Two Way ANOVA

Many experiments involve the study of the effects of two or more factors. When there are several factors of interest in an experiment, a **factorial design** should be used. In such designs, factors are varied together. Specifically, by a factorial experiment we mean that in each complete trial or replicate of an experiment all possible combinations of the levels of the factors are investigated. Thus, if there are two factors A and B with a levels of factor a and b levels of factor B, then each replicate contains all ab possible combinations. When factors are arranged in a factorial design they are often said to be **crossed.**
The effect of a factor is defined as the change in response produced by a change in the level of the factor. This is called a main effect because it refers to the primary factors in the study. Consider the data in Fig 1.

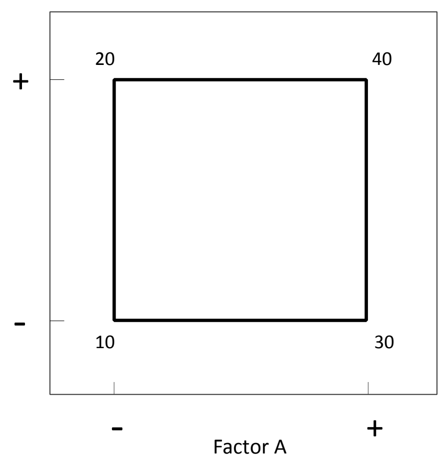

Fig. 1

In this factorial design, both the factors A and B have two levels, denoted by $-$ and $+$. These two levels are called *low* and *high*, respectively. The main effect of factor A is the difference between the average response at the high level of A and the average response at the low level of A.

$$A = \overline{y}_{A^+} - \overline{y}_{A^-} = \frac{30+40}{2} - \frac{10+20}{2} = 20$$

That is, changing factor A from the low level ($-$) to the high level ($+$) causes an average response increase of 20 units. Similarly, the main effect of B is

$$B = \overline{y}_{B+} - \overline{y}_{B-} = \frac{20+40}{2} - \frac{10+30}{2} = 10$$

In some experiments, the difference in response between the levels of one factor is not the same at all levels of the other factors. When this occurs, there is an interaction between the factors. For example, consider the data in Fig 2.

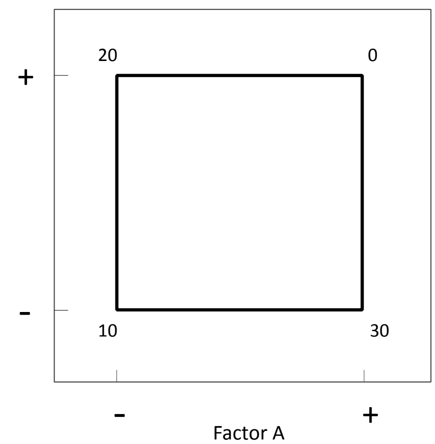

Figure 2

At the low level of factor B, the A effect is

$$A = 30 - 10 = 20$$

and at the high level of factor B, the A effect is

$$A = 0 - 20 = -20$$

Since the effect of A depends on the level chosen for factor B, there is an interaction between A and B.

When an interaction is large, the corresponding main effects have little meaning. For example, by using the data in Fig. 2, we find the main effect of A is

$$A = \frac{30+0}{2} - \frac{10+20}{2} = 0$$

and we would be tempted to conclude that there is no A effect. However, when we examine the main effect of A at different levels of B we see that this is not the case. The effect of factor A depends on the levels of factor B. Thus, knowledge of the AB interaction is more useful than knowledge of the main effect. A significant interaction can mask the significance of main effects.

The concept of interaction can be illustrated graphically. Fig. 3 plots the data shown in Fig. 1 against the levels of A for both levels of B. Note that the B^+ and the B^- lines are roughly parallel, indicating that factors A and B do not interact.

