

# ST431 Split Plot Design: Honors Credit

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## Contents

<b>Lawson Problems</b>	<b>1</b>
8.3 Seafood Company: Nested Split Plot . . . . .	1
8.5 Film Performance . . . . .	6
Code Appendix: . . . . .	12

## Lawson Problems

### 8.3 Seafood Company: Nested Split Plot

Kuehl (2000) reports the results of an experiment conducted at a large seafood company to investigate the effect of storage temperature and type of seafood upon bacterial growth on oysters and mussels. Three storage temperatures were studied (0 C, 5 C, and 10 C). Three cold storage units were randomly assigned to be operated at each temperature. Within each storage unit, oysters and mussels were randomly assigned to be stored on one of the two shelves. The seafood was stored for two weeks at the assigned temperature, and at the end of the time the bacterial count was obtained from a sample on each shelf. The resulting data (log bacterial count) is shown below.

Storage Unit	Temp.	Oysters	Mussels
1	0	3.6882	0.3565
2	0	1.8275	1.7023
3	0	5.2327	4.5780
4	5	7.1950	5.0169
5	5	9.3224	7.9519
6	5	7.4195	6.3861
7	10	9.7842	10.1352
8	10	6.4703	5.0482
9	10	9.4442	11.0329

- (a) What is the experimental unit for temperature?

**Storage** unit is the experimental unit for temperature.

- (b) Why was it necessary to include 9 storage units instead of 3?

There are three temperature levels and we need replication because only one storage unit per temperature would mean any difference in bacterial growth could be due to the unit itself, not the temperature. By selecting 3 storage units PER temperature, we are able to separate the temperature effect from the variation among units.

- (c) What is the experimental unit for seafood type?

**Shelf** is the experimental unit for seafood type. Within each storage unit, oysters and mussels are assigned to separate shelves.

- (d) Write the model for the data.

Split-plot design:

- whole-plot factor: temperature (applied to storage units)

- sub-plot factor: seafood type (applied within each unit to separate shelves)

$$y_{ijk} = \mu + T_i + S_k + (TS)_{ik} + U_{j(i)} + \epsilon_{ijk}$$

$y_{ijk}$ : log bacterial count for seafood type  $k$ , in storage unit  $j$  at temperature level  $i$

$\mu$ : overall log bacterial count mean

$T_i$ : fixed effect of temperature  $i$  applied at the wholeplot level

$S_j$ : fixed effect of seafood type  $k$  applied at the subplot level

$(TS)_{ij}$ : interaction effect between temperature (Wholeplot) and seafood (subplot)

$U_{j(i)}$ : **random** effect of storage unit  $j$  nested within temperature  $i$

$\epsilon_{ijk}$ : **random** error at wholeplot level

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: log_bacteria ~ temperature * seafood + (1 | storage_unit)
## Data: seafoodData
##
## REML criterion at convergence: 52.2
##
## Scaled residuals:
##      Min       1Q   Median       3Q      Max
## -1.2064 -0.4364  0.1485  0.4012  0.9421
##
## Random effects:
## Groups      Name      Variance Std.Dev.
## storage_unit (Intercept) 3.2050  1.7902
## Residual          0.9318  0.9653
## Number of obs: 18, groups: storage_unit, 9
##
## Fixed effects:
##              Estimate Std. Error    df t value Pr(>|t|)
## (Intercept)      2.2123     1.1743   7.4989  1.884  0.09878 .
## temperature5      4.2394     1.6607   7.4989  2.553  0.03585 *
## temperature10     6.5265     1.6607   7.4989  3.930  0.00495 **
## seafoodOysters     1.3705     0.7882   6.0000  1.739  0.13271
## temperature5:seafoodOysters 0.1568     1.1146   6.0000  0.141  0.89273
## temperature10:seafoodOysters -1.5431     1.1146   6.0000 -1.384  0.21553
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

The coefficients for the model are displayed in the output above.

