# STAT 401A - Statistical Methods for Research Workers Two-way ANOVA

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This is also referred to as a full factorial or fully crossed design.

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For all of these questions, we want to know

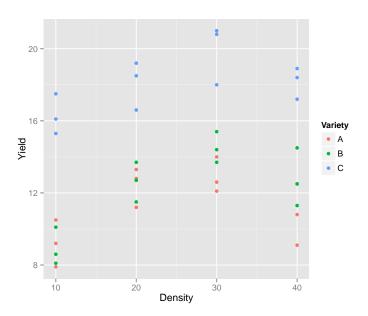
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- is there any effect and
- if yes, what is the nature of the effect.

Confidence intervals can answer these questions.



# Summary statistics

## Number of replicates

#### Mean Yield

```
Variety 10 20 30 40
1 A 9.20000 12.4333 12.90000 10.80000
2 B 8.93333 12.63333 14.50000 12.76667
3 C 16.300000 18.10000 19.93333 18.16667
```

## Standard deviation of yield

```
Variety 10 20 30 40

1 A 1.30000 1.096966 0.9848858 1.7000000

2 B 1.040833 1.101514 0.8544004 1.6165808

3 C 1.113553 1.345362 1.6772994 0.8736895
```

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	10	20	30	40
Α	$\mu_{11}$	$\mu_{12}$	$\mu_{13}$	$\mu_{14}$
В	$\mu_{21}$	$\mu_{22}$	$\mu_{23}$	$\mu_{24}$
С	$\mu_{31}$	$\mu_{32}$	$\mu_{33}$	$\mu_{34}$

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- The additive model:

$$\mu_i = \beta_0 + \beta_1 I(V_i = A) + \beta_2 I(V_i = B) + \beta_3 I(D_i = 10) + \beta_4 I(D_i = 20) + \beta_5 I(D_i = 30).$$

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The cell-means model:

$$\begin{split} \mu_i &= & \beta_0 + \beta_1 \mathrm{I}(V_i = A) + \beta_2 \mathrm{I}(V_i = B) \\ &+ \beta_3 \mathrm{I}(D_i = 10) + \beta_4 \mathrm{I}(D_i = 20) + \beta_5 \mathrm{I}(D_i = 30) \\ &+ \beta_6 \mathrm{I}(V_i = A) \mathrm{I}(D_i = 10) + \beta_7 \mathrm{I}(V_i = A) \mathrm{I}(D_i = 20) + \beta_8 \mathrm{I}(V_i = A) \mathrm{I}(D_i = 30) \\ &+ \beta_9 \mathrm{I}(V_i = B) \mathrm{I}(D_i = 10) + \beta_{10} \mathrm{I}(V_i = B) \mathrm{I}(D_i = 20) + \beta_{11} \mathrm{I}(V_i = B) \mathrm{I}(D_i = 30) \end{split}$$

 $eta_1$  is the expected difference in yield between varieties A and C at a density of 40

## **ANOVA Table**

ANOVA Table - Additive model

Source	SS	df	MS	F
Factor A	SSA	(I-1)	SSA/(I-1)	MSA/MSE
Factor B	SSB	(J-1)	SSB/(J-1)	MSB/MSE
Error	SSE	n-I-J+1	SSE/(n-I-J+1)	
Total	SST	n-1		

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Total	SST	n-1		

## ANOVA Table - Cell-means model

Source	SS	df	MS	
Factor A	SSA	I-1	SSA/(I-1)	MSA/MSE
Factor B	SSB	J-1	SSB/(J-1)	MSB/MSE
Interaction AB	SSAB	(I-1)(J-1)	SSAB /(I-1)(J-1)	MSAB/MSE
Error	SSE	n-IJ	$SSE/(n\text{-}\mathrm{IJ})$	
Total	SST	n-1		

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We will continue using the cell-means model to answer the scientific questions of interest.

