

ANCOVA

An alternative medical example for the same pre-test – post-test control group design

The design example that we will use is one for which ANCOVA is highly appropriate, but which is often neglected in favour of less satisfactory alternatives. The design is a pre-test - post-test control group design, which is frequently analysed with an ANOVA of gain scores or with a two-factor repeated measures ANOVA, in which the interaction between the IV (treatment) and test occasion (pre/post) tests the hypothesis that treatments have no effect. Todman and Dugard (1995) have shown why the ANCOVA should be preferred to both of these alternative analyses.

Normal adults have a platelet count in the range $150-400 \times 10^9/L$ blood. Aplastic anaemia is a serious illness that causes a drop in all blood cells, including platelets. A platelet reduction is life threatening when the count drops below 20. The condition can be treated with platelet transfusion, but this usually has only a short term effect. The illness is not curable but can usually be successfully treated to provide long or permanent remission. Patients normally receive a short course of antilymphocyte globulin (ATG), to be followed by further treatment in the long term. A study is carried out to investigate how best to get the initial treatment started. Sixty patients are recruited to the study, which is designed to compare the effect on platelet count one week after treatment (POSTCOUNT) with (1) transfusion, (2) a short course of ATG or (3) transfusion followed immediately by ATG, after 'taking out' any initial differences in the count by treating the pre-treatment count (PRECOUNT) as a covariate. Twenty patients are allocated at random to each of the three treatment conditions

(TREAT). A second factor, gender (SEX), is introduced to illustrate ANCOVA with more than one treatment factor.

The (fabricated) data

The two sets of test scores are recorded as PRECOUNT and POSTCOUNT. Actually, we have fabricated three versions of the data for this experiment in order to show the full power of the ANCOVA approach. Table 5.1 shows the first four rows of each treatment method for all three datasets; each has a set of PRECOUNT and POSTCOUNT scores, but the TREAT levels are only listed once. The full dataset, along with an extra variable called SEX which we use later, can be found on the book website (med.ancova.sav).

Table 5.1
The first four rows of each treatment method for the three datasets (the full dataset can be found as med.ancova.sav on the website)

	dataset 1		dataset 2		dataset 3	
treatment	precount1	postcount1	precount2	postcount2	precount3	postcount3
transfusion	106	110	136	128	119	141
transfusion	110	87	74	56	111	139
transfusion	85	76	95	107	145	166
transfusion	104	96	105	120	92	128
ATG	110	96	118	164	114	153
ATG	101	77	93	129	94	138
ATG	129	118	88	143	131	172
ATG	79	63	99	120	86	133
transfusion+ATG	111	112	74	127	96	161
transfusion+ATG	102	121	86	162	64	131
transfusion+ATG	130	126	121	162	76	144
transfusion+ATG	124	105	111	163	86	158

Graphical considerations: dataset 1

A first step in the analysis of the data, and one that is quite informative, is to plot POSTCOUNT (DV) against PRECOUNT (covariate) scores for all treatment (IV) groups on a single graph, in the same way as in our informal example illustrated in Figure 5.1. On the plot, the three treatment groups are distinguished by different symbols.

We begin by looking at a plot for the first dataset, shown in Figure 5.3(a). You can see that the POSTCOUNT scores increase with the PRECOUNT scores in an approximately linear way, with some random variation. However, the symbols for the three TREAT levels are thoroughly mixed together on the graph, with no discernible separation. For this dataset, as we shall see when we proceed to the analysis, neither of our two treatment methods had any effect, all three groups had similar POSTCOUNT (DV) results, which depended only on the PRECOUNT (covariate) scores.

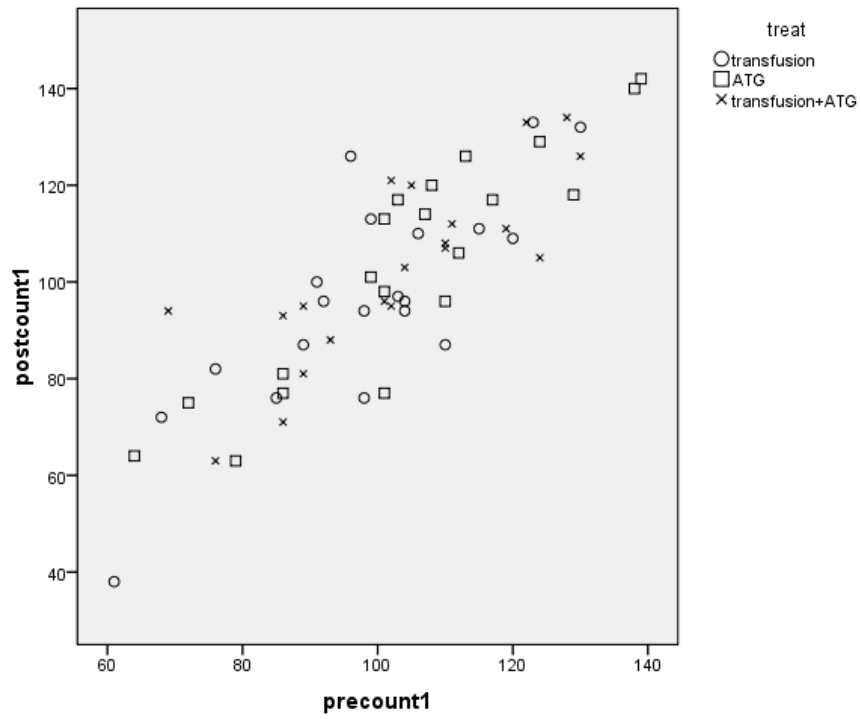
Graphical considerations: dataset 2

Now consider the second dataset, and the graph of POSTCOUNT against PRECOUNT scores, which is shown in Figure 5.3(b), again with the TREAT levels distinguished by different symbols. This time we can see that the points representing ATG mostly lie above those for the transfusion only group, and points representing transfusion+ATG mostly lie above those representing ATG. For all groups, we can still see that POSTCOUNT scores increase linearly with PRECOUNT scores, allowing for some random variation. Careful scrutiny of the graph shows us something else: if we were to fit regression lines of POSTCOUNT on PRECOUNT scores for each of the three TREAT levels, the lines would be approximately parallel.