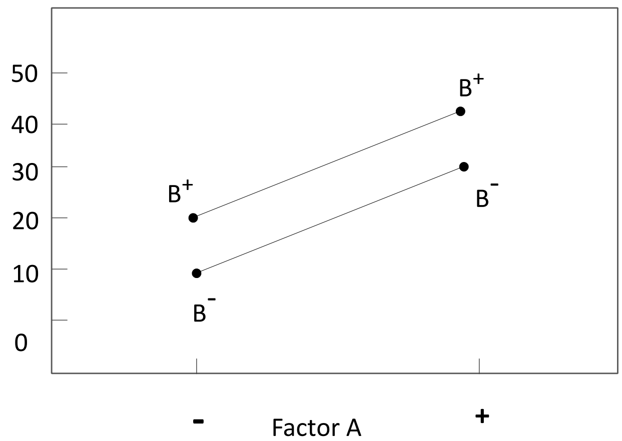

Fig. 3

Fig. 4 plots the data shown in Fig. 2. Note that the B^+ and B^- lines are not parallel, indicating that there is no interaction between factors A and B. Such graphical displays are often useful in presenting the results of experiments.

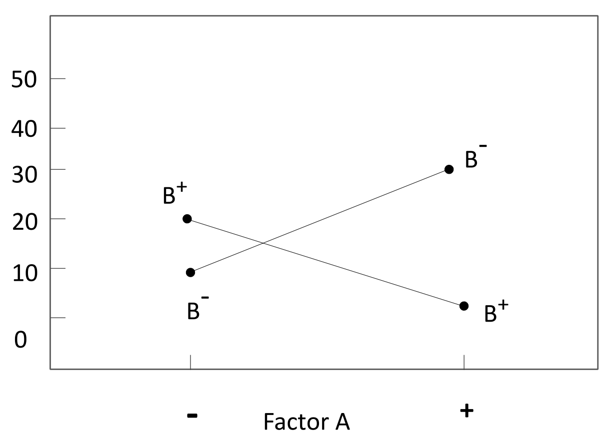

Fig. 4

Two-Way ANOVA

The simplest types of factorial designs involve only two factors or sets of treatments. Suppose that we have two factors denoted:

- A, that has $i = 1...a$ levels
- B, that has $j = 1...b$ levels

Let n_{ij} be the number of observations at level i of A and level j of B. For a balanced design $n_{ij} = n$.
Let observations of the response variable be denoted by y_{ijk}, $i = 1...a$, $j = 1...b$, $k = 1...n$
The table below illustrates the arrangement of a two factorial design

Factor B

	1	2	...	b
1	$y_{111}, y_{112}, ..., y_{11n}$	$y_{121}, y_{122}, ..., y_{12n}$		$y_{1b1}, y_{1b2}, ..., y_{1bn}$
2	$y_{211}, y_{212}, ..., y_{21n}$	$y_{221}, y_{222}, ..., y_{22n}$		$y_{2b1}, y_{2b2}, ..., y_{2bn}$
$\vdots$				
a	$y_{a11}, y_{a12}, ..., y_{a1n}$	$y_{a21}, y_{a22}, ..., y_{a2n}$		$y_{ab1}, y_{ab2}, ..., y_{abn}$

The effects model for the two-way ANOVA can be written as:

$$y_{ijk} = \mu + \alpha_i + \beta_j + (a\beta)_{ij} + \varepsilon_{ijk}$$

$\alpha_i$represents the treatment effect of factor A at level i
β_j represents the treatment effect of factor B at level j
$(\alpha\beta)_{ij}$ represents the interaction effect and ε_{ijk} ~NID$(0, \sigma^2)$.

Hypotheses

The hypotheses associated with a two-way ANOVA are:

$$\begin{aligned} H_0 &: \alpha_1 = \alpha_2 = ... = \alpha_a = 0 \\ H_A &: \text{at least one } \alpha_i \neq 0 \end{aligned}$$

$$\begin{aligned} H_0 &: \beta_1 = \beta_2 = ... = \beta_b = 0 \\ H_A &: \text{at least one } \beta_j \neq 0 \end{aligned}$$

$$
\begin{aligned}
H_0 &: \ (\alpha\beta)_{ij} = 0 \text{ for all } i, j \\
H_A &: \ \text{at least one } (\alpha\beta)_{ij} \neq 0
\end{aligned}
$$

