

# Design of Experiments

A statistical experiment in any field is performed to verify a particular hypothesis.

For Example, an agricultural experiment may be performed to verify the claim that a particular manure has got the effect of increasing the yield of paddy. Here the quantity of the manure used and the amount of yield are the two variables involved directly are called as *experimental variables*.

Fertility of the soil, the quantity of the seed used and the amount of rainfall, which also effect the yield of paddy - *extraneous variables*.

**Main Objective:** To control the extraneous variables and hence to minimise the experimental error so that the results of the experiments could be attributed only to the experimental variables.

# **Principles of Experimental Design**

- Randomisation
- Replication
- Local Control

# Analysis of Variance (ANOVA)

It is a powerful statistical tool in tests of significance. In parametric tests, we discussed the statistical tests relating to mean of a population or equality of means of two populations.

In situations, when we have three or more samples to consider at a time, an alternative procedure is needed for testing the hypothesis that all the samples are drawn from the same populations, which have the same mean.

*Analysis of variance* (ANOVA) was introduced by **R.A. Fisher** to deal the problem in the analysis of agricultural data. Variations in the observations are inherent in nature. The total variation in the observed data is due to the following two causes namely, (i) assignable causes, and (ii) chance causes.

By this technique, the total variation in the sample data can be bifurcated into variation between sample and variation within samples. The second kind of variation is due to experimental error.

## **1. Treatments:**

Various factors or methods that we adopted in a comparative experiment are termed as *treatments*. For example, in field experiments, different varieties of paddy seeds, different kinds of fertilizers, different methods of cultivation etc., are called treatments.

## **2. Experimental Unit**

A small area of experimental material is used for applying the treatment is called an *experimental unit*.

In agricultural experiments, a cultivated land, usually called as *experimental material* is divided into smaller areas of plots in which, different treatment can be applied in it. Such kind of plots are called experimental units.

## **3. Blocks**

In field experiments, the experimental material is firstly divided into relatively homogeneous divisions, known as *Blocks*. All the blocks are further divided into small plots of experimental units.

# One-way ANOVA

## Aim

To test the significance of the  $t$  – treatment effects based on the observations from  $n$  – experimental units.

## Source

Let  $y_{ij}$ , ( $i = 1, 2, \dots, t; j = 1, 2, \dots, r$ ) be the observations of  $t$  – treatments, each replicated with (equal number of replications)  $r$  - times in  $n$  – experimental units (*i.e.*,  $n = tr$ ).

In this design, treatments are allocated at random to the experimental units over the entire experimental material. That is, the entire experimental material is divided into  $n$  experimental units and the treatments are distributed completely at random over the units.

**Null Hypothesis ( $H_0$ ):** The  $k$  – treatments have equal effect.

$$i.e., \tau_1 = \tau_2 = \dots = \tau_k$$

**Alternative Hypothesis ( $H_1$ ):** The  $k$  – treatments do not have equal effect.

$$i.e., \tau_1 \neq \tau_2 \neq \dots \neq \tau_k$$

**Level of Significance ( $\alpha$ ) and Critical Region ( $F_\alpha$ )**

$$F > F_{\alpha, (k-1, n-k)} \text{ such that } P [F > F_{\alpha, (k-1, n-k)}] = \alpha$$

The critical values of  $F$  at level of Significance  $\alpha$  and degrees of freedom  $(k-1, n-k)$ , are obtained from Table.

**Method:**

1. Grand total of the all the observations,  $\mathbf{G} = \sum_{i=1}^k \sum_{j=1}^r y_{ij}$

2. Correction Factor,  $\mathbf{CF} = \mathbf{G}^2 / \mathbf{n}$

3. Total Sum of Squares,  $\mathbf{TSS} = \sum_{i=1}^k \sum_{j=1}^r y_{ij}^2 - \mathbf{CF}$

4. Sum of Squares between treatments,  $\mathbf{SSTr} = \frac{1}{r} \sum_{i=1}^k \mathbf{T}_i^2 - \mathbf{CF}$

$T_i$  - total of the  $i^{th}$ - treatment observations from all the replications( $r$ ).

