

University of Salahaddin
College of Agricultural Engineering Sciences
Dept. of Soil and Water



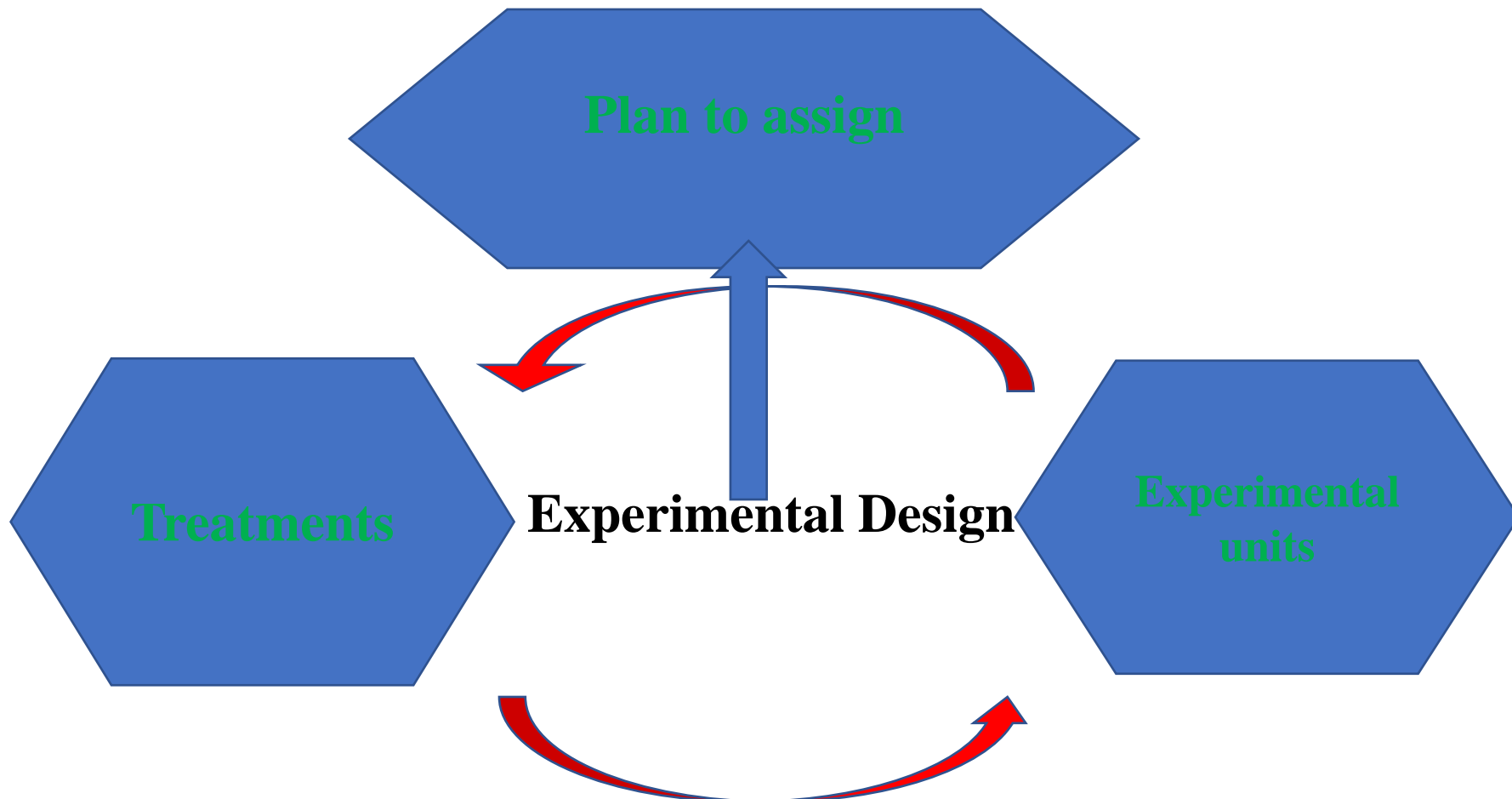
Academic year 2023-2024
3rd year Students
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Experimental Design and Analysis

Lect. Dr.Kazhin Sarbaz Rajab

Definition of experimental design:

It a part of biostatistics which refers to a plan for assigning subjects (treatment) to experimental condition and the statistical analysis associated with the plan.