We test these hypotheses using a two-way ANOVA. The total variance in a data set is partitioned into sums of squares:

$$SS_T = SS_A + SS_B + SS_{AB} + SS_E$$

SS_T is the Total sum of squares
SS_A Factor A sum of squares
SS_B Factor B sum of squares
SS_{AB} Interaction sum of squares
SS_E Error sum of squares

The table for a two-way ANOVA is shown below:

Source	Sum of Squares (SS)	*df*	Mean Square (MS)	F-ratio
Factor A	SS_A	$a-1$	$\frac{\mathbf{SS}_A}{a-1}$	$\frac{\text{Mean Square } A}{\text{Mean Square Error}}$
Factor B	SS_B	$b-1$	$\frac{\mathbf{SS}_B}{b-1}$	$\frac{\text{Mean Square } B}{\text{Mean Square Error}}$
Interaction AB	SS_{AB}	$(a-1)(b-1)$	$\frac{\mathbf{SS}_{AB}}{(a-1)(b-1)}$	$\frac{\text{Mean Square } AB}{\text{Mean Square Error}}$
Error	SS_E	$ab\,(n-1)$	$\frac{\mathbf{SS}_E}{ab(n-1)}$	
Total	SS_T	$abn-1$		

The sums of squares can be calculated by hand, we set up the notation as follows:

$$
\begin{aligned}
y_{...} &= \sum_{i=1}^{a}\sum_{j=1}^{b}\sum_{k=1}^{n} y_{ijk} & \overline{y}_{...} &= \tfrac{y_{...}}{abn} \\
y_{i..} &= \sum_{j=1}^{b}\sum_{k=1}^{n} y_{ijk} & \overline{y}_{i..} &= \tfrac{y_{i..}}{bn} \\
y_{.j.} &= \sum_{j=1}^{a}\sum_{k=1}^{n} y_{ijk} & \overline{y}_{.j.} &= \tfrac{y_{.j.}}{an} \\
y_{ij.} &= \sum_{k=1}^{n} y_{ijk} & \overline{y}_{ij.} &= \tfrac{y_{ij.}}{n}
\end{aligned}
$$

Then

$$
\begin{aligned}
\mathrm{SS}_T &= \sum_{i=1}^{a}\sum_{j=1}^{b}\sum_{k=1}^{n}(y_{ijk}-\overline{y}_{...})^2\\
\mathrm{SS}_A &= nb\sum_{i=1}^{a}\sum_{k=1}^{n}(\overline{y}_{i..}-\overline{y}_{...})^2\\
\mathrm{SS}_B &= na\sum_{j=1}^{b}\sum_{k=1}^{n}\left(\overline{y}_{.j.}-\overline{y}_{...}\right)^2\\
\mathrm{SS}_{AB} &= n\sum_{j=1}^{b}\sum_{i=1}^{a}\left(\overline{y}_{ij.}-\overline{y}_{i..}-\overline{y}_{.j.}+y_{ijk}\right)^2\\
\mathrm{SS}_E &= \sum_{i=1}^{a}\sum_{j=1}^{b}\sum_{k=1}^{n}\left(y_{ijk}-\overline{y}_{ij.}\right)^2
\end{aligned}
$$

$$\mathrm{SS}_T = \mathrm{SS}_A + \mathrm{SS}_B + \mathrm{SS}_{AB} + \mathrm{SS}_E$$

Interpretation of a two-way ANOVA depends on whether the interaction term between the two factors is significant. We will examine both cases using examples.

Example - Significant Interaction

An experiment was carried out to investigate the effect of pH and Catalyst Concentration on the Viscosity of a product. Three replicates of a design were performed, with the following results:

pH	**Catalyst Conc**	**Viscosity**	**Treatment Total**	**Treatment Mean**
5.6	2.5	$192, 199, 193$	$y_{11.} = 584$	$\overline{y}_{11.} = 194.6667$
5.6	2.7	$178, 185, 188$	$y_{12.} = 551$	$\overline{y}_{12.} = 183.6667$
5.9	2.5	$185, 193, 190$	$y_{21.} = 568$	$\overline{y}_{21.} = 189.3333$
5.9	2.7	$197, 196, 204$	$y_{22.} = 597$	$\overline{y}_{22.} = 189.3333$

To get a feel for the data we can summarise it across the different factors and write down the means using the notation introduced above.