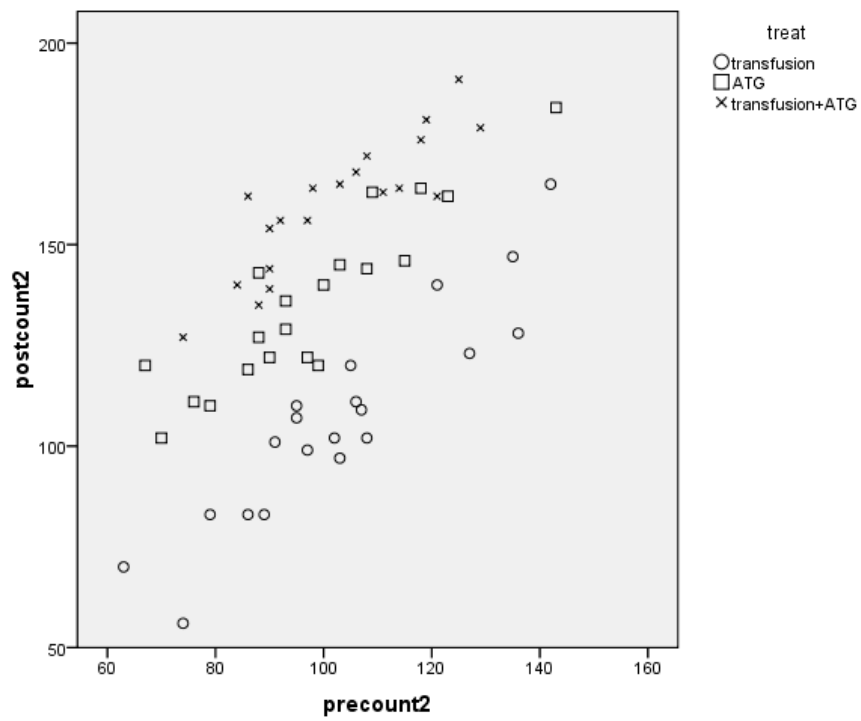
Graphical considerations: dataset 3

Now turn to the last dataset, graphed in Figure 5.3(c). Once again we can see the separation of the three TREAT levels, with transfusion+ATG higher than ATG, and this higher than the transfusion only group. This time however, if we were to fit regression lines of POSTCOUNT on PRECOUNT for each treatment group, the lines would not be parallel. The line for transfusion+ATG would be steeper than that for ATG, which in turn would be steeper than the one for transfusion only.

(a) Graph of dataset 1



(b) Graph of dataset 2



(c) Graph of dataset 3

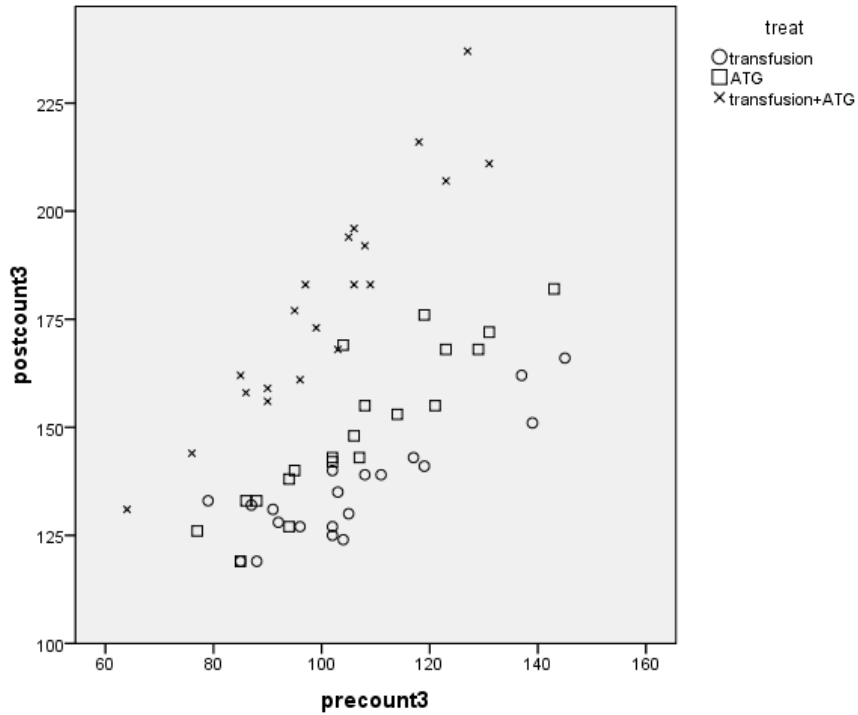


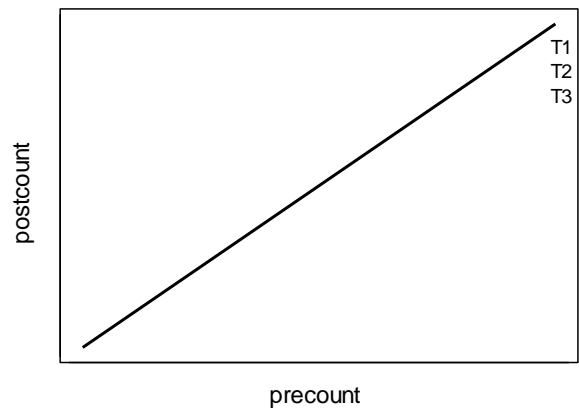
Figure 5.3. Graphs of the three datasets

