

MEAN SEPARATION TESTS (LSD AND Tukey's Procedure)

- If $H_0 = \mu_1 = \mu_2 = \dots \mu_n$ is rejected, we need a method to determine which means are significantly different from the others.
- We'll look at three separation tests during the semester:
 1. F-protected least significant difference (F-protected LSD)
 2. Tukey's Procedure
 3. Orthogonal linear contrasts (covered at the end of the semester)

F-protected Least Significant Difference

- The LSD we will use is called an F-protected LSD because it is calculated and used only when
- H_0 is rejected.
- Sometimes when one fails to reject H_0 and an LSD is calculated, the LSD will wrongly suggest that there are significant differences between treatment means.
- To prevent against this conflict, we calculate the LSD **only** when H_0 is rejected.

$$LSD = t_{\alpha/2} * s_{\bar{y}_1 - \bar{y}_2} \text{ and df for } t = \text{Error df}$$

$$\text{If } r_1 = r_2 \dots = r_n \text{ then } s_{\bar{y}_1 - \bar{y}_2} = \sqrt{\frac{2ErrorMS}{r}}$$

$$\text{If } r_i \neq r_{i'} \text{ then } s_{\bar{y}_1 - \bar{y}_2} = \sqrt{s^2 \left(\frac{1}{r_i} + \frac{1}{r_{i'}} \right)}$$

- If the difference between two treatment means is greater than the LSD, then those treatment means are significantly different at the $1 - \alpha\%$ level of confidence.

Example

Given an experiment analyzed as a CRD that has 7 treatments and 4 replicates with the following analysis

SOV	Df	SS	MS	F
Treatment	6	5,587,174	931,196	9.83**
Error	21	1,990,238	94,773	
Total	27	7,577,412		

and the following treatment means

Treatment	Mean
A	2,127
B	2,678
C	2,552
D	2,128
E	1,796
F	1,681
G	1,316

Calculate the LSD and show what means are significantly different at the 95% level of confidence.

Step 1. Calculate the LSD

$$\begin{aligned}LSD &= t_{\alpha/2} * s_{\bar{Y}_1 - \bar{Y}_2} \\&= 2.080 \sqrt{\frac{2ErrorMS}{r}} \\&= 2.080 \sqrt{\frac{2(94,773)}{4}} \\&= 452.8 \\&\cong 453\end{aligned}$$

Step 2. Rank treatment means from low to high

Treatment	Mean
G	1,316
F	1,681
E	1,796
A	2,127
D	2,128
C	2,552
B	2,678

Step 3. Calculate differences between treatment means to determine which ones are significantly different from each other.

If the difference between two treatment means is greater than the LSD, then those treatment means are significantly different at the 95% level of confidence.

$$\begin{aligned} \text{Treatment F vs. Treatment G} & \quad 1681 - 1316 = 365^{\text{ns}} \\ \text{Treatment E vs. Treatment G} & \quad 1796 - 1316 = 480^* \end{aligned}$$

Since E is significantly greater than Treatment G, then the rest of the means greater than that of Treatment E also are significantly different than Treatment G.

Thus there is no need to keep comparing the difference between the mean of Treatment G and Treatments with means greater than the mean of Treatment E.

$$\begin{aligned} \text{Treatment E vs. Treatment F} & \quad 1796 - 1681 = 115^{\text{ns}} \\ \text{Treatment A vs. Treatment F} & \quad 2127 - 1681 = 446^{\text{ns}} \\ \text{Treatment D vs. Treatment F} & \quad 2128 - 1681 = 447^{\text{ns}} \\ \text{Treatment C vs. Treatment F} & \quad 2552 - 1681 = 871^* \end{aligned}$$

*Therefore Treatment B must also be different from Treatment F

$$\begin{aligned} \text{Treatment A vs. Treatment E} & \quad 2127 - 1796 = 331^{\text{ns}} \\ \text{Treatment D vs. Treatment E} & \quad 2128 - 1796 = 332^{\text{ns}} \\ \text{Treatment C vs. Treatment E} & \quad 2552 - 1796 = 756^* \end{aligned}$$

*Therefore Treatment B must also be different from Treatment E

$$\begin{aligned} \text{Treatment D vs. Treatment A} & \quad 2128 - 2127 = 1^{\text{ns}} \\ \text{Treatment C vs. Treatment A} & \quad 2552 - 2127 = 425^{\text{ns}} \\ \text{Treatment B vs. Treatment A} & \quad 2678 - 2127 = 551^* \end{aligned}$$

