CHAPTER – III
MATERIALS AND METHODS

The field experiment was conducted at the Sagdividi Farm, Department of Seed Science and Technology, Junagadh Agricultural University, Junagadh during summer 2016 to study the “Characterization of okra (Abelmoschus esculentus (L.) Moench) genotypes through plant morphology and seed quality parameters” and the laboratory studies were carried out in the laboratory of Department of Seed Science and Technology, Junagadh Agricultural University, Junagadh in situated at 21.30°N latitude and 70.25°E longitudes with an altitude of 60 meters above the mean sea level. The soil of the experimental site was medium black, alluvial in origin and poor organic matter. Meteorological data on maximum and minimum temperature, relative humidity and rainfall of summer 2016 was obtained from the meteorological observatory, Junagadh Agricultural University, Junagadh. (Appendix I).

Detail of the material used and methodologies adopted in the present investigation are described in this chapter.

3.1 Treatment details (Genotypes)

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Genotype</th>
<th>Source</th>
<th>Genotype</th>
<th>Source</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Kashi Lalima</td>
<td>IIVR, Varansi</td>
<td>IC-43733</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>2</td>
<td>Kashi Kranti</td>
<td>IIVR, Varansi</td>
<td>KS-404</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>3</td>
<td>Kashi Vibhuti</td>
<td>IIVR, Varansi</td>
<td>131-10-1,2,3,4</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>4</td>
<td>Kashi Satdhari</td>
<td>IIVR, Varansi</td>
<td>29-10-1</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>5</td>
<td>Kashi Lila</td>
<td>IIVR, Varansi</td>
<td>EC-169417</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>6</td>
<td>Kashi Pragati</td>
<td>IIVR, Varansi</td>
<td>151-10-1,2,3</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>7</td>
<td>IC-45831</td>
<td>IIVR, Varansi</td>
<td>IC-43748</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>8</td>
<td>440-10-1</td>
<td>IIVR, Varansi</td>
<td>AOL-03-1</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>9</td>
<td>IC-169359</td>
<td>IIVR, Varansi</td>
<td>GJO-3</td>
<td>JAU, Junagadh</td>
</tr>
<tr>
<td>10</td>
<td>SC-35</td>
<td>IIVR, Varansi</td>
<td>204-10-1</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>11</td>
<td>323-10-1</td>
<td>IIVR, Varansi</td>
<td>JOL-10-18</td>
<td>JAU, Junagadh</td>
</tr>
<tr>
<td>12</td>
<td>IC-282240</td>
<td>IIVR, Varansi</td>
<td>IC-33345</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>13</td>
<td>EC-3016556</td>
<td>IIVR, Varansi</td>
<td>492-9-1</td>
<td>IIVR, Varansi</td>
</tr>
<tr>
<td>14</td>
<td>EC-329357</td>
<td>IIVR, Varansi</td>
<td>IC-169511</td>
<td>IIVR, Varansi</td>
</tr>
</tbody>
</table>
3.2 Experimental details

3.2.1 Experimental design and layout

The field experiment was laid out in a Randomized Block Design (RBD) with three replications and the laboratory experiment was laid out in a Completely Randomized Design (CRD) with four replications. The seeds were hand dibbled in the row at spacing of 45 × 30 cm. The experiment was surrounded by guard row to avoid damage and border effects. The recommended agronomical practices and plant protection measures were followed for the successful raising of the crop. The observations were recorded on five randomly selected plants in each entry of replication and their mean values were used for the statistical analysis.

3.3 Observation recorded

3.3.1 Characterization of okra genotypes through stem characteristics

(1) Stem Colour:

The colour of stem was observed on visual assessment basis at the 30 days after planting and the genotypes were grouped as green and red colour.

(2) Intensity of green colour on stem:

The observation was observed on visual assessment of intensity of green colour of stem at the 30 days after planting and the genotypes were grouped as light, medium and dark.

(3) Stem diameter (10 cm above ground level):

The average stem diameter of 5 randomly selected plants was measured in centimetre at the 70 days after planting.

