Chapter 4: Data Analysis for RCB designs with R

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Introduction

One of the three important principals for an experimental design is local control since either experimental units or conditions may not be uniform. Randomized complete block design is one of the most popular experimental designs used for local control.

Generate a RCB Design

It seems very easy to generate any RCB design by using R. The following R scripts show how to generate a RCB deisgn.

```
r=5 # 5 blocks
t=10 # 10 treatments
RCB=matrix(0, r, t)
for(i in 1:r)RCB[i,]=sample(1:t)
#rownames(RCB)=c("Block", "Treatment")
data.frame(RCB)
```

X1	X2	Х3	X4	X5	X6	X7	X8	X9	X10
8	6	7	10	4	2	5	9	3	1
3	10	4	8	7	9	2	1	6	5
10	3	8	9	6	5	4	2	7	1
10	2	1	7	6	5	4	8	3	9
8	10	5	7	9	6	4	2	3	1

Load an RCB data set

We may read a read data file from a hard drive using the following R codes.

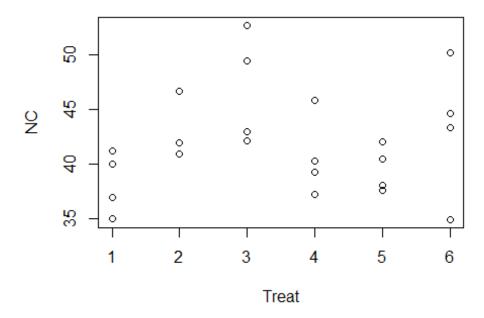
```
require(coursedata)
```

```
## Loading required package: coursedata

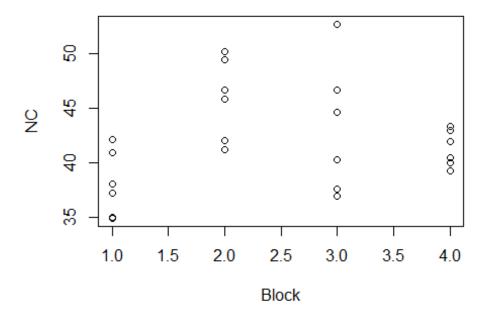
data(wheatrcb)
wheat=wheatrcb
```

We can visualize the data by using the following R codes

plot(NC~Treat,data=wheat)

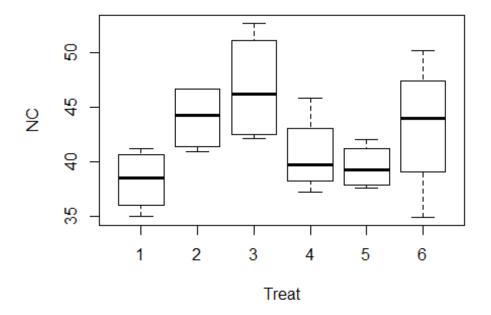


plot(NC~Block,data=wheat)

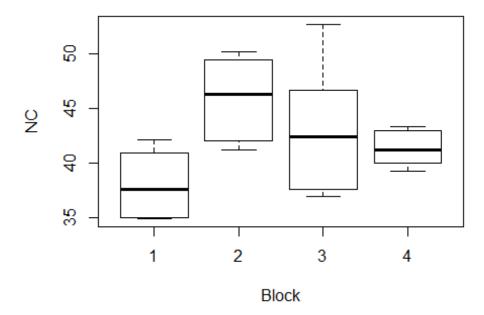


The above R codes treat Treat and Block as numerical values. So we need convert these two variables to categorical variables. Here we are using a very useful function, transform, to change the classes of variables with in a data frame. You will see some different results after variable conversion.

```
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
plot(NC~Treat,data=wheat)
```



plot(NC~Block,data=wheat)



Then we use any to run annova analysis when both Treat and Block are not converted to the categorical variables.

```
require(coursedata)
data(wheatrcb)
wheat=wheatrcb
mod=aov(NC~Treat+Block,data=wheat)
a=summary(mod)
```

