$
The five of the six genetic parameters viz. m, d', h, l and i were estimated following the relationship between generation means and gene effects.

\[ P_1 = m + d' - \frac{1}{4}h + l + \frac{1}{4}l \]
\[ P_2 = m - d' - \frac{1}{4}h + l + \frac{1}{4}l \]
\[ F_1 = m + \frac{1}{4}h - \frac{1}{4}l \]
\[ F_2 = m \]
\[ F_3 = m - \frac{1}{4}h + \frac{1}{16}l \]
\[ F_n = m - \left(\frac{1}{4} - 2^{-n} + \frac{1}{4}\right) h + \left(\frac{1}{4} - 2^{-n} + \frac{1}{4}\right) 2 l \]
\[ n = 1, 2, 3, 4 \]

and the expected values, components of variation obtained by least square computation are as follows:

\[ m = F_2 \]
\[ d' = \frac{1}{4} (F_1 - F_2) \]
\[ h = 1/6 \left(4 F_1 - 12 F_2 - 16 F_3\right) \]
\[ l = 1/6 \left(16 F_3 - 24 F_2 + 8 F_1\right) \]
\[ i = F_1 - F_2 + \left(\frac{1}{4}\right) (F_1 - F_2 + h) - \frac{1}{4} l \]

**Standard error of the estimates:**

For this purpose, variance of the estimates were calculated. Square root of the respective variance denotes
the standard error of the estimate.

\[ V(m) = VF_2 \]

\[ V(d') = \frac{1}{6} (VF_1 + VF_2) \]

\[ V(h) = \frac{1}{36} (VF_1 + 144 VF_2 + 256 VF_3) \]

\[ V(l) = \frac{1}{9} (256 VF_3 + 576 VF_2 + 64 VF_1) \]

\[ V(l) = VF_1 + VF_2 + \frac{1}{6} (VF_1 + VF_2 + Vh) + 1/16 Vl \]

Now S.E.m. for \( m = \sqrt{V_m} \)

\[ d' = \sqrt{Vd'} \]

\[ h = \sqrt{Vh} \]

\[ l = \sqrt{Vl} \]

\[ l = \sqrt{Vl} \]

't' value was obtained by dividing the values of respective estimate by S.E.m. of the same estimate.

Confidence interval was calculated by multiplying the S.E.m. of the respective estimate by the 't' value (Table 't' i.e. 1.96).

**Heterosis and Inbreeding depression:**

Since back cross data were not available, estimate of heterosis was calculated from the following formulae:

\[ \text{Heterosis (H)} = |(h) - (l)| - |(d')| \]
where \( d' = (d-j) \), which however, will give all the estimates of heterosis.

Let \( F_1 \) represents the mean of \( F_1 \) over three replications and \( mp \), the average of the two parents involved in the cross under considerations. The heterosis is measured as the proportion of deviation of the \( F_1 \) value from the midparental value.

\[
\text{Heterosis} \% = \frac{F_1 - mp}{mp} \times 100
\]

Similarly heterosis over better parent was calculated by the formula -

\[
\text{Heterosis} \% = \frac{F_1 - bp}{bp} \times 100
\]

Let \( F_2 \) represent the mean of the \( F_2 \) over three replications, inbreeding depression was given by the formula -

\[
\text{Inbreeding depression} \% = \frac{F_1 - F_2}{F_1} \times 100
\]

The variations amongst the means of parents, \( F_1 \), \( F_2 \) and \( F_3 \) generations obtained for different characters were tested through normal analysis of variance technique. The means were compared with critical difference at 5% level of significance. The analysis of variance are presented in appendix II to XI.