<table>
<thead>
<tr>
<th>Category</th>
<th>Diameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Small</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Medium</td>
<td>1-1.5</td>
</tr>
<tr>
<td>Large</td>
<td>&gt;1.5</td>
</tr>
</tbody>
</table>

(4) Stem: number of nodes at first flowering:

The numbers of nodes were calculated at the 60 days after planting.

<table>
<thead>
<tr>
<th>Category</th>
<th>No. of nodes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Few</td>
<td>&lt;5</td>
</tr>
<tr>
<td>Medium</td>
<td>5-8</td>
</tr>
<tr>
<td>Many</td>
<td>&gt;8</td>
</tr>
</tbody>
</table>
3.3.2 Characterization of okra genotypes through leaf characteristics

(1) Number of leaves per plant:
   Number of leaves per plant was counted at maximum growth stage.

(2) Length of leaf blade (cm):
   The length of 5 leaves were measured in centimetre of each genotype at the at 60 days after planting and the genotypes were grouped as small, medium and large.

(3) Width of leaf blade (cm):
   The width of 5 leaves were measured in centimetre of each genotype at the 60 days after planting and the genotypes were grouped as small, medium and large.

(4) Leaf vein colour:
   The colour of leaf vein was observed on visual assessment basis at the 60 days after planting and the genotypes were grouped as light green and Purple.

(5) Leaf blade (depth of lobbing):
   The average depth of lobbing was observed on visual assessment basis at the 60 days after planting and the genotypes were grouped as shallow, medium and deep.

3.3.3 Characterization of okra genotypes through flower characteristics

(1) Flower petal colour:
   The colour of flower petal was observed on visual assessment basis at the 50 days after planting and the genotypes were grouped as cream, yellow and purple.

(2) Flower length (cm):
   The length of 5 flowers were measured in centimetre of each genotype at the 50 days after planting and the genotypes were grouped as small, medium and large.

(3) Flower diameter (at the top of flower):
   The diameter of 5 flowers were measured in centimetre of each genotype at the 50 days after planting and the genotypes were grouped as small, medium and large.

(4) Pedicel length (cm):
   The length of 5 pedicels were measured in centimetre of each genotype at the 50 days after planting and the genotypes were grouped as short, medium and long.

(5) Days to 50% Flowering:
   The number of days taken from sowing to the emergence of first flower in 50 per cent of plant in each genotype was recorded and the genotypes were grouped as follows:
### Materials and methods

<table>
<thead>
<tr>
<th>Category</th>
<th>Number of days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Early days</td>
<td>&lt;35 days</td>
</tr>
<tr>
<td>Medium days</td>
<td>35 to 45</td>
</tr>
<tr>
<td>Late days</td>
<td>&gt;45 days</td>
</tr>
</tbody>
</table>

3.3.4 Characterization of okra genotype through fruit characteristics

1. **Fruit colour:**
   The colour of fruit was observed on visual assessment basis at the time of physiological maturity and the genotypes were grouped as green, red and purple.

2. **Fruit length (cm):**
   The length of 5 fruits was measured in centimetre of each genotype at the time of maturity and the genotypes were grouped as Small, Medium and Long.

3. **Fruit diameter (cm):**
   The diameter of 5 fruit was measured in centimetre of each genotype at the time of maturity and the genotypes were grouped as small, medium and large.

<table>
<thead>
<tr>
<th>Category</th>
<th>Diameter</th>
</tr>
</thead>
<tbody>
<tr>
<td>Small</td>
<td>&lt;1</td>
</tr>
<tr>
<td>Medium</td>
<td>1-1.5</td>
</tr>
<tr>
<td>Large</td>
<td>&gt;1.5</td>
</tr>
</tbody>
</table>

4. **Number of fruits per plant:**
   The total number of fruits of 5 selected plants from each genotype was counted at the time of maturity.

3.3.5 Characterization of okra genotypes through plant characteristics

1. **Plant: Number of branches:**
   The average plant branches of 5 randomly selected plants were calculated at the time of maturity.