- (e) Analyze the data to determine what temperature and type of seafood have significant effects.

```
## Type III Analysis of Variance Table with Satterthwaite's method
##              Sum Sq Mean Sq NumDF DenDF F value  Pr(>F)
## temperature    13.6637  6.8318     2     6  7.3318 0.02448 *
## seafood         3.7137  3.7137     1     6  3.9855 0.09289 .
## temperature:seafood 2.6476  1.3238     2     6  1.4207 0.31254
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

```
## temperature seafood emmean SE df lower.CL upper.CL
## 0 Mussels 2.21 1.17 7.5 -0.527 4.95
## 5 Mussels 6.45 1.17 7.5 3.712 9.19
## 10 Mussels 8.74 1.17 7.5 5.999 11.48
## 0 Oysters 3.58 1.17 7.5 0.843 6.32
## 5 Oysters 7.98 1.17 7.5 5.239 10.72
## 10 Oysters 8.57 1.17 7.5 5.827 11.31
##
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
```

The interaction and seafood term are not significant based on the ANOVA output. However, since the question is asking what temp AND seafood have significant effects we consider both main effects in our emmeans output. Temperature 10 and mussels result in the highest emmean of 8.74.

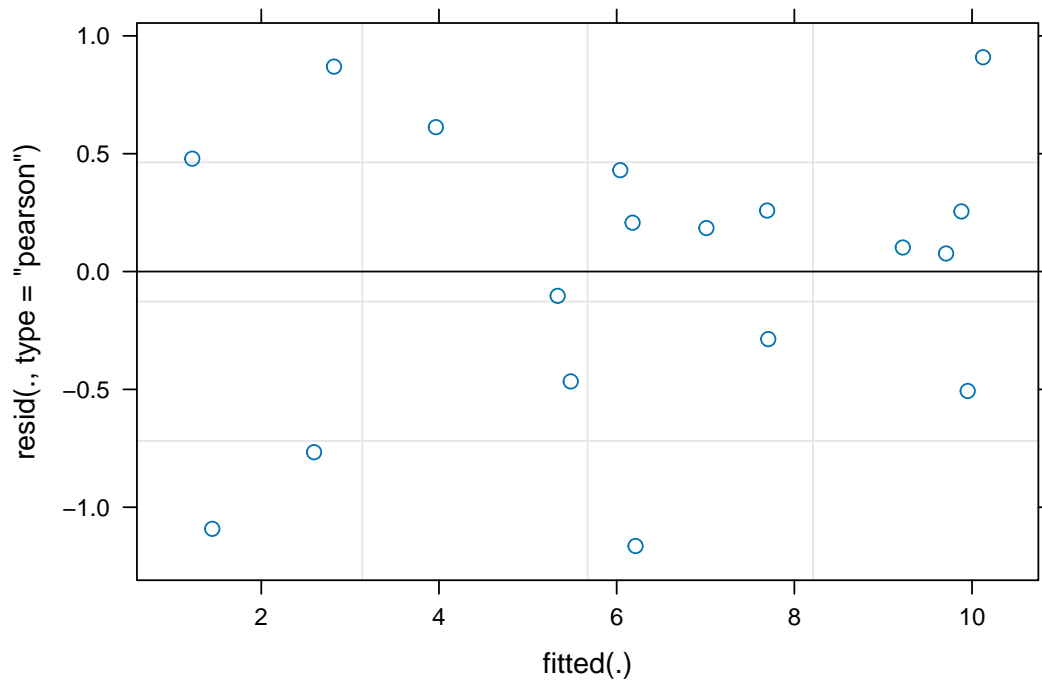
- (f) Interpret any significant effects.