# Two-way ANOVA using PROC GLM

```
DATA tomato;
  INFILE 'Ch13-tomato.csv' DSD FIRSTOBS=2;
  INPUT variety $ density yield;
PROC GLM DATA=tomato PLOTS=all;
  CLASS variety density;
  MODEL yield = variety|density / SOLUTION;
  LSMEANS variety / cl adjust=tukey;
  LSMEANS density / cl adjust=tukey;
  LSMEANS variety*density / cl adjust=tukey;
  RUN:
```

# Two-way ANOVA using PROC GLM

The GLM Procedure

Dependent Variable: yield

			Sun	ıof					
Source		DF	Squa	res	Mean	Square	F	Value	Pr > F
Model		11	422.3155	5556	38.3	923232		24.22	<.0001
Error		24	38.0400	0000	1.5	850000			
Corrected Total		35	460.3555	5556					
	R-Square	Coef	f Var	Root M	ISE	yield Me	an		
	0.917368	9.0	64568	1.2589	968	13.888	89		
Source		DF	Type 1	SS	Mean	Square	F	Value	Pr > F
variety		2	327.5972	2222	163.7	986111	1	103.34	<.0001
density		3	86.6866	667	28.8	955556		18.23	<.0001
variety*density		6	8.0316	6667	1.3	386111		0.84	0.5484
Source		DF	Type III	SS	Mean	Square	F	Value	Pr > F
variety		2	327.5972	2222	163.7	986111	1	103.34	<.0001
density		3	86.6866	6667	28.8	955556		18.23	<.0001
variety*density		6	8.0316	6667	1.3	386111		0.84	0.5484

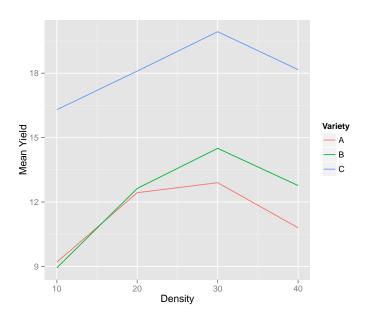
The Type I and Type III SS are equal because the design is balanced.

# Two-way ANOVA using PROC GLM

MODEL yield = variety|density / SOLUTION;

The GLM Procedure

Parameter		Estimate	Standard Error	t Value	Pr >  t
Intercept		18.16666667 B	0.72686542	24.99	<.0001
variety	A	-7.36666667 B	1.02794293	-7.17	<.0001
variety	В	-5.40000000 B	1.02794293	-5.25	<.0001
variety	C	0.00000000 B			
density	10	-1.8666667 B	1.02794293	-1.82	0.0819
density	20	-0.06666667 B	1.02794293	-0.06	0.9488
density	30	1.76666667 B	1.02794293	1.72	0.0986
density	40	0.00000000 B			
variety*density	A 10	0.26666667 B	1.45373083	0.18	0.8560
variety*density	A 20	1.70000000 B	1.45373083	1.17	0.2537
variety*density	A 30	0.3333333 B	1.45373083	0.23	0.8206
variety*density	A 40	0.00000000 B			
variety*density	B 10	-1.96666667 B	1.45373083	-1.35	0.1887
variety*density	B 20	-0.06666667 B	1.45373083	-0.05	0.9638
variety*density	B 30	-0.03333333 B	1.45373083	-0.02	0.9819
variety*density	B 40	0.00000000 B			
variety*density	C 10	0.00000000 B			



# Is the mean yield for variety A different from B on average?

```
LSMEANS variety / cl adjust=tukey;
                                      Least Squares Means
                          Adjustment for Multiple Comparisons: Tukey
                            Least Squares Means for effect variety
                             Pr > |t| for HO: LSMean(i)=LSMean(i)
                                  Dependent Variable: yield
                        i/j
                                                                     3
                                                  0.2249
                                                                < .0001
                                    0.2249
                                                                <.0001
                                    < .0001
                                                  <.0001
                                                  95% Confidence Limits
                     variety
                                yield LSMEAN
                                   11.333333
                                                   10.583245
                                                               12.083422
                                   12.208333
                                                   11.458245 12.958422
                                   18.125000
                                                   17.374912 18.875088
                             Least Squares Means for Effect variety
                                  Difference
                                                     Simultaneous 95%
                                                  Confidence Limits for
                                     Between
                                                   LSMean(i)-LSMean(j)
                                       Means
                                  -0.875000
                                                   -2.158534 0.408534
                                   -6.791667
                                                   -8.075201 -5.508132
                                   -5.916667
                                                   -7.200201 -4.633132
```

