## THE DESIGN AND MIXED-MODEL ANALYSIS OF EXPERIMENTS

# PRACTICAL XII SOLUTIONS

XII.1 Example XII.3, *Internal parasites in calves*, involved an experiment to compare two methods for the control of intestinal parasites in calves involved 60 calves. At the start of the grazing season the calves were randomly assigned to the two methods so that 30 calves received each method. The weights of each calf were measured at weeks 0, 2, 4, 6, 8, 10,12, 14, 16, 18 and 19.

What are the components of the study?

1. Observational unit a calf in a week

2. Response variable Weight

3. Unrandomized factors Calf, Week

4. Randomized factors Method

5. Type of study a repeated measurements study

with calves in a CRD

What is the experimental structure for this experiment?

Structure	Formula
unrandomized	60 Calf*11 Week
randomized	2 Method*Week

What are the terms derived from the experimental structure? Write out the Hasse diagram for each structure formula or use the rule for completely crossed structures.

Calf\*Week = Calf + Week + Calf.Week

Method\*Week = Method+ Week + Method.Week

The degrees of freedom can be computed using the rule for completely crossed structures.

Which factors would you suggested be regarded as fixed and which as random? What are the maximal expectation and variation models?

Fixed factors: Method and Week

Random factor: Calf

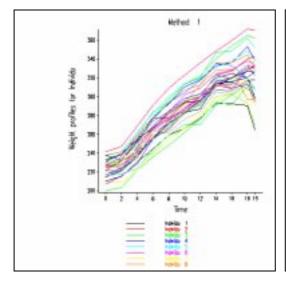
E[Y] = Method.Week

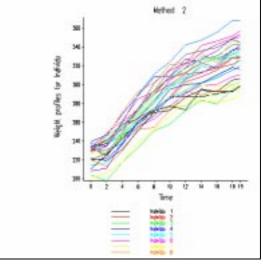
Var[Y] = Calf + Calf.Week

Write down the analysis of variance table, including the expected mean squares for the lines in it.

Source	df	E[MSq]
Calf	59	
Method	1	$\sigma_{\text{CW}}^2$ +11 $\sigma_{\text{C}}^2$ + $f_{\text{M}}(\psi)$
Residual	58	$\sigma_{\mathrm{CW}}^2$ +11 $\sigma_{\mathrm{C}}^2$
Week	10	$\sigma_{CW}^2 + f_{W}(\psi)$
Calf.Week	590	
Method.Week	10	$\sigma_{CW}^2 \qquad \qquad + f_{MW} \left( \psi \right)$
Residual	580	$\sigma_{\sf CW}^2$
Total	569	

a) The data are stored in the file *rmecalf.gsh* in the *G:\Disciplina\Genstat* directory. Obtain plots of the individual profiles for the two methods.

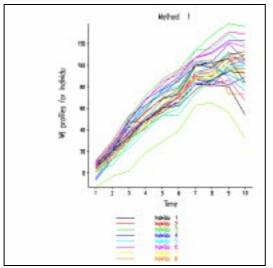


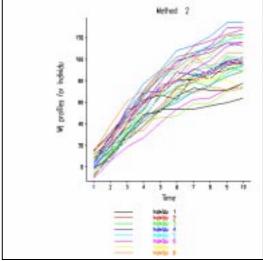


b) Note that the first observation is at week 0 and represents differences between the calves at the time of application of the treatment and not of any treatment effect. Use the following Genstat instructions to subtract the observation at week 0 from all other observations:

```
POINTER Wt; !p(Weight[2,4...18,19])
CALC #Wt=#Wt-Weight[0]
```

c) Having done that obtain plots of the individual difference profiles for the two methods. What problems do you think there would be in doing a single analysis of variance of all the measurements?





The variance is clearly increasing over time so that a transformation would have to be found to stabilize the variance.

d) The profiles are displaying some curvature so a linear relationship is likely to be inadequate. Fit orthogonal polynomials up to degree 6 to the profile for each calf. Analyse the resulting coefficients. What do you conclude?

The Genstat output below contains the commands, and associated output, to produce the diagram above and the analysis of the polynomials fitted to the differences from week 0.

It will be found that the coefficients of degree 3, 4 and 6 are significantly different between the treatments. It would appear that simple polynomials (up to degree 3) are inadequate in capturing the differences between the treatments.

```
Genstat 5 Release 4.1 (PC/Windows NT) 02 May 2000 14:43:45 Copyright 1998, Lawes Agricultural Trust (Rothamsted Experimental Station)
```

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

3 "Data taken from File: D:/ANALYSES/LM/REPEATMEASURE/RMECALF.GSH"

```
4 DELETE [redefine=yes] Weeks, Calves, Methods, Weights
     FACTOR [modify=yes;nvalues=660;levels=!(0,2,4,6,8,10,12,14,16,18,19)] Weeks
  6 READ Weeks; frepresentation=ordinal
                 Values
   Identifier
                          Missing
                                      Levels
        Weeks
                    660
 26 FACTOR [modify=yes;nvalues=660;levels=60] Calves
    READ Calves; frepresentation=ordinal
   Identifier
                 Values
                         Missing
                     660
       Calves
 53 FACTOR [modify=yes;nvalues=660;levels=2] Methods
 54 READ Methods; frepresentation=ordinal
   Identifier
                  Values
                           Missing
                                      Levels
      Methods
                    660
 73 VARIATE [nvalues=660] Weights
 74 READ Weights
   Identifier
                Minimum
                             Mean
                                     Maximum
                                                Values
                                                         Missina
                   198.0
                             283.5
                                      372.0
      Weights
                                                   660
110
111
-112
     **** separate data for Weeks and Calf profiles
-113
114
     SUBSET [CONDITION=Weeks==0] OLD=Calves, Methods; \
115
                                  NEW=Calf, Method
     FOR i=0,2...18,19
116
117
         SUBSET [CONDITION=Weeks==i] OLD=Weights; NEW=Weight[i]
118
     ENDFOR
119 PRINT Calf, Method, Weight[0,2...18,19]; FIELD=9; DEC=0
           Method Weight[0] Weight[2] Weight[4] Weight[6] Weight[8] Weight[10]
       1
                       210
                                215
                                       230
                                                  244
                                                            259
                                                                      266
               1
                        230
                                 240
                                          258
                                                   277
                                                             277
                                                                      293
       3
                        226
                                 233
                                          248
                                                   277
                                                             297
                                                                      313
                1
        4
                 1
                        233
                                 239
                                          253
                                                   277
                                                             292
                                                                      310
       5
                                 241
                                                   282
                1
                       238
                                          262
                                                             300
                                                                      314
                       225
       6
                1
                                 228
                                          237
                                                   261
                                                             271
                                                                      288
       7
                1
                        224
                                 225
                                          239
                                                   257
                                                             268
                                                                      290
                       237
                                         255
                                                   276
                                                             293
       8
                1
                                241
                                                                      307
                       237
       9
                1
                                 224
                                         234
                                                   239
                                                             256
                                                                      266
       10
                1
                       233
                                 239
                                          259
                                                   283
                                                             294
                                                                      313
                       217
                                          235
       11
                1
                                 222
                                                   256
                                                             267
                                                                      285
                       228
                                 223
                                          246
                                                             277
      12
                1
                                                   266
                       241
                                                   290
                                          268
      13
                1
                                 247
                                                             309
                                                                      323
       14
                 1
                        221
                                 221
                                          240
                                                   253
                                                             273
                                                                      282
                       217
                                                   259
      15
                1
                                 220
                                          235
                                                             262
                                                                      276
      16
                1
                       214
                                 221
                                          237
                                                   256
                                                             271
                                                                      283
      17
                1
                        224
                                 231
                                          241
                                                   256
                                                             265
                                                                      283
      18
                 1
                        200
                                 203
                                          221
                                                   236
                                                             248
                                                                      262
      19
                       238
                                 232
                                          252
                                                   268
                                                             285
                1
                                                                      298
       20
                1
                        230
                                 222
                                          243
                                                   253
                                                             268
                                                                      284
       21
                1
                        217
                                 224
                                          242
                                                   265
                                                             284
                                                                      302
       22
                       209
                                 209
                                          221
                                                   238
                                                             256
                                                                      267
                1
       23
                1
                       224
                                 227
                                          245
                                                   267
                                                             279
                                                                      294
       24
                1
                        230
                                 231
                                          244
                                                   261
                                                             272
                                                                      283
       25
                                                   243
                                                             259
                 1
                        216
                                 218
                                          223
                                                                      270
       26
                1
                       231
                                 239
                                          254
                                                   276
                                                             294
                                                                      304
                       207
                                                   255
                                                             275
       2.7
                1
                                 216
                                          228
                                                                      285
       28
                 1
                        227
                                 236
                                          251
                                                   264
                                                             276
                                                                      287
       29
                1
                       221
                                 232
                                          251
                                                   274
                                                             284
                                                                      295
                       233
       30
                1
                                 238
                                         254
                                                   266
                                                             282
                                                                      294
                 2
                        233
                                 224
                                          245
                                                   258
                                                             271
                                                                      287
       31
                                                   273
                2
                       231
                                                             290
       32
                                 238
                                          260
                                                                      300
       33
                2
                       232
                                237
                                         245
                                                   265
                                                             285
                                                                      298
                       239
       34
                 2
                                 246
                                          268
                                                   288
                                                             308
                                                                      309
       35
                2
                        215
                                 216
                                          239
                                                   264
                                                             282
                                                                      299
```