```
## temperature emmean SE df lower.CL upper.CL
## 0 2.90 1.11 6 0.191 5.60
## 5 7.22 1.11 6 4.509 9.92
## 10 8.65 1.11 6 5.946 11.36
##
## Results are averaged over the levels of: seafood
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
```

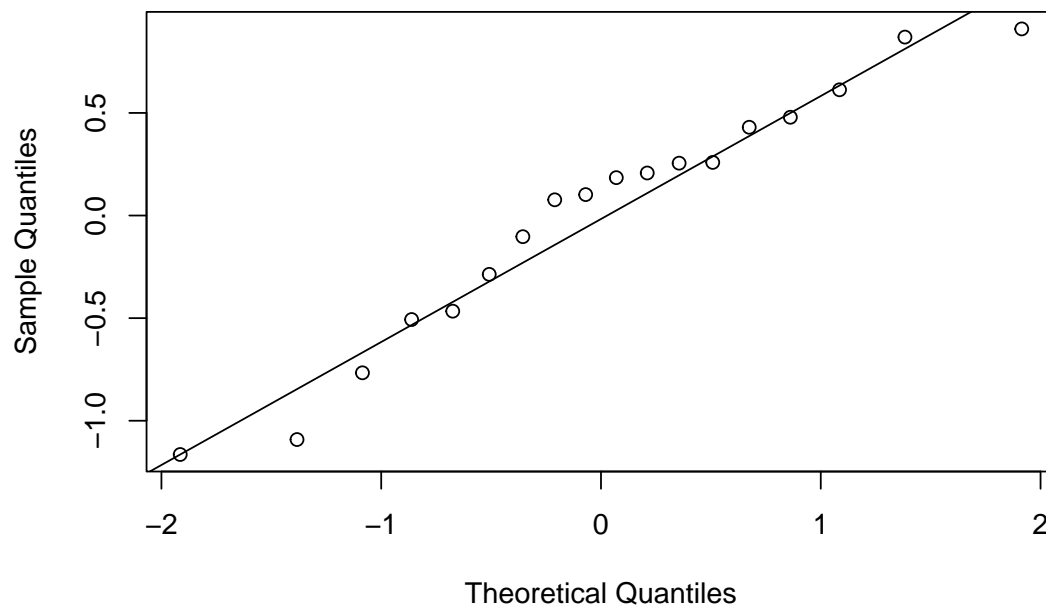
```
## seafood emmean SE df lower.CL upper.CL
## Mussels 5.80 0.678 7.5 4.22 7.38
## Oysters 6.71 0.678 7.5 5.13 8.29
##
## Results are averaged over the levels of: temperature
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
```

As the main effect temperature increases from 0, 5, 10, the emmean increases as well. Indicating a higher temperature results in a higher log bacterial count. For the seafood main effect, oysters have a higher emmean than mussels, indicating oysters generally result in a higher log bacterial count.

- (g) Check the assumptions of the model you used for analysis.



**Normal Q-Q Plot**



The fitted vs residual plot is generally equally dispersed above and below the horizontal line. **Linearity and homoscedasticity** (constant variance) assumptions of the model **are met**. The QQ plot tests for normality of the errors. In the QQ plot there is slightly heavy tails in the model, however since this is a smaller sample size slightly heavy tails aren't problematic, especially when the homoscedasticity and

linearity assumptions are met. Since the rest of the points generally follow the line, the **normality assumptions of errors are met**.

## 8.5 Film Performance

Ramirez and Tobias (2007) present the data shown below from an experiment to compare performance, based on a quality characteristic, of a new and old film type and three manufacturing pressures. One film type was selected at random and it was run through the manufacturing process randomly assigning each pressure to 1/3-rd of the roll. Quality measurements were made on each third of the roll. Next, the other film type was run through the process again randomly assigning each pressure to 1/3-rd of the roll. This two-step experimental process was repeated eight different times.

Time Block	Film Type	P1	P2	P3
1	Old	15.39	15.49	15.39
1	New	15.40	15.62	15.14
2	Old	15.97	15.79	14.99
2	New	15.25	15.37	15.55
3	Old	15.88	15.91	15.48
3	New	15.92	15.26	15.43
4	Old	15.36	15.51	15.47
4	New	15.30	15.53	15.66
5	Old	15.86	15.19	14.93
5	New	15.42	15.03	15.26
6	Old	15.53	15.61	15.49
6	New	15.32	15.55	15.50
7	Old	15.91	16.06	15.53
7	New	15.75	15.54	15.68
8	Old	16.03	15.55	15.49
8	New	15.75	15.31	15.62

- (a) Was this a completely randomized or randomized block design in the whole plots?

This is a **\*randomized block design at the whole-plot level**. Each time block (1 to 8) is a block. Within each time block, both Old and New film types are run. Meaning, film type is randomly assigned within each block. Making it a randomized complete block design for the whole plot factor.

- (b) What is the model for the data?

Split-plot design:

- whole-plot factor: temperature (applied to storage units)

- sub-plot factor: seafood type (applied within each unit to separate shelves)

$$\text{Mixed Effects Model: } y_{ijk} = \mu + F_i + P_j + (FP)_{ij} + B_k + (FB)_{ik} + \epsilon_{ijk}$$

$y_{ijk}$  = quality measurement for film type  $i$ , pressure  $j$ , block  $k$

$\mu$  = overall baseline mean

$F_i$  = fixed effect of film type

$P_j$  = fixed effect of pressure

$(FP)_{ij}$  = interaction of film type and pressure

$B_k$  = **random** effect of block (time)

$FB_{ik}$  = **random** whole-plot error (block x film)

$\epsilon_{ijk}$  = **random** residual error at the subplot level