5. Error Sum of Squares (Sum of Squares within treatments),  $\mathbf{ESS} = \mathbf{TSS} - \mathbf{SSTr}$

## Anova Table:

<i>Source of Variation</i>	<i>Degrees of freedom</i>	<i>Sum of Squares</i>	<i>Mean Sum of Squares</i>	<i>F-statistic</i>
<i>Treatments</i>	$k - 1$	$SSTr$	$s_{Tr}^2 = \frac{SSTr}{k - 1}$	$F = \frac{s_{Tr}^2}{s_E^2}$
<i>Error</i>	$n - k$	$SSE$	$s_E^2 = \frac{SSE}{n - k}$	
<i>Total</i>	$n - 1$	$TSS$		

## Conclusion:

If  $|F_{cal}| \leq |F_{\alpha, (k-1, n-k)}|$ , we conclude that the data do not provide us any evidence against the null hypothesis  $H_0$ , and hence it may be accepted at  $\alpha\%$  level of significance. Otherwise reject  $H_0$  or accept  $H_1$ .

# Example 1

The following data denotes the four “tropical feed stuffs *A, B, C, D*” tried on **20** chicks is given below. All the twenty chicks are treated alike in all respects except the feeding treatments and each feeding treatment is given to five chicks. Test whether all the four feedstuffs are alike in weight gain of the chicks at 5% level of significance.

A	55	49	42	21	52
B	61	112	30	89	63
C	42	97	81	95	92
D	169	137	169	85	154

*Solution:*

*Aim:* To test all the four feedstuffs are equal in weight gain of chicks.

*H<sub>0</sub>:* The four feedstuffs are equal in weight gain of chicks.

*H<sub>1</sub>:* The four feedstuffs are not equal in weight gain of chicks.

*Level of Significance:*  $\alpha = 0.05$  and *Critical value:*  $F_{0.05, (3,16)} = 3.24$

Number of treatments,  $k = 4$  &  $n = 20$

$$T_1 = 219, T_2 = 355, T_3 = 407, T_4 = 714$$

Grand Total,  $G = 1695$

$$CF = 1695^2/20 = 143651.25$$

$$TSS = 55^2 + \dots + 154^2 - CF = 181445 - 143651.25 = 37793.75$$

$$SSTr = \frac{1}{5}(219^2 + \dots + 714^2) - CF = 26234.95$$

$$ESS = TSS - SSTr = 11558.80$$

## Anova Table:

<i>Source of Variation</i>	<i>Degrees of freedom</i>	<i>Sum of Squares</i>	<i>Mean Sum of Squares</i>	<i>F-statistic</i>
<i>Treatments</i>	3	26234.95	8744.98	12.11
<i>Error</i>	16	11558.80	722.42	
<i>Total</i>	19	37793.75	-	

**Conclusion:** Since  $F > F_{0.05, (3,16)}$ , we conclude that the data provide us evidence against the null hypothesis  $H_0$  and in favor of  $H_1$ . Hence,  $H_1$  is accepted at 5% level of significance. That is, the four feedstuffs are not equal in weight gain of chicks.

## Example 2

The following tables gives the yields of wheat from **16** plots, all of the approximately equal fertility, when **4** varieties of wheat were cultivated in a completely randomised fashion. Test the hypothesis that the varieties are not significantly different.