36	2	236	226	242	255	263	277
37	2	219	229	246	265	279	292
38	2	231	245	270	292	302	321
39	2	230	228	243	255	272	276
40	2	232	240	247	263	275	286
41	2	234	237	259	289	311	324
42	2	237	235	258	263	282	304
43	2	229	234	254	276	294	315
44	2	220	227	248	273	290	308
45	2	232	241	255	276	293	309
46	2	210	225	242	260	272	277
47	2	229	241	252	265	274	285
48	2	204	198	217	233	251	258
49	2	220	221	236	260	274	295
50	2	233	234	250	268	280	298
51	2	234	234	254	274	294	306
52	2	200	207	217	238	252	267
53	2	220	213	229	252	254	273
54	2	225	239	254	269	289	308
55	2	236	245	257	271	294	307
56	2	231	231	237	261	274	285
57	2	208	211	238	254	267	287
58	2	232	248	261	285	292	307
59	2	233	241	252	273	301	316
60	2	221	219	231	251	270	272
aiah+[12]	Weight[14]	Weight	161 Weigh	+[10] Wai	ah+[10]		
277	292	292	.10) Weigh 290	264	giic[19]		
300	323	327	340	343			
322	340	354	365	362			
318	333	336	353	338			

Woight[12]	Woight[14]	Woight[16]	l Woight[	18] Weight[19]
weight[12] 277	292	292	290	16) Weight[19] 264
300	323	327	340	343
322	340	354	365	362
318	333	336	353	338
319	331	338	348	338
300	316	319	333	330
304	313	310	318	318
312	336	336	344	328
276	300	302	293	269
320	347	348	362	352
295	317	315	308	301
300	312	308	328	333
336	348	359	372	370
292	307	306	317	318
284	305	303	315	317
287	314	316	320	298
295	314	313	328	334
276	294	291	311	310
303	320	324	320	327
290	316	314	330	330
309	324	328	338	334
281	295	301	309	289
312	328	329	297	297
294	318	320	333	338
270	290	301	314	297
317	335	333	319	307
296	314	319	330	330
297	315	309	313	294
300	323	319	333	322
295	310	320	327	326
287	287	290	293	297
311	313	317	321	326
304	319	317	334	329
327	324	327	336	341
307	321	328	332	337
290	299	300	308	310
299	299	298	300	290
322	334	323	337	337
277	289	289	300	303
294	302	308	319	326
342	347	355	368	368
318	327	336	349	353
323	341	346	352	357
322	326	330	342	343

```
330
                    326
                            329
     310
                                       330
     273
              295
                      292
                               305
                                       306
     303
              308
                      315
                               328
                                       328
     272
              283
                     279
                              295
                                       298
     300
              301
                             318
                     310
                                      316
     308
              319
                      318
                               336
                                       333
                             349
                     343
     318
              334
                                       350
                     282
                             284
     284
             282
                                      288
                    294
327
     293
              289
                              292
                                       298
                              347
     313
              324
                                       344
                     328
                             328
     317
             327
                                      325
                     307
                              315
     291
              301
                                       320
     306
              312
                      320
                              337
                                       338
                              328
     312
              323
                      318
                                       329
     332
              336
                     339
                              348
                                       345
     287
              294
                      292
                              292
                                       299
120 FACTOR [LEV=30] Individuals
121 CALC Individuals=NEWLEVELS(Calf; !v((1...30)2))
122
     DREPMEASURES [GROUPS=Method, Individuals] DATA=Weight
******* Warning (Code HG 20). Statement 130 in Procedure DREPMEAS
Command: DGRAPH [WINDOW= window; KEYWINDOW= keywindow; TITLE= Title_] GroupMns
Key window full
****** Warning (Code HG 20). Statement 130 in Procedure DREPMEAS
Command: DGRAPH [WINDOW= window; KEYWINDOW= keywindow; TITLE= Title_] GroupMns
Key window full
123 DREPMEASURES [GROUPS=Method] DATA=Weight
124
     **** analyse differences from week 0 polynomial coefficients to degree 6
-125
-126
127 POINTER Wt; !p(Weight[2,4...18,19])
128 CALC #Wt=#Wt-Weight[0]
129 DREPMEASURES [GROUPS=Method,Individuals] DATA=Wt
****** Warning (Code HG 20). Statement 130 in Procedure DREPMEAS
Command: DGRAPH [WINDOW= window; KEYWINDOW= keywindow; TITLE= Title_] GroupMns
Key window full
****** Warning (Code HG 20). Statement 130 in Procedure DREPMEAS
Command: DGRAPH [WINDOW= window; KEYWINDOW= keywindow; TITLE= Title_] GroupMns
Key window full
130 DREPMEASURES [GROUPS=Method] DATA=Wt
131 VORTHPOL [MAXDEGREE=6] Wt; CONTRAST=pol
132 BLOCK Calf
133 TREAT Method
134 FOR k=0...6
     ANOVA [FPROB=Y; PSE=LSD] pol[k]
135
 136
       APLOT METHOD=fit, normal
137 ENDFOR
137.....
**** Analysis of variance ****
Variate: pol[0]
Source of variation d.f.
                               s.s.
                                         m.s. v.r. F pr.
Calf stratum
                                           0.1 0.00 0.982
                                 0.1
Method
                         1
Residual
                        58
                               7743.5
                                          133.5
                               7743.6
Total
                        59
```