Graphical considerations: comparison of the three datasets

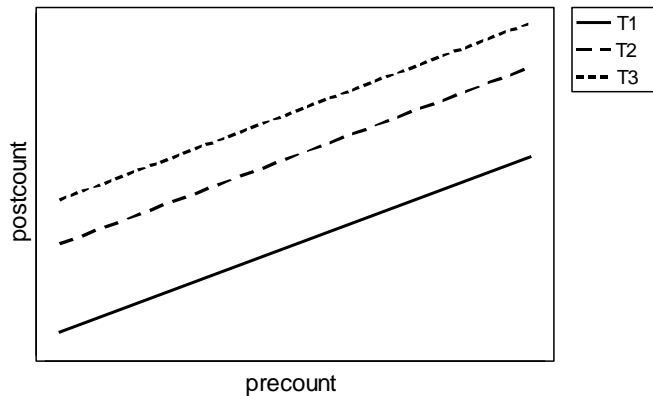
Figure 5.4 shows a diagrammatic representation of our three datasets. In the first version of our experiment (dataset 1) the POSTCOUNT scores do not show any difference among the treatment groups, as in Figure 5.4(a). In the second version (dataset 2) the graph makes it obvious that there are differences among the treatments, but the effect is quite simple: each treatment method has raised the regression line of POSTCOUNT on PRECOUNT but the lines are parallel, as in Figure 5.4(b). Here, applying Treatment 3 (transfusion+ATG) instead of Treatment 1 (transfusion only) would just add a constant amount to the POSTCOUNT scores (except for random variation). Similarly for Treatment 2. In the terminology used in many statistics texts, this is referred to as *homogeneity of regression slopes*.

In the last version (dataset 3), we see in Figure 5.4(c) that for Treatment 3 the difference in POSTCOUNT scores resulting from unit difference in PRECOUNT scores is greater than is the case for Treatment 2 (the line is steeper). For Treatment 2 the difference in POSTCOUNT scores resulting from unit difference in PRECOUNT scores is greater than is the case for Treatment 1 (transfusion only). The three treatment programs spread the POSTCOUNT scores across a wider range as well as raising them on average (i.e. there is an interaction between treatment levels (the IV) and PRECOUNT scores (the covariate)). This is referred to as *non-homogeneity of regression slopes*.

(a) A diagrammatic representation of dataset 1, showing no differences among the treatment groups



(b) A diagrammatic representation of dataset 2, showing parallel lines for the three treatment groups



(c) A diagrammatic representation of dataset 3, showing lines for the three treatment groups which are not parallel

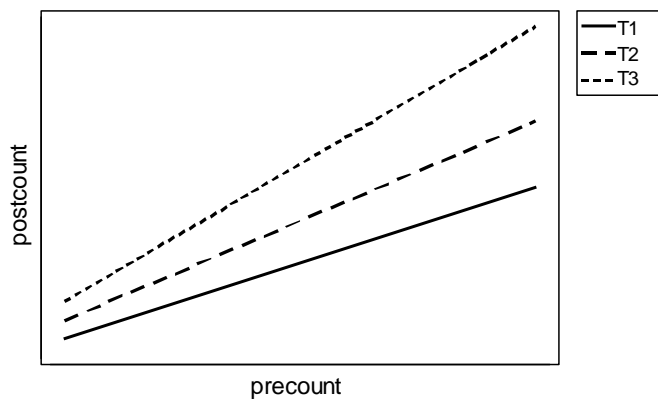


Figure 5.4. Diagram of the three data sets (adapted from Dugard & Todman 1995)

ANCOVA: assumptions

The analysis of covariance allows us to quantify the above observations. Before we begin, two assumptions are needed, and both are easily checked by looking at the graphs. First, we assume that the relationship between the DV (POSTCOUNT) and the covariate (PRECOUNT) is approximately linear within each level of the IV (TREAT). If this is not the case, then almost certainly complex processes are at work and these will

require careful investigation within each level of the IV before any attempt is made to compare these levels. The other assumption is that the variance of residual POSTCOUNT scores is approximately the same over the full range of PRECOUNT scores and in each IV level. In other words, the points do not become more widely scattered about their lines as we move from left to right, nor as we move from one level of the IV to another.

In addition to the assumptions that are specific to ANCOVA, we need the same assumption of random allocation to treatments as is needed for any analysis at all. It is not uncommon for statistics texts to refer to the requirement of an assumption of homogeneity of regression, but this is not, in fact, necessary, as will be seen from our analysis of dataset 3, illustrated in Figure 5.3(c).

Power considerations

As usual, in a real experiment the sample size should be based on considerations of expected effect size and required power. For a regression analysis with just one IV, the rule of thumb suggested for that would give a sample size of about 58 ($50 + 8*1$), since testing for the significance of the model as a whole is the same as testing for the (only) individual IV. However, what we have here is a design that combines ANOVA (with one factor at three levels) and regression. SPSS SamplePower gives the power as just below 0.8 (0.77 in fact) with 20 cases per level of the factor and one covariate, if the effect size is large. This is sufficient for our purpose in this example.