If you pay a little attention to the anova results, the degrees of freedom for both treatment and block are 1. It is because the codes used for these two effects are integers or numerical rather than categorical variables. Using str(datafile) you can check the class of each variable in a datafile.

```
str(wheat)
## 'data.frame': 24 obs. of 3 variables:
## $ Treat: int 2 5 4 1 6 3 1 3 4 6 ...
## $ Block: int 1 1 1 1 1 2 2 2 2 ...
## $ NC : num 40.9 38 37.2 35 34.9 ...
```

In order to obtain appropriate results, we first need to factorize these two factors: treatment and block. Please read the following R scripts as your reference to try. You will see the big differences for block and treatment after they are converted to factors.

```
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
str(wheat)

## 'data.frame': 24 obs. of 3 variables:
## $ Treat: Factor w/ 6 levels "1","2","3","4",..: 2 5 4 1 6 3 1 3 4 6 ...
## $ Block: Factor w/ 4 levels "1","2","3","4": 1 1 1 1 1 1 2 2 2 2 ...
## $ NC : num 40.9 38 37.2 35 34.9 ...
wheat
```

Treat	Block	NC
2	1	40.89
5	1	37.99
4	1	37.18
1	1	34.98
6	1	34.89
3	1	42.07
1	2	41.22
3	2	49.42
4	2	45.85
6	2	50.15
5	2	41.99

```
2
       2
               46.69
6
       3
               44.57
3
       3
               52.68
5
       3
               37.61
1
       3
               36.94
2
       3
               46.65
4
       3
               40.23
2
       4
               41.90
4
       4
               39.20
6
       4
               43.29
5
       4
               40.45
3
       4
               42.91
1
       4
               39.97
```

head(wheat)

Treat	Block	NC	
2	1	40.89	
5	1	37.99	
4	1	37.18	
1	1	34.98	
6	1	34.89	
3	1	42.07	

Now you may see the results look different after data conversion. The SS, MS, DF, and ftests are totally different compared to the data analysis shown above. Therefore, it is very important to check and/change the type of each variable before you use the built-in functions like aov or lm.

```
mod=aov(NC~Treat+Block,data=wheat)
summary(mod)
##
              Df Sum Sq Mean Sq F value Pr(>F)
                                 5.592 0.00419 **
## Treat
              5 201.3
                         40.26
              3 197.0
                                 9.120 0.00112 **
## Block
                         65.67
## Residuals
              15 108.0
                          7.20
## ---
## Signif. codes: 0 '***' 0.001 '**' 0.05 '.' 0.1 ' ' 1
```

Based on the above ANOVA results, we can conclude that both treatment and block effects are significant. Therefore, multiple mean comparsons among treatments are needed, but we will show the multiple comparisons among treatments later.

You may treat this RCB design data set to a CR design data set. By doing so, you can use a reduced anova model to run the data analysis, with only treatment effect included in the model.

Without including block effects in the model, you will see that the MSE increased from 7.2 to 16.9, more than the double. It is very reasonable that the F-test for treatment effect is not significant due to a large MSE using this reduced model for data analysis.

You may also run another reduced model with only block effect included. Again, the MSE for this reduced model slightly increased; however, the F-test for block effect was significant, but not so significant as in the full model.

