

PRACTICAL MANUAL

# BREEDING OF VEGETABLE, TUBER AND SPICE CROPS

Course No. HVS-301; Credit Hrs. 3(2+1)

For B.Sc. (Horticulture) III-year (1<sup>st</sup> Semester)



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2020

College of Horticulture and Forestry  
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**Syllabus: Breeding of Vegetable, Tuber and Spice Crops**

**Practical:** Floral biology and pollination mechanism in self- and cross-pollinated vegetables, tuber crops and spices. Working out phenotypic and genotypic heritability, genetic advance. GCA, SCA, combining ability, heterosis, heterobeltosis, standard heterosis, GxE interactions (stability analysis) Preparation and uses of chemical and physical mutagens. Polyploidy breeding and chromosomal studies. Techniques of F1 hybrid seed production. Maintenance of breeding records.

**Name of Student** .....

**Roll No.** .....

**Batch** .....

**Session** .....

**Semester** .....

**Course Name:** .....

**Course No.:** .....

**Credit** .....

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in the year.....in the respective lab/field of College.

Date:

Course Teacher

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**Experiment No. 1**

**Objective: To study floral biology, emasculation and pollination of Tomato.**

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

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**Calyx:** .....

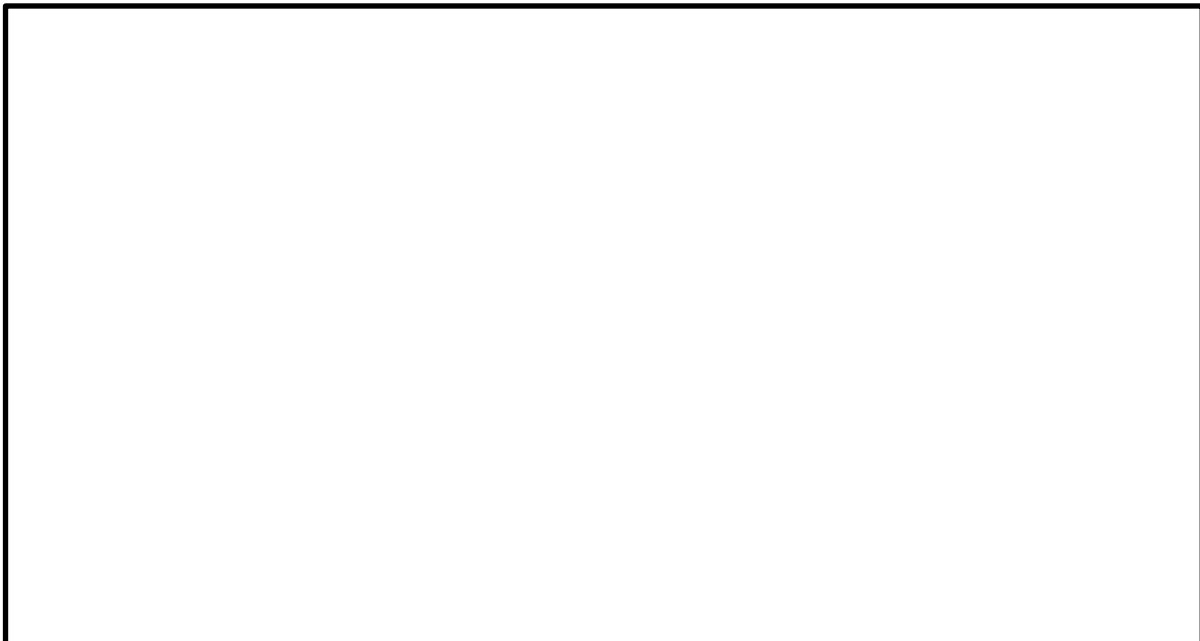
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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Floral formula:** .....

**Draw flower structure and floral diagram:**





## Experiment No. 2

**Objective:** To study floral biology, emasculation and pollination of Cucumber.

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

**Male flower**.....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Floral formula:** .....

**Male flower**.....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Floral formula:** .....



### Experiment No. 3

**Objective:** To study floral biology, emasculation and pollination of Indian bean.

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Fruit:** .....

**Floral formula:** .....

**Emasculation and crossing techniques:** .....

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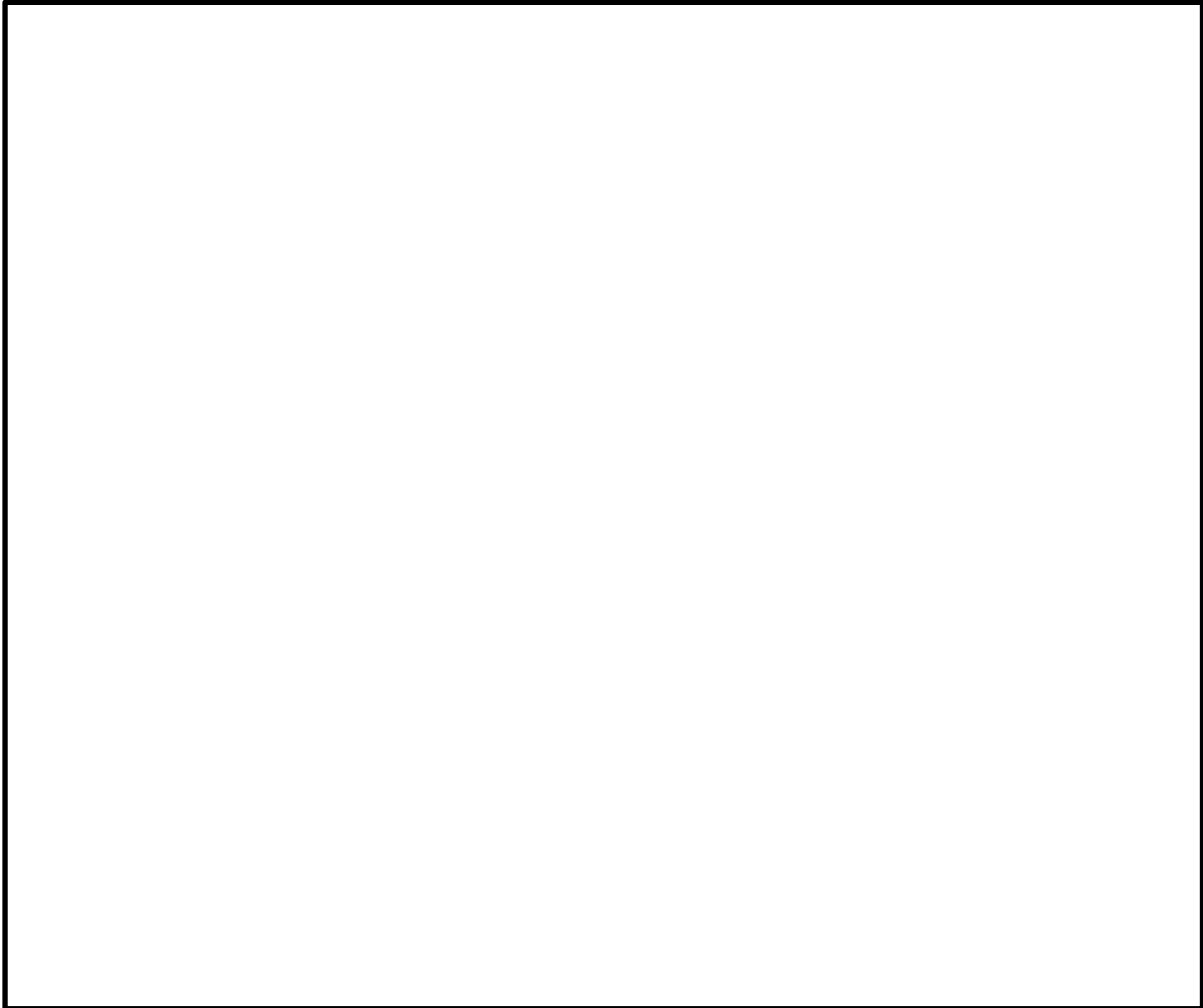
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**Draw flower structure and floral diagram:**



## Experiment No. 4

**Objective:** To study floral biology, emasculation and pollination of Okra.

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Fruit:** .....