Plot No.	1	2	3	4	5	6	7	8	9	10
Variety	A	B	D	C	B	C	A	D	B	D
Yield	32	34	29	31	33	34	34	26	36	30
Plot No.	11	12	13	14	15	16				
Variety	A	C	B	A	B	C				
Yield	33	35	37	35	35	32				

<b>A</b>	<b>B</b>	<b>C</b>	<b>D</b>
32	34	31	26
34	33	34	30
33	36	35	
35	37	32	
	35		

# Two-way ANOVA & Randomized Block Design

## Aim

To test the significance of the  $k$  treatment effects and the significance of the  $r$  block effects based on the observations from  $n$  experimental units.

## Null Hypotheses

$H_0(1)$ : The  $k$  treatments have equal effect. *i.e.*,  $\mathbf{H}_0: \mathbf{t}_1 = \mathbf{t}_2 = \dots = \mathbf{t}_k$ .

$H_0(2)$ : The  $r$  blocks have equal effect. *i.e.*,  $\mathbf{H}_0: \mathbf{b}_1 = \mathbf{b}_2 = \dots = \mathbf{b}_r$ .

## Alternative Hypotheses

$H_1(1)$ : The  $k$  treatments do not have equal effect.

*i.e.*,  $\mathbf{H}_1: \mathbf{t}_1 \neq \mathbf{t}_2 \neq \dots \neq \mathbf{t}_k$ .

$H_1(2)$ : The  $r$  blocks do not have equal effect.

*i.e.*,  $\mathbf{H}_1: \mathbf{b}_1 \neq \mathbf{b}_2 \neq \dots \neq \mathbf{b}_r$ .

## Level of Significance ( $\alpha$ ) and Critical Region ( $F_\alpha$ )

$F_1 > F_{\alpha, (k-1), (k-1)(r-1)}$  such that

$$P [F_1 > F_{\alpha, (k-1), (k-1)(r-1)}] = \alpha.$$

$F_2 > F_{\alpha, (r-1), (k-1)(r-1)}$  such that

$$P [F_2 > F_{\alpha, (r-1), (k-1)(r-1)}] = \alpha.$$

The critical values of  $F$  at level of Significance  $\alpha$  and degrees of freedom  $[(k-1), (k-1)(r-1)]$  and for  $[(r-1), (k-1)(r-1)]$ , are obtained from Table.

## Method:

1. Grand total of the all the observations,  $\mathbf{G} = \sum_{i=1}^k \sum_{j=1}^r y_{ij}$
2. Correction Factor,  $\mathbf{CF} = \mathbf{G}^2 / \mathbf{n}$
3. Total Sum of Squares,  $\mathbf{TSS} = \sum_{i=1}^k \sum_{j=1}^r y_{ij}^2 - \mathbf{CF}$
4. Sum of Squares between treatments,  $\mathbf{SSTr} = \frac{1}{r} \sum_{i=1}^k \mathbf{T}_i^2 - \mathbf{CF}$

$T_i$  - total of the  $i^{th}$ - treatment observations from all the replications.

5. Sum of Squares between blocks,  $\mathbf{SSB} = \frac{1}{k} \sum_{j=1}^r \mathbf{B}_j^2 - \mathbf{CF}$

$B_j$  - total of the  $j^{th}$ - block observation.

5. Error Sum of Squares (Sum of Squares within treatments),  $\mathbf{ESS} = \mathbf{TSS} - \mathbf{SSTr} - \mathbf{SSB}$

## Anova Table:

<i>Source of Variation</i>	<i>Degrees of freedom</i>	<i>Sum of Squares</i>	<i>Mean Sum of Squares</i>	<i>F-statistic</i>
<i>Treatments</i>	$k - 1$	$SSTr$	$s_{tr}^2 = SSTr / k - 1$	$F_1 = s_{tr}^2 / s_E^2$
<i>Blocks</i>	$r - 1$	$SSB$	$s_B^2 = SSB / r - 1$	$F_2 = s_B^2 / s_E^2$
<i>Error</i>	$(k - 1) (r - 1)$	$ESS$	$s_E^2 = ESS / (k-1) (r-1)$	
<i>Total</i>	$n - 1$	$TSS$		

## Example 2

The following result shows the yield of three varieties of paddy manure in four plots each using RBD layout.

