## THE DESIGN AND MIXED-MODEL ANALYSIS OF EXPERIMENTS

## PRACTICAL VI SOLUTIONS

VI.1 It is desired to run a wine-tasting experiment in which the differences between six wines are to be evaluated by scoring them on a 20 point scale. It is decided to have 6 expert judges evaluate the wines by evaluating a glass of wine on each of six consecutive occasions. It is desired to be able to isolate judge differences in scoring and differences between occasions so that a Latin square is to be employed. Use Genstat, with 224533 as the seed, to obtain a suitable randomized layout for the experiment.

The following information was supplied in response to questions from Genstat:

How many rows and columns are there in the Latin square?	6
How many treatment factors (or mutually orthogonal Latin	1
squares) do you want to generate? (up to 3 available)	
What would you like to call the treatment factor?	Wines
Give the identifier to be used for the row factor?	Occasion
Give the identifier to be used for the column factor?	Judge
Seed for randomization (0 for none)?	224533
What would you like to print?	design
Do you want to check the design by ANOVA	yes

The generated design is gven in the following output. For example on the first occasion, wines 1, 6, 4, 2, 5, 3 will be tasted by jusges 1–6, respectively.

```
Genstat 5 Release 4.1 (PC/Windows NT) 28 March 2000 16:01:25
Copyright 1998, Lawes Agricultural Trust (Rothamsted Experimental Station)

Genstat 5 Fourth Edition - (for Windows)
Genstat 5 Procedure Library Release PL11
```

3 DESIGN

\*\*\* Treatment combinations on each unit of the design \*\*\*

Judge	1	2	3	4	5	6
Occasion						
1	1	6	4	2	5	3
2	3	5	6	1	4	2
3	2	4	5	3	6	1
4	6	2	3	4	1	5
5	4	3	1	5	2	6
6	5	1	2	6	3	4

Treatment factors are listed in the order: Wines

3	
***** Analysis of variance Source of variation	
Occasion stratum	5
Judge stratum	5
Occasion.Judge stratum Wines Residual	5 20

35

Total

**VI.2** In the lecture, a Latin square example was discussed that involved 4 areas and 4 intervals in investigating the bias of samplers in selecting wheat samples. Discuss the circumstances in which the factors Area and Interval are likely to be regarded as fixed and those in which they are likely to be regarded as random.

Areas would be fixed if the 4 areas were not representative of a larger population of areas and one expects a systematic difference between the areas. For example, if the areas were of 4 specific types (for example sandy, marshy, stoney and shaded) and these were the only 4 types of area of interest. One expects the pattern in the deviations of the area means from the grand mean to be quite irregular and their distribution is likely to uninformative. The best model would appear to be that each brand has a different mean value.

Intervals might be random if it was felt that there was unlikely to be systematic effects and that the observed intervals were representative of a larger population. For example, one might argue that practice and fatigue would not affect the response because the samplers are experienced. Also, perhaps the intervals occur randomly throughout a short period of time and so there are breaks in between. In this case one expects the deviations to vary randomly above and below the grand mean and to be described using a probability distribution with some variance.

In this example, we went for the most likely classification of Areas as random and Intervals as fixed.

VI.3 The following data are from a Latin Square experiment designed to investigate the moisture content of turnip greens. The experiment involved the measurement of the percent moisture content of five leaves of different sizes from each of five plants. The treatments were time of measurement in days since the beginning of the experiment.

				Plant		
		1	2	3	4	5
-	Α	5 6.67	2 5.40	3 7.32	1 4.92	4 4.88
	В	4 7.15	5 4.77	2 8.53	3 5.00	1 6.16
Leaf Size	С	1 8.29	4 5.40	5 8.50	2 7.29	3 7.83
(A = smallest,	D	3 8.95	1 7.54	4 9.99	5 7.85	2 5.83
E = largest)	Е	2 9.62	3 6.93	1 9.68	4 7.08	5 8.51

Classify the factors Leaf Size and Plant as either fixed or random.

It is most likely that Leaf Size will be fixed whereas Plants will be random. There may well be systematic differences between leaves of different sizes and so this is best modelled using different means for each size. Plants on the other hand are most likely just 5 plants selected from many plants of this type and a probability distribution with some variance is likely to be an appropriate model.