F-protected LSD when $r_j \neq r_{j'}$

$$LSD = t_{.05/2; error df} \sqrt{S^2 \left(\frac{1}{r_j} + \frac{1}{r_{j'}} \right)}$$

Given:

SOV	Df	SS	MS	F
Treatment	3	0.978	0.326	6.392**
Error	13	0.660	0.051	
Total	16	1.638		

And

Treatment	n	Mean
A	5	2.0
B	3	1.7
C	5	2.4
D	4	2.1

How many LSD's do we need to calculate?

Step 1. Calculate the LSD's.

$$\text{LSD \#1) Treatment A or C vs. Treatment B: } 2.160 \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{3} \right)} = 0.356 \cong 0.4$$

$$\text{LSD \#2) Treatment A or C vs. Treatment D: } 2.160 \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{4} \right)} = 0.327 \cong 0.3$$

$$\text{LSD \#3) Treatment A vs. C: } 2.160 \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{5} \right)} = 0.309 \cong 0.3$$

$$\text{LSD \#4) Treatment B vs. D: } 2.160 \sqrt{0.051 \left(\frac{1}{3} + \frac{1}{4} \right)} = 0.373 \cong 0.4$$

Step 2. Write down the means in order from low to high.

Treatment	n	Mean
B	3	1.7
A	5	2.0
D	4	2.1
C	5	2.4

Step 3. Calculate differences between treatment means to determine which ones are significantly different from each other.

If the difference between two treatment means is greater than the LSD, then those treatment means are significantly different at the 95% level of confidence.

Treatment A vs. Treatment B (LSD #1) $2.0 - 1.7 = 0.3^{ns}$
 Treatment D vs. Treatment B (LSD #4) $2.1 - 1.7 = 0.4^{ns}$
 Treatment C vs. Treatment B (LSD #1) $2.4 - 1.7 = 0.7^*$

Treatment D vs. Treatment A (LSD #2) $2.1 - 2.0 = 0.1^{ns}$
 Treatment C vs. Treatment A (LSD #3) $2.4 - 2.0 = 0.4^*$

Treatment C vs. Treatment D (LSD #2) $2.4 - 2.1 = 0.3^{ns}$

Step 4. Place lowercase letters behind treatment means to show which treatments are significantly different.

Step 4.1. Write letters horizontally

B A D C

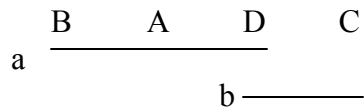
Step 4.2. Under line treatments that are not significantly different.

B A D C

Step 4.3. Ignore those lines that fall within the boundary of another line.

B A D C

Step 4.4 Label each line, beginning with the top one, with lowercase letters beginning with “a.”



Step 4.5 Add lowercase letters behind the respective means.

Treatment	n	Mean
B	3	1.7 a
A	5	2.0 a
D	4	2.1 ab
C	5	2.4 b

F-protected LSD with Sampling when $r_j s_k \neq r_j' s_{k'}$ or $r_j s_k = r_j' s_{k'}$

$$LSD = t_{.05/2, error df} \sqrt{s^2 \left(\frac{1}{r_j s_k} + \frac{1}{r_j' s_{k'}} \right)}$$

If $r_j s_k = r_j' s_{k'}$: $LSD = t_{.05/2, error df} \sqrt{\frac{2s^2}{rs}}$

Tukey’s Procedure

- This test takes into consideration the number of means involved in the comparison.
- Tukey’s procedure uses the distribution of the **studentized range statistic**.

$$q = \frac{\bar{y}_{\max} - \bar{y}_{\min}}{\sqrt{MS \text{ Error}/r}}$$

Where \bar{y}_{\max} and \bar{y}_{\min} are the largest and smallest treatment means, respectively, out of a group of **p** treatment means.

- Appendix Table VII, pages 621 and 622, contains values of $q_\alpha(p, f)$, the upper α percentage points of q where f is the number of degrees of freedom associated with the Mean Square Error.

