Statistical analysis

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Data table

```
setDT(isumm)
```

Trials 1-4 (treatment effects)

Subset.

```
isumm1 <- isumm[new.ID %in% as.character(1:4)]</pre>
```

Separate digestate type from treatment

```
isumm1[, dig.ID := substr(treat1, 1, 1)]
isumm1[, dig.treat := gsub('^[ABC]', '', treat1)]
```

Fit mixed-effects model.

```
m1 <- lmer(e.rel.150 ~ dig.treat + (1|dig.ID), data = isumm1)
m2 <- lmer(e.rel.150 ~ dig.treat + (1|dig.ID/dig.treat), data = isumm1)</pre>
```

```
summary(m1)
```

```
## Linear mixed model fit by REML ['lmerMod']
## Formula: e.rel.150 ~ dig.treat + (1 | dig.ID)
     Data: isumm1
##
##
## REML criterion at convergence: -115.8
##
## Scaled residuals:
              1Q Median
##
      Min
                               3Q
                                      Max
## -1.7502 -0.6560 0.1046 0.4727 2.5661
##
## Random effects:
## Groups Name
                        Variance Std.Dev.
## dig.ID (Intercept) 0.010730 0.10359
                       0.003904 0.06248
## Residual
```

```
## Number of obs: 61, groups: dig.ID, 3
##
## Fixed effects:
##
                        Estimate Std. Error t value
## (Intercept)
                         0.43276
                                    0.06161
                                              7.024
## dig.treat Acid
                        -0.09302
                                    0.02951 - 3.152
## dig.treat Dis
                        -0.01167
                                    0.03943 - 0.296
## dig.treat Dis + acid -0.08434
                                    0.03943 - 2.139
## dig.treat MF liquid
                        -0.31010
                                    0.03943 -7.864
## dig.treat MF slurry
                         0.07933
                                    0.03943 2.012
## dig.treat Sep-D
                         -0.30184
                                    0.02609 -11.570
                                    0.03943 -8.427
## dig.treat Sep-D + acid -0.33230
                        -0.12468
## dig.treat Sep-S
                                    0.02769 -4.503
                                    0.04179 -4.922
## dig.treat Sep-S + acid -0.20570
##
## Correlation of Fixed Effects:
##
              (Intr) dg.trA dg.trD dg.D+a dg.MFl dg.MFs dg.S-D d.S-D+a dg.S-S
## dig.tretAcd -0.095
## dig.treatDs -0.062 0.148
## dg.trtDs+ac -0.062 0.148
                            0.163
## dg.trtMFlqd -0.062 0.148 0.163 0.163
## dg.trtMFslr -0.062 0.148 0.163 0.163 0.163
## dig.trtSp-D -0.127  0.182  0.198  0.198  0.198  0.198
## dg.trSp-D+a -0.062 0.148 0.163 0.163 0.163 0.163 0.198
## dig.trtSp-S -0.099 0.261 0.168 0.168 0.168 0.168 0.207 0.168
## dg.trSp-S+a -0.076 0.217 0.055 0.055 0.055 0.055 0.070 0.055
                                                                      0.210
summary(m2)
## Linear mixed model fit by REML ['lmerMod']
## Formula: e.rel.150 ~ dig.treat + (1 | dig.ID/dig.treat)
     Data: isumm1
##
## REML criterion at convergence: -122.4
##
## Scaled residuals:
##
      Min
           1Q Median
                              3Q
                                     Max
## -2.0310 -0.5222 0.1171 0.4283 2.5808
##
## Random effects:
## Groups
                    Name
                               Variance Std.Dev.
## dig.treat:dig.ID (Intercept) 0.003286 0.05733
## dig.ID
                    (Intercept) 0.011537 0.10741
## Residual
                                0.003057 0.05529
## Number of obs: 61, groups: dig.treat:dig.ID, 15; dig.ID, 3
## Fixed effects:
##
                          Estimate Std. Error t value
## (Intercept)
                         0.420186 0.071655
                                             5.864
                         -0.076441 0.060889 -1.255
## dig.treat Acid
## dig.treat Dis
                         0.006601 0.079240
                                              0.083
## dig.treat Dis + acid -0.066065 0.079240 -0.834
## dig.treat MF liquid -0.291832 0.079240 -3.683
                                              1.232
## dig.treat MF slurry
                        0.097601
                                   0.079240
```

```
## dig.treat Sep-D
                        -0.281026 0.059784 -4.701
## dig.treat Sep-D + acid -0.314032  0.079240 -3.963
## dig.treat Sep-S
                        -0.110120 0.060313 -1.826
## dig.treat Sep-S + acid -0.190816 0.081197 -2.350
## Correlation of Fixed Effects:
              (Intr) dg.trA dg.trD dg.D+a dg.MFl dg.MFs dg.S-D d.S-D+a dg.S-S
## dig.tretAcd -0.288
## dig.treatDs -0.220 0.306
## dg.trtDs+ac -0.220 0.306
                           0.314
## dg.trtMFlqd -0.220 0.306 0.314 0.314
## dg.trtMFslr -0.220 0.306
                           0.314 0.314 0.314
## dig.trtSp-D -0.303 0.300 0.300 0.300 0.300 0.300
## dg.trSp-D+a -0.220 0.306 0.314 0.314 0.314 0.314 0.300
## dig.trtSp-S -0.291 0.423 0.312 0.312 0.312 0.312 0.306 0.312
## dg.trSp-S+a -0.217 0.330 0.152 0.152 0.152 0.152 0.157 0.152
```