Relative efficiency

It is very easy to calculate relative efficient (RE) using R from ANAVA analysis of different models. We just focus on using mean square error (MSE) from a full model and a reduced model. The MSE calculated from the first model is from a full model and the MSE from the second model is a reduced model (a CR design). We can extract MSE from the results obtained by aov analysis for these two models. Please pay a little attention on how I extracted a MSE from an AOV analysis: to find the index to locate MSE from each ANOVA table so that MSE from an ANOVA table can be automatically located.

```
require(coursedata)
data(wheatrcb)
wheat=wheatrcb
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
# A full model
mod1=aov(NC~Treat+Block,data=wheat)
a=summary(mod1)
dfe1=mod$df.residual ## degree of freedom for MSE
```

```
rn1=nrow(a[[1]])  ## which row where MSE is stored
mse1=a[[1]][rn1,3] ## Locate the MSE from this ANOVA table

# A reduced model
mod2=aov(NC~Treat,data=wheat)
a=summary(mod2)
dfe2=mod2$df.residual ## degree of freedom for MSE
rn2=nrow(a[[1]]) ## which row where MSE is stored
mse2=a[[1]][rn2,3] ## Locate the MSE from this ANOVA table

# Calculate relative efficiency (RE)
RE=mse2/mse1
RE
## [1] 2.353307
```

The RE calculated from above is 2.35, showing that the RCB design for the wheat experiment is estimated to be slightly more than twice as efficient as a CR design. Or we can conclude that the CR design will require twice replications for each treatment as compared with a RCB design. We can also conclude that an RCB design is highly needed to reduce experimental errors.

Pair-wise mean comparison

Once a F-test for treatment effects is significant, it is very reasonable to conduct pair-wise mean comparisons between treatments. Since the model for an RCB design is different from that for a CR design, the procedures in a CR design may not all work for RCB design or other complicated designs because more factors are used for modelling. Now let us try LSD test among these treatments. Again, we need load the package agricolae before we can use the function LSD.test. Again, it is very very important to provide the right MSE and its degrees of freedom (DF). Using the following R codes can automatically locate the right MSE and its corresponding DF.

LSD test

```
require(agricolae)
## Loading required package: agricolae

require(coursedata)
data(wheatrcb)
wheat=wheatrcb
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block)) ## convert th
e variables
mod=aov(NC~Treat+Block,data=wheat)
a=summary(mod1)
dfe=mod$df.residual
rn=nrow(a[[1]])
mse=a[[1]][rn,3]
```

```
res=LSD.test(wheat$NC, wheat$Treat, DFerror=dfe, MSerror=mse)
res
## $statistics
                    CV MSerror
##
         Mean
                                     LSD
##
     42.07167 6.378131 7.200561 4.044299
##
## $parameters
     Df ntr t.value alpha
##
                                test
                                          name.t
##
         6 2.13145 0.05 Fisher-LSD wheat$Treat
##
## $means
     wheat$NC
                   std r
                              LCL
                                       UCL
                                             Min
##
                                                   Max
## 1 38.2775 2.839324 4 35.41775 41.13725 34.98 41.22
## 2 44.0325 3.073352 4 41.17275 46.89225 40.89 46.69
## 3 46.7700 5.129659 4 43.91025 49.62975 42.07 52.68
## 4 40.6150 3.712811 4 37.75525 43.47475 37.18 45.85
## 5 39.5100 2.078012 4 36.65025 42.36975 37.61 41.99
## 6 43.2250 6.304525 4 40.36525 46.08475 34.89 50.15
##
## $comparison
## NULL
##
## $groups
##
    trt
          means
                   Μ
## 1
       3 46.7700
## 2
      2 44.0325 ab
       6 43.2250 abc
## 3
## 4
      4 40.6150 bcd
       5 39.5100 cd
## 5
## 6
      1 38.2775
```