VII. Percentage Points of the Studentized Range Statistic (continued)

f	$q_{0.05}(p, f)$																			
	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	
1	18.1	26.7	32.8	37.2	40.5	43.1	45.4	47.3	49.1	50.6	51.9	53.2	54.3	55.4	56.3	57.2	58.0	58.8	59.6	
2	6.09	8.28	9.80	10.89	11.73	12.43	13.03	13.54	13.99	14.39	14.75	15.08	15.38	15.65	15.91	16.14	16.36	16.57	16.77	
3	4.50	5.88	6.83	7.51	8.04	8.47	8.85	9.18	9.46	9.72	9.95	10.16	10.35	10.52	10.69	10.84	10.98	11.12	11.24	
4	3.93	5.00	5.76	6.31	6.73	7.06	7.35	7.60	7.83	8.03	8.21	8.37	8.52	8.67	8.80	8.92	9.03	9.14	9.24	
5	3.64	4.60	5.22	5.67	6.03	6.33	6.58	6.80	6.99	7.17	7.32	7.47	7.60	7.72	7.83	7.93	8.03	8.12	8.21	
6	3.46	4.34	4.90	5.31	5.63	5.89	6.12	6.32	6.49	6.65	6.79	6.92	7.04	7.14	7.24	7.34	7.43	7.51	7.59	
7	3.34	4.16	4.68	5.06	5.35	5.59	5.80	5.99	6.15	6.29	6.42	6.54	6.65	6.75	6.84	6.93	7.01	7.08	7.16	
8	3.26	4.04	4.53	4.89	5.17	5.40	5.60	5.77	5.92	6.05	6.18	6.29	6.39	6.48	6.57	6.65	6.73	6.80	6.87	
9	3.20	3.95	4.42	4.76	5.02	5.24	5.43	5.60	5.74	5.87	5.98	6.09	6.19	6.28	6.36	6.44	6.51	6.58	6.65	
10	3.15	3.88	4.33	4.66	4.91	5.12	5.30	5.46	5.60	5.72	5.83	5.93	6.03	6.12	6.20	6.27	6.34	6.41	6.47	
11	3.11	3.82	4.26	4.58	4.82	5.03	5.20	5.35	5.49	5.61	5.71	5.81	5.90	5.98	6.06	6.14	6.20	6.27	6.33	
12	3.08	3.77	4.20	4.51	4.75	4.95	5.12	5.27	5.40	5.51	5.61	5.71	5.80	5.88	5.95	6.02	6.09	6.15	6.21	
13	3.06	3.73	4.15	4.46	4.69	4.88	5.05	5.19	5.32	5.43	5.53	5.63	5.71	5.79	5.86	5.93	6.00	6.06	6.11	
14	3.03	3.70	4.11	4.41	4.64	4.83	4.99	5.13	5.25	5.36	5.46	5.56	5.64	5.72	5.79	5.86	5.92	5.98	6.03	
15	3.01	3.67	4.08	4.37	4.59	4.78	4.94	5.08	5.20	5.31	5.40	5.49	5.57	5.65	5.72	5.79	5.85	5.91	5.96	
16	3.00	3.65	4.05	4.34	4.56	4.74	4.90	5.03	5.15	5.26	5.35	5.44	5.52	5.59	5.66	5.73	5.79	5.84	5.90	
17	2.98	3.62	4.02	4.31	4.52	4.70	4.86	4.99	5.11	5.21	5.31	5.39	5.47	5.55	5.61	5.68	5.74	5.79	5.84	
18	2.97	3.61	4.00	4.28	4.49	4.67	4.83	4.96	5.07	5.17	5.27	5.35	5.43	5.50	5.57	5.63	5.69	5.74	5.79	
19	2.96	3.59	3.98	4.26	4.47	4.64	4.79	4.92	5.04	5.14	5.23	5.32	5.39	5.46	5.53	5.59	5.65	5.70	5.75	
20	2.95	3.58	3.96	4.24	4.45	4.62	4.77	4.90	5.01	5.11	5.20	5.28	5.36	5.43	5.50	5.56	5.61	5.66	5.71	
24	2.92	3.53	3.90	4.17	4.37	4.54	4.68	4.81	4.92	5.01	5.10	5.18	5.25	5.32	5.38	5.44	5.50	5.55	5.59	
30	2.89	3.48	3.84	4.11	4.30	4.46	4.60	4.72	4.83	4.92	5.00	5.08	5.15	5.21	5.27	5.33	5.38	5.43	5.48	
40	2.86	3.44	3.79	4.04	4.23	4.39	4.52	4.63	4.74	4.82	4.90	4.98	5.05	5.11	5.17	5.22	5.27	5.32	5.36	
60	2.83	3.40	3.74	3.98	4.16	4.31	4.44	4.55	4.65	4.73	4.81	4.88	4.94	5.00	5.06	5.11	5.15	5.20	5.24	
120	2.80	3.36	3.69	3.92	4.10	4.24	4.36	4.47	4.56	4.64	4.71	4.78	4.84	4.90	4.95	5.00	5.04	5.09	5.13	
∞	2.77	3.32	3.63	3.86	4.03	4.17	4.29	4.39	4.47	4.55	4.62	4.68	4.74	4.80	4.84	4.98	4.93	4.97	5.01	