<sup>\*</sup> MESSAGE: the following units have large residuals.

```
Calf 9 -34.9 s.e. 11.4
**** Tables of means ****
Variate: pol[0]
Grand mean 63.9
  Method 1 2 63.8 63.9
*** Least significant differences of means (5% level) ***
Table
                Method
rep.
d.f.
                    58
                  5.97
l.s.d.
       40.0 I
           I
е
           Ι
i
          I
d
          I
       0.0 I
u
а
           Ι
1
           Ι
s
           I
      -40.0 I
       63.840 63.852 63.864 63.876 63.888 63.900 63.912
                                  fitted values
                                   Normal plot
       40.0 I
r
е
i
                                 *22*2
**222222**
d
          I
       0.0 I
u
           Ι
а
           Ι
           Ι
          I *
      -40.0 I
         -2.4
                -1.6 -0.8 0.0 0.8 1.6 2.4
```

expected Normal quantiles

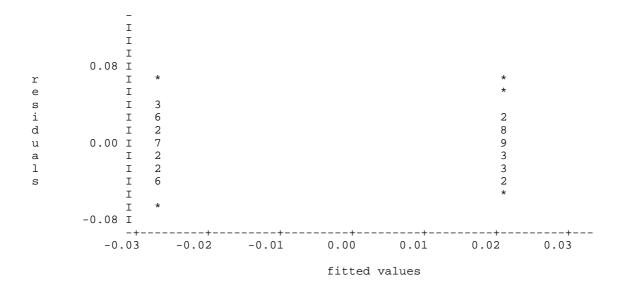
```
137.....
**** Analysis of variance ****
Variate: pol[1]
Source of variation d.f. s.s.
                                    m.s. v.r. F pr.
Calf stratum
                                 4.119 1.09 0.302
3.796
                     1
                          4.119
Method
Residual
                     58
                        220.182
                     59
                         224.302
Total
***** Tables of means *****
Variate: pol[1]
Grand mean 10.86
          1
  Method
         11.12 10.60
*** Least significant differences of means (5% level) ***
                Method
                   30
rep.
d.f.
                   58
                1.007
l.s.d.
          I
          I
       4.0 I*
          Ι2
          I3
е
s
          Ι2
i
          I*
d
          I2
       0.0 I2
u
          Ι6
а
1
          Ι5
          I3
s
          T *
      -4.0 I
                                10.9 11.0 11.1 11.2
         10.6 10.7 10.8
                                 fitted values
                                  Normal plot
       4.0 I
r
е
          I
                                     *2
*2*2*
**2*
2*
i
          Ι
                              * 222*
* 22*22*
·*
d
          Ι
       0.0 I
u
а
          I
1
          Т
S
          Ι
          I
      -4.0 I *
                -1.6 -0.8 0.0 0.8 1.6 2.4
         -2.4
                            expected Normal quantiles
```

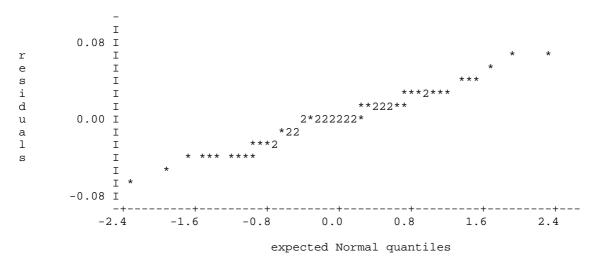
```
137.....
**** Analysis of variance ****
Variate: pol[2]
Source of variation d.f. s.s.
                                     m.s. v.r. F pr.
Calf stratum
                          0.0083 0.0083 0.05 0.829
10.2117 0.1761
                      1
Method
Residual
                     58
                         10.2117
                     59
                          10.2199
Total
* MESSAGE: the following units have large residuals.
Calf 23
             -1.220
                   s.e. 0.413
             -1.012 s.e. 0.413
Calf 26
**** Tables of means ****
Variate: pol[2]
Grand mean -1.090
  Method
              1
         -1.102 -1.078
*** Least significant differences of means (5% level) ***
Table
                Method
rep.
d.f.
                   58
                0.2169
l.s.d.
          I
          Ι
          I
       1.5 I
          I
r
е
          Ι
s
i
          Т
                                                         7
d
                 7
      0.0 I
                                                         5
u
                                                         9
          Ι
1
          I
                                                         3
s
          Ι
          Ι
      -1.5 I
       -1.105 -1.100 -1.095 -1.090 -1.085 -1.080 -1.075
```

fitted values

```
Normal plot
           Т
       1.5 I
r
           Ι
           Ι
е
           Ι
s
                                               **2*** *
i
           I
                                         *222*2
d
                                    222222
       0.0 I
u
                           *2**2*22*2
а
           Ι
1
           Ι
           Т
S
           Ι
           Ι
       -1.5 I
                          -0.8 0.0 0.8 1.6
         -2.4
                  -1.6
                                                              2.4
                              expected Normal quantiles
137.....
**** Analysis of variance ****
Variate: pol[3]
Source of variation d.f. s.s. m.s. v.r. F pr.
Calf stratum
Method
                       1
                          0.31775
                                    0.31775
                                            24.60 <.001
Residual
                      58
                           0.74931
                                     0.01292
                           1.06706
Total
                      59
* MESSAGE: the following units have large residuals.
Calf 9
                    s.e. 0.112
s.e. 0.112
             -0.380
Calf 12
              0.278
***** Tables of means *****
Variate: pol[3]
Grand mean -0.057
           1
  Method
                     2
          -0.130
                 0.016
*** Least significant differences of means (5% level) ***
Table
                 Method
rep.
                    30
                    58
d.f.
                 0.0587
l.s.d.
           Ι
           Ι
           Ι
        0.4 I
           I
           I
е
S
           Ι
i
           Ι
                  7
d
           Ι
u
        0.0 I
                  2
                                                            9
а
           Т
1
           Ι
                  5
                  2
           Ι
S
           Ι
       -0.4 I
                               -0.06 -0.03 0.00 0.03
        -0.15
                -0.12 -0.09
                                   fitted values
```

r e s i d u a 1 s	0.4 I I I I I I I I I I I I I I I I I I I	*** ; * *	-	2*22 *222222	2*2	* *** *	*
	-2.4	-1.6	-0.8	0.0	0.8	1.6	2.4
			expec	ted Normal	quantile	es	
137							
**** Aı	nalysis of v	rariance ***	* * *				
Variate	: pol[4]						
Source	of variation	d.f.	s.s.	m.s.	v.r.	F pr.	
Calf st: Method Residua: Total		58		0.0349516 0.0008208	42.58	<.001	
* MESSA	GE: the foll	owing units	s have larg	e residuals			
Calf 7	0.0	731 s.e.	0.0282				
**** To	ables of mea	ns ****					
Variate	: pol[4]						
Grand me	ean -0.0029	)					
Metho		2 0.0212					
*** Lea:	st significa	nt differer	nces of mea	ns (5% leve	1) ***		
Table rep. d.f. l.s.d.		Method 30 58 0.01481					