The Moisture contents have been saved in the Genstat spreadsheet file *LSTurn.gsh* in the directory *G:\Disciplina\Genstat*. Add the factors Size, Plant and Time to this spreadsheet.

Analyze the data using Genstat, including diagnostic checking and the examination of mean differences. You will find that there is an outlier. Set the value for this observation missing by replacing the value with Genstat's missing value indicator, an asterisk ('\*'). What efect does this have on the analysis?

```
Genstat 5 Release 4.1 (PC/Windows NT)
                                                   28 March 2000 17:46:40
Copyright 1998, Lawes Agricultural Trust (Rothamsted Experimental Station)
                Genstat 5 Fourth Edition - (for Windows)
                Genstat 5 Procedure Library Release PL11
     "Data taken from File: D:/ANALYSES/LM/ONEFAC/LSTURNALL.GSH"
     DELETE [redefine=yes] Size, Plant, Time, Moisture
     FACTOR [modify=yes;nvalues=25;levels=5] Size
   6 READ Size; frepresentation=ordinal
   Identifier
               Values Missing
                                   Levels
                   25
   8 FACTOR [modify=yes;nvalues=25;levels=5] Plant
  9 READ Plant; frepresentation=ordinal
   Identifier
                Values Missing
        Plant.
                  2.5
 11 FACTOR [modify=yes;nvalues=25;levels=5] Time
12 READ Time; frepresentation=ordinal
   Identifier Values Missing
                  25
 14 VARIATE [nvalues=25] Moisture
 15 READ Moisture
   Identifier Minimum
                           Mean Maximum Values Missing
     Moisture 4.770
                           7.204 9.990 25
 18
 19 PRINT Size, Plant, Time, Moisture
                 Plant
                                    Moisture
                              Time
                    6.670
          1
          1
```

```
4
                              3
                                    5.000
                             1
         2
                   5
                                    6.160
                   1
                             1
                                    8.290
         3
                   2
                                    5.400
                             4
                   3
                              5
                                    8.500
                             2
                                    7.290
                   4
                                    7.830
                   5
                             3
                              3
                   1
                                    8.950
                             1
                                    7.540
                   2
         4
                   3
                             4
                                   9.990
                                   7.850
5.830
         4
                   4
                             5
                   5
                              2
                                    9.620
         5
                             2
                   1
         5
                   2
                             3
                                    6.930
                             1
4
         5
                   3
                                    9.680
                                    7.080
         5
                    4
                             5
                                   8.510
 20 BLOCK Plant*Size
 21 TREAT POL(Time; 2)
 22 ANOVA [FPROB=Y; PSE=LSD] Moisture
22.....
**** Analysis of variance ****
Variate: Moisture
Source of variation d.f. s.s. m.s. v.r. F pr.
Plant stratum
                      4
                           28.8853
                                     7.2213 10.71
                       4
                         23.7081
                                     5.9270 8.79
Size stratum
Plant.Size stratum
Time
                       4
                           0.6273
                                    0.1568 0.23 0.915
 Lin
                       1
                           0.1512
                                    0.1512
                                            0.22 0.644
                                     0.0929
                                              0.14 0.717
0.28 0.758
                       1
                           0.0929
 Ouad
                       2
 Deviations
                            0.3831
                                     0.1916
Residual
                      12
                           8.0879
                                     0.6740
Total
                      24
                           61.3086
* MESSAGE: the following units have large residuals.
Plant 5 Size 4
                       -1.77 s.e. 0.57
***** Tables of means *****
Variate: Moisture
Grand mean 7.20
                     2
                  7.33 7.21 6.90
            7.32
                                         7.26
*** Least significant differences of means (5% level) ***
Table
                   Time
rep.
                     5
                    12
d.f.
                  1.131
l.s.d.
 23 CALC FP=7.2213/0.6740 & pP=1-FPROB(FP; 4; 12) : PRINT FP,pP
```