Duncan Test

You can also try Duncan test for pair-wise comparisons among treatments. Again, you need to define which factor to be used for pair-wise comparisons. The duncan test function needs the aov results as a list for data input for the first component. The second component is the variable name of the treatment.

```
require(agricolae)
require(coursedata)
data(wheatrcb)
wheat=wheatrcb
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
mod=aov(NC~Treat+Block,data=wheat)
res=duncan.test(mod, "Treat",main="Nitrogen content with different treatment"
)
res
```

```
## $statistics
##
                   CV MSerror
        Mean
     42.07167 6.378131 7.200561
##
##
## $parameters
     Df ntr alpha
##
                   test name.t
##
         6 0.05 Duncan Treat
##
## $Duncan
##
        Table CriticalRange
## 2 3.014325
                  4.044299
## 3 3.159826
                  4.239517
## 4 3.250248
                 4.360835
## 5 3.311848
                 4.443484
               4.502754
## 6 3.356024
##
## $means
##
         NC
                 std r
                         Min
                               Max
## 1 38.2775 2.839324 4 34.98 41.22
## 2 44.0325 3.073352 4 40.89 46.69
## 3 46.7700 5.129659 4 42.07 52.68
## 4 40.6150 3.712811 4 37.18 45.85
## 5 39.5100 2.078012 4 37.61 41.99
## 6 43.2250 6.304525 4 34.89 50.15
##
## $comparison
## NULL
##
## $groups
   trt
          means
                  Μ
## 1
     3 46.7700
## 2 2 44.0325 ab
## 3 6 43.2250 abc
## 4
     4 40.6150 bcd
## 5
      5 39.5100 cd
## 6 1 38.2775
```

Tukey Test

You can try Tukey test for pair-wise comparisons among treatments as well. The procedure is very similar to the Duncan test.

```
require(agricolae)
require(coursedata)
data(wheatrcb)
wheat=wheatrcb
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
mod=aov(NC~Treat+Block,data=wheat)
res=HSD.test(mod, "Treat",main="NC with different treatment")
res
```

```
## $statistics
##
                   CV MSerror
                                    HSD
        Mean
    42.07167 6.378131 7.200561 6.164724
##
##
## $parameters
    Df ntr StudentizedRange alpha test name.t
##
##
                   4.594735 0.05 Tukey Treat
##
## $means
##
         NC
                 std r
                         Min
                               Max
## 1 38.2775 2.839324 4 34.98 41.22
## 2 44.0325 3.073352 4 40.89 46.69
## 3 46.7700 5.129659 4 42.07 52.68
## 4 40.6150 3.712811 4 37.18 45.85
## 5 39.5100 2.078012 4 37.61 41.99
## 6 43.2250 6.304525 4 34.89 50.15
##
## $comparison
## NULL
##
## $groups
## trt
          means M
## 1 3 46.7700 a
## 2
      2 44.0325 ab
## 3 6 43.2250 ab
## 4 4 40.6150 ab
## 5 5 39.5100 b
## 6 1 38.2775 b
```

Linear mixed model approaches to analyzing one-factor experimental designs

Linear mixed model approaches are much complicated and but they are very powerful and suitable for unbalaced and/or missing data. You need install and load the minque R package before you can use some important R functions.

Install and load minque

You can use the function of install.packages such as install.packages ("minque") and then the function require like require(minque).

```
require(minque)
## Loading required package: minque
## Loading required package: klaR
## Loading required package: MASS
```

```
## Loading required package: Matrix
```

With the minque package you can run various kinds of data analyses

Case 1: Both treatment and block effects fixed

In this case we consider treatment effects as fixed.

```
require(coursedata)
data(wheatrcb)
wheat=wheatrcb
wheat=transform(wheat,Treat=factor(Treat),Block=factor(Block))
res1=lmm(NC~Treat+Block,data=wheat)[[1]]
res1$Var
## $NC
##
             Est
                       SE Chi_sq
                                    P_value
## V(e) 7.200561 2.629273
                             7.5 0.00308495
res1$FixedEffect
## $NC
##
                  Est
                             SE
                                   z value
                                                P value
## mu
           42.071667 0.6247409 67.3425872 0.000000e+00
## Treat(2) 1.960833 1.2360804 1.5863316 1.126641e-01
## Treat(5) -2.561667 1.2360804 -2.0724111 3.822712e-02
## Treat(4) -1.456667 1.2360804 -1.1784563 2.386148e-01
## Treat(1) -3.794167 1.2360804 -3.0695145 2.144070e-03
## Treat(6) 1.153333 1.2360804 0.9330569 3.507906e-01
## Treat(3) 4.698333 1.2360804 3.8009934 1.441171e-04
## Block(1) -4.071667 0.9811066 -4.1500758 3.323653e-05
## Block(2) 3.815000 0.9811066 3.8884664 1.008796e-04
## Block(3) 1.041667 0.9811066 1.0617263 2.883600e-01
## Block(4) -0.785000 0.9811066 -0.8001169 4.236430e-01
res1$RandomEffect ##no results shown
## NULL
```