\*\*\*\*\* Analysis of variance \*\*\*\*\*

Variate: pol[5]

Source of variation d.f. s.s. m.s. v.r. F pr.

Calf stratum

1 0.0000081 0.0000081 0.03 0.863 58 0.0155930 0.0002688 Method

Residual

59 0.0156011 Total

\* MESSAGE: the following units have large residuals.

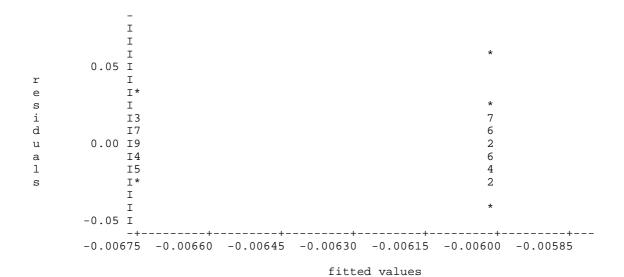
0.0574 s.e. 0.0161 -0.0454 s.e. 0.0161 Calf 23 Calf 25

```
**** Tables of means ****
Variate: pol[5]
Grand mean -0.0064

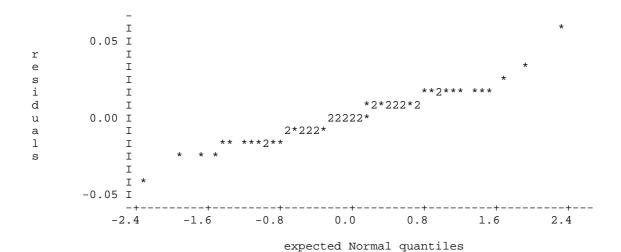
Method 1 2
-0.0060 -0.0067
```

\*\*\* Least significant differences of means (5% level) \*\*\*

Table	Method
rep.	30
d.f.	58
l.s.d.	0.00847



Normal plot



137.....

\*\*\*\*\* Analysis of variance \*\*\*\*

Variate: pol[6]

Source of variation d.f. s.s. m.s. v.r. F pr.

Calf stratum

Method 1 0.00045260 0.00045260 9.82 0.003

58 0.00267195 0.00004607

Total 59 0.00312455

\* MESSAGE: the following units have large residuals.

Calf 23 0.02241 s.e. 0.00667

\*\*\*\* Tables of means \*\*\*\*

Variate: pol[6]

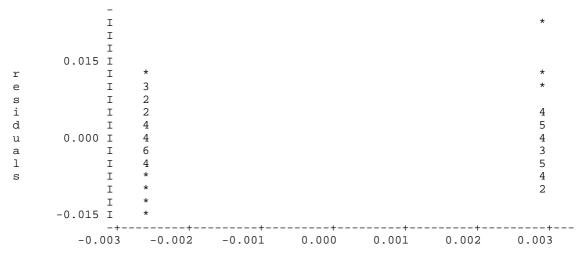
Residual

Grand mean 0.00012

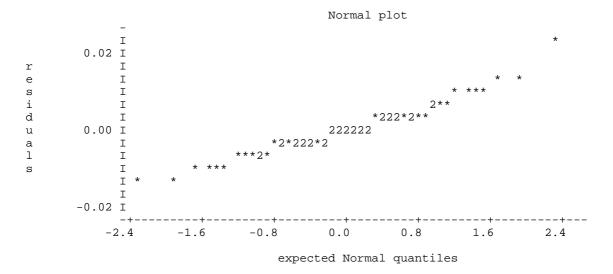
Method 1 2 0.00287 -0.00262

\*\*\* Least significant differences of means (5% level) \*\*\*

Table Method rep. 30 d.f. 58 l.s.d. 0.003508



fitted values



XII.2 In an experiment to study the influence of various strains of the Newcastle Disease virus on the growth of chickens, 200 chicks were divided into 20 groups in such a way as to ensure that the group averages were nearly equal. These 20 groups were then divided into four blocks of five and the five treatments (four strains of virus and an uninfected control) were randomized to the groups within a block. The average weight of the chicks in a group was measured at the start of the experiment and at weekly intervals thereafter.

What are the components of the study?

1. Observational unit a group of chicks

2. Response variable Weight

3. Unrandomized factors Block, Group, Week

4. Randomized factors Virus

5. Type of study a repeated measures experiment

with groups in an RCBD

What is the experimental structure for this experiment?

Structure	Formula
unrandomized	(4 Block/5 Group)*5 Week
randomized	5 Virus*Week

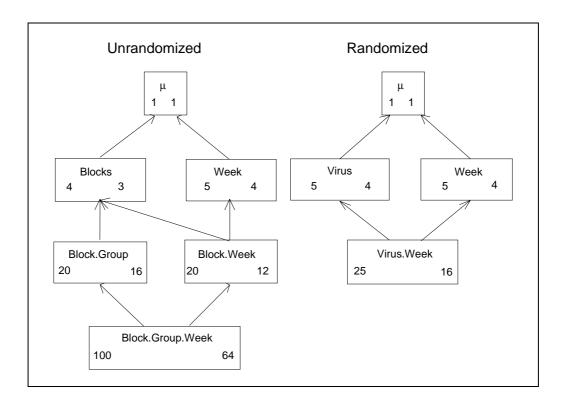
What are the terms derived from the experimental structure? Write out the Hasse diagram for each structure formula or use the rule for completely crossed structures.

(Block/Group)\*Week = (Block + Block.Group)\*Week

= Block + Block.Group + Block.Week

+ Block.Group.Week

Virus\*Week = Virus + Week + Virus.Week



Which factors would you suggested be regarded as fixed and which as random? What are the maximal expectation and variation models?