<table>
<thead>
<tr>
<th>Category</th>
<th>Branch</th>
</tr>
</thead>
<tbody>
<tr>
<td>Few</td>
<td>&lt;2</td>
</tr>
<tr>
<td>Medium</td>
<td>2-4</td>
</tr>
<tr>
<td>Many</td>
<td>&gt;4</td>
</tr>
</tbody>
</table>
(2) **Plant height (cm):**

The average plant height of 5 randomly selected plants was measured in centimetre at the time of maturity.

<table>
<thead>
<tr>
<th>Category</th>
<th>Height</th>
</tr>
</thead>
<tbody>
<tr>
<td>Short</td>
<td>&lt;90</td>
</tr>
<tr>
<td>Medium</td>
<td>90-120</td>
</tr>
<tr>
<td>Tall</td>
<td>&gt;12</td>
</tr>
</tbody>
</table>

### 3.3.6 Characterization of okra genotypes through seed characteristics

(1) **Seed colour:**

The colour of seed was observed on visual assessment basis after harvesting and the genotypes were grouped as green and brown.

(2) **Seed hairiness:**

The hairiness of seed was observed on visual assessment basis after harvesting and the genotypes were grouped as present or absent of hair in seed.

(3) **Number of seeds per fruit:**

It was determined at maturity by counting the total number of seeds in average of 5 randomly selected pods of 5 plants.

(4) **Seed yield per plant (g):**

The seed yield per plant was calculated in grams by extracting seeds of average 5 randomly selected plants at the time of maturity.

(5) **100-seed weight (g):**

It was determined by weighing 100 dry seeds as sample from the bulk of each accession.

### 3.3.7 Characterization of okra genotypes through seed germination and seedling characteristics

(1) **Speed of Germination:**

Germination percentage was calculated as per ISTA (International Seed Testing Association) Rules (Anon., 1993).

\[
\text{Speed of germination} = \frac{n_1}{d_1} + \frac{n_2}{d_2} + \ldots + \frac{n_N}{d_N}
\]

Where, \( n = \) no. of seed germinated

\( d = \) no. of days to germinated
(2) Seed Germination percentage:

The germination test was conducted as per the ISTA (International Seed Testing Association) Rules (Anon., 1993) using petri plate methods. Petri plate was placed in a seed germinator at constant temperature of 25± 1°C and 95± 1 per cent relative humidity final count on normal seedlings was recorded on eighth day and percent germination was computed by following formulation:

\[
\text{Germination percentage (\%)} = \frac{\text{Number of seeds germinated}}{\text{Total number of seeds kept for germination}} \times 100
\]

(3) Shoot length (cm):

Five normal seedlings were selected at random from the germination test. The length between the collar region and the tip of the shoot was measured as shoot length in centimeter.

(4) Root length (cm):

Five normal seedlings were selected at random from the germination test. The length between the collar region and the tip of the primary root was measured as root length in centimeter.

(5) Seedling length (cm):

Five normal seedlings were selected at random from the germination test. The length measured from the tip of root to the tip of shoot of seedling in centimeter.

\[
\text{Seedling length} = \text{Shoot length (cm)} + \text{Root length (cm)}
\]

(6) Seedling fresh weight (mg):

The seedling after measuring length was weighed in mg separately and averaged.

(7) Seedling dry weight (mg):

The seedling after measuring length was dried in shade for 12 hour and then hot air oven dried at 100 °C for 24 hour and weighed in mg separately and averaged.

(8) Seedling vigour index I:

The vigour index (VI) was calculated according to Abdul Baki and Anderson (1973) and expressed in whole number by following formula:

\[
\text{VI} = \text{Germination percentage} \times \text{mean seedling length (cm)}
\]
(9) Seedling vigour index II:
The vigour index (VI) was calculated according to Abdul Baki and Anderson (1973) and expressed in whole number by following formula:
\[ VI = \text{Germination percentage} \times \text{seedling dry weight (mg)} \]

(10) Seed moisture content:
Seed moisture content was calculated as per ISTA (International Seed Testing Association) Rules (Anon., 1993).
\[
\text{% Moisture content} = \frac{\text{Weight of fresh seeds} - \text{Weight of dry seeds}}{\text{Weight of fresh seeds}} \times 100
\]