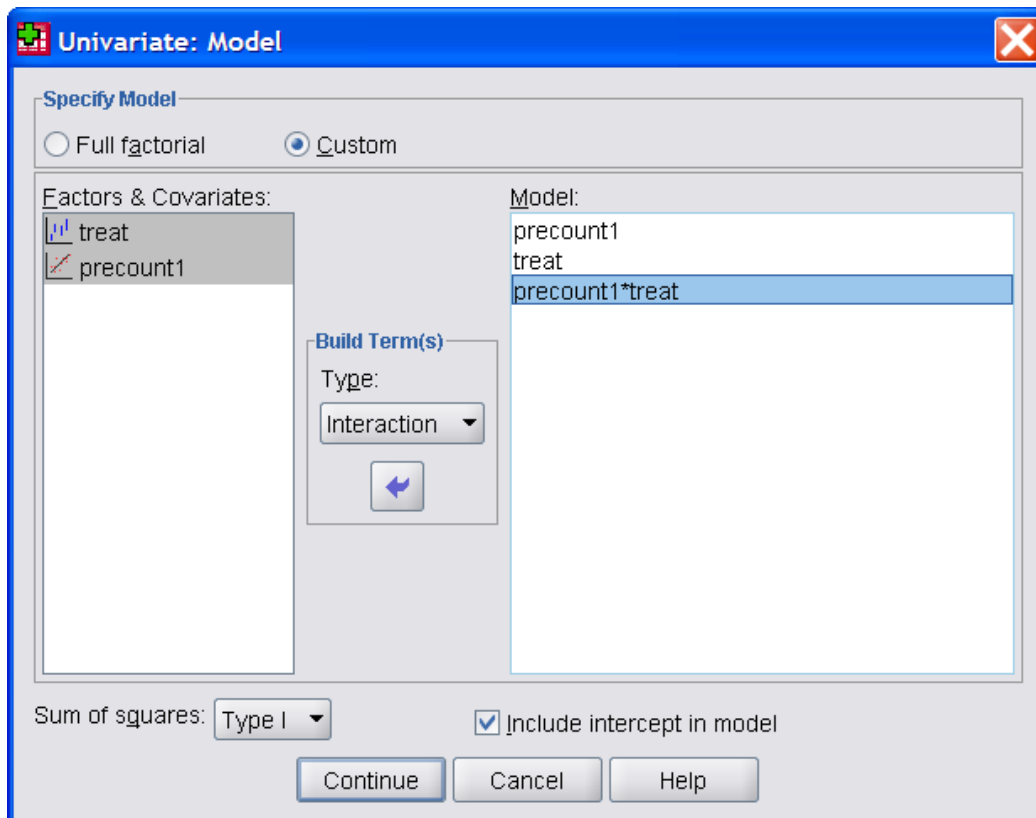
ANCOVA: requesting the analysis for dataset 1 in SPSS

We begin by considering dataset 1. From our graphical analysis we expect that there will be no significant differences among TREAT levels, but we expect that the

covariate (PRECOUNT) will have a significant effect. The most general model fits a regression of POSTCOUNT on PRECOUNT for each level of TREAT without applying any constraints to the parameters. This allows the slopes to be different, as in Figure 5.4(c). It is usually best to begin by fitting this most general model, then we can progressively eliminate higher order terms if they are nonsignificant. We will follow this approach with dataset 1. On the menu bar choose **Analyze**, then **General Linear Model**, then **Univariate**. We need a univariate analysis because we have only one DV or outcome (POSTCOUNT). We get the same dialog box as the one used for starting an ANOVA, SPSS Dialog Box 2.5. We are looking at the first dataset here, so our **Dependent Variable** is POSTCOUNT1. Our **Covariate** is PRECOUNT1 and our **Fixed Factor** is TREAT (fixed and random factors were discussed in Chapter 2 on ANOVA). Insert the variables in the appropriate boxes using the arrows.

Once the variables are entered we need to click the **Model** button to specify the model to be used, and again we get the same one as we used in ANOVA. This time look at the bottom to where **Sum of Squares** is set at the default, **Type III**. If we accept this, in the analysis each term will be considered in relation to all of the other terms listed in the model. This is usually what we want, but may not be appropriate if we have a clear idea of the order or hierarchy in which the terms can be expected to contribute to an explanation of the IV. In our experiment we do have, we are pretty sure the PRECOUNT score is important. The variation in the POSTCOUNT scores not explained by this may be explained by TREAT, and any remaining variation after we allow for that may be explained by the interaction term. With this order quite clear in our case, we opt for **Type I** sums of squares, which considers each term in relation to those already considered (above it in the list).

Now we must choose the **Custom** model, so that we can get the terms in the correct order, so click the radio button. To build the terms in the correct order, enter the covariate, PRECOUNT1 into the **Model** box first using the arrow, then the factor, TREAT, and finally the interaction term. To get this last one, select both the factor and the covariate together and use the arrow to get the interaction term, TREAT*PRECOUNT1, into the model. The result of all this is SPSS Dialog Box 5.1. Click **Continue**, then as usual we can use the **Options** button to request **Estimates of effect size** and **Observed power**. Accept the defaults for all other choices, and click **OK**.



SPSS Dialog Box 5.1. Specifying the model

ANCOVA: understanding the output for dataset 1

The output appears in SPSS Output 5.1. The first table (not shown here) just confirms that we had 20 participants at each level of TREAT. The second is the ANCOVA table. The column at the right of F shows the significance of each term in the model. First we note that the interaction term, TREAT*PRECOUNT1, has a large probability (0.672) of occurring by chance, so this term is not significant. The estimate of the effect size (partial eta squared) is also small. If this term is omitted from the model, we shall have a more powerful test of the main effects of TREAT and PRECOUNT1, so our next step is to go back to the model dialog box and remove the TREAT*PRECOUNT1 term. This gives the output shown in SPSS Output 5.2.