Fixed factors: Week and Virus

Random factors: Block and Group

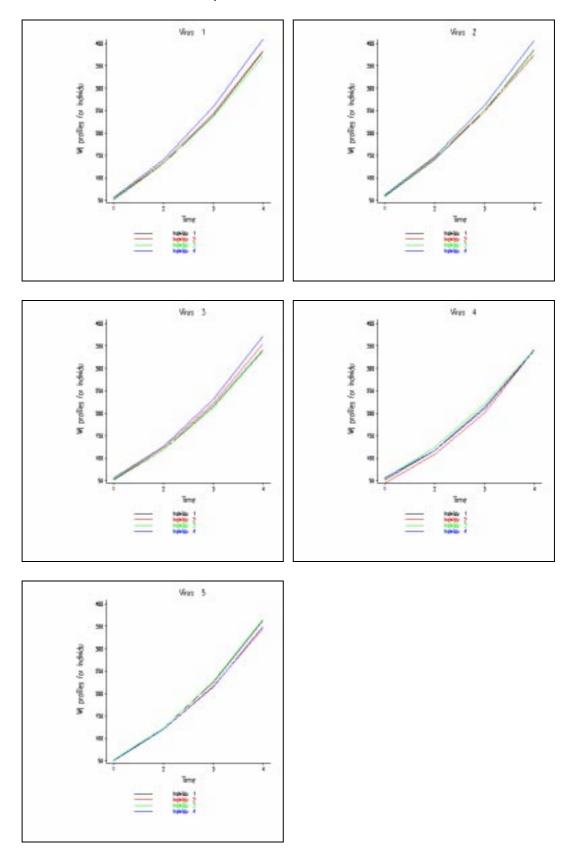
E[Y] = Virus.Week

Var[Y] = Block + Block.Group + Block.Week + Block.Group.Week

Write down the ANOVA table, including the E[MSq]s for the lines in it.

Source	df	E[MSq]
Block	3	$\sigma_{BGW}^2$ +5 $\sigma_{BG}^2$ +5 $\sigma_{BW}^2$ +25 $\sigma_{B}^2$
Block.Group	16	
Virus	4	$\sigma_{BGW}^2$ +5 $\sigma_{BG}^2$ + $f_{V}(\psi)$
Residual	12	$\sigma_{BGW}^2$ +5 $\sigma_{Bg}^2$
Week	4	$\sigma_{BGW}^2$ +5 $\sigma_{BW}^2$ + $f_{W}\left(\psi\right)$
Block.Week	12	$\sigma_{BGW}^2$ +5 $\sigma_{BW}^2$
Block.Group.Week	64	
Virus.Week	16	$\sigma_{BGW}^2 + f_{VW} \left( \Psi \right)$
Residual	48	$\sigma_{BGW}^2$
Total	99	

a) The differences from the initial reading are stored in the file *RMEChickVirusDiffs.gsh* in the *G:\Disciplina\Genstat* directory. Obtain plots of the individual difference profiles for the five treatments.



### The following is the Genstat output for producing these.

Genstat 5 Release 4.1 (PC/Windows NT) 03 May 2000 11:50:48 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/REPEATMEASURE/RMECHICKVIRUSDIFFS.GSH" 4 DELETE [redefine=yes] Weeks, Blocks, Groups, Viruses, Weights FACTOR [modify=yes;nvalues=80;levels=4] Weeks 6 READ Weeks; frepresentation=ordinal Identifier Values Missing Levels Weeks 80 10 FACTOR [modify=yes;nvalues=80;levels=4] Blocks 11 READ Blocks; frepresentation=ordinal Identifier Values Missing Levels Blocks 8.0 15 FACTOR [modify=yes;nvalues=80;levels=5] Groups 16 READ Groups; frepresentation=ordinal Identifier Values Missing Levels 8.0 Groups 20 FACTOR [modify=yes;nvalues=80;levels=5] Viruses 21 READ Viruses; frepresentation=ordinal Identifier Values Missing Levels Viruses 80 25 VARIATE [nvalues=80] Weights 26 READ Weights Identifier Minimum Mean Maximum Values Missing 194.3 45.1 Weights 410.6 8.0 34 35 -36 \*\*\*\* separate data for Weeks and Group profiles -37 38 SUBSET [CONDITION=Weeks==1] OLD=Blocks, Groups, Viruses; \ 39 NEW=Block, Group, Virus 40 FOR i=1...4 SUBSET [CONDITION=Weeks==i] OLD=Weights; NEW=Weight[i] 41 ENDFOR 43 PRINT Block, Group, Virus, Weight[1...4]; FIELD=9; DEC=2 Block Virus Weight[1] Weight[2] Weight[3] Weight[4] Group 1 52.00 130.85 240.15 382.55 1 1 1 2 59.30 140.60 249.40 387.10 118.35 

 3
 3
 52.05
 118.35
 217.15
 341.75

 4
 4
 51.50
 116.55
 213.55
 340.15

 5
 5
 50.35
 122.60
 225.80
 364.20

 1
 1
 54.30
 134.55
 245.15
 383.65

 2
 2
 60.65
 144.55
 248.35
 373.45

 3
 3
 53.40
 122.15
 223.15
 355.65

 4
 4
 45.10
 108.50
 201.50
 342.40

 5
 5
 50.00
 119.85
 217.15
 346.65

 1
 1
 54.45
 132.15
 237.35
 372.45

 2
 2
 59.85
 143.35
 253.05
 378.05

 3
 3
 52.50
 118.00
 212.10
 338.70

 4
 4
 54.80
 124.15
 221.15
 343.05

 5
 51.55
 122.95
 227.35
 366.35

 52.05 51.50 1 3 3 217.15 341.75 1 1 2 2 2 2 2 3 3 3

4	1	1	56.75	140.75	259.85	410.65
4	2	2	61.80	147.95	261.95	407.75
4	3	3	55.55	126.20	231.70	372.40
4	4	4	55.40	116.70	209.70	340.70
4	5	5	50.90	119.65	216.25	351.25

- 44 DUPLICATE OLD=Block; NEW=Individuals
- 45 DREPMEASURES [GROUPS=Virus, Individuals] DATA=Weight
- 46 DREPMEASURES [GROUPS=Virus] DATA=Weight
- b) Perform a repeated measurements ANOVA on the measurements from all 4 times. Perform diagnostic checking and, if necessary, identify a suitable transformation and analyse the transformed data.

The following output gives the repeated measurements ANOVA.

```
47
  -48 **** perform repeated measurements ANOVA
  -49
  50 DUPLICATE OLD=Weeks; NEW=Week
       BLOCK (Blocks/Groups)*Weeks
  52 TREAT Viruses*POL(Week;2)
  53 ANOVA [FPROB=Y; PSE=LSD] Weights
53.....
**** Analysis of variance ****
Variate: Weights
Source of variation d.f. s.s.
                                                          m.s. v.r. F pr.
                           3 808.34 269.45
Blocks stratum
Weeks stratum
Week
                                   3 1090989.69 363663.23
  Lin
                                  1 1072415.58 1072415.58
  Quad
                                 1 18571.47 18571.47
                                  1
  Deviations
                                         2.64
Blocks.Groups stratum
                                  4 11203.85 2800.96
12 1503.10 125.26
                                                                    22.36 <.001
Viruses
Residual
                                 12
                                                                      4.41
                                         439.20
                               9
Blocks.Weeks stratum
                                                         48.80
                                                                    1.72
Blocks.Groups.Weeks stratum

        Viruses.Week
        12
        3191.90
        265.99
        9.37
        <.001</th>

        Viruses.Lin
        4
        3016.34
        754.08
        26.57
        <.001</td>

        Viruses.Quad
        4
        167.51
        41.88
        1.48
        0.230

        Deviations
        4
        8.06
        2.01
        0.07
        0.990

        Residual
        36
        1021.55
        28.38

Total
                                  79 1109157.63
* MESSAGE: the following units have large residuals.
Blocks 4 Weeks 4
                                       6.20 s.e. 2.34
Blocks 2
            Groups 4 Weeks 4
                                                       8.56 s.e. 3.57
Blocks 2 Groups 4 Weeks 4 8.56 s.e. 3.57
Blocks 3 Groups 5 Weeks 4 8.97 s.e. 3.57
Blocks 4 Groups 5 Weeks 4 -8.90 s.e. 3.57
```