Check for interaction with odds ratio test (?).

```
anova(m2, m1, test = 'Chisq')
```

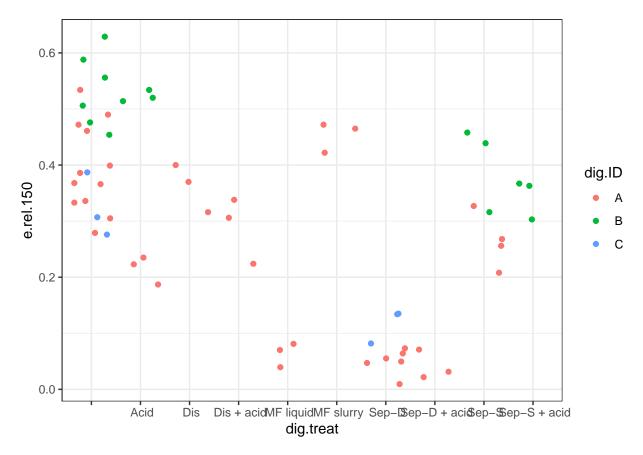
```
## refitting model(s) with ML (instead of REML)

## Data: isumm1
## Models:
## m1: e.rel.150 ~ dig.treat + (1 | dig.ID)
## m2: e.rel.150 ~ dig.treat + (1 | dig.ID/dig.treat)
## mpar AIC BIC logLik deviance Chisq Df Pr(>Chisq)
## m1 12 -140.87 -115.54 82.436 -164.87
## m2 13 -139.54 -112.10 82.769 -165.54 0.6652 1 0.4147
```

No evidence, good!

Plot to check.

```
ggplot(isumm1, aes(dig.treat, e.rel.150, colour = dig.ID)) +
geom_jitter(height = 0) +
theme_bw()
```



Hmm.

```
table(isumm1[, .(dig.ID, dig.treat)])
```

```
##
         dig.treat
              Acid Dis Dis + acid MF liquid MF slurry Sep-D Sep-D + acid
## dig.ID
##
        A 12
                 3
                      3
                                  3
                                             3
                                                        3
                                                               6
                                                                             3
                      0
                                  0
                                             0
       B 6
                 3
                                                        0
                                                               0
                                                                             0
##
       С 3
##
                 0
                                                               3
                                                                             0
##
        dig.treat
## dig.ID Sep-S Sep-S + acid
##
        Α
               4
                             3
##
       В
               3
        С
               0
                             0
##
```

Not close to balanced.

Marginal means.

```
mmeans <- emmeans(m1, 'dig.treat')
mmeans</pre>
```

```
dig.treat
                                  df lower.CL upper.CL
##
                  emmean
                             SE
##
                   0.433 0.0616 2.08
                                       0.1772
                                                 0.688
##
     Acid
                   0.340 0.0658 2.69
                                       0.1159
                                                 0.564
##
    Dis
                  0.421 0.0711 3.66
                                       0.2161
                                                 0.626
```

```
##
    Dis + acid
                  0.348 0.0711 3.66 0.1434
                                               0.553
##
    MF liquid 0.123 0.0711 3.66 -0.0823
                                               0.328
    MF slurry
                                               0.717
##
                 0.512 0.0711 3.66 0.3071
##
                  0.131 0.0638 2.39 -0.1049
    Sep-D
                                               0.367
##
    Sep-D + acid 0.100 0.0711 3.66 -0.1045
                                               0.305
##
    Sep-S
                  0.308 0.0651 2.57 0.0800
                                               0.536
##
    Sep-S + acid 0.227 0.0720 3.79 0.0228
                                               0.431
##
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
```

Need name for export.