The experimental design involves the following steps :

- 1- Formulation of a statistical hypothesis
- 2- Determination of independent variable (X) experimental conditions) to be used, the measuring or recording dependent variable (y).
- 3- limiting the requirements.
- 4- Selecting the suitable design for conducting the experiment.
- 5- Statistical analysis using suitable test

Basic terms in experimental design:

- 1- **Treatment** it is a condition under the control of the researcher like temp., moisture, amount of growth media...etc
- 2- **Experimental unit:** it is a smallest part in experiment which receives a treatment. Experimental unit may be human, small animal, large animal, pots, plant, Petri dish, tree.....etc.
- 3- **Simple experiment:** it is an experiment which includes one factor like studying four levels of temp. on the number of bacteria the factor is temp.
- 4- **Factorial experiment:** it is an experiment which includes two factors or more.
- 5- **Replicate:** Each experiment unit per treatment is a replicate
- 6- **Observation:** the experiment unit may be includes one observation or more for example one experiment unit may be includes two or more observations.

Principles of experimental design:

The Principles of experimental design includes:

1- Randomization. 2- Replication. 3-Local control

Functions of randomization:

The most important functions of randomization are:

1- To eliminate the effect of bias.

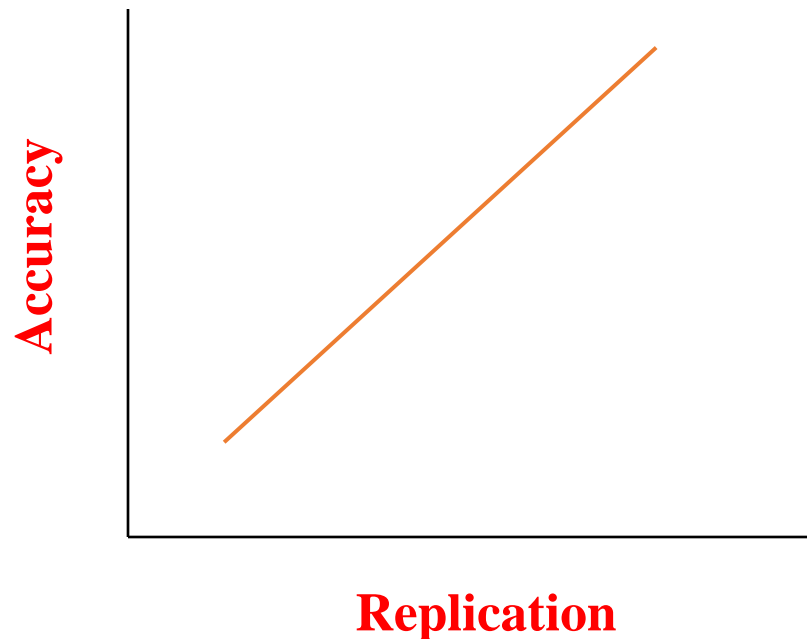
2- Protection from unusual events.

3- Error associate with adjacent experimental units are removed by randomization

Functions of replication:

The function of replication is to reduce standard error (SE) or increasing accuracy of data or experiments.

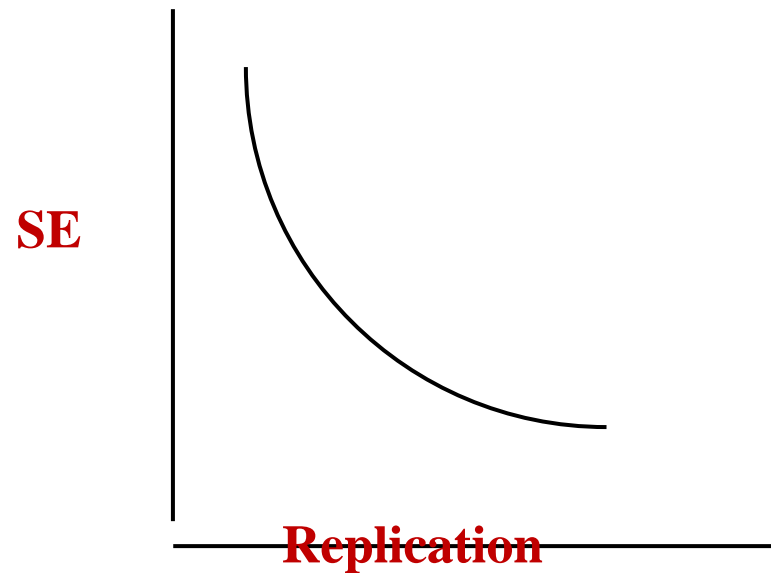
Increase in replication causes increase in accuracy .