3.4 Statistical analysis
The statistical analysis on following aspects was carried out:

1. Analysis of variance for Randomized Block Design (RBD) of experiment carried out at field.

2. Analysis of variance for Completely Randomized Design (CRD) of experiment carried out at laboratory.

The characters viz., stem diameter (cm), number of nodes at first flowering, number of leaves per plant, length of leaf blade (cm), width of leaf blade (cm), flower length(cm), flower diameter (cm), pedicel length (cm), days to 50% Flowering, fruit length (cm), Fruit diameter (cm), number of fruits per plant, number of branches per plant, plant height (cm), number of seeds per fruit, seed yield per plant (g), 100-seed weight (g) was analysed by Randomized Block Design (RBD).

The characters viz., speed of germination, germination percentage, shoot length (cm), root length (cm), seedling length (cm), seedling fresh weight (mg), seedling dry weight (mg), seedling vigour index, seed moisture content were analysed by Completely Randomized Design (CRD).
3.4.1 Randomized Block Design

Analysis of variance for Randomized Block Design was computed as per the method of Cochran and Cox (1957), which is based on the following mathematical model:

\[ Y_{ij} = \mu + g_i + r_j + e_{ij} \]

Where,

- \( Y_{ij} \) = Phenotypic expression of \( i^{th} \) genotype in \( j^{th} \) replication,
- \( \mu \) = Population mean,
- \( g_i \) = An effect of \( i^{th} \) genotype,
- \( r_j \) = An effect of \( j^{th} \) replication and
- \( e_{ij} \) = Un controlled variation associated with \( i^{th} \) genotype and \( j^{th} \) replication.

The form of analysis of variance as presented in the Table 3.1 was constructed for individual characters viz., stem diameter (cm), number of nodes at first flowering, number of leaves per plant, length of leaf blade (cm), width of leaf blade (cm), flower length (cm), flower diameter (cm), pedicel length (cm), days to 50% Flowering, fruit length (cm), Fruit diameter (cm), number of fruits per plant, number of branches per plant, plant height (cm), number of seeds per fruit, seed yield per plant (g), 100-seed weight (g).

### Table 3.1: Analysis of variance for Randomized Block Design

<table>
<thead>
<tr>
<th>Sources of variation</th>
<th>D.f.</th>
<th>Mean square</th>
<th>Expected mean squares</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replications</td>
<td>(r-1)</td>
<td>( M_r )</td>
<td>( \sigma_e^2 + g\sigma_{e_g}^2 )</td>
</tr>
<tr>
<td>Genotypes</td>
<td>(t-1)</td>
<td>( M_g )</td>
<td>( \sigma_e^2 + r\sigma_{e_r}^2 )</td>
</tr>
<tr>
<td>Error</td>
<td>(r-1) (t-1)</td>
<td>( M_e )</td>
<td>( \sigma_e^2 )</td>
</tr>
</tbody>
</table>

Where,

\( R \) = Number of replications,
\( G \) = Number of genotypes,
**Materials and methods**

\[ M_r = \text{Mean sum of square due to replications}, \]
\[ M_g = \text{Mean sum of square due to genotypes}, \]
\[ M_e = \text{Mean sum of square due to error}, \]
\[ g\sigma^2_e = \text{Expected genotypic variance}, \]
\[ r\sigma^2_e = \text{Expected replication variance and} \]
\[ \sigma^2_e = \text{Expected error variance}. \]

Mean squares due to different sources were tested against error mean square \((M_e)\) by calculating ‘F’ values.