Case 2: Random block effects

In this case we consider block effects as random.

```
res1=lmm(NC~Treat|Block,data=wheat)[[1]]
res1$Var

## $NC
## Est SE Chi_sq P_value
## V(Block) 9.744569 8.947018 1.186229 0.13804572
## V(e) 7.200561 2.629273 7.500000 0.00308495
```

```
res1$FixedEffect
## $NC
##
                  Est
                            SE
                                  z_value
                                               P_value
## mu
            42.071667 1.662511 25.3060951 0.0000000000
## Treat(2) 1.960833 1.236080 1.5863316 0.1126641103
## Treat(5) -2.561667 1.236080 -2.0724111 0.0382271197
## Treat(4) -1.456667 1.236080 -1.1784563 0.2386147539
## Treat(1) -3.794167 1.236080 -3.0695145 0.0021440697
## Treat(6) 1.153333 1.236080 0.9330569 0.3507905836
## Treat(3) 4.698333 1.236080 3.8009934 0.0001441171
res1$RandomEffect
## $NC
##
                   Pre
                             SE
                                   z value
                                             P value
## Block(1) -3.8419557 2.550892 -1.5061224 0.1320358
## Block(2) 3.5997693 2.550892 1.4111806 0.1581914
## Block(3) 0.9828990 2.550892 0.3853158 0.7000035
## Block(4) -0.7407127 2.550892 -0.2903740 0.7715301
```

Case 3: Random treatment and block effects model

In this case we consider both treatment effects and block effects as random.

```
res1=lmm(NC~1|Treat+Block,data=wheat)[[1]]
res1$Var
## $NC
##
                 Est
                           SE
                                Chi sq
                                          P value
## V(Treat) 8.265679 6.400028 1.667990 0.09826401
## V(Block) 9.744569 8.947018 1.186229 0.13804572
           7.200561 2.629273 7.500000 0.00308495
## V(e)
res1$FixedEffect
## $NC
##
           Est
                     SE z value P value
## mu 42.07167 2.028245 20.74289
res1$RandomEffect
## $NC
##
                   Pre
                             SE
                                   z value
                                              P value
## Treat(2) 1.7768689 2.378282 0.7471229 0.45498939
## Treat(5) -2.3213323 2.378282 -0.9760543 0.32903756
## Treat(4) -1.3200029 2.378282 -0.5550237 0.57887844
## Treat(1) -3.4381998 2.378282 -1.4456653 0.14827110
## Treat(6) 1.0451282 2.378282 0.4394467 0.66033790
## Treat(3) 4.2575380 2.378282 1.7901737 0.07342598
## Block(1) -3.8419557 2.550892 -1.5061224 0.13203576
## Block(2) 3.5997693 2.550892 1.4111806 0.15819138
```

```
## Block(3) 0.9828990 2.550892 0.3853158 0.70000346
## Block(4) -0.7407127 2.550892 -0.2903740 0.77153015
```