8.530

3

FP

10.71 0.0006232

```
0.001483
      8.794
  25 AGRAPH [GRAPH=line] XFACTOR=Time; BAR=*
                                    Means for Time
           I
        7.5 I
           I
           Ι
           Ι
           Ι
           Ι
        7.2 I
           Ι
           Ι
           Ι
           Ι
           Ι
        6.9 I
                                 3.0
                        2.0
                                                   5.0 6.0
          0.0
                1.0
                                          4.0
  26 APLOT METHOD=fit, normal
           Ι
           I
           Ι
        2.0 I
r
           Ι
е
           Ι
s
           I
d
           Ι
        0.0 I
u
а
           I
1
           Ι
       -2.0 I
           4.0
                   5.0
                            6.0
                                      7.0
                                               8.0
                                                      9.0 10.0
                                     fitted values
                                     Normal plot
           I
        2.0 I
r
           Ι
           Ι
е
S
i
           Ι
d
           Ι
        0.0 I
u
а
1
           Ι
           Ι
           Ι
           Ι
       -2.0 I
          -2.4
                 -1.6
                           -0.8
                                     0.0
                                               0.8 1.6 2.4
                               expected Normal quantiles
```

24 CALC FS=5.9270/0.6740 & pS=1-FPROB(FS; 4; 12) : PRINT FS,pS

```
27 "
 -28 **** Tukey''s one-degree-of-freedom-for-non-additivity.
-29 **** It is the term designated covariate in the following analysis
 -30
  31 AKEEP [FIT=Fit]
32 CALC ResSq=Fit*Fit
   33 ANOVA [PRINT=*] ResSq; RES=ResSq
   34 COVAR ResSq
35 ANOVA [PRINT=A; FPROB=Y] Moisture
                                                                "A computational trick"
35.....
**** Analysis of variance (adjusted for covariate) ****
Variate: Moisture
Covariate: ResSq
Source of variation d.f. s.s.
                                                             m.s. v.r. cov.ef. F pr.
                                     4 28.8853 7.2213 9.93
Plant stratum
                                     4 23.7081 5.9270 8.15
Size stratum
Plant.Size stratum

    4
    0.6273
    0.1568
    0.22
    1.00
    0.924

    1
    0.1512
    0.1512
    0.21
    1.00
    0.657

    1
    0.0929
    0.0929
    0.13
    1.00
    0.728

    2
    0.3831
    0.1916
    0.26
    1.00
    0.773

    1
    0.0853
    0.0853
    0.12
    0.738

    11
    8.0026
    0.7275
    0.93

Time
  Lin
   Quad
  Deviations
Covariate
Residual
                                    24
                                            61.3086
Total
```

Step 1: Set up hypotheses

- a)  $H_0$ :  $\tau_1 = \tau_2 = \tau_3 = \tau_4 = \tau_5$  $H_1$ : at least one pair of population time means is different
- b)  $H_0$ :  $\sigma_P^2 = 0$  $H_1$ :  $\sigma_P^2 > 0$
- c)  $H_0$ :  $\gamma_1 = \gamma_2 = \gamma_3 = \gamma_4 = \gamma_5$  $H_1$ : at least one pair of population size means is different

## Step 2: Calculate test statistics

The analysis of variance table for a Latin Square is:

Source	df	SSq	MSq	E[MSq]	F	Prob
Plant	4	28.8853	7.2213	$\sigma_{PS}^2 + 5\sigma_{P}^2$	10.71	<0.001
Size	4	23.7081	5.9270	$\sigma_{PS}^2 + f_{S}(\psi)$	8.79	0.002
Plant.Size	16					
Times	4	0.6273	0.1568	$\sigma_{PS}^2 + f_{\!\scriptscriptstyle T} \! \left( \psi \right)$	0.23	0.915
Linear	1	0.1512	0.1512		0.22	0.644
Quadratic	1	0.0929	0.0929		0.14	0.717
Deviations	2	0.3831	0.1916		0.28	0.758
Residual	12	8.0879	0.6740	$\sigma_{ t PS}^2$		
Non-additivity	1	0.0853	0.0853		0.12	0.738
Deviations	11	8.0026	0.7275			
Total	24	61.3806				

Step 3: Decide between hypotheses

There analysis indicates that there are no significant differences between the treatments, in spite of a very effective Latin Square (both Plant and Size are significant). Further, there is no evidence of transformable non-additivity. However, the plot of residuals-versusfitted-values indicates that there is an observation for which has an extremely low residual. The Normal Probability plot is also exhibiting The warning message unde rthe analysis of the same problem. variance table indicates that this observation is Plant 5 and Size 4 or the 20<sup>th</sup> observation. This observation is set to missing and the analysis repeated to see what effect the outlier has on the analysis.