# Is the mean yield at density 10 different from density 20 on average?

```
LSMEANS density / cl adjust=tukey;
```

Least Squares Means

Adjustment for Multiple Comparisons: Tukey

density yield LSMEAN 95% Confidence Limits 10 11.477778 10.611650 12.343905 14.388889 13.522762 15.255016 20 15.777778 14.911650 16.643905 30 13.911111 14.777238 40 13.044984

Least Squares Means for Effect density Difference Simultaneous 95% Confidence Limits for Between LSMean(i)-LSMean(j) i Means -4.548299 -1.273923-2.911111 -4.300000 -5.937188 -2.662812 1 -2.433333 -4.070521 -0.796145 -1.388889 -3.026077 0.248299 0.477778 -1.159410 2.114966 1.866667 0.229479 3.503855

# Is mean yield different for particular combinations?

LSMEANS variety\*density / cl adjust=tukey;

variety	density	yield LSMEAN	95% Confiden	ce Limits
A	10	9.200000	7.699824	10.700176
A	20	12.433333	10.933157	13.933510
A	30	12.900000	11.399824	14.400176
A	40	10.800000	9.299824	12.300176
В	10	8.933333	7.433157	10.433510
В	20	12.633333	11.133157	14.133510
В	30	14.500000	12.999824	16.000176
В	40	12.766667	11.266490	14.266843
C	10	16.300000	14.799824	17.800176
C	20	18.100000	16.599824	19.600176
C	30	19.933333	18.433157	21.433510
C	40	18.166667	16.666490	19.666843

# Is mean yield different for particular combinations?

LSMEANS variety\*density / cl adjust=tukey;

Least Squares Means for Effect variety\*density

		Difference	Simultane	ous 95%
		Between	Between Confidence Limits 1	
i	j	Means	LSMean(i)-L	SMean(j)
1	2	-3.233333	-6.939704	0.473037
1	3	-3.700000	-7.406371	0.006371
1	4	-1.600000	-5.306371	2.106371
1	5	0.266667	-3.439704	3.973037
1	6	-3.433333	-7.139704	0.273037
1	7	-5.300000	-9.006371	-1.593629
1	8	-3.566667	-7.273037	0.139704
1	9	-7.100000	-10.806371	-3.393629
1	10	-8.900000	-12.606371	-5.193629
1	11	-10.733333	-14.439704	-7.026963
1	12	-8.966667	-12.673037	-5.260296
2	3	-0.466667	-4.173037	3.239704
2	4	1.633333	-2.073037	5.339704
2	5	3.500000	-0.206371	7.206371
2	6	-0.200000	-3.906371	3.506371
2	7	-2.066667	-5.773037	1.639704
2	8	-0.333333	-4.039704	3.373037
2	9	-3.866667	-7.573037	-0.160296
2	10	-5.666667	-9.373037	-1.960296
2	11	-7.500000	-11.206371	-3.793629
2	12	-5.733333	-9.439704	-2.026963
3	4	2.100000	-1.606371	5.806371
3	5	3.966667	0.260296	7.673037
3	6	0.266667	-3.439704	3.973037

```
tomato = read.csv("Ch13-tomato.csv")
tomato$Density = factor(tomato$Density)
m = lm(Yield~Variety*Density, tomato)
anova(m)
Analysis of Variance Table
Response: Yield
              Df Sum Sq Mean Sq F value Pr(>F)
Variety
              2 327.60 163.799 103.3430 1.608e-12 ***
              3 86.69 28.896 18.2306 2.212e-06 ***
Density
Variety:Density 6 8.03 1.339
                                  0.8445
                                           0.5484
Residuals
           24 38.04
                         1.585
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
lsmeans(m, pairwise~Variety)
$1smeans
Variety 1smean
                       SE df lower.CL upper.CL
        11.33333 0.3634327 24 10.58325 12.08342
        12.20833 0.3634327 24 11.45825 12.95842
        18.12500 0.3634327 24 17.37491 18.87509
Results are averaged over the levels of: Density
Confidence level used: 0.95
$contrasts
contrast estimate
                          SE df t.ratio p.value
 A - B -0.875000 0.5139715 24 -1.702 0.2249
A - C -6.791667 0.5139715 24 -13.214 <.0001
B - C -5.916667 0.5139715 24 -11.512 <.0001
Results are averaged over the levels of: Density
P value adjustment: tukev method for a family of 3 means
```