Missing data case

A nice feature of linear model approaches is flexibity to deal with missing data. Now let us manually create a missing data set from the above wheat data.

```
n=nrow(wheat)
id=sample(n,2)
wheat1=wheat[-id,]
res1=lmm(NC~1|Treat+Block,data=wheat1)[[1]]
res1$Var
## $NC
##
                 Est
                           SE
                                Chi_sq
                                           P value
## V(Treat) 8.674201 6.724680 1.663856 0.098541772
## V(Block) 7.826003 7.400057 1.118433 0.145128078
## V(e)
           6.511680 2.552020 6.510551 0.005361813
res1$FixedEffect
## $NC
                     SE z_value P_value
           Est
## mu 41.67471 1.921438 21.68933
res1$RandomEffect
## $NC
##
                             SE
                                   z value
                                              P value
                   Pre
## Treat(2) 2.2042876 2.460433 0.8958941 0.37030934
## Treat(5) -2.0270947 2.460433 -0.8238771 0.41000938
## Treat(4) -0.9932246 2.460433 -0.4036787 0.68644897
## Treat(1) -3.1802575 2.460433 -1.2925599 0.19616330
## Treat(6) -0.7692829 2.296273 -0.3350136 0.73761481
## Treat(3) 4.7655722 2.460433 1.9368833 0.05275961
## Block(1) -3.4789198 2.262582 -1.5375886 0.12414924
## Block(2) 2.9676854 2.244452 1.3222318 0.18609096
## Block(3) 1.3595394 2.262582 0.6008797 0.54792011
## Block(4) -0.8483050 2.244452 -0.3779565 0.70546294
```

Then we can try one more time by repeating the above process.

```
n=nrow(wheat)
id=sample(n,2)
wheat1=wheat[-id,]
res1=lmm(NC~1|Treat+Block,data=wheat1)[[1]]
res1$Var

## $NC
## Est SE Chi_sq P_value
```

```
## V(Treat) 9.839131 7.505821 1.718372 0.094951390
## V(Block) 10.585735 9.728398 1.184021 0.138269462
## V(e)
             6.982848 2.738113 6.503731 0.005382418
res1$FixedEffect
## $NC
##
           Est
                   SE z_value P_value
## mu 42.29713 2.14301 19.73725
res1$RandomEffect
## $NC
                                              P value
##
                   Pre
                             SE
                                   z value
## Treat(2)
            1.6403982 2.634150 0.6227428 0.53345356
## Treat(5) -2.1764320 2.572333 -0.8460927 0.39750101
## Treat(4) -1.5574578 2.634150 -0.5912562 0.55434877
## Treat(1) -3.7447259 2.634150 -1.4216066 0.15514047
## Treat(6) 0.8847965 2.634150 0.3358944 0.73695046
## Treat(3)
            4.9534210 2.572333 1.9256533 0.05414768
## Block(1) -4.1087280 2.669230 -1.5392933 0.12373270
## Block(2) 3.7472254 2.645998 1.4161861 0.15672104
## Block(3) 0.8006381 2.669230 0.2999510 0.76421455
## Block(4) -0.4391356 2.645998 -0.1659622 0.86818671
```

You can compare the these results among different missing data cases and compare with the results from the complete data set.