Catalyst Concentration

	2.5	2.7	$\sum$	Mean
5.6	$192, 199, 193$	$178, 185, 188$	$y_{1..} = 1135$	$\overline{y}_{1..} = 189.1667$
5.9	$185, 193, 190$	$197, 196, 204$	$y_{2..} = 1165$	$\overline{y}_{2..} = 194.1667$
$\sum$	$y_{.1.} = 1152$	$y_{.2.} = 1148$	$y_{...} = 2300$	
Mean	$\overline{y}_{.1.} = 192$	$\overline{y}_{.2.} = 191.3333$		$\overline{y}_{...} = 191.6667$

To determine whether pH and Catalyst Concentration affect the Viscosity of the product and whether the two factors interact, we can carry out a two-way ANOVA. Let Factor A represent pH and let Factor B represent Catalyst Concentration.
First write down the model and state the hypotheses:

$$y_{ijk} = \mu + \alpha_i + \beta_j + (a\beta)_{ij} + \varepsilon_{ijk}$$

y_{ijk} represents the viscosity of replicate k at level i of pH and at level j of Catalyst Concentration $\quad i = 1, 2,\ j = 1, 2,\ k = 1, 2, 3$
α_i represents the treatment effect of pH at level i
β_j represents the treatment effect of Catalyst Concentration at level j
$(\alpha\beta)_{ij}$ represents the interaction effect and ε_{ijk} ~NID$(0, \sigma^2)$.

The hypotheses associated with this two-way ANOVA are:

$$\begin{aligned} H_0 &: \alpha_1 = \alpha_2 = ... = \alpha_a = 0 \\ H_A &: \text{at least one } \alpha_i \neq 0 \end{aligned}$$

$$\begin{aligned} H_0 &: \beta_1 = \beta_2 = ... = \beta_b = 0 \\ H_A &: \text{at least one } \beta_j \neq 0 \end{aligned}$$

$$\begin{aligned} H_0 &: (\alpha\beta)_{ij} = 0 \text{ for all } i, j \\ H_A &: \text{at least one } (\alpha\beta)_{ij} \neq 0 \end{aligned}$$

Before fitting the model, we can examine the relationship between Viscosity and pH and Viscosity and Catalyst Concentration (Fig. 5).

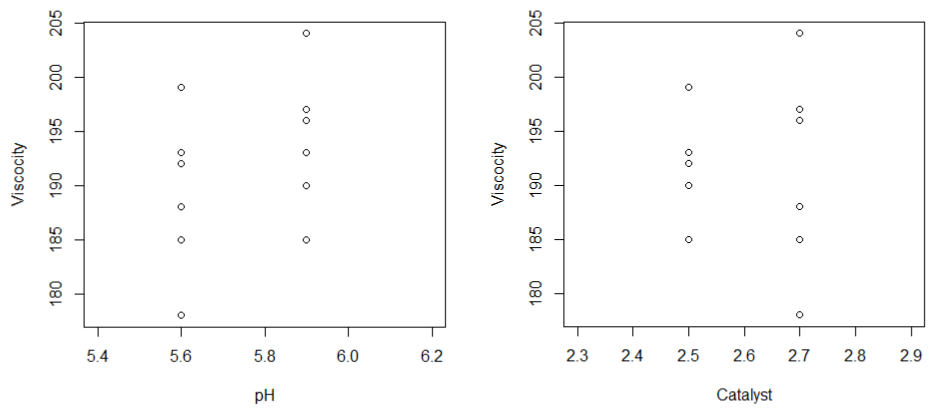

Fig. 5

We can also create interaction plots to check for an interaction visually (Fig. 6). To draw the interaction plot by hand we need to calculate the average viscosity for each run.

$$\begin{aligned}
\text{Factor } A \text{ 5.6 Factor } B \text{ 2.5} &= \frac{192+199+193}{3} = 194.67 \\
\text{Factor } A \text{ 5.9 Factor } B \text{ 2.5} &= \frac{185+193+190}{3} = 189.33 \\
\text{Factor } A \text{ 5.6 Factor } B \text{ 2.7} &= \frac{178+185+188}{3} = 183.67 \\
\text{Factor } A \text{ 5.9 Factor } B \text{ 2.7} &= \frac{197+196+204}{3} = 199
\end{aligned}$$