**Floral formula:** .....

**Emasculation and crossing techniques:** .....

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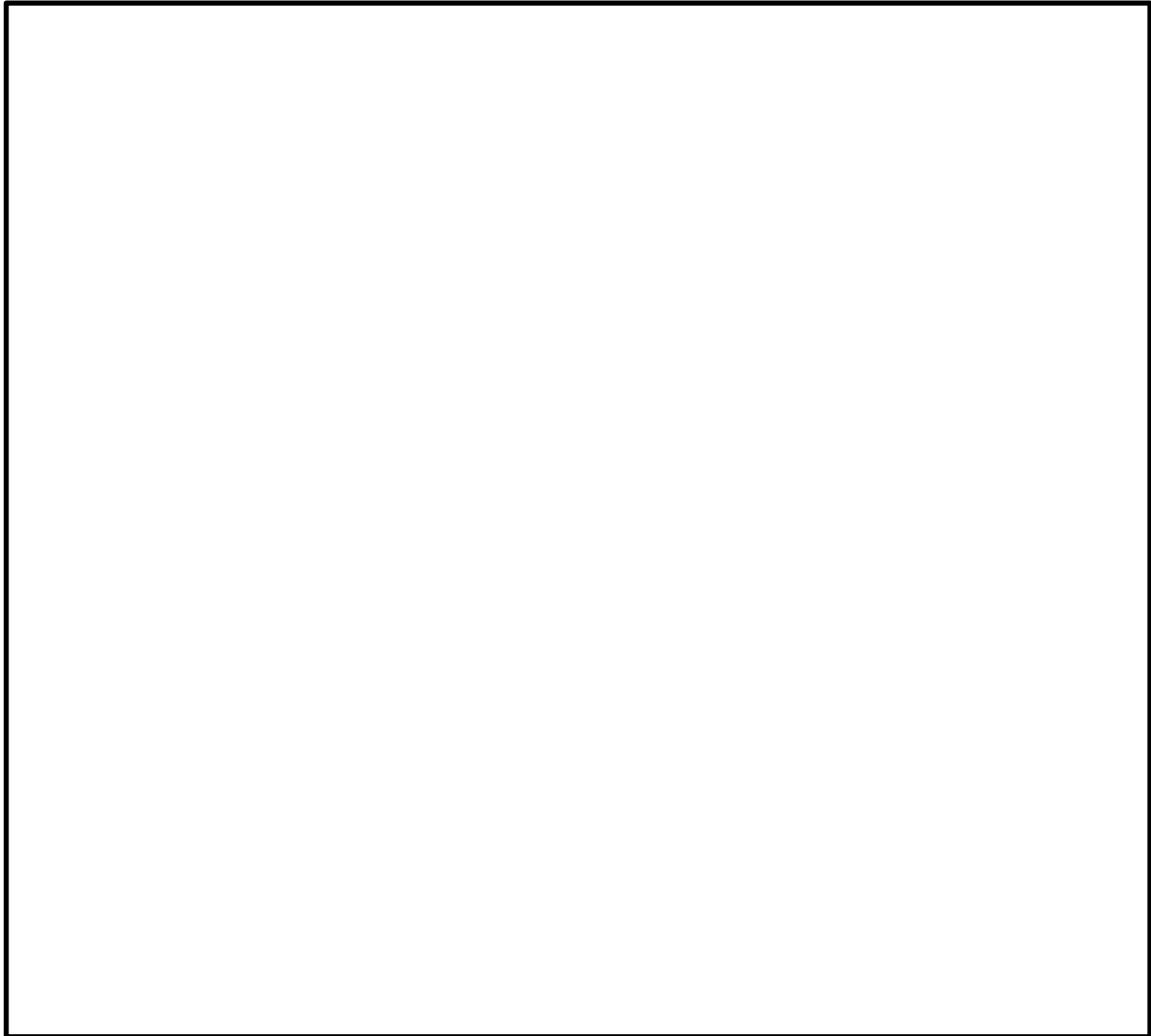
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**Draw flower structure and floral diagram:**



**Experiment No. 5**

**Objective: To study floral biology, emasculation and pollination of Cauliflower.**

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Fruit:** .....

**Floral formula:** .....

**Emasculation and crossing techniques:** .....

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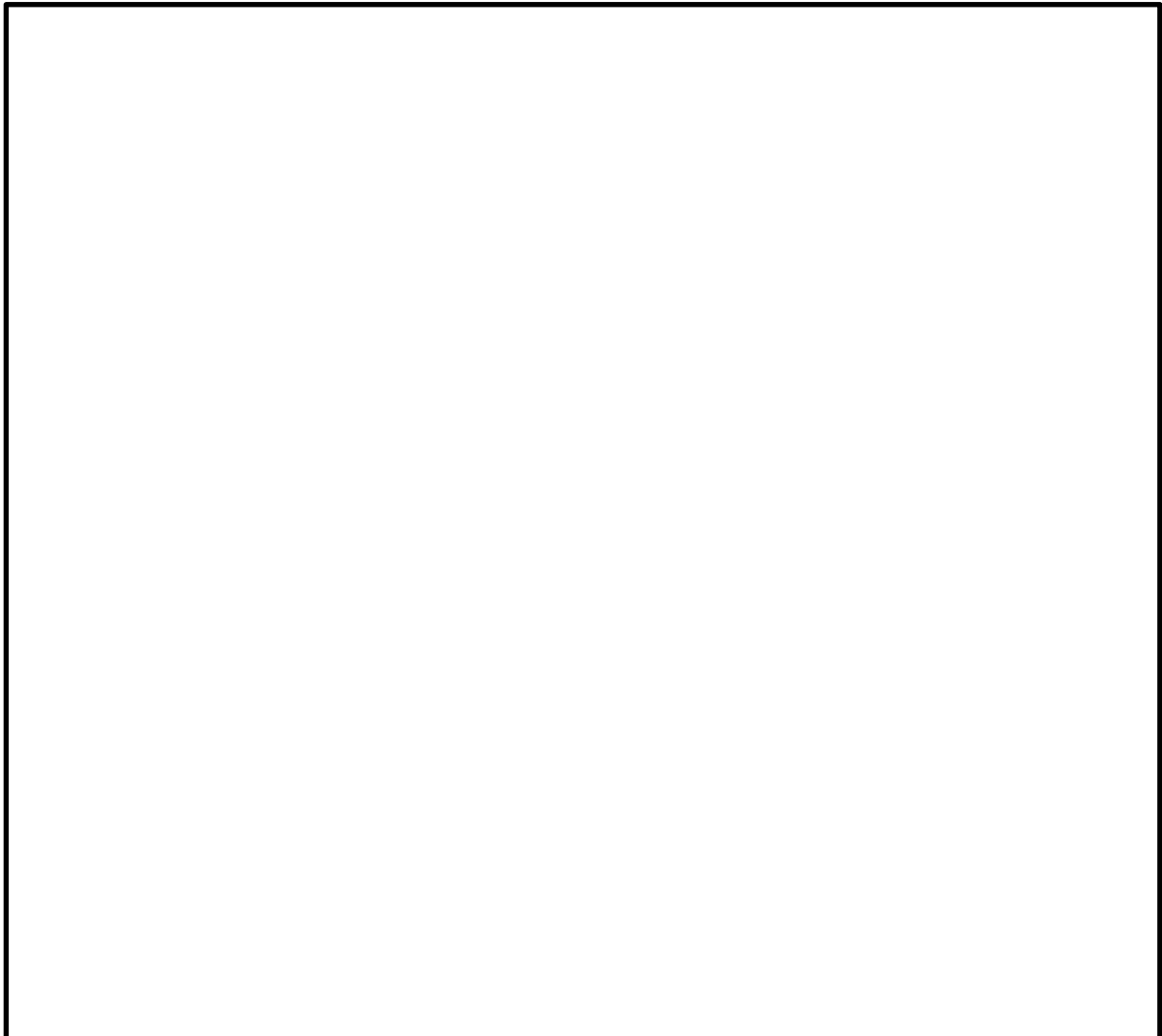
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**Draw flower structure and floral diagram:**





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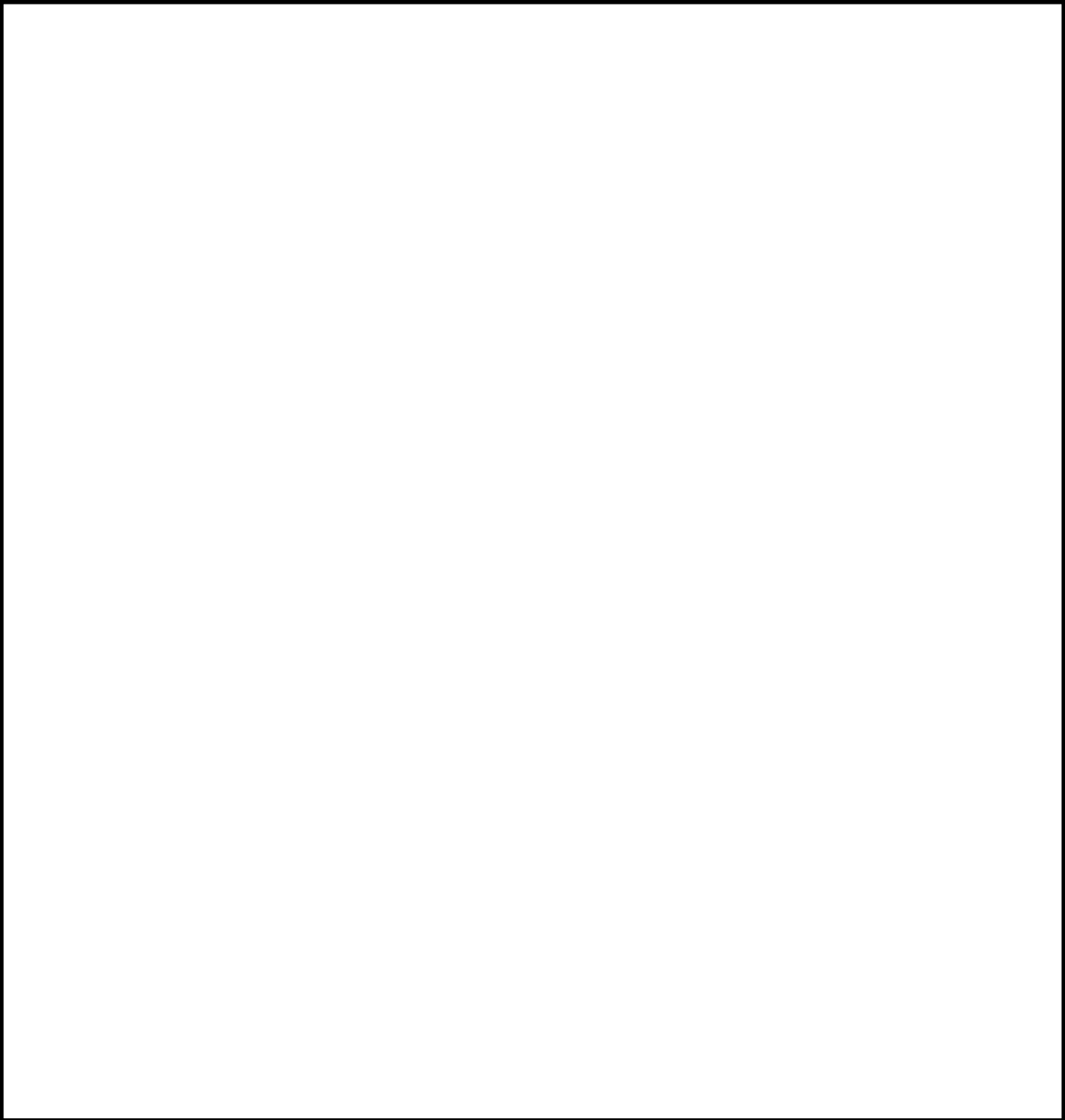
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**Draw flower structure and floral diagram:**



## Experiment No. 7

**Objective:** To study floral biology, emasculation and pollination of Coriander.

**Materials Required:** .....

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**Family:** .....