library(lsmeans)

```
lsmeans(m, pairwise~Density)
$1smeans
 Density 1smean SE df lower.CL upper.CL
 10
        11.47778 0.4196559 24 10.61165 12.34391
        14.38889 0.4196559 24 13.52276 15.25502
 20
 30
        15.77778 0.4196559 24 14.91165 16.64391
 40
        13 91111 0 4196559 24 13 04498 14 77724
Results are averaged over the levels of: Variety
Confidence level used: 0.95
$contrasts
                           SE df t.ratio p.value
 contrast
           estimate
 10 - 20 -2.9111111 0.5934831 24 -4.905 0.0003
 10 - 30 -4.3000000 0.5934831 24 -7.245 <.0001
 10 - 40 -2.4333333 0.5934831 24 -4.100 0.0022
 20 - 30 -1.3888889 0.5934831 24 -2.340 0.1169
 20 - 40 0.4777778 0.5934831 24 0.805 0.8514
 30 - 40 1 8666667 0 5934831 24 3 145 0 0213
```

Results are averaged over the levels of: Variety P value adjustment: tukey method for a family of 4 means

### lsmeans(m, pairwise~Variety\*Density)

### \$1smeans

Variety	Density	lsmean	SE	df	lower.CL	upper.CL
A	10	9.200000	0.7268654	24	7.699824	10.70018
В	10	8.933333	0.7268654	24	7.433157	10.43351
C	10	16.300000	0.7268654	24	14.799824	17.80018
A	20	12.433333	0.7268654	24	10.933157	13.93351
В	20	12.633333	0.7268654	24	11.133157	14.13351
C	20	18.100000	0.7268654	24	16.599824	19.60018
A	30	12.900000	0.7268654	24	11.399824	14.40018
В	30	14.500000	0.7268654	24	12.999824	16.00018
C	30	19.933333	0.7268654	24	18.433157	21.43351
A	40	10.800000	0.7268654	24	9.299824	12.30018
В	40	12.766667	0.7268654	24	11.266490	14.26684
C	40	18.166667	0.7268654	24	16.666490	19.66684

### Confidence level used: 0.95

```
$contrasts
contrast estimate
                              SE df t.ratio p.value
A.10 - B.10 0.26666667 1.027943 24
                                      0.259 1.0000
A.10 - C.10 -7.10000000 1.027943 24
                                     -6.907 <.0001
A,10 - A,20 -3.23333333 1.027943 24
                                     -3.145 0.1284
A.10 - B.20 -3.43333333 1.027943 24
                                     -3.340 0.0873
A.10 - C.20 -8.90000000 1.027943 24
                                     -8.658 <.0001
A,10 - A,30 -3.70000000 1.027943 24
                                     -3.599 0.0507
A,10 - B,30 -5.30000000 1.027943 24
                                     -5.156 0.0013
A.10 - C.30 -10.73333333 1.027943 24 -10.442 <.0001
A,10 - A,40 -1.60000000 1.027943 24 -1.557 0.9085
A,10 - B,40 -3.56666667 1.027943 24
                                     -3.470 0.0668
A.10 - C.40 -8.96666667 1.027943 24
                                     -8.723 <.0001
B.10 - C.10 -7.36666667 1.027943 24
                                     -7.166 <.0001
B,10 - A,20 -3.50000000 1.027943 24
                                     -3.405 0.0764
```

# Summary

• Use LSMEANS to answer questions of scientific interest.

## Summary

- Use LSMEANS to answer questions of scientific interest.
- Check model assumptions

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- Use LSMEANS to answer questions of scientific interest.
- Check model assumptions
- Consider alternative models, e.g. treating density as continuous