- As the number of means involved in a comparison increases, the studentized range statistic increases.
- The basis behind Tukey's Procedure is that in general, as the number of means involved in a test increases, the smaller or less likely is the probability that they will be alike (i.e. the probability of detecting differences increases).
- Tukey's Procedure accounts for this fact by increasing the studentized range statistic as the number of treatments (p) increases, such that the probability that the means will be alike remains the same.
- If $r_i = r_j$, Tukey's statistic = $T_\alpha = q_\alpha(p, f) \sqrt{\frac{MS\ Error}{r}}$
- Two treatments means are considered significantly different if the difference between their means is greater than T_α .

Example (using the same data previously used for the LSD example)

Given an experiment analyzed as a CRD that has 7 treatments and 4 replicates with the following analysis

SOV	Df	SS	MS	F
Treatment	6	5,587,174	931,196	9.83**
Error	21	1,990,238	94,773	
Total	27	7,577,412		

and the following treatment means

Treatment	Mean
A	2,127
B	2,678
C	2,552
D	2,128
E	1,796
F	1,681
G	1,316

Calculate used Tukey's procedure to show what means are significantly different at the 95% level of confidence.

Step 1. Calculate Tukey's statistic.

$$T_{\alpha} = q_{\alpha}(p, f) \sqrt{\frac{MS \text{ Error}}{r}}$$

$$T_{0.05} = q_{0.05}(7, 21) \sqrt{\frac{94,773}{4}}$$

$$= 4.60 \sqrt{23,693.26}$$

$$= 708.06$$

$$\approx 708$$

Step 2. Rank treatment means from low to high

Treatment	Mean
G	1,316
F	1,681
E	1,796
A	2,127
D	2,128
C	2,552
B	2,678

Step 3. Calculate differences between treatment means to determine which ones are significantly different from each other.

If the difference between two treatment means is greater than T_{α} , then those treatment means are significantly different at the 95% level of confidence.

Treatment F vs. Treatment G	$1681 - 1316 = 365^{\text{ns}}$
Treatment E vs. Treatment G	$1796 - 1316 = 480^{\text{ns}}$
Treatment A vs. Treatment G	$2127 - 1316 = 811^*$

Since A is significantly greater than Treatment G, then the rest of the means greater than that of Treatment A also are significantly different than Treatment G.

Thus there is no need to keep comparing the difference between the mean of Treatment G and Treatments with means greater than the mean of Treatment A.

Treatment E vs. Treatment F	$1796 - 1681 = 115^{\text{ns}}$
Treatment A vs. Treatment F	$2127 - 1681 = 446^{\text{ns}}$
Treatment D vs. Treatment F	$2128 - 1681 = 447^{\text{ns}}$
Treatment C vs. Treatment F	$2552 - 1681 = 871^*$

*Therefore Treatment B must also be different from Treatment F

Treatment A vs. Treatment E	$2127 - 1796 = 331^{\text{ns}}$
Treatment D vs. Treatment E	$2128 - 1796 = 332^{\text{ns}}$
Treatment C vs. Treatment E	$2552 - 1796 = 756^*$

*Therefore Treatment B must also be different from Treatment E

Treatment D vs. Treatment A	$2128 - 2127 = 1^{\text{ns}}$
Treatment C vs. Treatment A	$2552 - 2127 = 425^{\text{ns}}$
Treatment B vs. Treatment A	$2678 - 2127 = 551^{\text{ns}}$

Step 4. Place lowercase letters behind treatment means to show which treatments are significantly different.

Step 4.1. Write letters horizontally

G F E A D C B

Step 4.2. Under line treatments that are not significantly different.

G F E A D C B

Step 4.3. Ignore those lines that fall within the boundary of another line.