**Botanical name:** .....

**Chromosome no.:** .....

**Floral biology and floral structure:**

**Inflorescence:** .....

**Flower:** .....

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**Calyx:** .....

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**Corolla:** .....

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**Androecium:** .....

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**Gynoecium:** .....

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**Floral formula:** .....

**Emasculation and crossing techniques:** .....

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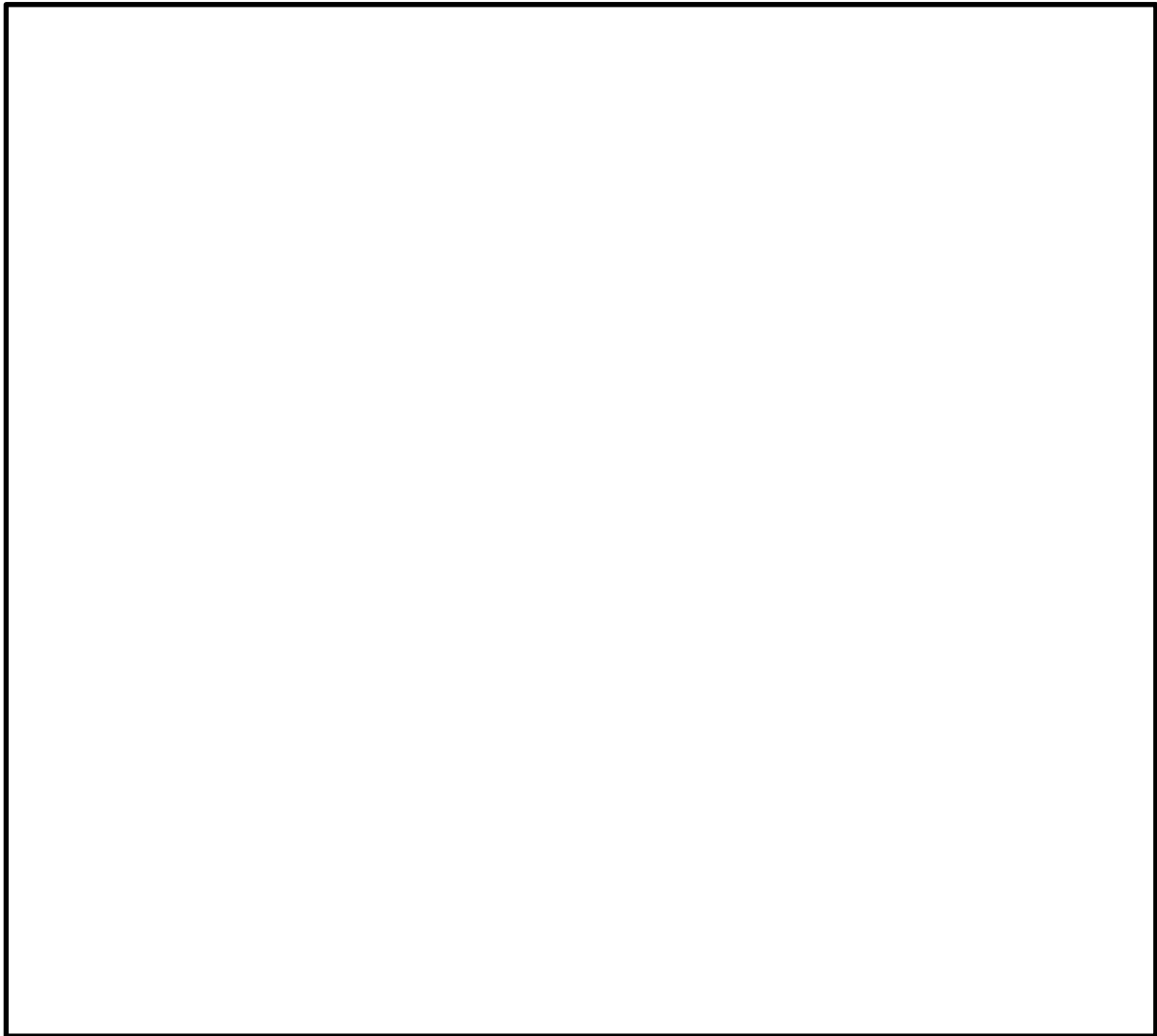
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**Draw flower structure and floral diagram:**







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**Experiment No. 9**

**Objective: Estimation of heritability and genetic advance.**

**Heritability:** .....

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**Problem no.-1:** Genotypic and phenotypic variances and covariances of two characters (x and y) are given below:

$$GV_x = 3.252, PV_x = 5.044, G Cov_{xy} = 1.657$$

$$GV_y = 4.728, PV_y = 5.520, P Cov_{xy} = 2.142$$

Calculate heritability of X and Y and their coheritability.

**Solution:** .....

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**Problem no.-2:** Following estimates were obtained from generation mean analysis:

$$VF_1 = 0.051, VF_2 = 0.218, D = 0.084$$

Calculate heritability in broad sense and narrow sense.

**Solution:** .....

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**Problem no.-3:** Calculate broad sense heritability from the following estimates obtained from generation mean analysis:

$D = 0.842, H = 1.465, E = 0.072$

**Solution:**

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**Problem-4:** For two plant characters (x and y), the estimates of genotypic and phenotypic covariances were 0.536 and 0.629, respectively.

**Solution:**

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**Genetic advance:** It is the measure of the progress of selection. The following formula is used for calculation of genetic advance.

Genetic advance under selection:  $(GS) = (K)(H)(SDp)$

Where K = Selection differential – a constant (2.06)

H= Heritability of the character under selection ( $Vg/Vp$ )

SDp= Phenotypic standard deviation ( $\sqrt{VP}$ )

Thus

$$GS = \frac{Vg}{(\sqrt{Vp} \sqrt{Vp})} \times \sqrt{VP} \times K$$
$$= \frac{Vg}{\sqrt{Vp}} \times K$$

Where  $Vg$ = Genotypic variance

$Vp$ = Phenotypic variance

$\sqrt{VP}$ = Phenotypic standard deviation

**Problem 1:** Calculate genetic advance from the following estimates  
MSS treatments = 16.47, MSS error= 2.83,  $\bar{X}$  = 11.68, replications= 3

**Solution:** .....

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## Experiment No. 11

**Objective: Estimation of GxE interactions.**

**Introduction:** .....

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**Environment and its kinds:**

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**1. Micro environment:** .....

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**Macro environment:** .....

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**Conventional procedures for the measurement of interaction:** .....

**ANOVA**

**1. Genotypes tested at one location for a single year**

<b>Source of variation</b>	<b>d.f.</b>



**Experiment No. 12**

**Objective: To study the effect of mutagens to induce chemical mutation.**

**Introduction**.....  
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**Materials Required:** .....  
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**Procedure:** .....  
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**Observation:** .....  
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**Percentage of germination**

**Untreated:** ..... **Treated:** .....



## Experiment No. 13

**Objective: To study the effect of mutagens to induce physical mutation.**

**Gamma-Garden:** Gamma gardens or atomic gardens are a type of induced mutation breeding where radioactive sources particularly gamma rays from cobalt -60 or Caesium-137 are used to induce desirable mutations in crop plants.

### Salient features of Gamma Garden

- Rarely Caesium-137 is also used as the source of radiation.
- The strength of Co is 200 curies.
- The source of radiation is located at the center.
- The area is divided into concentric circles with varying intensity of radiation.
- Plants to be irradiated are arranged as concentric circles around the radiation source.
- The intensity of radiation decreases as one move away from source of the radiation.
- The radially arranged plants in gamma garden can be grouped into three sectors.

### Layout of Gamma Garden

**Sector – I:** .....

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**Sector – II:** .....

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**Sector – III:** .....

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**Advantages of Gamma Garden: .....**

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**Disadvantages of Gamma Garden: .....**

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## Experiment No. 14

**Objective:** To study the techniques of inducing polyploidy.

**Introduction:** .....

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**Materials Required:** .....

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**Procedure:** .....

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Colchicines treatment may be imposed on seeds, seedlings or growing shoot apex.

**Seed treatment:** .....

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**Seedlings treatment:** .....

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**Shoot apex:** .....

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**Experiment No. 15**

**Object: To study hybrid seed production of tomato.**

**Botany:** Tomato is a typical day neutral plant. It requires temperature of 15-20° C for fruit setting. It is self-pollinated crop and self-fertilization is favoured by the position of receptive stigma within the cone anthers and the normal pendant position of the flower.

**Method of seed production:** .....

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**Stages of seed production:** .....

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**Season:** .....

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**Land requirement:**

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**Isolation requirement:** .....

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**Seed rate:** .....

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**Nursery:** .....

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**Transplanting:** .....

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**Rouging:** .....

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**Planting ratio:** .....

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**Different seed extraction methods**

	<b>Fermentation</b>	<b>Acid</b>	<b>Alkali</b>
<b>Method</b>			
<b>Salient features</b>			



**Object: Maintenance of breeding records.**

**Introduction:** .....

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**Origin of Seed:** .....

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**Pedigree of Seed:** .....