Why?

Because increase increase in replication causes decrease in standard error (SX⁻)

$$SX^{-} = \sqrt{\frac{S^2}{n}} = \frac{S}{\sqrt{n}}$$



Classification of experimental designs:

In general the experimental designs can be divided in to two classes: **1- systematic nature designs** as shown from the schemes below:

B	C	A
B	C	A
B	C	A
B	C	A
B	C	A
B	C	A

C	C	C	C	A	A	A	A	B	B	B	B
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A	A	A	A
B	B	B	B
C	C	C	C

B	B	C	C	A	A
B	B	C	C	A	A

A,B,C= Treatments

2- Modern experimental design or (Random nature designs)

nature design: the following schemes represent some random design:

B	A	B
A	C	A
C	B	C
C	A	C
A	B	B
B	C	A

A	B	C	A
B	B	A	B
C	A	C	C

C	A	B	C	A	B	A	C	B	C	B	A
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B	A	B	C	A	C
A	B	C	B	C	A

A,B,C= Treatments

Completely randomized design (CRD)

CRD: This Design can be use if environmental conditions are fairly uniform or experimental units are homogenous ,for this reason CRD can be use in :

- 1- Laboratory experiments.
- 2-Green house experiments.
- 3-Glass house experiments.
- 4-Pot experiments.

Advantages of CRD:-

- 1- It is very simple design
- 2- It is a flexible design or there are no any restriction on the number of treatments or replication.
- 3- Statistical analysis is very easy.
- 4- It is used in case of equal or unequal replication
- 5- The missing value does not affect on the statistical analysis.

Disadvantages:

The disadvantage of CRD can be summarized as follows:

- 1- The experimental units must be homogenous.
- 2- It is used in laboratory experiment green house experiments and pot experiments.
- 3- It can not be used in field experiments.
- 4- It is less accurate than other designs.

Construction or layout of CRD:

The following steps are including for layout CRD

1- Preparing the requirements of the experiment like treatments and experimental units..... etc.

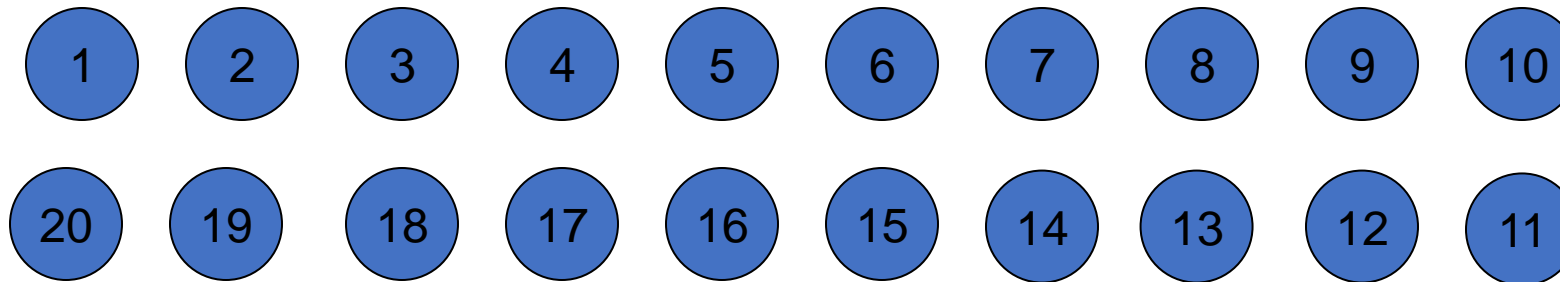
For example for studying the effect of 5 different temperatures (5, 10, 15, 20 and 25 C-) on growth radiuses (cm) of fungi using (4) replication.

$N = 5 * 4 = 20$ (Petri dishes) , because $N = t * r$, while

$N =$ Number of experimental units , $t =$ Number of treatments.

2- Number the experimental units from (1 to N) in a zigzag from.

3- Random assigning of experimental unit to treatments or assigning treatments randomly to experimental units as follow:



Statistical model:

$$X_{ij} = \mu + T_i + E_{ij}$$

X_{ij} = value of any experimental unit.