G F E A D C B

Step 4.4 Label each line, beginning with the top one, with lowercase letters beginning with "a."

G F E A D C B
 _____ a
 _____ b
 _____ c

Step 4.5 Add lowercase letters behind the respective means.

Treatment	Mean
G	1,316 a
F	1,681 ab
E	1,796 ab
A	2,127 bc
D	2,128 bc
C	2,552 c
B	2,678 c

Tukey-Kramer Procedure

- Used for unbalanced data (i.e., $r_i \neq r_j$).

- $$T_\alpha = \frac{q_\alpha(p, f)}{\sqrt{2}} \sqrt{\text{Error MS} \left(\frac{1}{r_i} + \frac{1}{r_j} \right)}$$

Example

Given:

SOV	Df	SS	MS	F
Treatment	3	0.978	0.326	6.392**
Error	13	0.660	0.051	
Total	16	1.638		

And

Treatment	n	Mean
A	5	2.0
B	3	1.7
C	5	2.4
D	4	2.1

How many T_α values do we need to calculate?

Step 1. Calculate the T_α values.

$$\text{Where } T_\alpha = \frac{q_\alpha(p, f)}{\sqrt{2}} \sqrt{\text{Error MS} \left(\frac{1}{r_i} + \frac{1}{r_j} \right)}$$

$$\text{And } q_\alpha(p, f) = q_{0.05}(4, 13) = 4.15$$

$$T_\alpha \text{ #1) Treatment A or C vs. Treatment B: } \frac{4.15}{\sqrt{2}} \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{3} \right)} = 0.48 \cong 0.5$$

$$T_\alpha \text{ #2) Treatment A or C vs. Treatment D: } \frac{4.15}{\sqrt{2}} \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{4} \right)} = 0.445 \cong 0.4$$

$$T_\alpha \text{ #3) Treatment A vs. C: } \frac{4.15}{\sqrt{2}} \sqrt{0.051 \left(\frac{1}{5} + \frac{1}{5} \right)} = 0.415 \cong 0.4$$

$$T_\alpha \text{ #4) Treatment B vs. D: } \frac{4.15}{\sqrt{2}} \sqrt{0.051 \left(\frac{1}{3} + \frac{1}{4} \right)} = 0.508 \cong 0.5$$

Step 2. Write down the means in order from low to high.

Treatment	n	Mean
B	3	1.7
A	5	2.0
D	4	2.1
C	5	2.4

Step 3. Calculate differences between treatment means to determine which ones are significantly different from each other.

If the difference between two treatment means is greater than the T_α -value, then those treatment means are significantly different at the 95% level of confidence.

$$\text{Treatment A vs. Treatment B } (T_\alpha \text{ value \#1}) \quad 2.0 - 1.7 = 0.3^{\text{ns}}$$

$$\text{Treatment D vs. Treatment B } (T_\alpha \text{ value \#4}) \quad 2.1 - 1.7 = 0.4^{\text{ns}}$$

$$\text{Treatment C vs. Treatment B } (T_\alpha \text{ value \#1}) \quad 2.4 - 1.7 = 0.7^*$$

Treatment D vs. Treatment A (T_α value #2) $2.1 - 2.0 = 0.1^{ns}$
 Treatment C vs. Treatment A (T_α value #3) $2.4 - 2.0 = 0.4^{ns}$

Step 4. Place lowercase letters behind treatment means to show which treatments are significantly different.

Step 4.1. Write letters horizontally

B A D C

Step 4.2. Under line treatments that are not significantly different.

B A D C

Step 4.3. Ignore those lines that fall within the boundary of another line.

B A D C

Step 4.4 Label each line, beginning with the top one, with lowercase letters beginning with “a.”

a B A D C

 b _____

Step 4.5 Add lowercase letters behind the respective means.

Treatment	n	Mean
B	3	1.7 a
A	5	2.0 ab
D	4	2.1 ab
C	5	2.4 b

Tukey’s Procedure with Sampling

$$T_\alpha = q_\alpha(p, f) * s_{\bar{y}} \quad \text{where } s_{\bar{y}} = \sqrt{\frac{s^2}{rs}}$$

Tukey Kramer Procedure with Sampling

$$T_\alpha = \frac{q_\alpha(p, f)}{\sqrt{2}} \sqrt{s^2 \left(\frac{1}{r_j s_k} + \frac{1}{r_j s_{k'}} \right)}$$