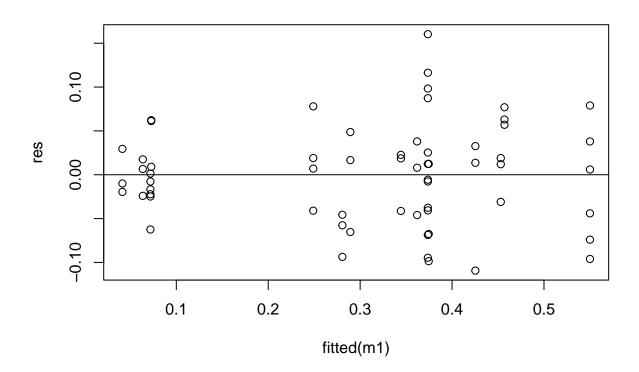
Dunnett's test, all compared to untreated.

```
contrast(mmeans, 'dunnett', ref = '')
```

```
##
   contrast
                     estimate
                                      df t.ratio p.value
                                 SE
##
    Acid -
                     -0.0930 0.0296 49.2 -3.145 0.0211
##
    Dis -
                      -0.0117 0.0395 49.1 -0.296 0.9978
## ( Dis + acid) -
                    -0.0843 0.0395 49.1 -2.136 0.2161
##
   MF liquid -
                      -0.3101 0.0395 49.1 -7.855 <.0001
##
   MF slurry -
                      0.0793 0.0395 49.1
                                          2.010 0.2714
## ( Sep-D) -
                      -0.3018 0.0262 49.4 -11.514 <.0001
## ( Sep-D + acid) -
                      -0.3323 0.0395 49.1 -8.418 <.0001
## ( Sep-S) -
                      -0.1247 0.0277 49.2 -4.495 0.0004
##
   ( Sep-S + acid) -
                      -0.2057 0.0420 49.5 -4.895 0.0001
##
## Degrees-of-freedom method: kenward-roger
## P value adjustment: dunnettx method for 9 tests
```

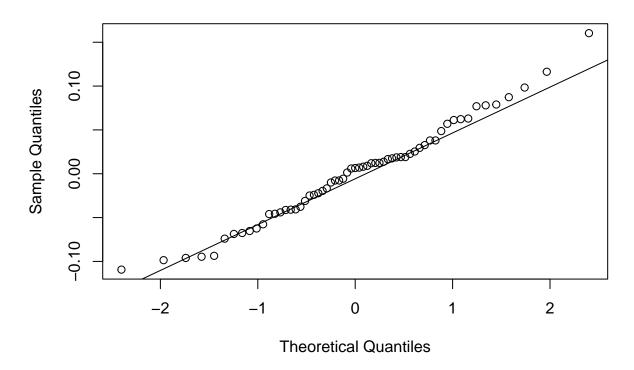
Residuals.

```
res <- resid(m1)
plot(fitted(m1), res)
abline(0,0)</pre>
```



qqnorm(res)
qqline(res)

Normal Q-Q Plot



Trials 5-10 (app tech on winter wheat)