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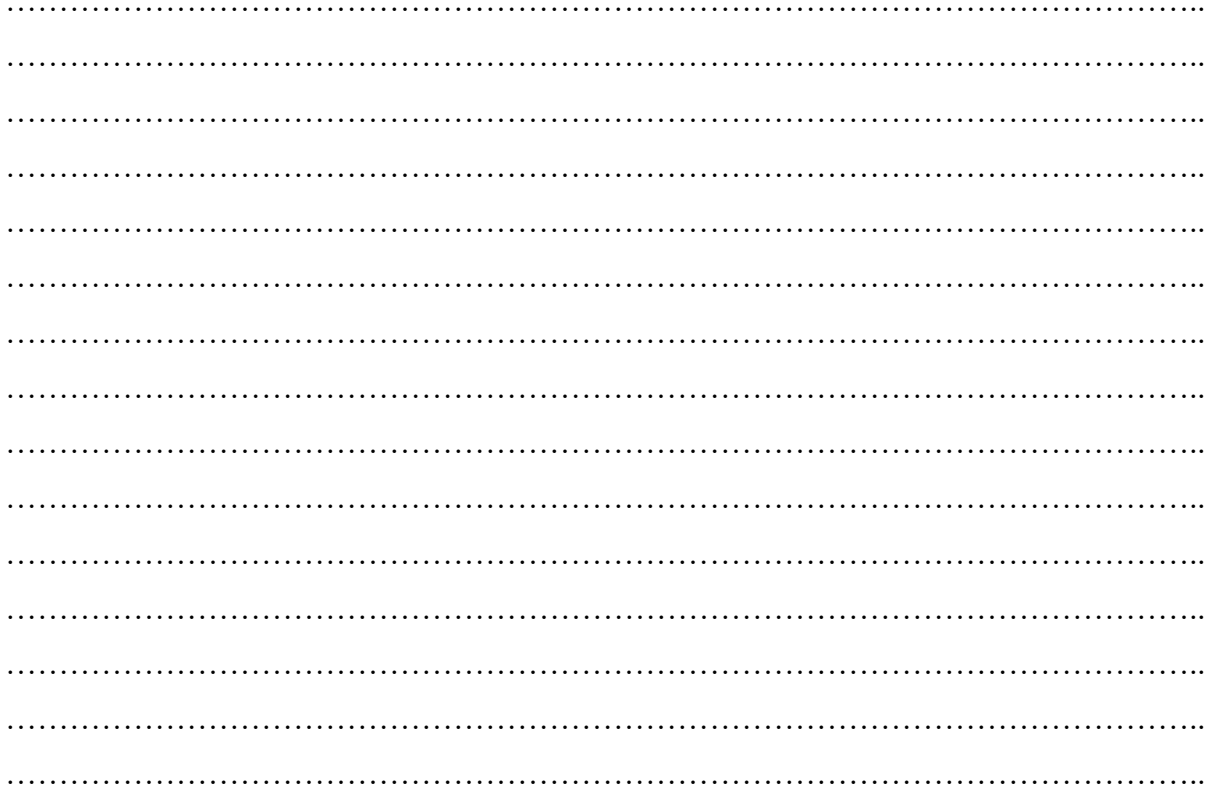
**Types of records:** .....

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**FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF TOMATO**

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Solanaceae

**Botanical name:** *Solanum lycopersicum* (*Lycopersicon esculentum* L.)

**Chromosome no.:** 2n = 24

**Floral biology and floral structure:**

**Inflorescence:** Extra-axillary helicoid cymes. Extra axillary position is due to fusion.

**Flower:** Ebracteate; pedicellate, complete, hermaphrodite, actinomorphic, pentamerous, hypogynous, small and white.

**Calyx:** Sepals 5, gamosepalous, pentafid, valvate, persistent, green, hairy, inferior.

**Corolla:** Petals 5, gamopetalous, rotate, valvate, five lobed, white, inferior.

**Androecium:** Stamens 5, polyandrous epipetalous, filaments shorts, equal in length, anthers long and conniving, basifixed, ditheous, and dehiscence by apical pores.

**Gynoecium:** Bicarpellary, syncarpous, ovary superior, bilocular, axile placentation, placentae swollen, ovules many in each loculus, ovary obliquely placed; style simple, hairy; stigma bilobed.

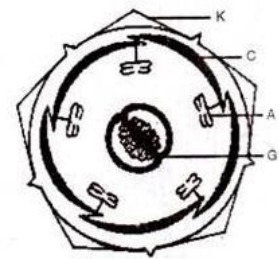
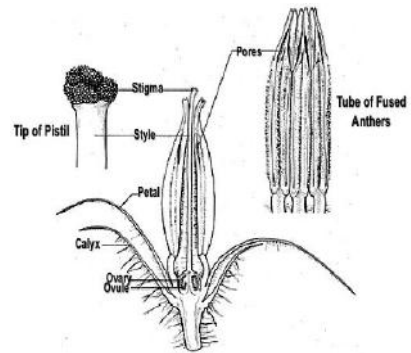
**Fruit:** Berry.

**Floral formula:**  $\oplus \text{♀ } K_{(5)} C_{(5)} A_{(5)} G_{(2)}$

**Emasculation and pollination technique:**

Emasculation is usually done one day prior to anthesis/flower opening. At this stage, the sepals have started to separate and the anthers and corolla are beginning to change from light to dark yellow. The stigma is fully receptive at this stage allowing for pollination even immediately after emasculation.

Anthers are removed as a group with or without the surrounding corolla, by inserting forceps between the sepals to grip the base of the anthers and / or petals which are then removed by a firm but steady pull. If anthers seem reluctant to part company from flower receptacle as a group, it is advisable to remove a single one first by careful manipulation of the forceps. Following this, the remaining four may be gripped firmly without any fear of damaging the style. Pollen is best applied in experimental crosses by slitting the inside of the anthers of mature flowers of the male parent with the forceps in such a way that a small amount of pollen is collected at the tip of the forceps. This can then be lightly applied to the stigmatic surface and should be visible as a white covering. Forceps should be sterilized by dipping in alcohol or methylated spirit after each pollination. Pollen may be collected in large amounts by inverting the mature flower and tapping pollen into the thumbnail (Watts, 1980). Protection of pollinated flowers by wrapping with cotton or small pollination bags is essential.



**FLORAL BIOLOGY, EMASCULATION AND POLLINATION IN CUCUMBER**

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Cucurbitaceae

**Botanical name:** *Cucumis sativus* L.

**Chromosome no.:** 2n = 14

**Floral biology and floral structure:**

**Inflorescence:** Female flowers solitary axillary but male flowers in cymose.

**Flower:** Unisexual.

**Male flower:** Bracteate, pedicellate, actinomorphic, incomplete, staminate, yellow.

**Calyx:** Sepals 5, gamosepalous, green, hairy, lobes linear or leafy, imbricate aestivation.

**Corolla:** Petals 5, gamopetalous, campanulate, yellow, imbricate aestivation.

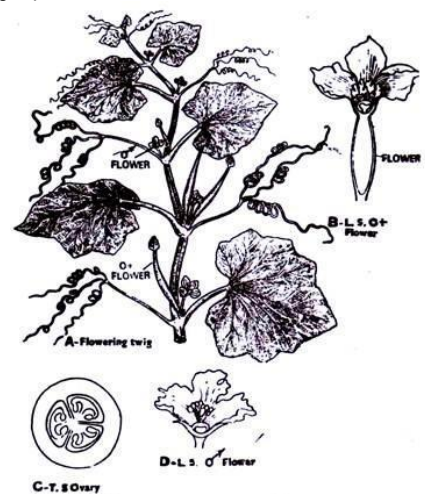
**Androecium:** Stamens 5, two united in two pairs and one free, anthers twisted spirally mono and ditheous, extrorse.

**Gynoecium:** Absent.

**Floral formula:**  $\text{Br } \oplus \text{♀ } K (5) C (5) A_{3\text{std}} G (3)$

**Female flower:** Bracteate, pedicellate, pistillate, actinomorphic, incomplete, epiygynous.

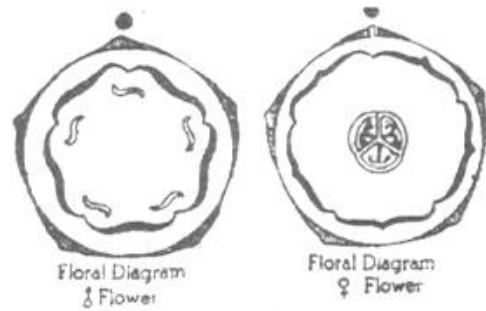
**Calyx:** Sepals 5, gamosepalous, green, hairy, lobes linear, imbricate aestivation.



**Corolla:** Petals 5, gamopetalous, campanulate, yellow, imbricate aestivation.

**Androecium:** Absent.

**Gynoecium:** Tricarpellary, syncarpous, ovary inferior, unilocular, parietal placentation, ovules many on each placentum, style one stigma 3 forked. **Br. ♂ K (5) C (5) A (2) + (2) + 1 G<sub>0</sub>**



**Floral formula:**

**Emasculation and crossing techniques:** In monoecious plants, the pistillate and staminate flowers are covered with butter paper bags one prior to anthesis. In andromonoecious flowers, the bisexual flower is emasculated a day before flower opening by anthers and petals with forceps. Pollen from the bagged male flower is dusted on the stigma of the female emasculated flowers. The pistilled flower is bagged after pollination.

### FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF INDIAN BEAN

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Fabaceae

**Botanical name:** *Lablab purpureus* (L.) Sweet

**Chromosome no.:**  $2n = 22$

**Floral biology and floral structure:**

**Inflorescence:** Solitary axillary.

**Flower:** Bracteate, pedicellate, complete, hermaphrodite, zygomorphic pentamerous, hypogynous.

**Calyx:** Sepals 5, gamosepalous, pentapartite, companulate, odd sepal anterior, imbricate aestivation, green, hairy.

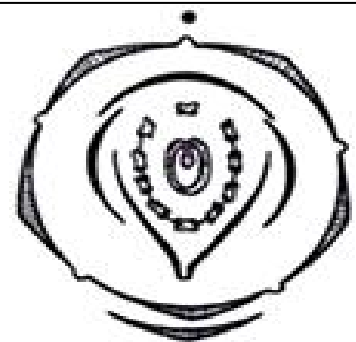
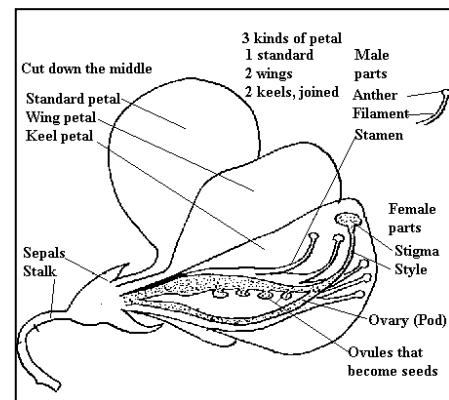
**Corolla:** Petals 5, polypetalous, papilionaceous, consisting of a large posterior petal – the vexillum or standard, two lateral-alae or wings and two inner fused to form a boat shaped structure the keel or carina, vexillary aestivation.

**Androecium:** Stamens 10, diadelphous, nine are fused by the lower halves of their filaments to form a tube round the ovary and tenth posterior one free, anthers basifixed, introrse, dithealous, enclosed in the keel.

**Gynoecium:** Monocarpellary, ovary superior, unilocular, hairy, elongated, laterally compressed, marginal placentation ovules many, style long, stigma hairy.

**Fruit:** Legume.

**Floral formula:** **Br % ♂ K(5) C1+2+(2) A(9)+1 G<sub>1</sub>**



**Emasculation and crossing techniques:**

For emasculation, the flower bud chosen should have developed to the stage just before anther dehiscence, indicated by extension of petals beyond sepals. Flowers can be emasculated any time. The first step in emasculation is to tear away with the forceps the tip of the sepal from in front of the keel. The fore finger is positioned behind the flower and thumb in front and light pressure is applied. This spreads the standard and wings to expose the keel. The exposed keel is slightly open by the tips of the forceps. Pressure can be applied by the thumb and finger on the keel for increased exposure of the pistil and stamens. The 10 stamens are pulled out.

Pollen can be obtained throughout the day, preferably from a freshly opened flower. For pollen collection, it is more convenient to pick the male flowers, remove the standard and wings, pull back the keel so that the style protrudes and use the pollen-covered styler brush as an applicator to transfer the pollen to the stigma of the emasculated bud. Older flowers and other flower buds not used in crossing are removed to increase the pod set after crossing.

### FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF OKRA.

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Malvaceae

**Botanical name:** *Abelmoschus esculentus* L. (Moench)

**Chromosome No.:**  $2n=130$

**Floral biology and floral structure:**

**Inflorescence:** Solitary axillary.

**Flower:** Ebracteate, pedicellate, complete, actinomorphic, hermaphrodite, pentamerous, hypogynous, large, showy, red, mucilaginous.

**Epicalyx:** Five to seven, green, linear.

**Calyx:** Sepals 5, gamosepalous, ovate, campanulate, valvate aestivation.

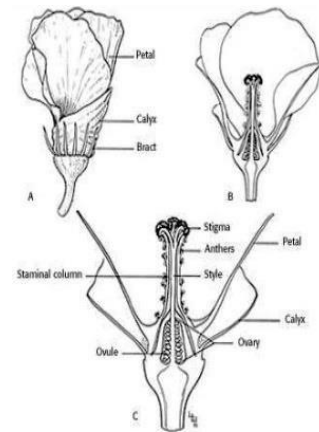
**Corolla:** Petals 5, polypetalous, mucilaginous, twisted, red, united at the base and adnate to the staminal tube.

**Androecium:** Stamens numerous, forming a tube, monadelphous, epipetalous, anthers monothealous, yellow, reniform, extrorse, transversely attached to the filament, pollen grains multiporate.

**Gynoecium:** Pentacarpellary, syncarpous, superior, pentalocular, axile placentation, many ovules in each loculus; style passing through the staminal tube; stigma 5 capitate.

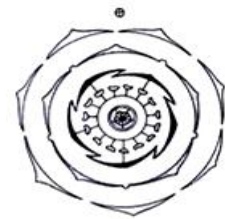
**Fruit:** Capsule.

**Floral**  $\oplus \text{Epi}_{5-7} \text{K}_{(5)} \text{C}_5 \text{A}_{(5)} \text{G}_{(5)}$  **formula:**



**Emasculation and crossing techniques:**

- Matured flower buds of the desired female parent are emasculated one day prior to flower opening and bagged with butter paper cover.
- The dehisced pollen plant is also bagged.
- Pollen from male parent is smeared on the sticky stigmatic surface of the emasculated flower of the female parent, the next day.
- The crossed flower is bagged with butter cover to prevent natural cross pollination.
- The flower is properly labelled indicating the seed and the pollen parents and also date of crossing.
- The butter paper bags are removed after a week of crossing and the fruits are allow to develop to maturity.



**FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF CAULIFLOWER**

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Brassicaceae

**Botanical name:** *Brassica oleracea* var. *botrytis* L.

**Chromosome No.:** 2n=18

**Floral biology and floral structure:**

**Inflorescence:** A corymbose-raceme.

**Flower:** Ebracteate, pedicellate, complete, actinomorphic, hermaphrodite, cruciform, tetramerous, hypogynous and yellow.

**Calyx:** Sepals 4 (2 + 2) in two whorls, outer whorl antero-posterior, the two lateral one saccate, green, polysepalous, inferior.

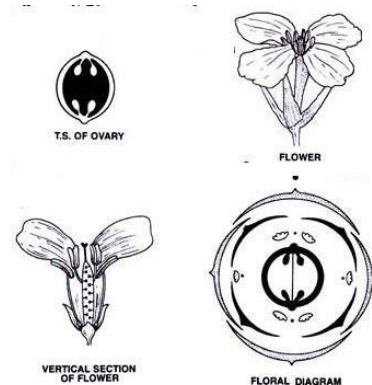
**Corolla:** Petals four, polypetalous, cruciform, valvate, inferior, yellow.

**Androecium:** Stamens six, tetradynamous, in two whorls, the outer with two short lateral stamens and inner with four long stamens arranged in two median pairs. Basifixed, polyandrous, introrse. Four green nectaries are present, on the inner side of each short stamen and a similar one at the base but outside each pair of long median stamens, inferior.

**Gynoecium:** Bicarpellary, syncarpous, superior, unilocular becoming bilocular by the development of false septum called-replum; parietal placentation, style short, stigma bilobed.

**Fruit:** Siliqua.

**Floral formula:**  $\oplus \text{K}_{2+2} \text{C}_{4x} \text{A}_{2+4} \text{G}_{(2)}$



**Emasculation and crossing techniques:** The self-compatible varieties of cauliflower can be selfed by simply bagging the flower-stalk. Selfing is also done by caging some plants with flies in cages or by isolation planting of lines having decreased level of self-incompatibility. With self-incompatible plants, bud pollination gives better results. In this system, the pollination is carried out in buds before 2-4 days of opening, with emasculation or without emasculation.

The flowers may be emasculated by removing 6 stamens using a pair of forceps. In self-compatible cauliflowers (European types), the stamens are removed before the opening of the buds as the flowers are already fertile in the bud stage, crossing can be done at the same time. In self-incompatible types, emasculation may be omitted. When pollination cages are available, crosses between self-incompatible types can be made by insects such as honey bees, bumble bees and flies.

## FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF ONION

**Material required:** Forcep, needle, brush, butter paper bags, scissors, label tags, pencil etc.

**Family:** Liliaceae

**Botanical name:** *Allium cepa* L.

**Chromosome No.:**  $2n=2x=16$

**Floral biology and floral structure:**

**Inflorescence:** Cymose umbel enclosed by 2 or 3 membranous bracts arranged in an umbellate fashion on an erect leafless scape.

**Flower:** Pedicellate, small, hermaphrodite, hypogynous, complete, actinomorphic, trimerous, white, bracteate.

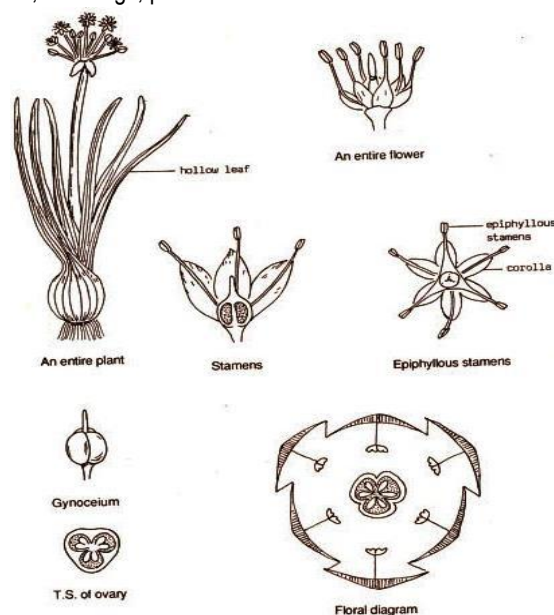
**Perianth:** Tepals 6, in 2 whorls of 3 each, gamophyllous, united at the base, white, inferior.

**Androecium:** Stamens 6, polyandrous, in two whorls of 3 each, epiphyllous, filament narrow, dilated at the base, anthers dithecos, dorsifixed.

**Gynoecium:** Tricarpellary, syncarpous, ovary superior, trilocular, axile placentation, 2 ovules per loculus; style short filiform, stigma minute.