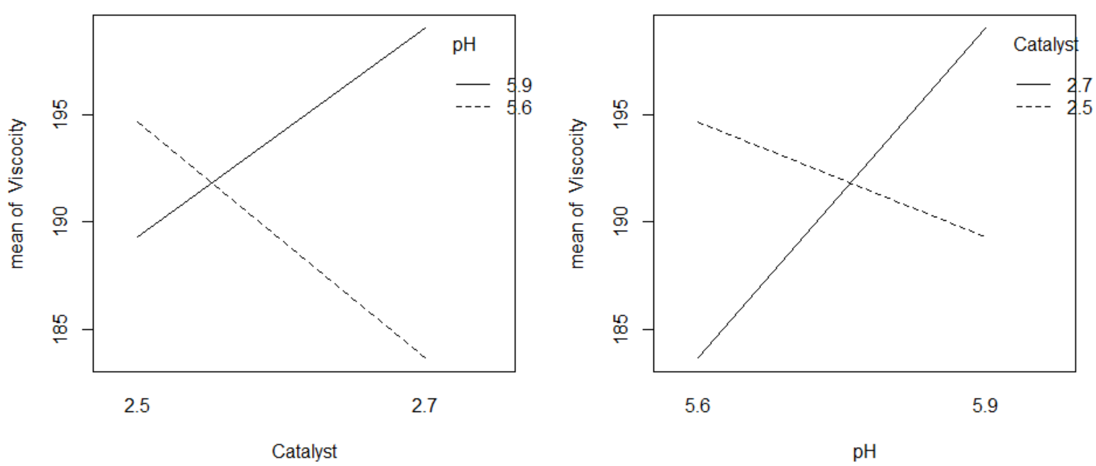

Fig. 6

If we fit a two-way ANOVA model on the Viscosity data using R, we obtain the following ANOVA table:

```
             Df   Sum Sq   Mean Sq   F value   Pr(>F)
Catalyst      1      1.3       1.3     0.070   0.79778
pH            1     75.0      75.0     3.947   0.08218 .
Catalyst:pH   1    320.3     320.3    16.860   0.00341 **
Residuals     8    152.0      19.0
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Interpretation In this example, the interaction term is significant at the 1% level but the main effects are not significant. Considering our results in the context of our hypotheses, we reject H_0 for the interaction term (p = 0.003) and conclude that the interaction term is significantly different to 0. If the interaction term is significant, interpretation of the main effects without considering the interaction is not meaningful. The significant interaction tells us that the effect that Catalyst Concentration has on Viscosity depends on the pH the experiment is run at (or vice versa). When pH is lower (5.6), an increase in Catalyst Concentration decreases the Viscosity but when pH is higher (5.9), an increase in Catalyst Concentration increases the Viscosity. Since the interaction term is significant, if we want to further explore the effects of pH and Catalyst Concentration on the Viscosity we need to examine each of the Factors, whilst holding the other Factor constant. For example, to investigate the effect of pH on the Viscosity we examine the effect of changing pH from 5.6 to 5.9 for the groups where Catalyst Concentration is 2.5 and again on the data where Catalyst Concentration is 2.7, this is referred to as analysing the **simple effects.** Comparisons of all pairwise group means can be examined using a post hoc test, for example, Tukey's Honest

Significant Difference (HSD). The output for the Viscosity example is shown below.