<i>Block</i>	<i>Paddy Varieties</i>			<i>Total</i>
	<i>ADT36</i>	<i>IR20</i>	<i>PONNI</i>	
I	46.2	48.5	54.3	149
II	48.4	52.6	57.0	158
III	44.3	51.4	53.3	149
IV	49.1	53.5	51.4	154
Total	188	206	216	610

*Aim:* 1. To test the yield of all the three varieties of paddy are equal.  
2. To test the yield in all the four blocks are equal.

$H_0(1)$ : The yields of all the three varieties of paddy are homogeneous.

$H_0(2)$ : The yields in all the four blocks are homogeneous.

$H_1(1)$ : The yields of all the three varieties of paddy are not homogeneous.

$H_1(2)$ : The yields in all the four blocks are not homogeneous.

*Level of Significance:*  $\alpha = 0.05$

*Critical values:*  $F_{0.05,(2,6)} = 5.14$  and  $F_{0.05,(3,6)} = 4.76$

No. of treatments,  $t = 3$ ; No. of Blocks,  $r = 4$ , Grand total,  $G = 610$

$$CF = 610^2/12 = 31008.33$$

$$TSS = 46.2^2 + \dots + 51.4^2 - CF = 31153.86 - 31008.33 = 145.53$$

$$SST = \frac{1}{4} (188^2 + 206^2 + 216^2) - CF = 100.67$$

$$BSS = \frac{1}{3} (149^2 + 158^2 + 149^2 + 154^2) - CF = 19.003$$

$$ESS = TSS - SST - BSS = 25.857$$

*ANOVA Table:*

<i>Sources of variation</i>	<i>Degrees of freedom</i>	<i>Sum of squares</i>	<i>Mean sum of squares</i>
Treatments	2	100.67	50.335
Blocks	3	19.003	6.334
Error	6	25.857	4.3095
Total	11	145.53	–

*Test Statistics:*

1. 
$$F_1 = \frac{SST/(t-1)}{ESS/(t-1)(r-1)} = \frac{50.335}{4.3095} = 11.68$$

2. 
$$F_2 = \frac{SSB/(r-1)}{ESS/(t-1)(r-1)} = \frac{6.334}{4.3095} = 1.47$$

*Conclusions:*

1. Since,  $F_1 > F_{0.05,(2,6)}$ , we conclude that the data provide us any evidence against the null hypothesis  $H_0(1)$  and in favor of  $H_1(1)$ . Hence  $H_1(1)$  is accepted at 5% level of significance. That is, the yields of all the three varieties of paddy are not homogeneous.
2. Since,  $F_2 < F_{0.05,(3,6)}$ , we conclude that the data do not provide us any evidence against the null hypothesis  $H_0(2)$ , and hence it may be accepted at 5% level of significance. That is, the yields in all the four blocks are homogeneous.

# Latin Square Design ( LSD )

## Aim

To test the significance of the  $m$  treatment effects,  $m$  row effects and  $m$  column effects based on the observations from  $m$  square ( $m^2$ ) experimental units.

## Null Hypotheses

$H_0(1)$ : The  $m$  treatments have equal effect. *i.e.*,  $H_0(1): \tau_1 = \tau_2 = \dots = \tau_m$

$H_0(2)$ : The  $m$  rows have equal effect. *i.e.*,  $H_0(2): \beta_1 = \beta_2 = \dots = \beta_m$

$H_0(3)$ : The  $m$  columns have equal effect. *i.e.*,  $H_0(3): \nu_1 = \nu_2 = \dots = \nu_m$

## Alternative Hypotheses

$H_1(1)$ : The  $m$  treatments do not have equal effect.