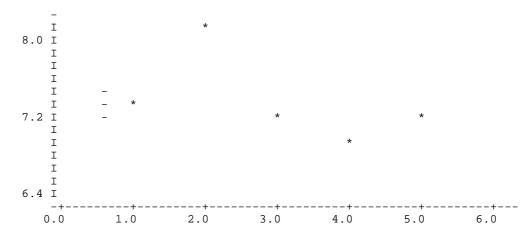
<sup>36 &</sup>quot;Analysis with Plant 5 Size 4 missing"

<sup>37</sup> CALC Moisture\$[20]=!(\*)

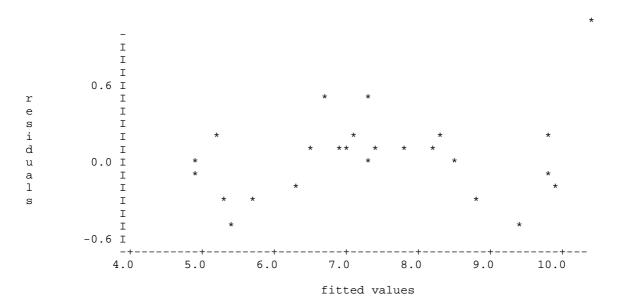
<sup>38</sup> COVAR 39 ANOVA [FPROB=Y; PSE=LSD] Moisture

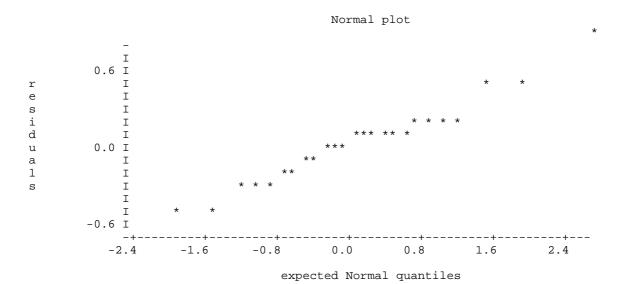
Mariate: Moisture					
Source of variation	d.f.(m.v.)	s.s.	m.s.	v.r.	F pr.
Plant stratum	4	26.9192	6.7298	47.60	
Size stratum	4	31.9976	7.9994	56.58	
Plant.Size stratum					
Time	4	3.7668			
Lin	1	0.8292	0.8292 0.0185	5.87	0.034
Quad	1				
Deviations	2	2.9190		10.32	0.003
Residual	11(1)	1.5551	0.1414		
Total	23(1)	59.3432			
* MESSAGE: the follow	ing units have	large resid	duals.		
Plant 5 Size 1					
ranco size i	-0.535	s.∈. U.∠49			
***** Tables of means	****				
Variate: Moisture					
Grand mean 7.351					
Time 1	2	3 4	5		
	8.072 7.20				
*** Least significant	differences o	f means (5%	level) ***	k	
Table	Time				
rep.	5				
d.f.	11				
l.s.d. 0	.5234				
(Not adjusted for mis	sing values)				
***** Missing values	****				
Variate: Moisture					
Unit estimate					
20 9.519					
Max. no. iterations 4					
40 CALC FP=6.7298/	0.1414 & pP=1-	FPROB(FP; 4	; 11) : PRI	INT FP,p	P
FP 47.59 0.000000	рР 705				
41 CALC FS=7.9994/	0.1414 & pS=1-	FPROB(FS; 4	; 11) : PRI	INT FS,p	S
FS 56.57 0.000000	pS 288				
30.37 0.000000	۵00				
42 AGRAPH [GRAPH=1	ine] XFACTOR=T	ime; BAR=*			