\*\*\*\* Tables of means \*\*\*\*

Variate: Weights

Grand mean 194.29

Viruses	1 205.47	2 211.07	3 186.92	4 180.31	5 187.68
Week	1 54.11	2 127.52	3 230.59	4 364.94	
Viruses 1 2 3 4	Week	1 54.37 60.40 53.37 51.70	2 134.57 144.11 121.17 116.47	3 245.62 253.19 221.03 211.47	4 387.32 386.59 352.12 341.57
5		50.70	121.26	221.64	357.11

\*\*\* Least significant differences of means (5% level) \*\*\*

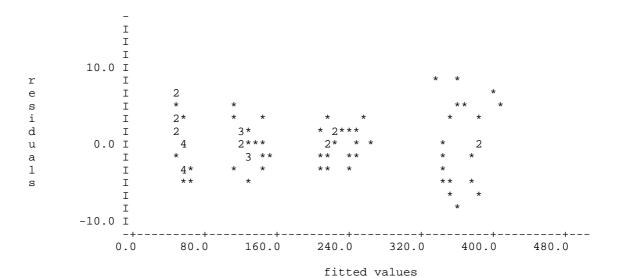
Table	Viruses	Week	Viruses Week
			WCCIL
rep.	16	20	4
l.s.d.	8.621	*	*
d.f.	12	*	*
Except Week d.f.	when comparing means with	h the same	level(s) of 10.483 29.34

54 CALC pBG=1-FPROB(125.26/28.38; 12; 36)

55 & pBW=1-FPROB(48.80/28.38; 9; 36) 56 PRINT pBG,pBW

pBW рBG 0.0002568 0.1203

57 APLOT METHOD=fit, normal

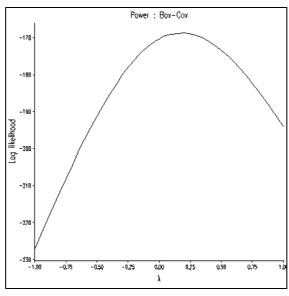


```
10.0 I
                                                    * * *
r
           Ι
                                             2***
*2222
2
е
           Т
i
           Ι
                                         23322
d
           Ι
                                     33433*
u
        0.0 I
                                 *2333
           Т
а
                            *22222
1
           I
s
           Ι
           Ι
           Ι
      -10.0 I
              -2.0 -1.0 0.0 1.0 2.0 3.0
         -3.0
                               expected Normal quantiles
 58
 -59 **** Tukey''s one-degree-of-freedom-for-non-additivity.
 -60 **** It is the term designated covariate in the following analysis
 -61
 62 TREAT Viruses*Week
 63 AKEEP [FIT=Fit]
 64 CALC ResSq=Fit*Fit
    ANOVA [PRINT=*] ResSq; RES=ResSq
 65
 66 COVAR ResSq
                                        "A computational trick"
 67 ANOVA [PRINT=A; FPROB=Y] Weights
****** Warning (Code AN 40). Statement 1 on Line 67
Command: ANOVA [PRINT=A; FPROB=Y] Weights
Stratum variance cannot be estimated
Weeks stratum has zero residual sum of squares or degrees of freedom
67.....
**** Analysis of variance (adjusted for covariate) ****
Variate: Weights
Covariate: ResSq
Source of variation
                   d.f.
                                       m.s. v.r. cov.ef. F pr.
                             s.s.
                             808.34
                                      269.45
Blocks stratum
Weeks stratum
                       3 1090989.69 363663.23
Week
                                                     1.00
Blocks.Groups stratum
                          11203.85
                                    2800.96
                                             22.36
Viruses
                       4
                                                      1.00 <.001
Residual
                       12
                           1503.10
                                      125.26
                                             11.49
                                                      1.00
Blocks.Weeks stratum
                       9
                            439.20
                                      48.80
                                              4.48
Blocks.Groups.Weeks stratum
Viruses.Week
                            3191.90
                                      265.99
                                                      1.00 <.001
                      12
                                               24.41
                                     640.09
                                              58.73
Covariate
                             640.09
                        1
                                                            < .001
Residual
                       35
                             381.46
                                      10.90
                                                       2.60
                       79 1109157.63
Total
```

68 COVAR

The above analysis indicates that the assumptions are not met. In particular, Tukey's test for nonadditivity is significant. In addition, the residual-versus-fitted-values plot indicates that there might be variance heterogeneity.

In the following output, we use the Box-Cox procedure, implemented in YTRANSFORM, to find that the logarithmic transformation may satisfactory. The analysis of the log-transformed data is also given.



```
69 "
-70 **** search for a transformation and analyse
-71 "
72 YTRANSFORM [LOWER=-1; UPPER=1; TERMS=Viruses*Week] Weights; SAVE=s
73 CALC TWeights=LOG(Weights)
74 BLOCK (Blocks/Groups)*Weeks
75 TREAT Viruses*POL(Week;2)
76 ANOVA [FPROB=Y; PSE=LSD] TWeights
```

76.....

\*\*\*\* Analysis of variance \*\*\*\*

Variate: TWeights Source of variation d.f. m.s. v.r. F pr. s.s. Blocks stratum 3 0.0229820 0.0076607 Weeks stratum 3 40.7739383 13.5913128 Week Lin 1 39.9719058 39.9719058 Ouad 1 0.7850574 0.7850574 Deviations 1 0.0169750 0.0169750 Blocks.Groups stratum Viruses 4 0.3164797 0.0791199 22.73 <.001 Residual 12 0.0417637 0.0034803 6.97 9 0.0041444 0.0004605 Blocks.Weeks stratum Blocks.Groups.Weeks stratum 12 0.0302026 0.0025169 5.04 <.001 Viruses.Week Viruses.Lin 4 0.0180220 0.0045055 9.03 <.001 4 0.0111231 0.0027808 0.0010575 0.0002644 Viruses.Quad 5.57 0.001 0.53 0.715 Deviations 4 Residual 36 0.0179681 0.0004991

Total 79 41.2074789

\* MESSAGE: the following units have large residuals.