**Floral formula:**  $\text{Br.} \oplus \text{♀} \overline{\text{P}_{(3+3)}} \text{A}_{3+3} \text{G}_{(3)}$

**Fruit:** Capsule



## FLORAL BIOLOGY, EMASCULATION AND POLLINATION OF CORIANDER *CORIANDRUM SATIVUM*.

**Material required:** Forceps, needle, brush, butter paper bags, scissors, label tags, pencil etc

**Family:** Umbelliferae (Apiaceae).

**Botanical name:** *Coriandrum sativum*

**Chromosome No.:**  $2n=22$

**Floral biology and floral structure:**

**Inflorescence:** Compound umbel, involucre and involucel present.

**Flower:** Ebracteate, pedicellate, complete, actinomorphic, hermaphrodite, epigynous, cyclic, pentamerous.

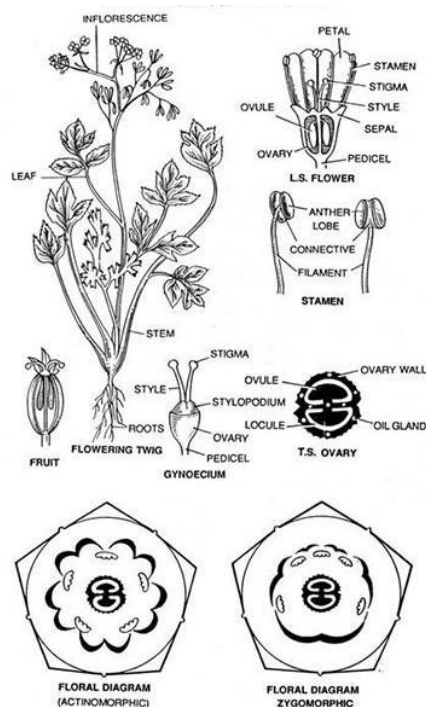
**Calyx:** Five sepals, calyx tube adnate to the ovary, gamosepalous, calyx teeth 5, small, unequal, slightly petaloid, superior.

**Corolla:** Five petals, polypetalous, white, superior, inner or the posterior petals are smallest and lobed, the anterior is largest and deeply bilobed, two lateral are medium sized and unequal bilobed.

**Androecium:** Five stamens, polyandrous, filament long, anther dorsifixed superior.

**Gynoecium:** Bicarpellary, syncarpous, ovary inferior, a small fleshy nectar secreting disc lying above it form the nectar, bilocular with in pendulous ovule in each loculus, style absent, stigma two, arising from the disc.

**Floral Formula:**  $\oplus \text{♂} \overline{\text{K}(5)} \text{C}_5 \text{A}_5 \text{G} \overline{(2)}$



**Emasculation and crossing techniques:** The inner flowers of the umbellets are staminate. The umbels of higher order usually contain more staminate flowers than the first ones, and their flowering period is shorter. After the flower opens, the white filaments are visible between the petals, because they are bent and the pollen sacs at their top are hidden in the centre of the flower. This stage is the best for artificial emasculation of the flowers, because the filaments are easy to distinguish, and they have not yet spread any pollen grains. Since the peripheral flowers of every umbellet reach this stage earlier than the central flowers, the latter should be removed so that their pollen will not lead to fertilization. When this process has finished, the two pistils become longer and separate from each other at the top. The former green colour sometimes changes to pink or violet too. This is the right moment for successful pollination. The stigma is receptive for pollination for a maximum period of 5 days. The plant can be artificially pollinated by placing pollen grains of the father plant on the stigma using a paintbrush or by carefully brushing the stigmas with flowering umbels of the father plant. Thus, the pollination behaviour of coriander is that of a facultative cross-pollinator. The protandry and the observations made suggest that geitonogamy is common and xenogamy is possible.



## PHYSICAL MUTATION

### Layout of a gamma garden

#### Sector – I:

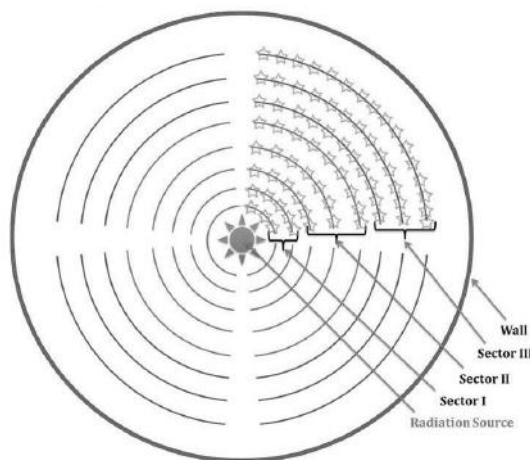
- They are plants nearest to the central radiation source.
- Plants in the sector-I usually die immediately due to the dose of radiation.
- They are not used in further experiments

#### Sector – II:

- This include plants located next to the sector-I
- These plants develop severe tumors, malformations and other abnormalities.
- These plants are also not used in further experiments.

#### Sector – III

- They include plants located next to sector II.
- They are the actual plants of interest in Gamma gardens.
- They may have random mutations not severe enough to damage the crop plant.
- The variations obtained in the sector III are used in further breeding experiments.
- They can be used as a source of variation in hybridization or can be directly released as a variety.



#### Advantages of Gamma Garden

- Gamma gardens can produce large amount of variations within a short time.
- Desirable mutants can be released directly as a new variety.
- Gamma gardens are good examples of the peaceful use of atomic energy for human welfare.

#### Disadvantages of Gamma Garden:

- High initial investment required.
- Other cheapest mutation methods are now available.
- Chances of undesirable mutations are very high.
- Mutations are random; we cannot predict the effects of mutations.

## ESTIMATION OF HETEROISIS, INBREEDING DEPRESSION, HERITABILITY AND GENETIC ADVANCE

### A. Heterosis and Inbreeding

Various types of heterosis are estimated as follows:

1. Mid-parent heterosis  $= \frac{F_1 - MP}{MP} \times 100$
2. Heterobeltiosis  $= \frac{F_1 - BP}{BP} \times 100$
3. Useful or economic heterosis  $= \frac{F_1 - CC}{CC} \times 100$
4. Standard heterosis  $= \frac{F_1 - SH}{SH} \times 100$

Where,

- $F_1$  = Mean value of a particular cross,
- $MP$  = Mean of two parents involved in the cross,
- $BP$  = Mean value of better parent of a cross,
- $CC$  = Mean value of a commercial cultivar,
- $SH$  = Mean value of standard (check) hybrid.

**Problem No.-1:** In black gram, grain yields of parents ( $P_1$  and  $P_2$ ), their  $F_1$  and  $F_2$  progenies are given below:

Parent 1	Parent 2	F <sub>1</sub> hybrid	F <sub>2</sub> Progeny
18.94	22.69	29.38	15.18

Calculate average heterosis, heterobeltiosis and inbreeding depression.

#### Solution:

1. Mid-parent heterosis  $= \frac{F_1 - MP}{MP} \times 100$   
 Here, value of  $F_1$  = 29.38  
 Mean of parents (MP)  $= \frac{18.94 + 22.69}{2}$   
 $= 20.81$   
 $= \frac{29.38 - 20.81}{20.81} \times 100$

- = 41.12 % answer.**
2. Heterobeltiosis =  $\frac{F_1 - BP}{BP} \times 100$   
 Better parent value = 22.69  
 =  $\frac{29.38 - 22.69}{22.69} \times 100$   
**= 29.48% answer.**
3. Inbreeding depression =  $\frac{F_1 - F_2}{F_1} \times 100$   
 =  $\frac{29.38 - 15.18}{29.38} \times 100$   
**= 48.33% answer.**

**Problem no.-2:** In cotton, yield data per plant (g) for parents (P<sub>1</sub> and P<sub>2</sub>), their F<sub>1</sub> and F<sub>2</sub> progeny, and for a commercial cultivar and hybrid are given below:

P <sub>1</sub>	P <sub>2</sub>	F <sub>1</sub>	F <sub>2</sub>	Check cultivar	Check hybrid
50	40	90	35	70	80

Calculate useful heterosis, standard heterosis and inbreeding depression.

**Solution:**

1. Useful heterosis =  $\frac{F_1 - CC}{CC} \times 100$   
 Thus =  $\frac{90 - 70}{70} \times 100$   
**= 28.57% answer.**
2. Standard heterosis =  $\frac{F_1 - SH}{SH} \times 100$   
 Thus =  $\frac{90 - 80}{80} \times 100$   
**= 12.50% answer.**
3. Inbreeding depression =  $\frac{F_1 - F_2}{F_1} \times 100$   
 Thus =  $\frac{90 - 35}{90} \times 100$   
**= 61.11% answer.**

B. **Heritability:** Heritability is an index of the transmission of character from parents to their offspring. Heritability is of two types, viz. broad sense heritability and narrow sense heritability. Broad sense heritability is the percentage ratio of genotypic variance to the phenotypic variance, whereas narrow sense heritability is the ratio of additive variance to the phenotypic variance.

**Problem no.-1:** Genotypic and phenotypic variances and covariances of two characters (x and y) are given below:

$GV_x = 3.252, PV_x = 5.044, G Cov_{xy} = 1.657$

$GV_y = 4.728, PV_y = 5.520, P Cov_{xy} = 2.142$

Calculate heritability of X and Y and their coheritability.

**Solution:**

Heritability of X =  $\frac{GV_x}{PV_x} \times 100$   
 =  $\frac{3.252}{5.044} \times 100 = 64.47\% \text{ answer.}$

Heritability of Y =  $\frac{GV_y}{PV_y} \times 100$   
 =  $\frac{4.720}{5.520} \times 100 = 85.51\% \text{ answer.}$

Coheritability of X and Y characters =  $\frac{G Cov_{xy}}{P Cov_{xy}} \times 100$   
 =  $\frac{1.657}{2.142} \times 100 = 77.35\% \text{ answer.}$

**Problem no.-2:** Following estimates were obtained from generation mean analysis:

$VF_1 = 0.051, VF_2 = 0.218, D = 0.084$

Calculate heritability in broad sense and narrow sense.