*i.e.*,  $H_1(1): \tau_1 \neq \tau_2 \neq \dots \neq \tau_m$

$H_1(2)$ : The  $m$  rows do not have equal effect

*i.e.*,  $H_1(2): \beta_1 \neq \beta_2 \neq \dots \neq \beta_m$

$H_1(3)$ : The  $m$  columns do not have equal effect.

*i.e.*,  $H_1(3): \nu_1 \neq \nu_2 \neq \dots \neq \nu_m$

## Level of Significance ( $\alpha$ ) and Critical Region

$$F_i > F_{\alpha, (m-1), (m-1)(m-2)} \text{ such that } P [F_i > F_{\alpha, (m-1), (m-1)(m-2)}] = \alpha$$

for  $i = 1, 2, 3$ . The critical values of  $F$  at level of Significance  $\alpha$  and degrees of freedom  $(m - 1, (m - 1)(m - 2))$  are obtained from Table

## Method

Calculate the following, based on the observations.

1. Grand total of all the observations,  $G = \sum_{j=1}^m \sum_{k=1}^m y_{ijk}$

2. Correction Factor,  $CF = \frac{G^2}{m^2}$

3. Total Sum of Squares,  $TSS = \sum_{j=1}^m \sum_{k=1}^m y_{ijk}^2 - CF$

4. Sum of Squares between Treatments,  $SST = \frac{1}{m} \sum_{i=1}^m T_i^2 - CF$   
 $T_i$  be the total of the  $i^{\text{th}}$  treatment observations.

5. Sum of Squares between Rows,  $SSR = \frac{1}{m} \sum_{j=1}^m R_j^2 - CF$   
 $R_j$  be the total of the  $j^{\text{th}}$  row observations.

6. Sum of Squares between Columns,  $SSC = \frac{1}{m} \sum_{k=1}^m C_k^2 - CF$

$C_k$  be the total of the  $k^{\text{th}}$  column observations.

7. Error Sum of Square,  $ESS = TSS - SST - SSR - SSC$ .

## Analysis of Variance Table

<i>Sources of variation</i>	<i>Degrees of freedom</i>	<i>Sum of squares</i>	<i>Mean sum of squares</i>
Treatments	$m-1$	$SST$	$SST/(m-1)$
Rows	$m-1$	$SSR$	$SSR/(m-1)$
Columns	$m-1$	$SSC$	$SSC/(m-1)$
Error	$(m-1)(m-2)$	$ESS$	$ESS/(m-1)(m-2)$
Total	$m^2 - 1$	$TSS$	-

## Test Statistics

$$1. \quad F_1 = \frac{SST/(m-1)}{ESS/(m-1)(m-2)}$$

$$2. \quad F_2 = \frac{SSR/(m-1)}{ESS/(m-1)(m-2)}$$

$$3. \quad F_3 = \frac{SSC/(m-1)}{ESS/(m-1)(m-2)}$$

The statistic  $F_1, F_2, F_3$  follows  $F$  distribution with  $(m-1), (m-1)(m-2)$  degrees of freedom.

## Conclusions

If  $F_i \leq F_{\alpha, (m-1), (m-1)(m-2)}$ , we conclude that the data do not provide us any evidence against the null hypothesis  $H_0(i)$ , and hence it may be accepted at  $\alpha\%$  level of significance. Otherwise reject  $H_0(i)$  or accept  $H_1(i)$  for  $i = 1, 2, 3$ .

**Example**

1. An experiment was carried out to determine the effect of claying the ground on the field of barley grains; amount of clay used were as follows. *A*: No clay, *B*: Clay at 100 per acre. *C*: Clay at 200 per acre, *D*: Clay at 300 per acre. The yields were in plots of 10 square meters and the layout and yields were as follows. Analyze all the effects at 5% level of significance.