```
## Linear mixed model fit by REML. t-tests use Satterthwaite's method [
## lmerModLmerTest]
## Formula: response ~ film * pressure + (1 | block) + (1 | block:film)
## Data: filmData
##
## REML criterion at convergence: 7.7
##
## Scaled residuals:
##      Min       1Q   Median       3Q      Max
## -1.6732 -0.7949  0.1622  0.7583  1.5445
##
## Random effects:
## Groups      Name                Variance Std.Dev.
## block:film (Intercept) 0.0000    0.0000
## block      (Intercept) 0.0144    0.1200
## Residual                    0.0435    0.2086
## Number of obs: 48, groups:  block:film, 16; block, 8
##
## Fixed effects:
##              Estimate Std. Error    df t value Pr(>|t|)
## (Intercept)   15.51375   0.08507 32.07592 182.356 <2e-16 ***
## filmOld        0.22750   0.10428 35.00000   2.182  0.0359 *
## pressureP2    -0.11250   0.10428 35.00000  -1.079  0.2880
## pressureP3    -0.03375   0.10428 35.00000  -0.324  0.7481
## filmOld:pressureP2 0.01000   0.14747 35.00000   0.068  0.9463
## filmOld:pressureP3 -0.36125   0.14747 35.00000  -2.450  0.0194 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
## optimizer (nloptwrap) convergence code: 0 (OK)
## boundary (singular) fit: see help('isSingular')
```

The coefficients for the model are displayed in the output above.

- (c) Analyze the data to determine if type or pressure have any effect on the quality characteristic measured.

```
## Type III Analysis of Variance Table with Satterthwaite's method
##           Sum Sq Mean Sq NumDF DenDF F value  Pr(>F)
## film      0.14630 0.14630     1    35  3.3634 0.07517 .
## pressure  0.36765 0.18383     2    35  4.2261 0.02270 *
## film:pressure 0.35790 0.17895     2    35  4.1140 0.02485 *
## ---
## Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```

Based on the ANOVA output of the model above, film type is not significant but pressure and film:pressure are significant.

