Example Analysis of an Unbalanced Two-Factor Experiment

An experiment was conducted to study the effect of storage time and storage temperature on the amount of active ingredient present in a drug at the end of storage. A total of 16 vials of the drug, each containing approximately 30 mg/mL of active ingredient were assigned (using a completely randomized design) to the following treatments:

1) Storage for 3 months at 20° C
2) Storage for 3 months at 30° C
3) Storage for 6 months at 20° C
4) Storage for 6 months at 30° C

Six of the 16 vials were damaged during shipment to the laboratory where the active ingredient was measured. Accurate measures of the amount of active ingredient could be obtained only for the 10 undamaged vials. The table below shows the amount of active ingredient lost during storage (in tenths of mg/mL) for each of the undamaged vials.

Storage Time	Storage Temperature 20°C	30°C
3 months	2 5	9 12 15
6 months	6 6 7 7	16

```
time=factor(rep(c(3,6),each=5))
temp=factor(rep(c(20,30,20,30),c(2,3,4,1)))
y=c(2,5,9,12,15,6,6,7,7,16)
d=data.frame(time,temp,y)
d
   time temp  y
1     3   20  2
2     3   20  5
3     3   30  9
4     3   30 12
5     3   30 15
6     6   20  6
7     6   20  6
8     6   20  7
9     6   20  7
10    6   30 16

o=lm(y~time+temp+time:temp,data=d)
```

```
model.matrix(o)
   (Intercept) time6 temp30 time6:temp30
1            1     0      0            0
2            1     0      0            0
3            1     0      1            0
4            1     0      1            0
5            1     0      1            0
6            1     1      0            0
7            1     1      0            0
8            1     1      0            0
9            1     1      0            0
10           1     1      1            1
attr(,"assign")
[1] 0 1 2 3
attr(,"contrasts")
attr(,"contrasts")$time
[1] "contr.treatment"

attr(,"contrasts")$temp
[1] "contr.treatment"
```

```
coef(o)
 (Intercept)          time6          temp30  time6:temp30
         3.5            3.0             8.5           1.0
vcov(o)
             (Intercept)     time6    temp30  time6:temp30
(Intercept)     1.958333 -1.958333 -1.958333      1.958333
time6          -1.958333  2.937500  1.958333     -2.937500
temp30         -1.958333  1.958333  3.263889     -3.263889
time6:temp30    1.958333 -2.937500 -3.263889      8.159722
```

```
#Cell means are
#
#            temp 20    temp 30
#            -------------------------
# time 3     mu         mu+temp30
# time 6     mu+time6   mu+time6+temp30+time6:temp30
#
#Time main effects?
#
#     (mu+mu+temp30)/2
#    -(mu+time6+mu+time6+temp30+time6:temp30)/2
#     ----------------------------
#    -time6-time6:temp30/2
#
#    H0:time6+time6:temp30/2=0
```

```
test=function(lmout,C,d=0){
  b=coef(lmout)
  V=vcov(lmout)
  dfn=nrow(C)
  dfd=lmout$df
  Cb.d=C%*%b-d
  Fstat=drop(t(Cb.d)%*%solve(C%*%V%*%t(C))%*%Cb.d/dfn)
  pvalue=1-pf(Fstat,dfn,dfd)
  list(Fstat=Fstat,pvalue=pvalue)
}
```

```
Ctime=matrix(c(
0,1,0,.5
),nrow=1,byrow=T)
```

```
test(o,Ctime)
$Fstat
[1] 6.005106
$pvalue
[1] 0.04975481
```

```
#The R function anova will produce tests for
#the presence of time main effects,
#temp main effects, and time-by-temp interaction.
#However, these are "Type I Tests" that do not
#in general match the "Type III Tests" above.

anova(o)
Analysis of Variance Table

Response: y
          Df Sum Sq Mean Sq F value    Pr(>F)
time       1   0.10   0.100  0.0255 0.878292
temp       1 158.42 158.420 40.4477 0.000709 ***
time:temp  1   0.48   0.480  0.1226 0.738243
Residuals  6  23.50   3.917
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.'
0.1 ' ' 1
```

```
> library(car)
> Anova(o,type='II')
Anova Table (Type II tests)

Response: y
          Sum Sq Df F value   Pr(>F)
time       23.52  1  6.0051 0.049755 *
temp      158.42  1 40.4477 0.000709 ***
time:temp   0.48  1  0.1226 0.738243
Residuals  23.50  6
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
```