Means for Time



43 APLOT METHOD=fit, normal





```
**** Tukey''s one-degree-of-freedom-for-non-additivity.
 -45
     **** It is the term designated covariate in the following analysis
 -46
 -47
  48 AKEEP [FIT=Fit]
 49 CALC ResSq=Fit*Fit
50 ANOVA [PRINT=*] ResSq; RES=ResSq
  51 COVAR ResSq
                                          "A computational trick"
  52 ANOVA [PRINT=A; FPROB=Y] Moisture
52.....
**** Analysis of variance (adjusted for covariate) ****
Variate: Moisture
Covariate: ResSq
                    d.f.(m.v.)
Source of variation
                                             m.s. v.r. cov.ef. F pr.
                                   s.s.
Plant stratum
Covariate
                        1
                                  0.0051
                                           0.0051
                                                     0.00
                                                                  0.982
                        3
                                            8.9714
Residual
                                 26.9141
                                                     76.89
                                                             0.75
Size stratum
                                          12.5785
                                 12.5785
                                                      1.94
Covariate
                        1
                                                                  0.258
                        3
                                                             1.24
Residual
                                 19.4191
                                             6.4730
                                                     55.48
Plant.Size stratum
                                                           1.00 0.005
1.00 0.027
Time
                        4
                                  3.5191
                                            0.8798
                                                     7.54
                                                    6.69
                                  0.7804
                                            0.7804
 Lin
                        1
                                                    0.11
  Quad
                                 0.0129
                                           0.0129
                                                            1.00 0.746
                                 2.7258
                                            1.3629 11.68
0.3883 3.33
                        2
                                                            1.00 0.002
 Deviations
                                                    3.33
Covariate
                        1
                                  0.3883
                                                                  0.098
                                                             1.21
Residual
                       10(1)
                                  1.1668
                                           0.1167
Total
                       23(1)
                                 59.3432
```

	ORIGINAL ANALYSIS				MISSING VALUE ANALYSIS				
Source	df	MSq	F	Prob	df	MSq	F	Prob	
Plant	4	7.2213	10.71	<0.001	4	6.7298	47.60	< 0.001	
Size	4	5.9270	8.79	0.002	4	7.9994	56.58	< 0.001	
Plant.Size	16				15				
Times	4	0.1568	0.23	0.915	4	0.9417	6.66	0.006	
Linear	1	0.1512	0.22	0.644	1	0.8292	5.87	0.034	
Quadratic	1	0.0929	0.14	0.717	1	0.0185	0.13	0.724	
Deviations	2	0.1916	0.28	0.758	2	1.4595	10.32	0.003	
Residual	12	0.6740			11 <sup>†</sup>	0.1414			
Non-additivity	1	0.0853	0.12	0.738	1	0.3883	3.33	0.098	
Deviations	11	0.7275			10	0.1167			
Total	24				23				

<sup>&</sup>lt;sup>†</sup>The Residual degrees of freedom have been reduced by one to take account of the missing value.

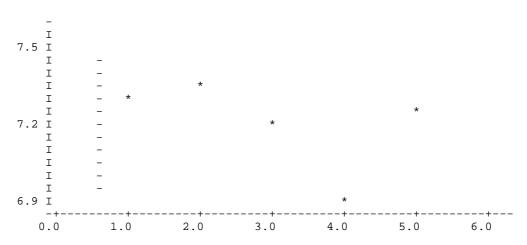
The analysis now indicates that there are differences between the Times and that the Times means exhibit a linear trend from which there are significant deviations. Consequently we can only examine treatment differences using a multiple comparisons procedure.

The Times means (adjusted for the missing value) and accompanying plot are:

		Time			
1	2	3	4	5	s.e.d
7.318	8.072	7.206	6.900	7.260	0.2378

25 AGRAPH [GRAPH=line] XFACTOR=Time; BAR=\*

Means for Time



It is clear that the trend is not described by either a straight line or a quadratic curve.

VI.4 An experimenter wants to investigate the effects of four different rations on the apparent consumption of total carbohydrates (as a percentage) by calves. He has available four calves of around 280 kg. He plans to use a Latin square for two reasons. Firstly, so that each calf receives the four rations, one in each of four periods. Secondly, so that differences, such as climatic differences, between the periods are eliminated from treatment differences. The experimenter is willing to run a 5% chance of making a type I error and would like to have a 95% chance of detecting any difference of 7.5% or more in the apparent consumption between rations. A variance of 10% for the animal-period combinations is expected in the experiment. Will the Latin square have the desired power?

ANOVAPower.xls is used to compute the power and the values in the cells below the headings are as follows:

sample	•		DF	central	no. values			lambda	power
size (r)		-ator	denomin- ator	r	in a mean (m)		deviation		
4	0.05	3	6	4.7571	4	7.5	3.162278	11.25	0.5155

The power is well below the desired 95%.