The standard error of mean (S. Em.) was calculated using following formula.

\[ S.\text{Em} = \sqrt{\frac{\text{ErrorMS}}{r}} \]

The critical difference (C.D.) to compare the mean of any two genotypes was calculated using following formula.

\[ \text{C.D.} = S.\text{Em} \times \sqrt{2} \times t \]

Where,

\[ t = \text{Table value of ‘t’ at 5% level of significant and error degree of freedom} \]

The coefficient of variation (C.V.) was determined according to the following formula.

\[ \text{C. V.} (%) = \frac{\text{ErrorMS}}{\bar{x}} \times 100 \]

Where,

\[ \bar{x} = \text{General mean} \]

**3.4.2 Genetic variability, heritability and genetic advance**

(1) **Phenotypic coefficient of variation (PCV %)**

The phenotypic coefficient of variation, which measures the magnitude of phenotypic variation present in a particular character, was estimated as per the formula suggested by Burton (1952)
Materials and methods

\[ \text{PCV} (\%) = \frac{\sqrt{\sigma^2_p}}{\bar{X}} \times 100 \]

Where,
\[
\sigma^2_p = \text{Phenotypic variance} \\
\bar{X} = \text{Mean of the character}
\]

(2) Genotypic coefficient of variation (GCV %)

The genotypic coefficient of variation, which measures the magnitude of genetic variation present in a particular character, was estimated as per the formula suggested by Burton (1952)

\[ \text{GCV} (\%) = \frac{\sqrt{\sigma^2_g}}{\bar{X}} \times 100 \]

Where,
\[
\sigma^2_g = \text{Genotypic variance} \\
\bar{X} = \text{Mean of the character}
\]

The GCV and PCV were classified as followed as suggested by Sivasubramanian and Madhvamenon (1973).

Low : (0 - 10%)
Moderate : (10.1-20%)
High : (>20%)

(3) Heritability (Broad sense)

It is the ratio of genotypic variance (\(\sigma^2_g\)) to the phenotypic variance(\(\sigma^2_p\)), was calculated according to formula suggested by Allard (1960).

\[ \text{Heritability} (h^2) = \frac{\sigma^2_g}{\sigma^2_p} \times 100 \]

Where,
\[
\sigma^2_g = \text{Genotypic variance} \\
\sigma^2_p = \text{Phenotypic variance}
\]
The range of heritability according to Robinson (1996).

Low : (0 - 50%)
Moderate : (50-70%)
High : (>70%)

(4) Genetic advance (GA)

The expected genetic advance under selection \( (G_s) \) was estimated as per the formula described by Allard (1960).

\[
G_s = k \times h^2 \times \sigma_p
\]

Where,

\( G_s \) = Genetic advance under selection
\( k \) = Selection differential (value of \( k \) at 5% selection intensity is 2.06)
\( \sigma_p \) = Phenotypic standard deviation
\( h^2 \) = Heritability value of the character

(5) Genetic advance expressed as percentage of mean

The genetic advance expressed as percentage of mean was computed as under:

\[
GA \text{ as } \% \text{ of mean} = \frac{\text{Genetic advance } (G_s)}{\text{Mean of character } (X)} \times 100
\]

The Genetic advance expressed as percentage of mean according to Johnson et al. (1955).

Low : (0 - 10%)
Moderate : (10.1-20%)
High : (>20%)

3.4.3 Correlation coefficient:

Correlation coefficient is the measurement of relationship between two or more series of variables. The genotypic correlation coefficient provides a measure of genotypic association between different characters, while phenotypic correlation includes both genotypic as well as environmental influences.
The phenotypic and genotypic correlation coefficients of all the pair of characters were worked out as per Al-Jibouri et al. (1958). The data were subjected to covariance analysis from which different components of mean sum of products were estimated. The format of analysis of covariance is given as under.

**Table: 3.2 The format of analysis of covariance between two characters**

<table>
<thead>
<tr>
<th>Source of Variation</th>
<th>d.f.</th>
<th>Mean of sum of products</th>
<th>Expected mean sum product</th>
</tr>
</thead>
<tbody>
<tr>
<td>Replications</td>
<td>(r-1)</td>
<td>MP&lt;sub&gt;r&lt;/sub&gt;</td>
<td></td>
</tr>
<tr>
<td>Genotypes</td>
<td>(g-1)</td>
<td>MP&lt;sub&gt;g&lt;/sub&gt;</td>
<td>Cov&lt;sub&gt;exy&lt;/sub&gt; + Cov&lt;sub&gt;gxy&lt;/sub&gt;</td>
</tr>
<tr>
<td>Error</td>
<td>(r-1)(g-1)</td>
<td>MP&lt;sub&gt;e&lt;/sub&gt;</td>
<td>Cov&lt;sub&gt;exy&lt;/sub&gt;</td>
</tr>
</tbody>
</table>