**Solution:**

Heritability =  $\frac{VF_2 - VF_1}{VF_2} \times 100$   
 =  $\frac{0.218 - 0.051}{0.218} \times 100 = \frac{0.167}{0.218} \times 100 = 76.60\% \text{ answer.}$

Heritability =  $\frac{\frac{1}{2D}}{VF_2} \times 100$   
 =  $\frac{\frac{0.084}{2}}{0.218} \times 100 = \frac{0.042}{0.218} \times 100 = 19.26\% \text{ answer.}$

**Problem no.-3:** Calculate broad sense heritability from the following estimates obtained from generation mean analysis:

$D = 0.842, H = 1.465, E = 0.072$

**Solution:**



$$\text{Heritability} = \frac{D}{D+H+E} \times 100$$

$$= \frac{0.842}{0.842+1.465+0.072} \times 100 = \frac{0.842}{2.379} = 35.39\% \text{ answer.}$$

**Problem-4:** For two plant characters (x and y), the estimates of genotypic and phenotypic covariances were 0.536 and 0.629, respectively.

**Solution:**

$$\text{Coheritability (\%)} = \frac{G \text{ Cov } xy}{P \text{ Cov } xy} \times 100 = \frac{0.536}{0.629} \times 100 = 85.21\% \text{ answer.}$$

C. **Genetic advance:** It is the measure of the progress of selection. The following formula is used for calculation of genetic advance.

Genetic advance under selection: (GS)=(K) (H) (SDp)

Where K = Selection differential – a constant (2.06)

H = Heritability of the character under selection (Vg/Vp)

SDp = Phenotypic standard deviation ( $\sqrt{VP}$ )

$$\text{Thus } GS = \frac{Vg}{(\sqrt{Vp} \sqrt{VP})} \times \sqrt{VP} \times K$$

$$= \frac{Vg}{\sqrt{Vp}} \times K$$

Where

Vg = Genotypic variance

Vp = Phenotypic variance

$\sqrt{VP}$  = Phenotypic standard deviation

**Problem 1:** Calculate genetic advance from the following estimates

MSS treatments = 16.47, MSS error = 2.83,  $\bar{X}$  = 11.68, replications = 3

**Solution:** For calculation of advance, first genotypic and phenotypic variances are calculated as follows:

$$\text{Genotypic variance (Vg)} = \frac{MS_{Str} - MS_{Se}}{\text{Replications}}$$

$$= \frac{16.47 - 2.83}{3} = 4.547$$

$$\text{Phenotypic variance (Vp)} = Vg + V_e$$

$$= 4.547 - 2.830 = 7.37$$

$$\text{Genetic advance} = \frac{Vg}{\sqrt{Vp}} \times K$$

$$= \frac{4.547}{\sqrt{7.377}} \times 2.06$$

$$= \frac{4.547}{2.716} \times 2.06$$

$$= 3.45$$

$$\text{Genetic advance as \% of mean} = \frac{G.A.}{\bar{X}} \times 100$$

$$= \frac{3.45}{11.68} \times 100$$

$$= 29.54\% \text{ Ans.}$$

**Problem 2:** Calculate genetic advance from the following estimates

$V_{F1} = 0.051$ ,  $V_{F2} = 0.218$

**Solution:** The following formula is used for calculate of genetic advance.

$$\text{Genetic advance} = \frac{Vg}{\sqrt{Vp}} \times K$$

Here  $Vp = V_{F2}$  and  $Vg = V_{F2} - V_{F1}$

$V_{F1} = V_e$ , K = Constant (2.06)

$$\text{Thus, } G.A. = \frac{0.218 - 0.051}{\sqrt{0.218}} \times 2.06$$

$$= \frac{0.167}{\sqrt{0.218}} \times 2.06$$

$$= \frac{0.167}{0.4669} \times 2.06$$

$$= 0.3577 \times 2.06$$

$$= 0.736862 \text{ Ans.}$$

## COMBINING ABILITY

**Combining ability:** The concept of combining ability as a measures of gene action was proposed by Sprague and Tatum in 1942 working on maize. Combining ability refers to the capacity or ability of a genotype to transmit superior performance to its crosses. The value of an inbred line depends on its ability to produce superior hybrids in combination with other inbreds.

**Types of combining ability:** There are two types of combining ability viz., general combining ability (gca) and specific combining ability. A brief description of each type is given below-

1. **General combining ability (gca):** The average performance of a strain or genotype in a series of hybrid combinations is termed as gca. In other words, the crosses which have one parent in common are used for the calculation of gca. The main features of gca are given below-
  - The gca variance is primarily a function of the additive genetic variance, but if epistasis is present gca will also include additive x additive type of non-allelic interaction.
  - The gca is estimated from half sib families.
  - The gca variance has positive correlation with narrow sense heritability.
  - The gca helps in the selection of suitable (good general combiners) parents for hybridization.
2. **Specific combining ability (sca):** The performance of a parent in a specific cross is known as specific combining ability. Thus, sca refers to the deviation of a particular cross from the general combining ability. The main feature of specific combining ability is briefly presented below.
  - The gca variance is mainly a function of dominance variance, but if epistasis is present, it would also include additive x dominance and dominance x dominance types of non-allelic interactions.
  - The sca is estimated from full sib families.
  - The sca variance has positive association with heterosis or hybrid vigour.
  - The sca helps in the identification of superior cross combination for commercial exploitation of heterosis.

**Estimation:** The analysis of data is carried out as per the biometrical technique used. The biometrical techniques which are commonly used for the estimation of combining ability as given below:

1. Diallel Cross Analysis – Griffing (1956)
2. Partial Diallel Analysis – Kempthorne and Curnow (1961)
3. Line x Tester analysis - Kempthorne (1957)

**Interpretation of results:** The following interpretation can be drawn from the estimates of gca and sca variances and effects.

- If gca variances are higher than sca variances, it means that there is predominance of additive gene action and progeny selection will be effective for the genetic improvement of such traits.
- If sca variances are higher than gca variances, it means that there is preponderance of non-additive gene action (dominance and epistasis) and therefore, heterosis breeding may be rewarding.
- If both gca and sca variances are of equal magnitude it shows that additive and non-additive genes are equally important in the expression of character. In such situation, reciprocal recurrent selection may be resorted for population improvement.

## ESTIMATION GXE INTERACTIONS (STABILITY ANALYSIS)

**Introduction:** A phenotype is the result of an interplay of a genotype and its environment. A specified genotype does not exhibit the same phenotypic characteristics under all environments and different genotypes respond differently to a specified environment. This variation arising from the lack of correspondence between the genetic and non-genetic effects is known as the genotype x environment interaction. In other words, the failure of a genotype to give the same phenotypic performance when tested under different environments is the replication of the genotype x environment interaction. Thus, the genotype A may be superior to the genotype B under one environment but inferior to it under another. The result is the change in the selective ranking, reducing the correlation between the genotype and environment. When we talk of a genotype x environment interaction, we may take into consideration the environments of different kinds.

### Environment and its kinds

Environment is some total of physical, chemical and biological factors. The individuals or populations or species do not live in a vacuum, but are surrounded and affected by these factors. Comstock and Moll (1963) have classified the environment into two categories.

1. **Micro environment:** Micro environment is the environment of a single organism, as opposed to that of another growing at the same time and in almost the same place. It includes physical and chemical attributes of soil, climatic variables (temperature and humidity), solar radiation, insects-pest and diseases.
2. **Macro environment:** Macro environment is the environment which is associated with a general location and period of time and a collection of micro environments.

Allard and Bradshaw (1964) coined the terms predictable and unpredictable environment. The predictable environment includes the permanent features of the environment such as climate, soil type and day length. It also includes what are called controllable variables (Perkins and Jinks, 1971) eg. the level of fertilizer application, sowing dates, sowing density and method of harvesting.

The unpredictable or the uncontrollable environment includes weather fluctuation, such as differences between seasons in terms of the amount and distribution of rainfall and the prevailing temperature. For the uncontrollable variables, a low level of interaction would be desirable so as to have the maximum uniformity of performance over number of seasons. In contrast, for the controllable variables a high level of interaction will be desirable to produce the maximum increase in performance.

**Conventional procedures for the measurement of interaction:** The interaction between a genotype and its environment contributes to the total variance which can be isolated and tested for significance.

### **ANOVA**

#### **1. Genotypes tested at one location for a single year**

Source of variation	d.f.
Replication	(r-1)
Genotypes	(g-1)
Error	(r-1) (g-1)
Total	(rg-1)

Where g and r stand for the no. of genotype and replications, respectively.

#### **2. Genotypes tested at different locations in the same year**

Source of variation	d.f.
Replication within environment	n(r-1)
Environment	(n-1)
Genotypes	(g-1)
GxE	(g-1) (n-1)
Residual error	n(r-1) (g-1)

Where g, n and r stand for the no. of genotype, environment and replications, respectively.

#### **3. Genotypes grown at different locations over a no. of year**

Source of variation	d.f.
Replication within locations and years	ly(r-1)
Year	(y-1)
Locations	(l-1)
Years x Locations	(y-1) (l-1)
Genotypes	(g-1)
Genotypes x years	(g-1) (y-1)
Genotypes x location	(g-1) (l-1)
Genotypes x location x year	(g-1) (l-1) (y-1)
Residual error	ly(r-1) (g-1)

Where g, y, l and r stand for the no. of genotype, year, location and replications, respectively

## **HYBRID SEED PRODUCTION IN TOMATO**

**Method of seed production:** Seed to Seed.

**Stages of seed production:** Breeder seed → Foundation Seed I → Foundation Seed II → Certified Seed

**Season:** May - June and November - December

**Land requirement:** Selection of suitable land for tomato seed production is important where the previous crop should not be the same variety to avoid the contamination due to the volunteer plants. If the land has not been grown with tomato crop itself or even any other solanaceous vegetable crops like chilli, capsicum, brinjal etc. It will be very helpful to avoid certain diseases common to all members of Solanaceae.

**Isolation requirement:** For Seed production of tomato, varieties require minimum of 50 m for foundation seed and 25 m for certified seed. For hybrid seed production, it requires minimum of 200 m for foundation (parental line increase) and 100 m for certified hybrid seeds.