Tests of Between-Subjects Effects

Dependent Variable: postcount1

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	20453.195 ^a	5	4090.639	28.846	.000	.728	144.229	1.000
Intercept	611858.017	1	611858.017	4314.605	.000	.988	4314.605	1.000
precount1	20304.245	1	20304.245	143.178	.000	.726	143.178	1.000
treat	35.184	2	17.592	.124	.884	.005	.248	.068
treat * precount1	113.766	2	56.883	.401	.672	.015	.802	.112
Error	7657.788	54	141.811					
Total	639969.000	60						
Corrected Total	28110.983	59						

a. R Squared = .728 (Adjusted R Squared = .702)

b. Computed using alpha = .05

SPSS Output 5.1. Analysis for dataset 1 with interaction term included

Now we can look at the significance of the main effects. We see that the IV, TREAT, has a high probability (0.881) of occurring by chance; the different treatments had no significant effect (and the estimated effect size is small). This is what we expected after looking at the graph in Figure 5.3(a). The covariate (PRECOUNT1) has a very small probability, less than 0.001, so this certainly has a significant effect. Again, this is what we expected from our graphical analysis. The Corrected Model and Intercept terms are usually of no interest and can be ignored at this stage.

Tests of Between-Subjects Effects

Dependent Variable: postcount1

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	20339.429 ^a	3	6779.810	48.854	.000	.724	146.561	1.000
Intercept	611858.017	1	611858.017	4408.905	.000	.987	4408.905	1.000
precount1	20304.245	1	20304.245	146.308	.000	.723	146.308	1.000
treat	35.184	2	17.592	.127	.881	.005	.254	.069
Error	7771.555	56	138.778					
Total	639969.000	60						
Corrected Total	28110.983	59						

a. R Squared = .724 (Adjusted R Squared = .709)

b. Computed using alpha = .05

SPSS Output 5.2. Analysis for dataset 1 without the interaction term

Reducing the model and understanding the new output

If we now omit the IV, TREAT, our model will be reduced to the familiar simple regression of POSTCOUNT1 on PRECOUNT1, putting all the data from set 1 together. Of course SPSS offers simpler ways to do this, but to follow our model reduction process to its conclusion we will do it with **General Linear Model** again. Just remove the factor (TREAT) from the **Model** box. This time click the **Options** button again and check the box for **Parameter estimates** as well as those for effect size and observed power already ticked. The results are shown in SPSS Output 5.3.

Tests of Between-Subjects Effects

Dependent Variable: postcount1

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	20304.245 ^a	1	20304.245	150.850	.000	.722	150.850	1.000
Intercept	611858.017	1	611858.017	4545.786	.000	.987	4545.786	1.000
precount1	20304.245	1	20304.245	150.850	.000	.722	150.850	1.000
Error	7806.739	58	134.599					
Total	639969.000	60						
Corrected Total	28110.983	59						

a. R Squared = .722 (Adjusted R Squared = .718)

b. Computed using alpha = .05

Parameter Estimates

Dependent Variable: postcount1

Parameter	B	Std. Error	t	Sig.	95% Confidence Interval		Partial Eta Squared	Noncent. Parameter	Observed Power ^a
					Lower Bound	Upper Bound			
Intercept	-3.049	8.602	-.354	.724	-20.267	14.169	.002	.354	.064
precount1	1.021	.083	12.282	.000	.855	1.188	.722	12.282	1.000

a. Computed using alpha = .05

SPSS Output 5.3. Analysis for dataset 1 using only PRECOUNT1 as regressor

Here we see that the probability for the covariate is less than 0.001 and the estimated effect size is large. As expected, the POSTCOUNT1 scores are significantly dependent on the PRECOUNT1 scores, and the second table gives us the regression coefficient, 1.021. The intercept does not differ significantly from zero and the slope is close to 1, so in our first dataset the POSTCOUNT scores would be well estimated just from the PRECOUNT scores.

ANCOVA: requesting the analysis for dataset 2 in SPSS

Now we turn our attention to our second dataset, where we expect to find that the different treatments have a significant effect. Once again set up the model exactly as in SPSS Dialog Box 5.1, except that PRECOUNT1 and POSTCOUNT1 are replaced by PRECOUNT2 and POSTCOUNT2.

ANCOVA: understanding the output for dataset 2

The result is shown in SPSS Output 5.4. All we need to note from this is that the interaction term is not significant, with a probability of 0.427 and a small estimated effect size. We do not reject the null hypothesis that the three regression lines all have the same slope (i.e. they are parallel). For this dataset we do not need the most general model illustrated in Figure 5.4(c): the simpler model with parallel lines illustrated in Figure 5.4(b) adequately describes these data, as we had already guessed from looking at our graph (Figure 5.3(b)). Because the regression lines are parallel, we know that the effects of the treatments are the same for all values of the PRECOUNT. Estimating the TREAT effect is just equivalent to estimating the vertical separation of the regression lines, and testing for significant differences among TREAT levels is equivalent to testing the hypothesis that the distances between the lines are zero.

Tests of Between-Subjects Effects

Dependent Variable: postcount2								
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	50182.693 ^a	5	10036.539	114.724	.000	.914	573.619	1.000
Intercept	1078164.150	1	1078164.150	12324.074	.000	.996	12324.074	1.000
precount2	20074.203	1	20074.203	229.460	.000	.809	229.460	1.000
treat	29957.162	2	14978.581	171.214	.000	.864	342.429	1.000
treat * precount2	151.328	2	75.664	.865	.427	.031	1.730	.191
Error	4724.157	54	87.484					
Total	1133071.000	60						
Corrected Total	54906.850	59						

a. R Squared = .914 (Adjusted R Squared = .906)

b. Computed using alpha = .05

SPSS Output 5.4. Analysis for dataset 2 with the interaction term included

Testing the treatment effect and understanding the output

This, of course, is what we want to do next. We can fit the model with parallel lines by removing the interaction term from the model. Use the **Options** button to request **Parameter estimates** again. The results are in SPSS Output 5.5.