<i>Row</i> \ <i>Column</i>	<i>I</i>	<i>II</i>	<i>III</i>	<i>IV</i>	<i>Total</i>
<i>I</i>	<i>D</i> 34.7	<i>A</i> 35.6	<i>B</i> 38.2	<i>C</i> 35.5	144
<i>II</i>	<i>C</i> 38.2	<i>D</i> 34.4	<i>A</i> 42.8	<i>B</i> 37.6	153
<i>III</i>	<i>A</i> 36.4	<i>B</i> 37.2	<i>C</i> 41.7	<i>D</i> 36.7	152
<i>IV</i>	<i>B</i> 39.7	<i>C</i> 38.8	<i>D</i> 40.3	<i>A</i> 38.2	157
<i>Total</i>	149	146	163	148	606

## **Solution**

$H_0(1)$ : The yields under four types of clay are equal.

$H_0(2)$ : All the four rows have equal yields.

$H_0(3)$ : All the four columns have equal yields.

$H_1(1)$ : The yields under four types of clay are not equal.

$H_1(2)$ : All the four rows do not have equal yields.

$H_1(3)$ : All the four columns do not have equal yields.

*Level of Significance:*  $\alpha = 0.05$  and *Critical value:*  $F_{0.05,(3,6)} = 4.76$

*Calculations:*

$m = \text{No. of treatments} = \text{No. of rows} = \text{No. of columns} = 4$

No. of experimental units,  $n = 16$ .  $T_1=153$   $T_2=152.7$   $T_3= 154.2$   $T_4= 146.1$

$$1. \quad G = \sum_{j=1}^m \sum_{k=1}^m y_{ijk} = 606$$

$$2. \quad CF = \frac{G^2}{m^2} = \frac{606^2}{4^2} = 22952.25$$

$$3. \quad TSS = \sum_{j=1}^m \sum_{k=1}^m y_{ijk}^2 - CF = 23038.58 - CF = 86.33$$

$$4. \quad SST = \frac{1}{m} \sum_{i=1}^m T_i^2 - CF = \frac{1}{4} (153^2 + 152.7^2 + 154.2^2 + 146.1^2) - CF = 10.035$$

$$5. \quad SSR = \frac{1}{m} \sum_{j=1}^m R_j^2 - CF = \frac{1}{4} (144^2 + 153^2 + 152^2 + 157^2) - CF = 22.25$$

$$6. \quad SSC = \frac{1}{m} \sum_{k=1}^m C_k^2 - CF = \frac{1}{4} (149^2 + 146^2 + 163^2 + 148^2) - CF = 45.25$$

$$7. \quad ESS = TSS - SST - SSR - SSC = 8.795$$

*ANOVA Table:*

<i>Sources of variation</i>	<i>Degrees of freedom</i>	<i>Sum of squares</i>	<i>Mean sum of squares</i>
Treatments	3	10.035	3.345
Rows	3	22.25	7.4167
Columns	3	45.25	15.08
Error	6	8.795	1.4658
Total	15	86.33	—

*Test Statistics:*

$$1. \quad F_1 = \frac{SST/(m-1)}{ESS/(m-1)(m-2)} = 2.28$$

$$2. \quad F_2 = \frac{SSR/(m-1)}{ESS/(m-1)(m-2)} = 5.06$$

$$3. \quad F_3 = \frac{SSC/(m-1)}{ESS/(m-1)(m-2)} = 10.29$$

*Conclusions:* Since  $F_1 < F_{0.05, (3,6)}$ , we conclude that the data do not provide us any evidence against the null hypothesis  $H_0(1)$ , and hence it may be accepted at 5% level of significance. That is, all the four types of clay have equal yields.

Since  $F_2, F_3 > F_{0.05, (3,6)}$ , we conclude that the data provide us evidence against the null hypotheses  $H_0(2)$  and  $H_0(3)$  and in favor of  $H_1(2)$  and  $H_1(3)$ . Hence,  $H_1(2)$  and  $H_1(3)$  are accepted at 5% level of significance. That is, all the four rows have not equal yields and all the four columns have not equal yields.