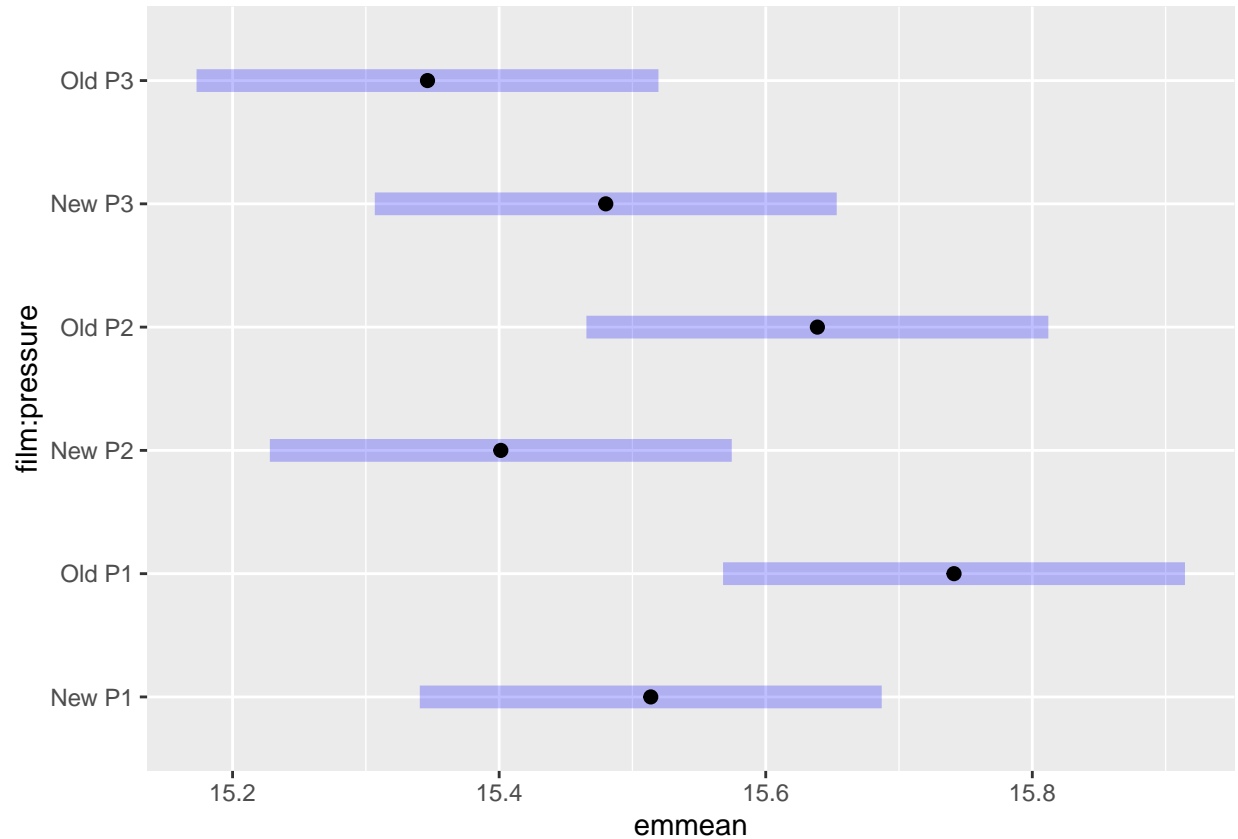
- (d) Describe any significant differences you find and interpret what these differences mean by referring to tables or graphs of means or multiple comparison tests.

```
## contrast estimate      SE df t.ratio p.value
## New - Old      -0.11 0.0602  7  -1.834  0.1093
##
## Results are averaged over the levels of: pressure
## Degrees-of-freedom method: kenward-roger
```

```
## contrast estimate      SE df t.ratio p.value
## P1 - P2        0.107 0.0737 28   1.458  0.3261
## P1 - P3        0.214 0.0737 28   2.907  0.0187
## P2 - P3        0.107 0.0737 28   1.449  0.3301
##
## Results are averaged over the levels of: film
## Degrees-of-freedom method: kenward-roger
## P value adjustment: tukey method for comparing a family of 3 estimates
```

```
## contrast      estimate      SE    df t.ratio p.value
## New P1 - Old P1 -0.2275 0.104 31.5  -2.182  0.2744
## New P1 - New P2  0.1125 0.104 28.0   1.079  0.8856
## New P1 - Old P2 -0.1250 0.104 31.5  -1.199  0.8341
## New P1 - New P3  0.0338 0.104 28.0   0.324  0.9995
## New P1 - Old P3  0.1675 0.104 31.5   1.606  0.6007
## Old P1 - New P2  0.3400 0.104 31.5   3.260  0.0292
## Old P1 - Old P2  0.1025 0.104 28.0   0.983  0.9196
## Old P1 - New P3  0.2612 0.104 31.5   2.505  0.1531
## Old P1 - Old P3  0.3950 0.104 28.0   3.788  0.0087
## New P2 - Old P2 -0.2375 0.104 31.5  -2.278  0.2331
## New P2 - New P3 -0.0788 0.104 28.0  -0.755  0.9727
## New P2 - Old P3  0.0550 0.104 31.5   0.527  0.9946
## Old P2 - New P3  0.1588 0.104 31.5   1.522  0.6530
## Old P2 - Old P3  0.2925 0.104 28.0   2.805  0.0861
## New P3 - Old P3  0.1338 0.104 31.5   1.283  0.7920
##
## Degrees-of-freedom method: kenward-roger
## P value adjustment: tukey method for comparing a family of 6 estimates
```





While all combinations fall in a fairly narrow range (15.3–15.7), there is some evidence that:

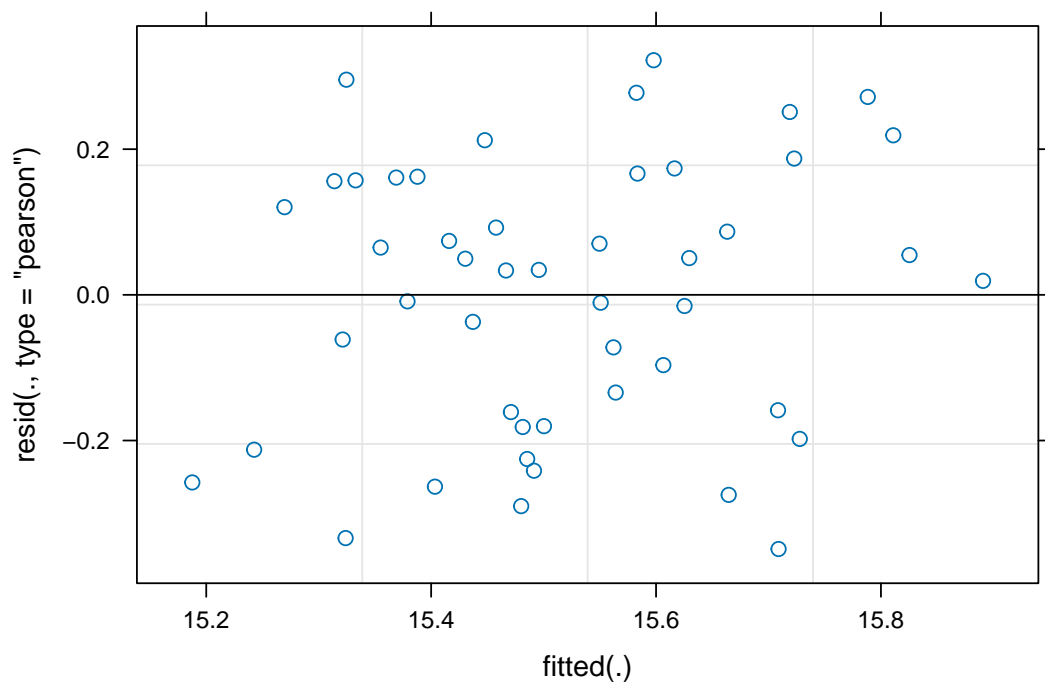
- P1 (especially New P1) may give slightly better film quality.
- P3 (especially Old P3) may yield lower quality.

No strong interaction appears. The order of best to worst is similar for both films.

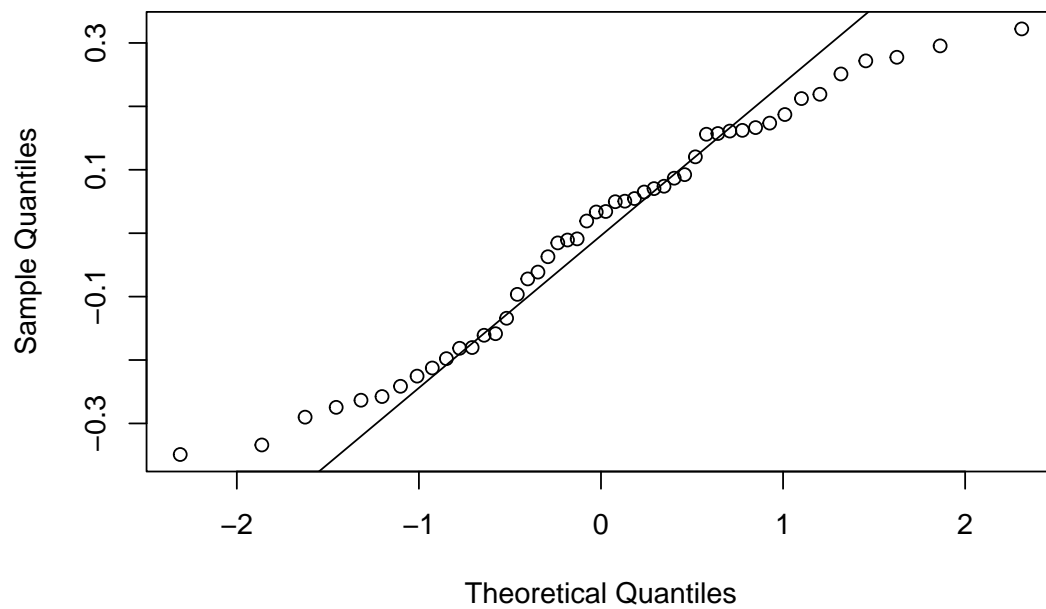
The differences are small but potentially meaningful for optimization.