Tests of Between-Subjects Effects

Dependent Variable: postcount2								
Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	50031.365 ^a	3	16677.122	191.554	.000	.911	574.662	1.000
Intercept	1078164.150	1	1078164.150	12383.832	.000	.995	12383.832	1.000
precount2	20074.203	1	20074.203	230.573	.000	.805	230.573	1.000
treat	29957.162	2	14978.581	172.045	.000	.860	344.089	1.000
Error	4875.485	56	87.062					
Total	1133071.000	60						
Corrected Total	54906.850	59						

a. R Squared = .911 (Adjusted R Squared = .906)

b. Computed using alpha = .05

Parameter Estimates

Dependent Variable: postcount2									
Parameter	B	Std. Error	t	Sig.	95% Confidence Interval		Partial Eta Squared	Noncent. Parameter	Observed Power ^a
					Lower Bound	Upper Bound			
Intercept	52.231	7.120	7.335	.000	37.967	66.495	.490	7.335	1.000
precount2	1.054	.067	15.815	.000	.921	1.188	.817	15.815	1.000
[treat=1]	-54.049	2.951	-18.314	.000	-59.961	-48.137	.857	18.314	1.000
[treat=2]	-19.285	2.969	-6.496	.000	-25.232	-13.338	.430	6.496	1.000
[treat=3]	0 ^b								

a. Computed using alpha = .05

b. This parameter is set to zero because it is redundant.

SPSS Output 5.5. Analysis for dataset 2 without the interaction term

We can see at once from the first table in SPSS Output 5.5 that the covariate (PRECOUNT2) is significant and has a large estimated effect size. This tells us that we

can reject the hypothesis of no linear relationship between PRECOUNT2 and POSTCOUNT2. Also, the significant TREAT effect tells us that, with POSTCOUNT2 scores adjusted to take account of PRECOUNT2 scores, the hypothesis of no difference between treatment methods can be rejected (the parallel lines have non-zero separations and the estimated effect size is large).

Using parameter values to obtain differences among treatment methods

The table of parameter estimates tells us that the slope of each of the (parallel) regression lines is 1.054. To get the intercepts for each of the regression lines we have to add the parameter called 'intercept' in the table to the value given for each level of TREAT. So for transfusion only (TREAT 1) the intercept is

$$52.231 + (-54.049) = -1.818.$$

So, to predict the POSTCOUNT2 scores for the transfusions using this model, we need the equation

$$\text{POSTCOUNT2} = -1.818 + 1.054 * \text{PRECOUNT2}.$$

Similarly, to get the intercept for ATG (TREAT 2) group we need

$$52.231 - 19.285 = 32.946,$$

and the prediction equation for this group is

$$\text{POSTCOUNT2} = 32.946 + 1.054 * \text{PRECOUNT2}.$$

For transfusion+ATG (TREAT 3) group the intercept is 52.231+0, so the prediction equation is

$$\text{POSTCOUNT2} = 52.231 + 1.054 * \text{PRECOUNT2}$$

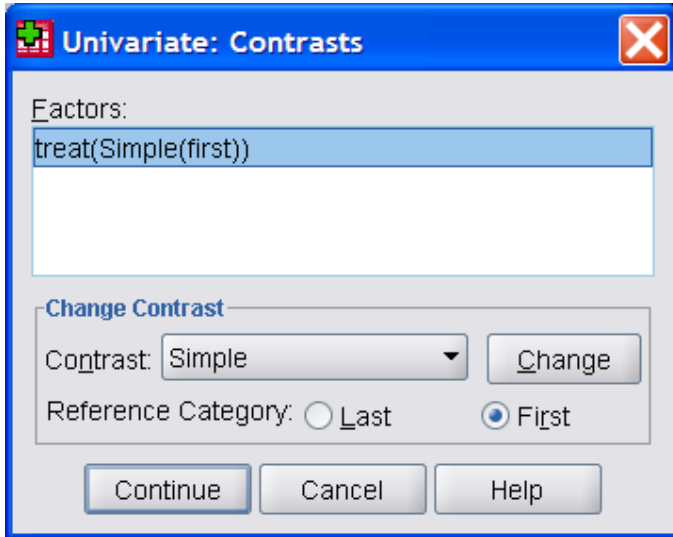
So ATG increases POSTCOUNT2 scores above the transfusion only scores by an estimated $32.946 - (-1.818) = 34.746$, (the difference between their intercepts) at all PRECOUNT2 levels. Transfusion+ATG increases POSTCOUNT2 scores above transfusion only scores by an estimated $52.231 - (-1.818) = 54.049$ at all PRECOUNT2 levels.

Note that statistics packages do not all present parameter estimates in the same way so if you use one other than SPSS you may have to spend a few minutes working out how to interpret them.

There are two simpler models but both would be rejected for this dataset. The first assumes no difference among treatments (a single regression line will do for all 60 observations, as in Figure 5.4(a)). This model would be rejected because of the significant TREAT effect in SPSS Output 5.5. The other simpler model assumes there is no dependence of POSTCOUNT2 on PRECOUNT2 so that the analysis of the data would reduce to a one-way ANOVA of POSTCOUNT2 by TREAT, ignoring PRECOUNT2 scores. This model would be rejected because of the significant covariate in SPSS Output 5.5.