```
Tukey multiple comparisons of means
    95% family-wise confidence level

Fit: aov(formula = Viscosity ~ Catalyst * pH)

$Catalyst
              diff       lwr     upr     p adj
2.7-2.5 -0.6666667 -6.469983 5.13665 0.7977791

$pH
        diff        lwr      upr     p adj
5.9-5.6    5 -0.8033165 10.80332 0.0821793

$`Catalyst:pH`
                      diff        lwr        upr      p adj
2.7:5.6-2.5:5.6 -11.000000 -22.397254  0.3972543  0.0585163
2.5:5.9-2.5:5.6  -5.333333 -16.730588  6.0639210  0.4808069
2.7:5.9-2.5:5.6   4.333333  -7.063921 15.7305877  0.6337900
2.5:5.9-2.7:5.6   5.666667  -5.730588 17.0639210  0.4337337
2.7:5.9-2.7:5.6  15.333333   3.936079 26.7305877  0.0110745
2.7:5.9-2.5:5.9   9.666667  -1.730588 21.0639210  0.0993067
```

Main effects

- The difference between mean Viscosity measurements for the groups where Catalyst Concentration is 2.5 and 2.7 is -0.6667 (across all levels of pH), this result is not significant ($p = 0.798$).
- The difference between the mean Viscosity measurements for the groups where pH is 5.6 and 5.9 is 5 (across all levels of Catalyst Concentration), this result is not significant ($p = 0.082$).

Simple effects

- The difference between the mean Viscosity measurements for the groups where Catalyst Concentration is 2.5 and 2.7 is 11 (when pH is held at 5.6), this result is not significant ($p = 0.058$).
- The difference between the mean Viscosity measurements for the groups where Catalyst Concentration is 2.5 and 2.7 is 9.667 (when pH is held at 5.9), this result is not significant ($p = 0.099$).
- The difference between the mean Viscosity measurements for the groups where pH is 5.6 and 5.9 is -5.333 (when Catalyst Concentration is held at 2.5), this result is not significant ($p = 0.48$).

- The difference between the mean Viscosity measurements for the groups where pH is 5.6 and 5.9 is 15.333 (when Catalyst Concentration is held at 2.7), this result is significant (p = 0.011).

Remaining comparisons

- The difference between the mean Viscosity measurements for the treatment group with pH 5.6, Catalyst Concentration 2.5 and the treatment group with pH 5.9 and Catalyst concentration 2.7 is 4.333, this result is not significant (p = 0.634).
- The difference between the mean Viscosity measurements for the treatment group with pH 5.6, Catalyst Concentration 2.7 and the treatment group with pH 5.9 and Catalyst concentration 2.5 is 5.667, this result is not significant (p = 0.434).

The ANOVA model

Recall, the effects model for the two-way ANOVA is of the form:

$$y_{ijk} = \mu + \alpha_i + \beta_j + (a\beta)_{ij} + \varepsilon_{ijk}$$

y_{ijk} represents the viscosity of replicate k at level i and at level j of Catalyst Concentration
α_i represents the treatment effect of Factor A at level i