Based on the estimated marginal means, film pressure level P1, particularly with the New film type, results in slightly higher average film quality than other combinations. The differences across film-pressure combinations are not large, and most confidence intervals overlap, indicating no strong interaction between film and pressure. However, Old P3 appears to perform slightly worse, suggesting that this combination may be less optimal.

- (e) Check the assumptions of equal variance and normality of the whole-plot and split-plot error terms as described in Section 5.9.



**Normal Q-Q Plot**



The fitted vs residual plot points are equally dispersed above and below the horizontal line. **Linearity and homoscedasticity** (constant variance) assumptions of the model **are met**. In the QQ plot there are slightly heavy tails in the model, however since this is a smaller sample size these tails aren't problematic, especially when the homoscedasticity and linearity assumptions are met. Since the rest of the points

generally follow the line, the **normality assumptions of errors are met**.

## Code Appendix:

```
knitr::opts_chunk$set(echo = F,
warning = F,
message = F)
knitr::include_graphics("C:\\Users\\19106\\Pictures\\Screenshots\\ST431SplitPlot8.3.png")
library(lmerTest)
seafoodData <- data.frame(
  storage_unit = rep(1:9, each = 2),
  temperature = rep(c(0, 0, 0, 5, 5, 5, 10, 10, 10), each = 2),
  seafood = rep(c("Oysters", "Mussels"), times = 9),
  log_bacteria = c(
    3.6882, 0.3565,
    1.8275, 1.7023,
    5.2327, 4.5780,
    7.1950, 5.0169,
    9.3224, 7.9519,
    7.4195, 6.3861,
    9.7842, 10.1352,
    6.4703, 5.0482,
    9.4442, 11.0329
  )
)
seafoodData$temperature <- factor(seafoodData$temperature)
seafoodData$seafood <- factor(seafoodData$seafood)

bacteriaModel <- lmer(log_bacteria ~ temperature * seafood + (1 | storage_unit), data = seafoodData)
summary(bacteriaModel, correlation=F)
library(emmeans)
anova(bacteriaModel)

# Marginal means for model
emmeans(bacteriaModel, ~ temperature + seafood)
emmeans(bacteriaModel, ~ temperature)
emmeans(bacteriaModel, ~ seafood)
plot(bacteriaModel) # default plot for lmer gives residuals vs fitted

qqnorm(resid(bacteriaModel)) #QQ plot
qqline(resid(bacteriaModel))
knitr::include_graphics("C:\\Users\\19106\\Pictures\\Screenshots\\ST431SplitPlot8.5.png")
library(lme4)
library(lmerTest)

# Create the dataset manually
block <- rep(1:8, each = 6)
film <- rep(c("Old", "New"), each = 3, times = 8)
pressure <- rep(c("P1", "P2", "P3"), times = 2 * 8)
response <- c(
  15.39, 15.49, 15.39, 15.40, 15.62, 15.14,
  15.97, 15.79, 14.99, 15.25, 15.37, 15.55,
  15.88, 15.91, 15.48, 15.92, 15.26, 15.43,
  15.36, 15.51, 15.47, 15.30, 15.53, 15.66,
  15.86, 15.19, 14.93, 15.42, 15.03, 15.26,
```

```

15.53, 15.61, 15.49, 15.32, 15.55, 15.50,
15.91, 16.06, 15.53, 15.75, 15.54, 15.68,
16.03, 15.55, 15.49, 15.75, 15.31, 15.62
)
filmData <- data.frame(
  block = factor(block),
  film = factor(film),
  pressure = factor(pressure),
  response = response
)
# Fit model with film and pressure as fixed effects, block and block:film as random
filmModel <- lmer(response ~ film * pressure + (1 | block) + (1 | block:film), data = filmData)

summary(filmModel, correlation=F)
library(lme4)
library(lmerTest)

# ANOVA to test fixed effects
anova(filmModel)
library(emmeans)

# Get marginal means
emm_film <- emmeans(filmModel, ~ film)
emm_pressure <- emmeans(filmModel, ~ pressure)
emm_interaction <- emmeans(filmModel, ~ film * pressure)

# Pairwise comparisons
pairs(emm_film)
pairs(emm_pressure)
pairs(emm_interaction)

# Plot means
plot(emm_interaction)
# Residuals vs fitted values (equal variance)
plot(filmModel)

# QQ plot (normality)
qqnorm(resid(filmModel))
qqline(resid(filmModel))

```