Using SPSS to obtain differences among treatment methods

We have already shown how to find the regression lines for the treatment groups from the table of parameter values, and this tells us how far apart the lines are (just compare their intercepts). However, we can obtain comparisons among treatments directly from SPSS by requesting specific contrasts. Suppose we want to compare each of the ATG and ATG+transfusion treatment methods with the transfusion only condition. We can use the **Contrasts** button to get the comparisons that interest us. To compare the two ATG treatment methods with transfusion only, set the **Contrasts** dialog box as in SPSS Dialog Box 5.2. Click the **Change** button after selecting **Simple** from the **Contrast** list and **First** as the reference category.



SPSS Dialog Box 5.2. Contrasts dialog box

The results can be seen in SPSS Output 5.6. Treatment ATG (TREAT level 2) raises scores by nearly 35 points above transfusion only. Treatment transfusion+ATG (TREAT level 3) raises scores by just over 54 points.

Contrast Results (K Matrix)

treat Simple Contrast ^a		Depende...	
		postcount2	
Level 2 vs. Level 1	Contrast Estimate	34.763	
	Hypothesized Value	0	
	Difference (Estimate - Hypothesized)	34.763	
	Std. Error	2.976	
	Sig.	.000	
	95% Confidence Interval for Difference	Lower Bound	28.802
		Upper Bound	40.725
Level 3 vs. Level 1	Contrast Estimate	54.049	
	Hypothesized Value	0	
	Difference (Estimate - Hypothesized)	54.049	
	Std. Error	2.951	
	Sig.	.000	
	95% Confidence Interval for Difference	Lower Bound	48.137
		Upper Bound	59.961

a. Reference category = 1

SPSS Output 5.6. Contrasts for dataset 2

ANCOVA: requesting the analysis for dataset 3 in SPSS

We turn now to consideration of dataset 3. Once again we begin by fitting the most general model, setting it up as in SPSS Dialog Box 5.1, but with PRECOUNT3 and POSTCOUNT3 replacing PRECOUNT1 and POSTCOUNT1.

ANCOVA: understanding the output for dataset 3

This results in the analysis summary shown in SPSS Output 5.7, in which we are again interested in the line of the first table giving the values for the SS, F and probabilities for the interaction term TREAT*PRECOUNT3. This term is highly significant, with $F(2,54) = 16.863$. This time the estimated effect size for the interaction is also much larger, though only about half those for the main effects. So this time we reject the null hypothesis that the three lines all have the same slope. There is no simpler model for this dataset. Looking now at the main effects we see that the TREAT factor is highly significant with $F(2,54) = 197.792$ and a probability less than 0.001. The covariate PRECOUNT3 is also significant with a very large F and a probability less than 0.001. However, we recall that interpreting significant main effects is not straightforward when there is a significant interaction.

Obtaining prediction equations for each of the treatment groups

To get the prediction equations for each of the treatment groups we have to combine the parameter values listed in the second column of the table of estimates. The intercept for transfusion only is $32.299+39.398 = 71.697$. The slope for transfusion only is $1.462+(-0.858) = 0.604$, so the prediction equation for transfusion only is $POSTCOUNT3 = 71.697+0.604*PRECOUNT3$.

Similarly, for ATG the equation is

$$\text{POSTCOUNT3} = 32.299 + 16.003 + (1.462 - 0.511) * \text{PRECOUNT3} \text{ or}$$

$$\text{POSTCOUNT3} = 48.302 + 0.951 * \text{PRECOUNT3}.$$

For transfusion+ATG the equation is

$$\text{POSTCOUNT3} = 32.299 + 1.462 * \text{PRECOUNT3}.$$

Tests of Between-Subjects Effects

Dependent Variable: postcount3

Source	Type III Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^b
Corrected Model	39106.436 ^a	5	7821.287	123.548	.000	.920	617.741	1.000
Intercept	1439021.067	1	1439021.067	22731.370	.000	.998	22731.370	1.000
precount3	11928.716	1	11928.716	188.431	.000	.777	188.431	1.000
treat	25042.704	2	12521.352	197.792	.000	.880	395.585	1.000
treat * precount3	2135.015	2	1067.508	16.863	.000	.384	33.726	1.000
Error	3418.498	54	63.306					
Total	1481546.000	60						
Corrected Total	42524.933	59						

a. R Squared = .920 (Adjusted R Squared = .912)

b. Computed using alpha = .05

Parameter Estimates

Dependent Variable: postcount3

Parameter	B	Std. Error	t	Sig.	95% Confidence Interval		Partial Eta Squared	Noncent. Parameter	Observed Power ^a
					Lower Bound	Upper Bound			
Intercept	32.299	11.120	2.905	.005	10.005	54.592	.135	2.905	.814
precount3	1.462	.109	13.415	.000	1.244	1.681	.769	13.415	1.000
[treat=1]	39.398	15.441	2.552	.014	8.441	70.356	.108	2.552	.707
[treat=2]	16.003	15.876	1.008	.318	-15.826	47.833	.018	1.008	.168
[treat=3]	0 ^b								
[treat=1] * precount3	-.858	.148	-5.796	.000	-1.154	-.561	.384	5.796	1.000
[treat=2] * precount3	-.511	.151	-3.375	.001	-.815	-.207	.174	3.375	.912
[treat=3] * precount3	0 ^b								

a. Computed using alpha = .05

b. This parameter is set to zero because it is redundant.

SPSS Output 5.7. Analysis for dataset 3 with the interaction term included

Focussing on the differences among slopes

It is easy to see from Figure 5.3(c) or Figure 5.4(c) that the effect of TREAT is not the same at all levels of the PRECOUNT3. For low PRECOUNT3 values (on the left of the figures) the differences among treatments are smaller than for the high PRECOUNT3 values (on the right). Another way of saying this is that we have a significant interaction between PRECOUNT3 and TREAT. This being the case, there is little point in trying to discuss differences among treatment methods in terms of the mean (or adjusted mean) effects. The important difference among treatment methods found here is the difference among the slopes of the regressions of POSTCOUNT3 on

PRECOUNT3. The increase in POSTCOUNT3 per unit increase in PRECOUNT3 is greatest for transfusion+ATG (TREAT 3) and least for transfusion only (TREAT 1).