μ = General mean . T_i = Treatment effect

i = from 1 to I^{th} treatment)

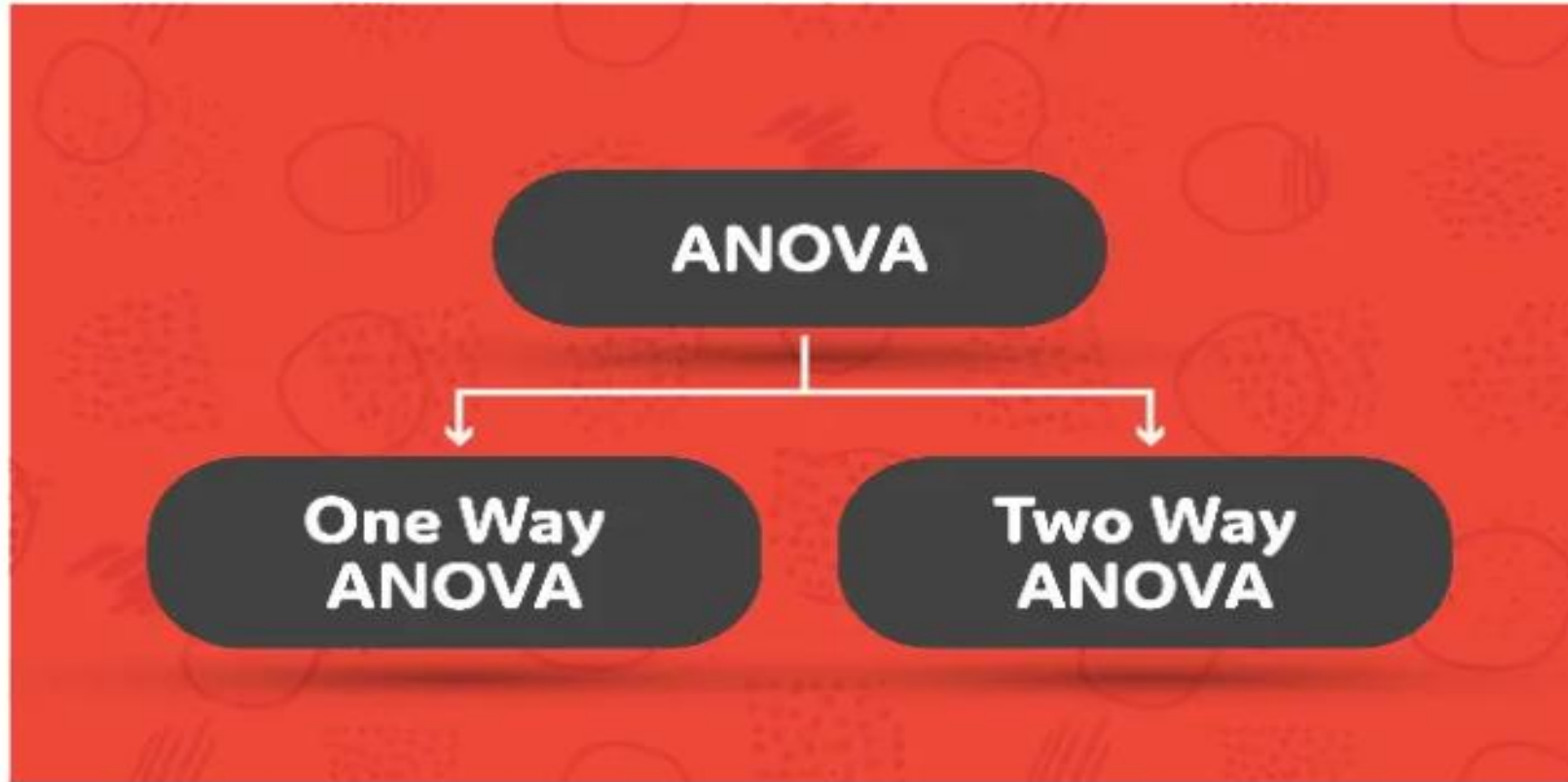
X_{ij} = the replications belong to a certain treatment.