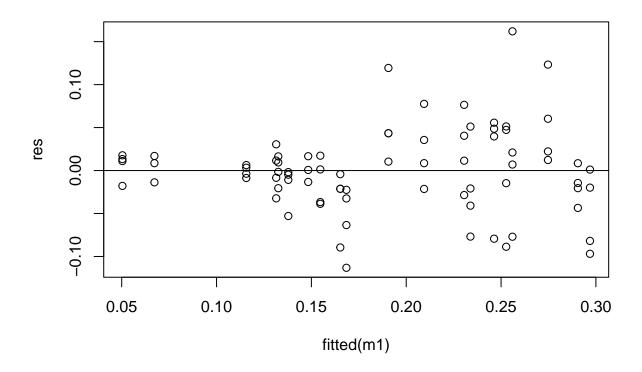
Subset.

```
setDT(isumm)
isumm1 <- isumm[new.ID %in% c('5', '6', '7', '8', '9', '10') & !treat1 %in% c('TH-4', 'TS1-4')]
Fit mixed-effects model.
m1 \leftarrow lmer(e.rel.150 \sim treat1 + (1|new.ID), data = isumm1)
summary(m1)
## Linear mixed model fit by REML ['lmerMod']
## Formula: e.rel.150 ~ treat1 + (1 | new.ID)
##
      Data: isumm1
##
## REML criterion at convergence: -203.3
##
## Scaled residuals:
                1Q Median
##
       Min
                                 ЗQ
                                         Max
## -2.2137 -0.4155 0.0176 0.3447
##
```

```
## Random effects:
## Groups Name
                        Variance Std.Dev.
## new.ID (Intercept) 0.004217 0.06494
## Residual
                        0.002613 0.05112
## Number of obs: 78, groups: new.ID, 6
##
## Fixed effects:
##
               Estimate Std. Error t value
## (Intercept) 0.214787 0.028492 7.539
## treat1TS1 -0.022150
                          0.014756 - 1.501
## treat1TS2 -0.006375
                          0.017576 -0.363
## treat1TS3 -0.087455
                          0.017576 -4.976
## Correlation of Fixed Effects:
##
            (Intr) tr1TS1 tr1TS2
## treat1TS1 -0.259
## treat1TS2 -0.217 0.420
## treat1TS3 -0.217 0.420 0.436
Marginal means.
mmeans <- emmeans(m1, 'treat1')</pre>
mmeans
                          df lower.CL upper.CL
##
  treat1 emmean
                     SE
          0.215 0.0285 5.99
                              0.1450
                                        0.285
## TS1
           0.193 0.0285 5.99
                               0.1229
                                         0.262
## TS2
           0.208 0.0301 7.36 0.1380
                                        0.279
## TS3
           0.127 0.0301 7.36 0.0569
                                        0.198
##
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
Need name for export.
Tukey's test
pairs(mmeans)
  contrast estimate
                          SE df t.ratio p.value
## TH - TS1
             0.02215 0.0148 69.0
                                    1.501 0.4424
              0.00638 0.0176 69.8
## TH - TS2
                                   0.362 0.9837
## TH - TS3
              0.08746 0.0176 69.8
                                  4.963 <.0001
## TS1 - TS2 -0.01577 0.0176 69.8 -0.895 0.8074
## TS1 - TS3 0.06531 0.0176 69.8
                                   3.706 0.0023
## TS2 - TS3 0.08108 0.0187 69.0
                                  4.344 0.0003
## Degrees-of-freedom method: kenward-roger
## P value adjustment: tukey method for comparing a family of 4 estimates
```

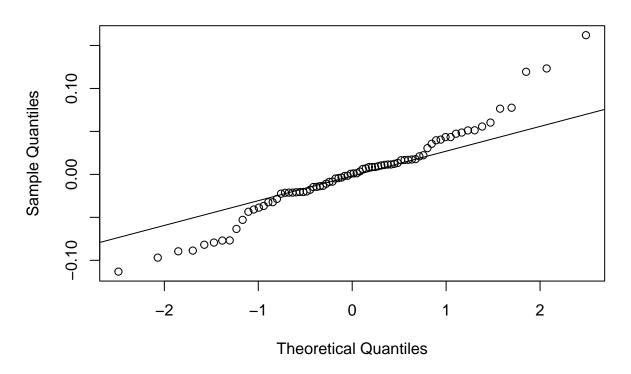
Residuals.

```
res <- resid(m1)
plot(fitted(m1), res)
abline(0,0)</pre>
```



qqnorm(res)
qqline(res)

Normal Q-Q Plot



Marginal means, export, save?

```
mmeans <- emmeans(m1, 'treat1')</pre>
mmeans
                             df lower.CL upper.CL
##
    treat1 emmean
                       SE
##
    TH
             0.215 0.0285 5.99
                                  0.1450
                                             0.285
             0.193 0.0285 5.99
                                             0.262
##
    TS1
                                  0.1229
    TS2
             0.208 0.0301 7.36
##
                                  0.1380
                                             0.279
##
    TS3
             0.127 0.0301 7.36
                                  0.0569
                                             0.198
##
## Degrees-of-freedom method: kenward-roger
## Confidence level used: 0.95
```

Tukey's test

pairs(mmeans)

```
##
   contrast
              estimate
                           SE
                                df t.ratio p.value
   TH - TS1
               0.02215 0.0148 69.0
                                     1.501 0.4424
   TH - TS2
               0.00638 0.0176 69.8
                                     0.362 0.9837
##
##
   TH - TS3
               0.08746 0.0176 69.8
                                     4.963
                                            <.0001
   TS1 - TS2 -0.01577 0.0176 69.8
                                    -0.895
                                           0.8074
##
   TS1 - TS3
               0.06531 0.0176 69.8
                                     3.706
                                            0.0023
   TS2 - TS3 0.08108 0.0187 69.0
                                     4.344
                                            0.0003
##
```

Degrees-of-freedom method: kenward-roger ## P value adjustment: tukey method for comparing a family of 4 estimates