Focussing on the individual slopes

Once we reject the hypothesis of parallel regressions as we have done here we may want to test other hypotheses about the slopes. For example, we may want to test the hypothesis that the slope is zero (i.e. POSTCOUNT3 does not depend on PRECOUNT3) for each level of TREAT. So far, all we know is that, since the slopes are not equal, they cannot all be zero. We may also want to test whether the slopes for ATG and transfusion+ATG are significantly different, or compare the slopes of either with that for transfusion only.

Testing the differences between pairs of slopes

From the list of parameter estimates shown in SPSS Output 5.7, we found the intercepts for the three regression lines, but these are rarely of interest when the lines are not parallel. If you want to test whether the slopes for the two ATG treatment methods (TREAT 2 and 3) are significantly different, obtain the t value for the difference between them using the slope estimates and standard errors from SPSS Output 5.7. The slope for TREAT = 2 is the sum of the B value for PRECOUNT3 and that for the interaction term [TREAT = 2]*PRECOUNT3, i.e. $1.462 - 0.511$. The slope for TREAT = 3 is the sum of the B value for PRECOUNT3 and that for the interaction term [TREAT = 3]*PRECOUNT3, i.e. $1.462 - 0$. So the difference between the slopes is -0.511.

$$t = \frac{-0.511}{0.151} = -3.384$$

The degrees of freedom for t is $40-4 = 36$ (total number of observations in the groups compared - total number of parameters fitted; 2 slopes and 2 intercepts). Clearly, in this case the slopes for methods A and B do differ significantly.

In the above examples using datasets 2 and 3, the differences among treatment groups are so large that even a one-way ANOVA of POSTCOUNT scores will reject the null hypothesis that mean POSTCOUNT scores do not differ among treatment groups. Even in these cases, however, the covariance analysis reveals important features of the data.

ANCOVA with more than one treatment factor

Adding another factor

So far we have considered data that can be well understood using graphs. With more variables involved, graphical analysis can only give a series of partial pictures. Here we introduce an extra factor into our experiment (extra covariates can also be used). We now reveal that both male and female patients participated in our study. It is possible that gender could influence scores, so we did have half male and half female participants in each of our treatment groups, and now we use this extra information. In the data list of Table 5.1, we have arranged the data so that the first ten in each treatment group were men. The extra column of data can be seen in the dataset med.ancova.sav on the book website.

Requesting the ANCOVA to include all interactions

We refer to this new factor as SEX, and include it in the model for an analysis of dataset 2. The model is set up as in SPSS Dialog Box 5.1, adding the terms in this order: PRECOUNT2, TREAT, TREAT*PRECOUNT2, SEX, PRECOUNT2*SEX, TREAT*SEX, TREAT*PRECOUNT2*SEX, so we have all second order and the third order interactions

included. We enter the terms in the model in the order we believe is correct, and again use Type 1 sums of squares.

Making sense of the output

The results appear in SPSS Output 5.8. We can see at once that the third order interaction TREAT*SEX*PRECOUNT2 is not significant, and neither are the second order interactions TREAT*SEX, SEX*PRECOUNT2 and TREAT*PRECOUNT2. Removing these interaction terms from the model will give us more power to detect the main effects. If we do this we get the second table in SPSS Output 5.8 in which we see that the main effect of SEX is also non-significant (and has a small estimated effect). We are back to the model we found before, treatment methods represented by parallel regression lines of POSTCOUNT2 on PRECOUNT2 scores.

Tests of Between-Subjects Effects

Dependent Variable: postcount2

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^a
Corrected Model	50556.975 ^a	11	4596.089	50.717	.000	.921	557.886	1.000
Intercept	1078164.150	1	1078164.150	11897.325	.000	.996	11897.325	1.000
precount2	20074.203	1	20074.203	221.515	.000	.822	221.515	1.000
treat	29957.162	2	14978.581	165.286	.000	.873	330.571	1.000
treat * precount2	151.328	2	75.664	.835	.440	.034	1.670	.185
sex	36.139	1	36.139	.399	.531	.008	.399	.095
sex * precount2	15.786	1	15.786	.174	.678	.004	.174	.069
treat * sex	72.091	2	36.046	.398	.674	.016	.796	.111
treat * sex * precount2	250.266	2	125.133	1.381	.261	.054	2.762	.283
Error	4349.875	48	90.622					
Total	1133071.000	60						
Corrected Total	54906.850	59						

a. R Squared = .921 (Adjusted R Squared = .903)

b. Computed using alpha = .05

Tests of Between-Subjects Effects

Dependent Variable: postcount2

Source	Type I Sum of Squares	df	Mean Square	F	Sig.	Partial Eta Squared	Noncent. Parameter	Observed Power ^a
Corrected Model	50068.130 ^a	4	12517.032	142.277	.000	.912	569.106	1.000
Intercept	1078164.150	1	1078164.150	12255.105	.000	.996	12255.105	1.000
precount2	20074.203	1	20074.203	228.176	.000	.806	228.176	1.000
treat	29957.162	2	14978.581	170.256	.000	.861	340.512	1.000
sex	36.765	1	36.765	.418	.521	.008	.418	.097
Error	4838.720	55	87.977					
Total	1133071.000	60						
Corrected Total	54906.850	59						

a. R Squared = .912 (Adjusted R Squared = .905)

b. Computed using alpha = .05

SPSS Output 5.8. Analysis for dataset 2 with SEX as an additional factor