**Seed rate:**

- i) F1 hybrid - Male parent 25 g/ha
- ii) Female parent 100 g/ha

Srimathi *et al.* (2000) reported that tomato seeds pelleted with ZnSO<sub>4</sub> (250 mg kg<sup>-1</sup> of seed) improved the initial seed quality in terms of germination, field emergence and the seed yield by 25.9%. The ZnSO<sub>4</sub> pelleted seeds also maintained their germination upto a period of 3 months without any reduction in germination.

**Nursery:** Sow the seeds in raised nursery bed of 20 cm height, in rows of 5 cm gap and covered with sand. Eight and ten nursery beds will be sufficient to transplant one acre. Apply 2 kg of DAP 10days before pulling out of seedling.

**Transplanting:** Transplanting should be done when the seedlings are 20-25 days old, preferably at evening time. Spacing is 60 x 45 cm (90 x 60 cm for female parent and 60 x 45 cm for male parent of hybrids).

**Roguing:** The roguing should be done based on the plant characters (determinate / indeterminate), leaf (cut leaf/potatoleaf), branching and spreading characters and also based on fruit size, shape and colour. The plants affected by early blight, leaf spot and mosaic (TMV) diseases should be removed from the seed production field.

**Planting ratio:** For hybrid seed production, the female and male parents are normally planted in the ratio of 12:1 or 12:2.

### Comparison of different seed extraction methods

	Fermentation	Acid	Alkali
<b>Method</b>	Mix fruit pulp with water 24 - 48 h	HCl @10ml / Kg of pulp (20-30 minutes)	Washing soda @ 900mg/ 4 l of water- equal volume – overnight soak
<b>Salient features</b>	<ul style="list-style-type: none"> <li>• Low cost</li> <li>• Unskilled labour</li> <li>• More time taken</li> <li>• Low seed recovery (0.5 to 0.6%)</li> <li>• Dull seed colour</li> <li>• Seed borne pathogens</li> </ul>	<ul style="list-style-type: none"> <li>• Cost is more</li> <li>• Skilled labour</li> <li>• Lesser time</li> <li>• High seed recovery (0.8 to 1 %)</li> <li>• Bright seed colour (market value higher).</li> <li>• Seed borne pathogen remove</li> <li>• Improper washing leads to injury to seeds</li> </ul>	<ul style="list-style-type: none"> <li>• Recovery 0.7 to 0.8 per cent</li> <li>• Lustre of the seeds will be lost</li> <li>• Improper washing leads to injury to seeds</li> </ul>

### MAINTENANCE OF BREEDING RECORDS

Crop improvement programmes are of long duration, conducted at more than one research station and involve the participation of scientific and supporting staff of several disciplines. The plant breeder has to observe and evaluate thousands of plants in several replications and multilocational trials every year. There will be numerous strains under each species. A systematic record keeping of all breeding activities carried out over years and locations is necessary to provide individual identity to seed lots held in the store and the breeding plots in the field. Unless complete, simple, accurate and easily retrievable records are maintained, evaluation of the breeding material is impossible. Most of the records kept at various stations are concerned with 'origin' and pedigree' of seed which are described below

**Origin of Seed:** Each seed lot of a breeding material has an origin. For example, an origin depicted as Hyd. 13 K-4001 relates to the harvest of plot number 4001 sown at Hyderabad station during Kharif season of year 2012. Standard abbreviations are used for each research station. Last two digits of the year denote the year. Crop seasons are Kharif (crop sown in April-July) Rabi (crop sown in October-December) and Zaid (spring) (Crop sown in February-March) abbreviated to K, R & Z respectively. A set of plot numbers are used 10 each type of yield trial. Plot numbers used in one location in one crop season in a year are not duplicated elsewhere. These plot numbers of a plot, of a given location, become the origin of the following crop season. Seed lots harvested are tagged using this number.

**Pedigree of Seed:** Pedigree of breeding materials describes its complete past breeding history. It has two major components.

- i. Name of the parental variety in an abbreviated form.
- ii. Detailed breeding programme executed in each generation it was grown.

Following the name of the parental variety, a specific research station code may be added to identify the station where the breeding programme was initiated. The pedigree also provides information on the number of generations of self pollination, sib pollination or selection carried out. For example, the pedigree of an inbred line in maize depicted as Cuba 11J-A 46 # - # - # denotes the following. The inbred line was derived from Cuba 11J by self pollination at Delhi centre (station code A). The 46<sup>th</sup> self-pollinated plant was selected which was later maintained by sib Pollination for the next three generations.

#### Types of records

1. Accession Register
2. Germplasm bank
3. Descriptive blank register
4. Cropping programme
5. Single plant selection register

#### Field Note books

1. Row test
2. Replicated row test
3. Preliminary/initial yield evaluation trial
4. Comparative yield/yield evaluation trial
5. Multiplication I, II trials
6. Quality observations note book
7. Record of crosses
8. F<sub>1</sub> generation
9. F<sub>2</sub> generation note book

There are different types of records such as accession record, project book, planting plan, planting list, record book for crosses and field book. The records have to be complete such that any new plant breeder, on studying them can understand the entire breeding material available.

**1. Accession record:** This is an important and continuing permanent record which provides information on all material received and tested. For every crop there is a separate crop-wise accession book. One line of each double page of the accession book is given to variety. There are eight columns on each double page. These show accession number, name of variety, date of receiving material, source/origin of seed, source number and complete address of seed donor, pedigree record, seed description and remarks respectively.

Numbering or assigning an accession number starts with '1' every year, Last two figures year (e.g. 13 for the year 2013) in which the variety was first recorded is subscribed to the previous identifying number.

**Proforma for accession register**

1.	Accession number
2.	Name/variety
3.	Date of receipt
4.	Source of seed
5.	Source number
6.	Pedigree record
7.	Description of material
8.	How disposed and to whom sent
9.	Feedback information
10.	Remark

**2. Project book/Basic record:** Every project is separately numbered and its name, objective, plan and duration are written on a specific page of the project book. It records the purpose and procedure of the complete breeding programme.

**3. Breeding book:** A separate breeding book is maintained in the research station for each crop season. It has planting plans or sowing plans for each experimental field giving location and other details. Planting plan is the elaborate plan of a breeding block or nursery which is made well in advance of the sowing time. Planting plan contains number of replications, size of plots, row and planting distances, number of plants and location of check rows and check plots. Every row or plot in the block can be identified by a row or plot number. Breeding book, in addition to sowing plan, lists the pedigree and origin of seed as well.

**4. Planting list:** Planting list or planting schedule contains the information regarding the location of varieties in different plots. It records sowing time, starting of germination etc. which is transferred to field note book afterwards.

**5. Record diary for crosses:** The detailed information regarding each cross is separately recorded under different heads. Name of the cross, objective, number of female heads used number of F<sub>1</sub> seeds obtained, number of seeds sown and plants harvested in F<sub>1</sub>, F<sub>2</sub> and advanced generations etc. is given. The criterion of selection is also mentioned.

**6. Field book:** Field books are permanent records made on standard note books. They are always taken to the field by the plant breeder for recording daily observations. For yield tests, printed sheets containing column headings with important characters row-wise may be bound together and used.

**Numbering and Labelling the Material**

For numbering selections, crosses, introductions and mutants' notations I, II, III and M respectively may be used, e.g.

I-13-18

II-13-1608

III-3-4251

M-13-5172

The first symbol denotes whether it is a selection, cross, mutant etc. The second number denotes the year in which selection, cross etc. were made. The third number indicates the number of the particular plant selected. After emasculation and artificial cross pollination, the tag labelled on the female parent must read as follows.

No. of crossing: Name of female parent x Name of male parent: Date of emasculation, Date of pollination: Initials of Breeder(s):
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A standard form of a field note book.

Each field note book should contain the following information:

**B. Yield trials**

1. **First page:**

- (a) Number and title of the project
- (b) Season of raising the crop
- (c) Unit under which the trial is being conducted

2. **Second page:**

- (a) A full plan of the field showing the location of the trial with the approach path
- (b) North/East direction should be specified

**3. Third page:**

- (a) Plan of the experiment
- (b) Experiment details:
  - i. Name of the experiment
  - ii. Season  
Number of varieties
  - iii. Design of the experiment
  - iv. Replication  
Size of the plot (Block/plot/row etc.,)
  - v. Spacing (between rows and within the row in cm)
  - vi. Date of sowing
  - vii. Date of harvest
  - viii. Name of the principal investigator

**4. Fourth page: Detail of cultural practices followed for the plot/field**

- (a) Dates of ploughing
- (b) Date of layout of the trial
- (c) Manurial schedule adopted: Basal/ Top dressing  
Irrigation schedules with date, from life irrigation onwards.

**5. Fifth page onwards:** One-page for each variant per replication is allotted. The following information have to be recorded in each page.

- 1. Date of germination
- 2. Date of gap filling
- 3. Initial stand on
- 4. Date of first flowering
- 5. Date of general flowering
- 6. Date of harvest
- 7. Final stand
- 8. Wet weight of grain
- 9. Wet weight of haulms/straw etc.
- 10. Dry weight of produce after cleaning
- 11. Yield per ha in kg

The page will also have additional information on observations about the variant, recorded by the breeder in relation to the object of the project.

The fifth page will also contain the following information and their modification depending upon the crop, e.g., Rice - number of tillers, date of ear head emergence etc., Sesame-number of branches, days to first flowering etc.

**B. Generation Study:** This field note book will contain in addition to details as per A(i),(ii), (iii) and (iv) the third page will contain the following information.

- (a) Plan of the segregating generation
- (b) Details of the generation
  - 1. Name of the generation
  - 2. Number of crosses
  - 3. Details of the cross-cross number, details of parents, number of families, and number of seeds sown
  - 4. Length of row
  - 5. Spacing (cm)
  - 6. Date of sowing
  - 7. Dates of harvest
  - 8. Name of the principal investigator