Extra Exercise 23

Data: Measurement of lymphocyte counts in blood samples of mice that had been exposed to one of four treatments (A−D). Four mice were selected from each of four litters, and within each litter the treatments were randomly allocated to the mice. The data therefore include a total of $4 \cdot 4 = 16$ mice. According to the description, the design is a completely randomized block design with litters as blocks. It seems sensible that treatment responses would be similar among mice from the same litter. The data form a two-way layout without replication, and the design is balanced.

Model: The two-way ANOVA model for these data can be written:

$$\text{lymph}_{ij} = \mu + \alpha_i + \beta_j + \varepsilon_{ij},$$

where the errors (ε_{ij}) are i.i.d. from $\text{N}(0, \sigma)$, and the indices i and j correspond to the row (e.g., treatment) and column (e.g., litter) factors, with $I = 4$, $J = 4$, and $N = 16$. Note that without replication in the data, an interaction term cannot be included in the model.

Because of the lack of replication, all observations have different means, and the model checking will have to be based on the residuals. We therefore move straight ahead to the analysis for the model.

```
ANOVA 'lymph' = treatment litter;
  Means treatment litter;
  GFourpack.
```

Factor Information

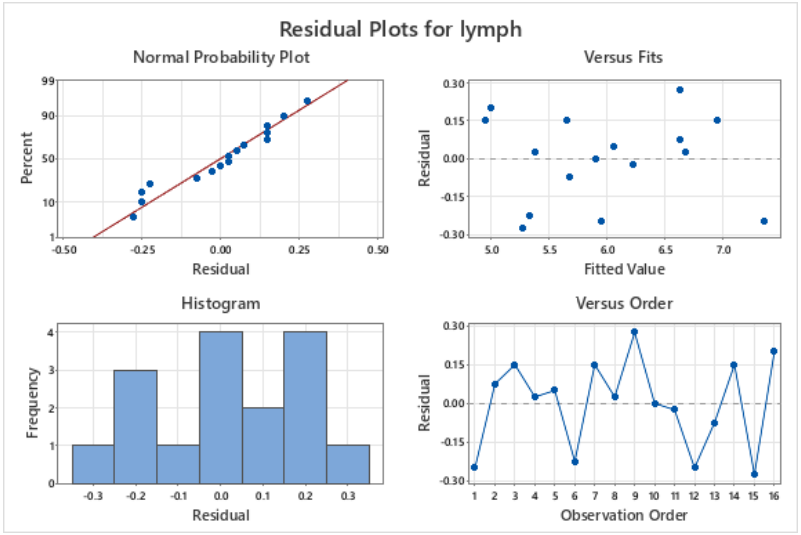
Factor	Type	Levels	Values
treatment	Fixed	4	A, B, C, D
litter	Fixed	4	1, 2, 3, 4

Analysis of Variance for lymph

Source	DF	SS	MS	F	P
treatment	3	1.3250	0.44167	8.83	0.005
litter	3	6.3950	2.13167	42.63	0.000
Error	9	0.4500	0.05000		
Total	15	8.1700			

Model Summary

S	R-sq	R-sq(adj)
0.223607	94.49%	90.82%



Comments:
The ANOVA table shows that both treatment and litter effects are clearly significant ($P = 0.005$ and $P < 0.001$, respectively). The residual plots look quite reasonable, considering the small dataset. There is some curvature around the line in the normal probability plot, but this could be attributed to the dependence (recall that residuals are not independent, and in a small design such as this one the dependence is substantial). The plot of residuals against fitted values does not show any alarming patterns. It can also be noted that the model explains a very large proportion of the variance in the data ($R^2 = 94.5\%$).

Pairwise comparisons among treatments will be based on the treatment means. If we wanted to perform unadjusted comparisons manually, we could use the LSD-value: $\text{LSD}_{0.95} = t^* \, s \sqrt{2/n} = 2.262 \cdot 0.2236 \sqrt{2/4} = 0.358$. Alternatively, we can in Minitab use the `Comparisons` menu after having fitted the model by the `General Linear Model` menu. The output for unadjusted (Fisher) comparisons is shown below.

Fisher Pairwise Comparisons: treatment

Grouping Information Using Fisher LSD Method and 95% Confidence

treatment	N	Mean	Grouping	
A	4	6.425	A	
C	4	6.025		B
D	4	5.750		B
B	4	5.700		B

Means that do not share a letter are significantly different.

Fisher Individual Tests for Differences of Means

Difference of treatment Levels	Difference of Means	SE of Difference	Individual 95% CI	T-Value	P-Value
B - A	-0.725	0.158	(-1.083, -0.367)	-4.59	0.001
C - A	-0.400	0.158	(-0.758, -0.042)	-2.53	0.032
D - A	-0.675	0.158	(-1.033, -0.317)	-4.27	0.002
C - B	0.325	0.158	(-0.033, 0.683)	2.06	0.070
D - B	0.050	0.158	(-0.308, 0.408)	0.32	0.759
D - C	-0.275	0.158	(-0.633, 0.083)	-1.74	0.116

Simultaneous confidence level = 82.15%

The pairwise comparisons show that treatment A is significantly different from the other three treatments, which in turn are not significantly different. A Bonferroni adjustment for six comparisons will make the comparison between A and C non-significant (because its unadjusted P-value is larger then $0.05/6$), reducing the conclusion to A being significantly different from B and D. There is presumably no particular interest in the litters, so it will suffice to note the strong differences between them determined by the overall test.