β_j represents the treatment effect of Factor B at level j
$(\alpha\beta)_{ij}$ represents the interaction effect and ε_{ijk} ~NID$(0, \sigma^2)$.

For a given data set:
$\widehat{\mu} = \overline{y}_{...}$
$\widehat{\alpha}_i = \overline{y}_{i..} - \overline{y}_{...}$
$\widehat{\beta}_j = \overline{y}_{.j.} - \overline{y}_{...}$
$\left(\widehat{a\beta}\right)_{ij} = \overline{y}_{ij.} - \overline{y}_{i..} - \overline{y}_{.j.} + \overline{y}_{...}$

For the Viscosity example:

- the overall mean, $\widehat{\mu}$ is 191.6667 the effect of pH on the Viscosity is α_i and this represents the difference between the treatment means and the overall mean.

- the mean for pH at 5.6 was $\overline{y}_{1..} = 189.1667$, therefore $\widehat{\alpha}_1 = 189.1667 - 191.6667 = -2.5$
- the mean for pH at 5.9 was $\overline{y}_{2..} = 194.1667$, therefore $\widehat{\alpha}_2 = 194.1667 - 191.6667 = 2.5$

The effect of Catalyst Concentration on the Viscosity is $\widehat{\beta}_j$ and this represents the difference between the treatment means and the overall mean.

- the mean for Catalyst Concentration at 2.5 was $\overline{y}_{.1.} = 192$, therefore $\widehat{\beta}_1 = 192 - 191.6667 = 0.3333$
- the mean for Catalyst Concentration at 2.7 was $\overline{y}_{.2.} = 191.3333$,therefore $\widehat{\beta}_2 = 191.3333 - 191.6667 = -0.3333$

The Interaction effect on the Viscosity $\left(\widehat{\alpha\beta}\right)_{ij}$ represents the difference between the treatment mean when levels i and j are the same and when levels i and j are different.

- For $i = j = 1$, pH = 5.6 and Catalyst Concentration = 2.5,the treatment mean, $\overline{y}_{11.}$ is 194.6667, therefore

$$\left(\widehat{\alpha\beta}\right)_{11} = 194.6667 - 189.1667 - 192 + 191.6667 = 5.1667$$

- For $i = 1$, $j = 2$, pH = 5.6 and Catalyst Concentration = 2.7,the treatment mean, $\overline{y}_{12.}$ is 183.6667

$$\left(\widehat{\alpha\beta}\right)_{12} = 183.6667 - 189.1667 - 191.3333 + 191.6667 = -5.1667$$

- For $i = 2$, $j = 1$, pH = 5.9 and Catalyst Concentration = 2.5,the treatment mean is, $\overline{y}_{21.}$ 189.3333

$$\left(\widehat{\alpha\beta}\right)_{21} = 189.3333 - 194.1667 - 192 + 191.6667 = -5.1667$$

- For $i = 2$, $j = 2$, pH = 5.9 and Catalyst Concentration = 2.7,the treatment mean, $\overline{y}_{22.}$ is 199

$$\left(\widehat{\alpha\beta}\right)_{22} = 199 - 194.1667 - 191.3333 + 191.6667 = 5.1667$$

Thus the fitted value when pH is 5.6 and Catalyst Concentration is 2.5 is:

$$\begin{aligned} y_{11k} &= \widehat{\mu} + \widehat{\alpha}_1 + \widehat{\beta}_1 + \left(\widehat{\alpha\beta}\right)_{11} + e_{11k} \\ y_{11k} &= 191.6667 - 2.5 + 0.3333 + 5.1667 + e_{11k} \\ &= 194.6667 + e_{11k} \end{aligned}$$

We can obtain a summary of the effects ANOVA model using R using the contrasts command (set to sum contrasts):