Where,

\[ r = \text{Number of replications} \]
\[ g = \text{Number of genotypes} \]
\[ \text{Cov} = \text{Covariance} \]

**1. Genotypic covariance (Cov<sub>(xy)g</sub>)**

Formula for calculating genotypic covariance is described as below:

\[ \text{Cov}_{(xy)g} = \frac{(M_g - M_e)}{r} \]

Where,

\[ M_g = \text{Mean sum of products due to genotypes between variable } x \text{ and } y \]
\[ M_e = \text{Mean sum of products due to error between variable } x \text{ and } y \]
\[ r = \text{Number of replications} \]

**2. Phenotypic covariance (Cov<sub>(xy)p</sub>)**

Formula for calculating genotypic covariance is described as below:

\[ \text{Cov}_{(xy)p} = \text{Cov}_{(xy)g} + \text{Cov}_{exy} \]

Where,

\[ \text{Cov}_{exy} = \text{Error covariance} = M_e \]
Now, genotypic and phenotypic variance and covariance were used for calculating the genotypic and phenotypic correlation coefficient, respectively (Al-Jibouri et al. 1958).

**a. Genotypic correlation coefficient \((r_{gxy})\)**

\[
    r_{gxy} = \frac{\text{Cov}(xy)_{g}}{\sqrt{\sigma_{gx}^2 \cdot \sigma_{gy}^2}}
\]

Where,

- \(\text{Cov}(xy)_{g}\) = Genotypic covariance between two character \(x\) and \(y\).
- \(\sigma_{gx}^2\) = Genotypic variance for character \(x\)
- \(\sigma_{gy}^2\) = Genotypic variance for character \(y\)

**b. Phenotypic correlation coefficient \((r_{pxy})\)**

\[
    r_{pxy} = \frac{\text{Cov}(xy)_{p}}{\sqrt{\sigma_{px}^2 \cdot \sigma_{py}^2}}
\]

Where,

- \(\text{Cov}(xy)_{p}\) = Phenotypic covariance between two character \(x\) and \(y\).
- \(\sigma_{px}^2\) = Phenotypic variance for character \(x\)
- \(\sigma_{py}^2\) = Phenotypic variance for character \(y\)

**c. Test of significance**

The significance of the correlation coefficient values at \(n-2\) degrees of freedom was tested by adopting the formula described by Panse and Sukhatme (1985).

**3.4.4 Path coefficient analysis**

Path coefficient is a standardized partial regression coefficient and measures the direct and indirect influence of one variable upon another thereby permitting the separation of the correlation coefficient into the component of direct and indirect effects. The original concept of path coefficient analysis was given by Smith (1936). The path coefficient analysis was carried-out as per the method suggested by Dewey and Lu (1959). Genotypic correlation coefficients of nine variables with seed yield
per plant were used to estimate the path coefficients for the direct effects of various independent characters on seed yield per plant.

The path coefficients were obtained by saving simultaneous equation which represents the basic relationship between correlation and path coefficients of the form given below:

\[ r_{ny} = P_{ny} + r_{n2}y_2 + r_{n3}y_3 + \ldots, + r_{nx}P_{xy} \]

Where,

\[ r_{ny} = \text{Correlation coefficient between one component Character and seed yield} \]

\[ P_{ny} = \text{Path coefficient between the character and seed yield} \]

The following genotypic correlation matrix was formed,

\[
\begin{pmatrix}
  r_{1y} \\
  r_{2y} \\
  r_{3y} \\
  \vdots \\
  r_{ny}
\end{pmatrix}
= 
\begin{pmatrix}
  1 & r_{12} & r_{13} & \ldots, & \ldots, & r_{1n} \\
  r_{21} & 1 & r_{23} & \ldots, & \ldots, & r_{2n} \\
  r_{31} & r_{32} & 1 & \ldots, & \ldots, & r_{3n} \\
  r_{n1} & r_{n2} & r_{n3} & \ldots, & \ldots, & 1
\end{pmatrix}
\begin{pmatrix}
  P_{1y} \\
  P_{2y} \\
  P_{3y} \\
  \vdots \\
  P_{ny}
\end{pmatrix}
\]