In general the mathematical models can be divided in to two **models or types**: 1- Fixed effects model (Model I)
2- Random effects model (Model II)

Types of CRD:

- 1- CRD with equal replication.
- 2- CRD with unequal replication.
- 3- CRD with more than one observation.
- 4- Factorial CRD

Statistical analysis (ANOVA)Using SPSS program:



Equations

df = Degree of freedom

$$\text{df } t = t - 1 \quad , \quad \text{df error} = t (r - 1)$$
$$\text{df total} = n - 1 \text{ or } [(t * r) - 1]$$

SS = Sum of square

$$\text{Total SS} = \sum y_{ij}^2 - \text{C.F}$$

C.F = Correlation factor

$$\text{C.F} = (G)^2 / t * r$$

Where:-G = summation of all values

n = number of experimental units

$$SS_t = \frac{\sum t_1^2 + \sum t_2^2 + \sum t_3^2 + \sum t_n^2}{r} - \text{C.F}$$

Equations

$$SS \text{ error} = SS \text{ total} - SS \text{ t}$$

Ms = mean square

$$Mst = SSt / dft$$

$$Mse = SSe / dfe$$

Calculate F

$$\text{Cal. F} = Mst / Mse$$

We compare Cal. F with tab. F, if cal. F is equal or greater than tab. F, it means that there was a significant difference between them, and vice versa.

Exp./ A green house experiment was conducted at Plymouth university ,Faculty of Science and Environment to test the effect of number of earthworm (0, 2, 4 and 8) earth worm/pot on nitrogen content (mg/g)of *Brassica rapi* using 3 replicates ,if you are given the following information , complete ANOVA table .

treats	r1	r2	r3	Σ treats
t1	2	3	4	9
t2	3	3	4	10
t3	4	3	4	11
t4	4	6	5	15

$$\text{Total SS} = \sum y_{ij}^2 - C.F$$

$$\sum x_{ij}^2 = 2^2 + 3^2 + 4^2 + \dots + 4^2 + 5^2 - (45)2 / 3 * 4 = 181 - 168.75 = 12.25$$

$$SS_t = \frac{\sum t1^2 + \sum t2^2 + \sum t3^2 + \sum t4^2}{r} - C.F$$

$$SS_{\text{treat.}} = \frac{9^2 + \dots + \dots + 15^2}{3} - 168.75 = 6.91$$

$$SS_{\text{Error}} = \text{Total SS} - \text{Treat. SS} = 12.25 - 6.91 = 5.34$$

$$M_{\text{st}} = SS_{\text{T}} / df_{\text{T}} = 6.91 / 3 = 2.30$$

$$MSE = SS_{\text{e}} / df_{\text{E}} = 5.34 / 8 = 0.67$$

$$\text{Cal. F} = 2.34 / 0.67 = 3.43$$

From special statistical tables we obtain:

$$\text{Tab. F}_{(0.01, 3, 8)} = 7.59$$

S.O.V.	df	SS	MS	Cal.F	Tab.F _{0.01}
Treatment	3	6.91	2.30	3.43 n.s	7.59
Error	8	5.34	0.67		
Total	11	12.25			

Since calculated F is less than Tab.F it means that there are no significance differences between treatments, or the earth worm not affected significantly on nitrogen content

Exp./ A green house experiment was conducted at Plymouth university ,Faculty of Science and Environment to test the effect of 4 species of earthworm on Nitrogen % of *Brassica rapi* using 3 replicates ,if you are given the following information , complete ANOVA table ,then test the significance of treatments.

treats	r1	r2	r3	Σ treats
t1	2	3	4	9
t2	3	3	4	10
t3	4	2	5	11
t4	7	6	7	20

$$\text{Total SS} = \sum y_{ij}^2 - C.F$$

$$\begin{aligned} \sum x^2_{ij} &= 2^2 + 3^2 + 4^2 + \dots + 5^2 + 7^2 - (50)2/3 * 4 = 242 - 208.33 = 33.67 \\ &= 242 - (2500/12) = 242 - 208.33 = 33.67 \end{aligned}$$

$$SS_t = \frac{\sum t1^2 + \sum t2^2 + \sum t3^2 + \sum t4^2}{r} - C.F$$

$$SS_{\text{treat.}} = \frac{9^2 + \dots + \dots + 20^2}{3} - 208.33 = (702/3) - 208.33 = 25.67$$

$$SS_{\text{Error}} = \text{Total SS} - \text{Treat. SS} = 33.67 - 25.67 = 8.00$$

$$M_{\text{st}} = SS_{\text{t}} / df_{\text{t}}$$

$$= 25.67 / 3 = 8.56$$

$$MSE = SS_{\text{e}} / df_{\text{E}}$$

$$= 8 / 8 = 1$$

$$\text{Cal. F} = 8 / 1 = 8$$

From special statistical tables we obtain:

$$\text{Tab. F}_{(0.01, 3, 8)} = 7.59$$

S.O.V.	df	SS	MS	Cal.F	Tab.F _{0.01}
Treatment	3	25.67	8.56	8.56**	7.59
Error	8	8	1.00		
Total	11	33.67			