```
Coefficients:
               Estimate   Std. Error   t value   Pr(>|t|)
(Intercept)    191.6667       1.2583   152.321   3.86e-15 ***
Catalyst1        0.3333       1.2583     0.265   0.79778
pH1             -2.5000       1.2583    -1.987   0.08218 .
Catalyst1:pH1    5.1667       1.2583     4.106   0.00341 **
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

Residual standard error: 4.359 on 8 degrees of freedom
Multiple R-squared:  0.723,   Adjusted R-squared:  0.6191
F-statistic: 6.959 on 3 and 8 DF,  p-value: 0.01278
```

Effect Size

To measure the effect size associated with a two-way ANOVA, we can use η_P^2 (partial Eta squared).

$$\eta_P^2 = \frac{\mathbf{SS_F}}{\mathbf{SS_F} + \mathbf{SS_E}}$$

Partial eta squared measures the proportion of the total variance in the response variable that is associated with changes in the factor of interest. The total variance considered is the variance that remains after excluding variance accounted for by other factors.
For the Viscosity example:

$$\text{For pH} : \eta_P^2 = \frac{SS_A}{SS_A + SS_E} = \frac{75}{75 + 152} = 0.33 \text{ or } 33\%$$

We can say that 33% of the variation in Viscosity was caused by pH. For small sample sizes, η_P^2 can be biased and overestimate the proportion of variance for small sample sizes. An alternative measure is ω^2

$$\omega^2 = \frac{SS_F - d.f._F\,(MS_E)}{SS_T + MS_E}$$

For the Viscosity example:

$$\text{For pH} : \omega^2 = \frac{SS_A - d.f._A\,(MS_E)}{SS_T + MS_E} = \frac{75 - (1 \times 19)}{548.3 + 19} = 0.099 \text{ or } 9.9\%$$

Example - no interaction

An experiment was run to investigate how the type of glass and the type of phosphorescent coating affects the brightness of a light bulb. The response variable is the current (in microamps) to obtain a specified brightness.

Glass Type	**Phosphor Type**	**Current**	**Treatment Total**	**Treatment Mean**
1	1	$278, 291, 285$	$y_{11.} = 854$	$\overline{y}_{11.} = 284.6667$
1	2	$297, 304, 296$	$y_{12.} = 897$	$\overline{y}_{12.} = 299$
2	1	$229, 235, 241$	$y_{21.} = 705$	$\overline{y}_{21.} = 235$
2	2	$259, 249, 241$	$y_{22.} = 749$	$\overline{y}_{22.} = 249.6667$

To get a feel for the data we can summarise it across the different factors and write down the treatment means using the correct notation.

Phosphor Type

	1	2	$\sum$	Mean
1	$278, 291, 285$	$297, 304, 296$	$y_{1..} = 1751$	$\overline{y}_{1..} = 291.8333$
2	$229, 235, 241$	$259, 249, 241$	$y_{2..} = 1454$	$\overline{y}_{2..} = 242.3333$
$\sum$	$y_{.1.} = 1559$	$y_{.2.} = 1646$	$y_{...} = 6410$	
Mean	$\overline{y}_{.1.} = 259.833$	$\overline{y}_{.2.} = 274.3333$		$\overline{y}_{...} = 267.0833$

An interaction plot indicates that the there is no interaction between Glass Type and Phosphor Type (Fig. 7).

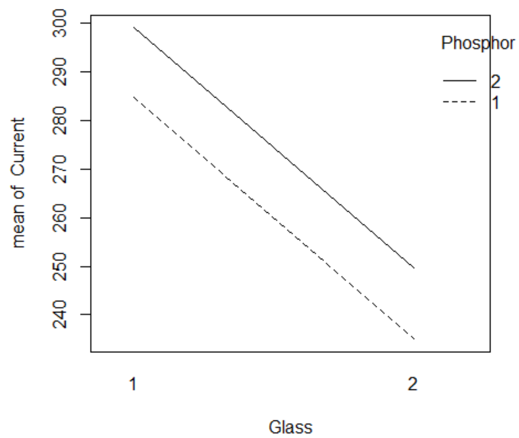

Fig. 7

Despite the plot, we start by fitting the full model including an interaction term:

$$y_{ijk} = \mu + \alpha_i + \beta_j + (a\beta)_{ij} + \varepsilon_{ijk}$$

The output obtained from R is shown below:

```
                Df  Sum Sq  Mean Sq   F value    Pr(>F)
Glass            1    7351     7351   164.569  1.29e-06 ***
Phosphor         1     631      631    14.121   0.00556 **
Glass:Phosphor   1       0        0     0.002   0.96661
Residuals        8     357       45
---
Signif. codes:  0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1
```

Since the interaction term is not significant (p = 0.96661) we can remove the interaction term from the model and rerun the analysis.

```
           Df  Sum Sq  Mean Sq  F value    Pr(>F)
Glass       1    7351     7351   185.10  2.63e-07 ***
Phosphor    1     631      631    15.88   0.00318 **
Residuals   9     357       40
```

Both Glass Type and Phosphor explained a significant amount of the variation in Current ($p < 0.001$ and $p = 0.003$ respectively). The output for Tukey's Honest Significant Difference (HSD) test is shown below (since there are just two levels for each factor this step is unnecessary to assess significance but does tell us the differences between the treatment means).

```
  Tukey multiple comparisons of means
    95% family-wise confidence level

Fit: aov(formula = Current ~ Glass + Phosphor)

$Glass
     diff       lwr       upr p adj
2-1 -49.5 -57.73054 -41.26946 3e-07

$Phosphor
    diff      lwr      upr     p adj
2-1 14.5 6.269461 22.73054 0.0031802
```

- The difference between the mean current for Glass type 1 and Glass type 2 is -49.5 (this effect is significant at $p < 0.001$)
- The difference between the mean current for Phosphor type 1 and Phosphor type 2 is 14.5 (this effect is significant at $p = 0.03$)

As in the case for a one way ANOVA, the following assumptions must hold:

- random sampling
- variances of different treatments are equal (homogeneity of variance)
- the error terms are independent from observation to observation and are normally distributed with zero mean and the same variance i.e. the error terms are i.i.d. random variables.