Where,

\[ r_{12}, r_{21} \text{ and so on} = \text{correlation between component characters} \]

\[ r_{1y}, r_{2y} \text{ and so on} = \text{correlation between component characters and yield} \]

\[ P_{12}, P_{21} \text{ and so on} = \text{Path coefficient of character on yield} \]

The technique given by Goulden (1962) was followed for inversion of the ‘B’ matrix using partitioning method of matrix inversion.

Path coefficients (\(P_{ij}\)) were obtained as follows:
\[ P_{ij} = (B^{-1}) \times (A) \]

The indirect effect for a particular character through other character was obtained by multiplication of direct path and particular correlation coefficient between those two characters, respectively.

Indirect effect = \( r_{ij} \times P_{ij} \)

Where,

\[ i = 1, \ldots, \ldots, n \]

\[ j = 1, \ldots, \ldots, n \]

\[ P_{ij} = P_{1y} + P_{2y} + \ldots + P_{ny} \]

The residual variable was computed from the following formula:

Residual variable (X) = 1 – \( R^2 \)

Where,

\[ R^2 = P_{1y} \times r_{1y} + P_{2y} \times r_{2y} + \ldots + P_{ny} \times r_{ny} \]

3.4.5 Completely Randomized Design

Analysis of variance for Completely Randomized Design was computed as per the method of Cochran and Cox (1957), which is based on the following mathematical model:

\[ Y_{ij} = \mu + g_i + e_{ij} \]

Where,

\[ Y_{ij} = \text{Phenotypic expression of i}^{\text{th}} \text{genotype in j}^{\text{th}} \text{replication}, \]

\[ \mu = \text{Population mean}, \]

\[ g_i = \text{An effect of i}^{\text{th}} \text{genotype and} \]

\[ e_{ij} = \text{Un controlled variation associated with i}^{\text{th}} \text{genotype and j}^{\text{th}} \text{replication}. \]

The form of analysis of variance as presented in the Table 3.2 was constructed for individual characters viz., speed of germination, germination percentage, shoot length (cm), root length (cm), seedling length (cm), seedling fresh weight (mg), seedling dry weight (mg), seed vigour index I, seedling vigour index II and seed moisture content.
Materials and methods

Table 3.3: Analysis of variance for Completely Randomized Design

<table>
<thead>
<tr>
<th>Sources of variation</th>
<th>D.f.</th>
<th>Mean square</th>
<th>Expected mean squares</th>
</tr>
</thead>
<tbody>
<tr>
<td>Genotypes</td>
<td>(t-1)</td>
<td>M_g</td>
<td>$\sigma^2_g + r\sigma^2_e$</td>
</tr>
<tr>
<td>Error</td>
<td>(r-1) (t-1)</td>
<td>M_e</td>
<td>$\sigma^2_e$</td>
</tr>
</tbody>
</table>

Where,

- $G$ = Number of genotypes,
- $M_g$ = Mean sum of square due to genotypes,
- $M_e$ = Mean sum of square due to error,
- $r\sigma^2_e$ = Expected replication variance and
- $\sigma^2_e$ = Expected error variance.

Mean squares due to different sources were tested against error mean square ($M_e$) by calculating ‘F’ values.

The standard error of mean (S. Em.) was calculated using following formula.

$$S.\text{Em} = \sqrt{\frac{\text{Error M.S}}{r}}$$

The critical difference (C.D.) to compare the mean of any two genotypes was calculated using following formula.

$$\text{C.D.} = S.\text{Em} \times \sqrt{2} \times t$$

Where,

- $T$ = Table value of ‘t’ at 5% level of significant and error degree of freedom.

The coefficient of variation (C.V.) was determined according to the following formula.

$$\text{C. V.} (%) = \sqrt{\frac{\text{Error M.S}}{\bar{x}}} \times 100$$

Where,

- $\bar{x}$ = General mean