Blocks	2	Groups	4	Weeks	1	-0.0601	s.e.	0.0150
Blocks	2	Groups	4	Weeks	4	0.0589	s.e.	0.0150
Blocks	4	Groups	4	Weeks	1	0.0467	s.e.	0.0150

\*\*\*\* Tables of means \*\*\*\*

Variate: TWeights

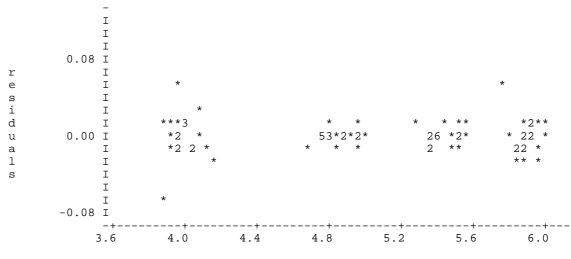
Grand mean 5.0422

Viruses	1 5.0897	2 5.1405	3 5.0087	4 4.9715	5 5.0006
Week	1 3.9883	2 4.8447	3 5.4378	4 5.8980	
Viruses 1 2 3 4 5	Week	1 3.9954 4.1009 3.9770 3.9422 3.9259	2 4.9017 4.9704 4.7969 4.7565 4.7979	3 5.5032 5.5339 5.3977 5.3535 5.4008	4 5.9586 5.9568 5.8633 5.8336 5.8778

\*\*\* Least significant differences of means (5% level) \*\*\*

Table		Vin	cuses		Wee	ek	Viruses	
							Week	
rep.			16		2	20	4	
l.s.d.		0.0	04544			*	*	
d.f.			12			*	*	
Except	when	comparing	means	with	the	same	level(s)	of
Week							0.05159	
d.f.							23.12	

#### 77 APLOT METHOD=fit, normal



fitted values

```
0.08 I
r
           Ι
е
           Т
i
           Ι
                                            222222****
d
           Ι
       0.00 I
                                  333334333332*
u
                         ***222232
а
           Т
1
           I
           Ι
s
           Ι
           Ι
      -0.08 T
         -3.0 -2.0 -1.0 0.0 1.0 2.0 3.0
                              expected Normal quantiles
 78
    **** Tukey''s one-degree-of-freedom-for-non-additivity.
 -79
 -80 **** It is the term designated covariate in the following analysis
 -81
 82 TREAT Viruses*Week
 83 AKEEP [FIT=Fit]
 84 CALC ResSq=Fit*Fit
 85
    ANOVA [PRINT=*] ResSq; RES=ResSq
                                       "A computational trick"
 86 COVAR ResSq
 87 ANOVA [PRINT=A; FPROB=Y] TWeights
******* Warning (Code AN 40). Statement 1 on Line 87
Command: ANOVA [PRINT=A; FPROB=Y] TWeights
Stratum variance cannot be estimated
Weeks stratum has zero residual sum of squares or degrees of freedom
87.....
**** Analysis of variance (adjusted for covariate) ****
Variate: TWeights
Covariate: ResSq
                   d.f.
Source of variation
                                      m.s. v.r. cov.ef. F pr.
                             s.s.
                      3 0.0229820 0.0076607
Blocks stratum
Weeks stratum
                      3 40.7739383 13.5913128
Week
                                                    1.00
Blocks.Groups stratum
Viruses
                      4 0.3164797 0.0791199 22.73 1.00 <.001
                      12 0.0417637 0.0034803
Residual
                                              7.03
                                                    1.00
                    9 0.0041444 0.0004605 0.93
Blocks.Weeks stratum
Blocks.Groups.Weeks stratum
              12 0.0302026 0.0025169
                                            5.08 1.00 <.001
Viruses.Week
Covariate
                      1 0.0006352 0.0006352 1.28
                                                           0.265
Residual
                      35 0.0173330 0.0004952
                                                     1.01
Total
                      79 41.2074789
 88 COVAR
```

It would appear that the assumptions underlying the log-transformed data are met, except for the presence of 3 outliers. The residual-versus-fitted-

values plots indicates that the variance is homogeneous and the normal probability plot is displaying a straight line trend (except for the outliers) indicating that the data are approximately normally distributed. Tukey's test for nonadditivity is nonsignificant.

In the repeated measures ANOVA the Deviations for Viruses. Weeks is not significant (p = 0.715) and the Viruses. Quad (p = 0.001) is significant. This indicates that the time trends are adequately described by a quadratic equation but that these differ between the Virus treatments.

c) The profiles are displaying some curvature so a linear relationship is likely to be inadequate. Fit orthogonal polynomials up to degree 3 to the profile for each group. Analyse the resulting coefficients. What do you conclude?

The Genstat output below contains the commands, and associated output, to produce the diagram above and the analysis of the polynomials fitted to the differences from week 0.

The assumptions underlying the analysis are not met for the linear coefficient as Tukey's test for nonadditivity is significant and the residual-versus-fitted-values plot is displaying curvature. The mean of the observations over time is also displaying some evidence of curvature, although Tukey's test for non-additivity is not significant in this case.

An analysis that corrects this problem is required. I have found that a quadratic regression of the logarithms of the weights on the logarithms of the times provides a satisfactory analysis in that the assumptions are met and 3<sup>rd</sup> and 4<sup>th</sup> degree coefficients are not significant. However, this curve does not have a particularly practical interpretation. Any other ideas for the analysis of this data set?

```
89
-90
     **** analyse differences from week 0 polynomial coefficients to degree 3
-91
 92 VORTHPOL [MAXDEGREE=3] Weight; CONTRAST=pol
 93 DELETE [REDEF=y] Fit,ResSq
94 BLOCK Block/Group
 95 TREAT Virus
 96 FOR k=0...3
 97
     ANOVA [FPROB=Y; PSE=LSD] pol[k]
      APLOT METHOD=fit, normal
 98
 99
      AKEEP [FIT=Fit]
      CALC ResSq=Fit*Fit
100
       ANOVA [PRINT=*] ResSq; RES=ResSq
101
      COVAR ResSq
                                                "A computational trick"
102
      ANOVA [PRINT=A; FPROB=Y] pol[k]
103
104
      COVAR
105 ENDFOR
```

```
105.....
**** Analysis of variance ****
Variate: pol[0]
                                      m.s. v.r. F pr.
Source of variation
                    d.f.
                            s.s.
Block stratum
                      3
                           202.08
                                     67.36 2.15
Block.Group stratum
                      4
                           2800.96
                                    700.24
                                           22.36 <.001
Virus
Residual
                      12
                            375.78
                                      31.31
                           3378.82
Total
                      19
***** Tables of means *****
Variate: pol[0]
Grand mean 194.3
   Virus
                              3
                 211.1
           205.5
                         186.9
                                180.3
                                        187.7
*** Least significant differences of means (5% level) ***
Table
                  Virus
rep.
                     4
d.f.
                    12
l.s.d.
                  8.62
           Ι
           Ι
           Ι
       10.0 I
r
           Ι
           I
е
           Ι
S
i
           Ι
d
           Ι
       0.0 I
u
а
           Т
1
           Ι
s
           Ι
           Ι
           Ι
      -10.0 I
                                  200.0 208.0 216.0 224.0
        176.0
                         192.0
                184.0
                                   fitted values
                                   Normal plot
           Ι
       10.0 I
r
           Ι
е
           Ι
           Ι
S
           Ι
i
d
           Ι
       0.0 I
u
а
1
           Т
s
           Ι
           Ι
           Ι
      -10.0 I
         -2.4
                 -1.6
                          -0.8
                                  0.0
                                         0.8 1.6 2.4
                             expected Normal quantiles
```