This is some idea to lead jackknife process which are integrated into mingue package.

```
res1=lmm.jack(NC~1|Treat+Block,data=wheat)[[1]]
res1$Var
## $NC
##
            Estimate
                           SE
                                    PValue
                                             2.5%LL 97.5%UL
## V(Treat) 8.195099 1.289060 0.0005267433 3.641791 12.74841
## V(Block) 9.715641 1.527788 0.0005255852 4.319083 15.11220
## V(e)
            7.296507 1.046848 0.0002614985 3.598758 10.99426
res1$PVar
## $NC
                Estimate
                                 SE
                                          PValue
                                                    2.5%LL
                                                             97.5%UL
## V(Treat)/VP 0.3254295 0.05087590 5.030255e-04 0.1457221 0.5051369
## V(Block)/VP 0.3858190 0.05606868 2.886427e-04 0.1877693 0.5838687
               0.2887515 0.02908983 1.522258e-05 0.1859984 0.3915046
## V(e)/VP
res1$FixedEffect
## $NC
      Estimate
                      SE PValue 2.5%LL 97.5%UL
## mu 42.07862 0.2263466 0 41.2791 42.87813
```

```
res1$RandomEffect
## $NC
                  Pre
##
                              SE
                                       PValue
                                                  2.5%LL
                                                             97.5%UL
## Treat(2)
            1.7714842 0.2699423 4.145948e-04 0.8179755
                                                         2.72499288
## Treat(5) -2.2382754 0.6034653 1.924481e-02 -4.3698771 -0.10667375
## Treat(4) -1.2911194 0.3468661 1.885902e-02 -2.5163436 -0.06589519
## Treat(1) -3.4257074 0.3512336 1.761293e-05 -4.6663589 -2.18505584
## Treat(6)
            1.0349789 0.5963467 3.817324e-01 -1.0714778 3.14143560
## Treat(3)
            4.1486390 0.4829286 4.990068e-05 2.4428053 5.85447277
## Block(1) -3.7963571 0.3561667 8.394372e-06 -5.0544334 -2.53828076
## Block(2) 3.5858482 0.2946617 2.733097e-06 2.5450238
                                                         4.62667257
## Block(3) 0.9768081 0.3405839 7.178237e-02 -0.2262257
                                                         2.17984200
## Block(4) -0.7662993 0.2060491 1.895259e-02 -1.4941200 -0.03847858
```

Please compare these results with the results without jackknife. The power with jackknife has been greatly improved, yet other results remain almost the same. This is the beauty of linear mixed model approaches integrated with jackknife methods.

You may also try a model with fixed treatment effects and random block effects.

```
res1=lmm.jack(NC~Treat|Block,data=wheat)[[1]]
res1$Var
## $NC
##
            Estimate
                           SE
                                    PValue
                                             2.5%LL 97.5%UL
## V(Block) 9.732036 1.422840 0.0003023116 4.706182 14.75789
## V(e)
            7.145094 1.108034 0.0004733303 3.231219 11.05897
res1$PVar
## $NC
##
                Estimate
                                 SE
                                          PValue
                                                    2.5%LL
                                                             97.5%UL
## V(Block)/VP 0.5760467 0.05209507 6.162131e-06 0.3920329 0.7600605
## V(e)/VP
               0.4239533 0.05209507 7.720345e-05 0.2399395 0.6079671
res1$FixedEffect
## $NC
##
             Estimate
                             SE
                                      PValue
                                                 2.5%LL
                                                            97.5%UL
## mu
            42.106171 0.2888862 8.881784e-16 41.0857477 43.12659520
## Treat(2) 1.946825 0.3414294 1.173674e-03 0.7408051 3.15284576
## Treat(5) -2.577648 0.4941621 2.205821e-03 -4.3231614 -0.83213508
## Treat(4) -1.459995 0.4038456 2.223398e-02 -2.8864866 -0.03350406
## Treat(1) -3.797184 0.5131014 1.640453e-04 -5.6095961 -1.98477239
## Treat(6)
            1.202675 0.8239168 5.263815e-01 -1.7076206 4.11297069
## Treat(3) 4.685327 0.7432622 5.613028e-04 2.0599257 7.31072903
res1$RandomEffect
## $NC
                              SE
                   Pre
                                       PValue
                                                  2.5%LL
                                                           97.5%UL
##
```

```
## Block(1) -3.7811056 0.3805671 1.510512e-05 -5.1253707 -2.436841

## Block(2) 3.5356619 0.3925568 3.394277e-05 2.1490460 4.922278

## Block(3) 1.0145633 0.5359516 3.107230e-01 -0.8785617 2.907688

## Block(4) -0.7691197 0.5270475 5.266079e-01 -2.6307930 1.092554
```

Conclusions

Please remember several key functions that can be used for one-factor ANOVA analysis:lm,aov,LSD.test,HSD.test, and duncan.test. Actually these functions can be used in more complicated experimental deisgn analysis.

You may also use minque package to analyze one-factor experimental designs. It can estimate all variance components and fixed effects and predict random effects.