Since calculated F is more than Tab.F it means that there are significance differences between treatments, or the earth worm affected significantly on nitrogen content

Home work :

Example: A green house experiment was conducted at Plymouth university, Faculty of Science and Environment to test the effect of number of earthworm (0, 2, 4 and 8) earth worm/pot on leaf area (cm²)of *Brassica rapi* using 5 replicates ,if you are given the following information , complete ANOVA table ,then test the significance effect of earthworm on leaf area value.

Earthworm density	r ₁	r ₂	r ₃	r ₄	r ₅
t ₁ =0	60	65	70	75	80
t ₂ =2	80	90	70	85	75
t ₃ =4	90	100	100	112	96
t ₄ =8	110	112	120	121	114

Homework :

Example: A green house experiment was conducted at Plymouth university ,Faculty of Science and Environment to test the effect of 4 species of earthworm (*Aporrecodea spp.*) on number of leaves /plant (*Brassica rapi*) using 5 replicates ,if you are given the following information , complete ANOVA table ,then test the significance effect of earthworm species on number of leaves at L.S =0.01.

Earthworm density	r ₁	r ₂	r ₃	r ₄	r ₅
t ₁ = control	3	4	5	5	3
t ₂ = <i>A. rosea</i>	5	5	6	5	7
t ₃ = <i>A. chlorotica</i>	7	8	7	8	9
t ₄ = <i>A. caliginosa</i>	15	15	13	15	14

Example ./ The Green house experiment was done at Plymouth university to study the effect of 5 species of earth worm on nitrogen concentration (mg/g dry matter of plant , Complete ANOVA table .

<i>Treatments</i>	r_1	r_2	r_3	r_4	Σ of t
t_1	12.3	13.1	12.9	13.2	51.5
t_2	14.6	15.5	13.5	14.2	57.8
t_3	13.5	14.6	13.8		31.9
t_4	12.9	13.0	14.0	14.0	53.9
t_5	13.9	12.8	13.7		40.4
					G=235.5

No. of exp. Units (n) = $\Sigma r_i = 4+4+3+4+3=18$

$df_t = t-1=4$ and $df_r = \Sigma r_i -1=18-1=17$

C.F. = $(235.5)^2/18=3081.125$

SST = $[(12.3)^2+(13.1)^2+\dots+(13.7)^2 - \mathbf{3081.125} = 10.50$

$$SSt = \frac{\sum t1^2}{r_1} + \frac{\sum t2^2}{r_2} + \frac{\sum t3^2}{r_3} + \dots + \frac{\sum t5^2}{r_5} - C.F$$

$$SS_t = \frac{(51.5)^2}{4} + \frac{(57.8)^2}{4} + \frac{(31.9)^2}{3} + \frac{(53.9)^2}{4} + \frac{(40.4)^2}{3} - 3081.125$$

$$= 5.48$$

$$SS_E = SSTotal - SStreat = 10.5 - 5.48 = 5.02$$

ANOVA Table

S.O.V.	df	SS	MS	Cal.F	Tab.F
Treatment	4	5.48	1.37	3.51 ^{n.s}	5.205 _(0.01)
Error	13	5.02	0.39		
Total	17	10.5			

Home work: The study was conducted to test the effect of [5]levels of iron on number of active nodules/plant ,complete ANOVA table if you are given the following information ,then compare between treatments using $LSD_{0.01}$ and DMRT :

Treatments	r1	r2	r3	r4
$t_1=0$ mg/pot	4	5	4	6
$t_2=2$ mg/pot	5	7	7	8
$t_3= 4$ mg/pot	6	7	9	9
$t_4=6$ mg/pot	10	12	14	14
$t_5=8$ mg/pot	7	8	8	9

A study was conducted to study the effect of 6 specie of Rhizobium ,on weight of active nodules/plant using 4 replicates. Complete .ANOVA table then compare between treatments using different tests

treats	r_1	r_2	r_3	r_4
t_1	7	8	7	6
t_2	6	5	6	5
t_3	4	4	3	4
t_4	3	3	3	2
t_5	2	1	1	1
t_6	1	2	2	1