105									
**** Analysis of va	riance (a	djusted for	covariate)	****					
Variate: pol[0] Covariate: ResSq									
Source of variation	d.f.	s.s.	m.s.	v.r.	cov.ef.	F pr.			
Block stratum	3	202.08	67.36	2.46					
Block.Group stratum Virus Covariate Residual	4 1 11	2800.96 74.63 301.14	700.24 74.63 27.38			<.001 0.127			
Total	19	3378.82							
105***** Analysis of variance *****									
Variate: pol[1]									
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.				
Block stratum	3	72.83	24.28	1.68					
Block.Group stratum Virus Residual	4 12	603.27 173.65	150.82 14.47	10.42	<.001				
Total	19	849.75							
**** Tables of mean	ıs ****								
Variate: pol[1]									
Grand mean 103.56									
Virus 1 110.99	2 108.76	3 99.61	4 96.46 101	5 .96					
*** Least significar	nt differe	nces of mean	ns (5% leve	l) ***					
Table rep. d.f. l.s.d.	Virus 4 12 5.861								
I I I I I I I I I I I I I I I I I I I	* * *	* * * * *	* *	* * * *	* *	*			
92.0	96.0	100.0	104.0 10 fitted valu	08.0 ues	112.0	116.0			

r e s i d u a 1 s	6.0 I I I I I 0.0 I I I I -6.0 I	* *	* * * * * *	* * * * * *		*	*
	-2.4		-0.8			1.6	2.4
			expect	ed Normal o	quantil	es	
105							
****	Analysis of va	ariance (ad	liusted for	covariate)	****		
Variat	te: pol[1] .ate: ResSq	.11.100 (00	1942	00 ( 01 1 0 0 0 )			
Source	e of variation	d.f.	s.s.	m.s.	v.r.	cov.ef.	F pr.
Block	stratum	3	72.83	24.28	2.32		
Block. Virus Covari Residu		4 1 11	603.27 58.58 115.07	150.82 58.58 10.46	14.42 5.60		<.001 0.037
Total		19	849.75				
****	Analysis of va						
Source	e of variation	d.f.	s.s.	m.s.	v.r.	F pr.	
Block	stratum	3	17.416	5.805	2.13		
Block. Virus Residu Total	Group stratum	4 12 19	41.878 32.691 91.985	10.469 2.724	3.84	0.031	

<sup>\*</sup> MESSAGE: the following units have large residuals.

Block 2 Group 4 3.26 s.e. 1.28

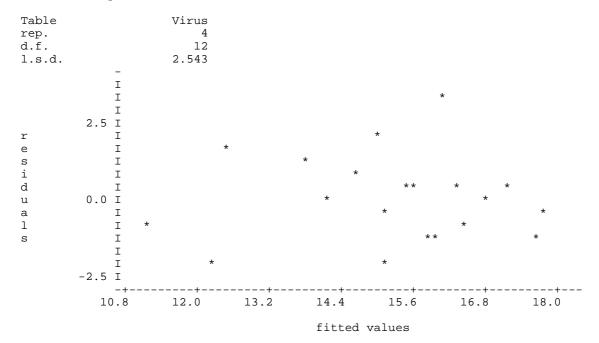
\*\*\*\*\* Tables of means \*\*\*\*\*

Variate: pol[2]

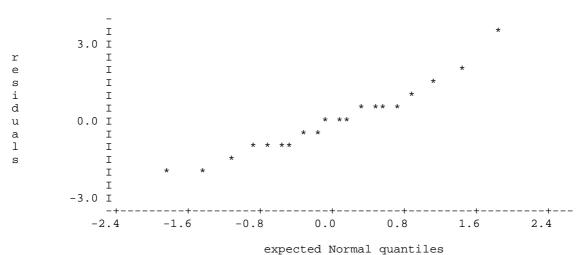
Grand mean 15.24

Virus 1 2 3 4 5
15.37 12.42 15.82 16.33 16.23

\*\*\* Least significant differences of means (5% level) \*\*\*

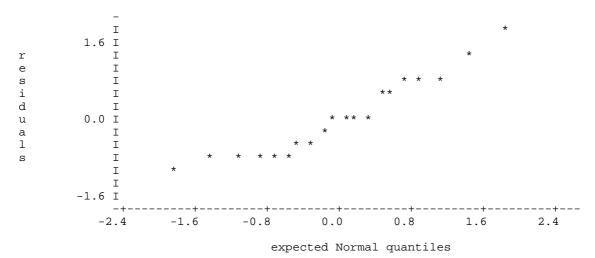


Normal plot



105			• • • • • • • • •			• • • • • • • • • • • • • • • • • • • •			
**** Analysis of va	riance (ad	justed for c	ovariate)	****					
Variate: pol[2] Covariate: ResSq									
Source of variation	d.f.	s.s.	m.s.	v.r.	cov.ef.	F pr.			
Block stratum	3	17.416	5.805	2.18					
Block.Group stratum Virus Covariate Residual	4 1 11	41.878 3.383 29.309	10.469 3.383 2.664			0.032 0.284			
Total	19	91.985							
105*  ***** Analysis of variance *****									
Variate: pol[3]									
Source of variation	d.f.	s.s.	m.s.	v.r.	F pr.				
Block stratum	3	2.979	0.993	0.95					
Block.Group stratum Virus Residual	4 12	4.475 12.513	1.119	1.07	0.412				
Total	19	19.967							
* MESSAGE: the follow	ving units	have large	residuals.						
Block 2 Group 4	:	1.80 s.e.	0.79						
**** Tables of means	5 ****								
Variate: pol[3]									
Grand mean 0.27									
Virus 1 -0.03	-0.17	3 -0.13	4 0.81 0.	5 88					
*** Least significant	differen	ces of means	(5% level	) ***					
Table rep. d.f. l.s.d.	Virus 4 12 1.573								

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	-0.8	-0.4	0.0	0.4	0.8	1.2	1.6
				fitted v	alues		



105.....

\*\*\*\*\* Analysis of variance (adjusted for covariate) \*\*\*\*\*

Variate: pol[3]
Covariate: ResSq

Source of variation	d.f.	s.s.	m.s.	v.r. c	ov.ef.	F pr.
Block stratum	3	2.9788	0.9929	1.04		
Block.Group stratum Virus Covariate Residual	4 1 11	4.4752 2.0270 10.4857	1.1188 2.0270 0.9532	1.17 2.13	1.00	0.375 0.173
Total	19	19.9667				