

Normal subjects were enrolled in a pilot study to compare blood levels among 4 doses (A, B, C, and D) of an antibiotic using a new aerosol delivery formulation. The response measure is a quantity called the area-under-the-curve (AUC) – a 4 period crossover design was used with a 3-day wash-out period between doses. Three subjects were randomized to each of four sequence groups ...

are there any differences in blood levels among the 4 doses?

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TITLE1 'Cross-Over Design';
TITLE2 'Antibiotic Blood Levels Following Aerosol Inhalation' ;
options nonumber nodate nocenter ls=80;
DATA INHAL1;
    INPUT SEQGRP $ PAT AUC1 AUC2 AUC3 AUC4 @@;
    DATALINES;
ABDC 102 2.31 3.99 11.75 4.78
ABDC 106 3.95 2.07 7.00 4.20
ABDC 109 4.40 6.40 9.76 6.12
BCAD 104 6.81 8.38 1.26 10.56
BCAD 105 9.05 6.85 4.79 4.86
BCAD 111 7.02 5.70 3.14 7.65
CDBA 101 6.00 4.79 2.35 3.81
CDBA 108 5.25 10.42 5.68 4.48
CDBA 112 2.60 6.97 3.60 7.54
DACB 103 8.15 3.58 8.79 4.94
DACB 107 12.73 5.31 4.67 5.84
DACB 110 6.46 2.42 4.58 1.37
;

DATA INHAL2; SET INHAL1;
    DOSE = SUBSTR(SEQGRP,1,1); PER = 1; AUC = AUC1; OUTPUT;
    DOSE = SUBSTR(SEQGRP,2,1); PER = 2; AUC = AUC2; OUTPUT;
    DOSE = SUBSTR(SEQGRP,3,1); PER = 3; AUC = AUC3; OUTPUT;
    DOSE = SUBSTR(SEQGRP,4,1); PER = 4; AUC = AUC4; OUTPUT;
RUN;

PROC SORT DATA = INHAL2; BY PAT PER;
DATA INHAL; SET INHAL2;
    KEEP PAT SEQGRP DOSE PER AUC CO;
    CO = LAG(DOSE); IF PER = 1 THEN CO = '0';
RUN;

PROC SORT DATA = INHAL; BY SEQGRP PER PAT;
PROC PRINT DATA = INHAL;
RUN;

PROC GLM DATA = INHAL;
    CLASS SEQGRP DOSE PER PAT CO;
    MODEL AUC = SEQGRP PAT(SEQGRP) DOSE PER CO;
    TEST H=SEQGRP E=PAT(SEQGRP);
RUN;
```

Cross-Over Design  
 Antibiotic Blood Levels Following Aerosol Inhalation

Obs	SEQGRP	PAT	DOSE	PER	AUC	CO
1	ABDC	102	A	1	2.31	0
2	ABDC	106	A	1	3.95	0
3	ABDC	109	A	1	4.40	0
4	ABDC	102	B	2	3.99	A
5	ABDC	106	B	2	2.07	A
6	ABDC	109	B	2	6.40	A
7	ABDC	102	D	3	11.75	B
8	ABDC	106	D	3	7.00	B
9	ABDC	109	D	3	9.76	B
10	ABDC	102	C	4	4.78	D
11	ABDC	106	C	4	4.20	D
12	ABDC	109	C	4	6.12	D
13	BCAD	104	B	1	6.81	0
14	BCAD	105	B	1	9.05	0
15	BCAD	111	B	1	7.02	0
16	BCAD	104	C	2	8.38	B
17	BCAD	105	C	2	6.85	B
18	BCAD	111	C	2	5.70	B
19	BCAD	104	A	3	1.26	C
20	BCAD	105	A	3	4.79	C
21	BCAD	111	A	3	3.14	C
22	BCAD	104	D	4	10.56	A
23	BCAD	105	D	4	4.86	A
24	BCAD	111	D	4	7.65	A
25	CDBA	101	C	1	6.00	0
26	CDBA	108	C	1	5.25	0
27	CDBA	112	C	1	2.60	0
28	CDBA	101	D	2	4.79	C
29	CDBA	108	D	2	10.42	C
30	CDBA	112	D	2	6.97	C
31	CDBA	101	B	3	2.35	D
32	CDBA	108	B	3	5.68	D
33	CDBA	112	B	3	3.60	D
34	CDBA	101	A	4	3.81	B
35	CDBA	108	A	4	4.48	B
36	CDBA	112	A	4	7.54	B
37	DACB	103	D	1	8.15	0
38	DACB	107	D	1	12.73	0
39	DACB	110	D	1	6.46	0
40	DACB	103	A	2	3.58	D
41	DACB	107	A	2	5.31	D
42	DACB	110	A	2	2.42	D
43	DACB	103	C	3	8.79	A
44	DACB	107	C	3	4.67	A
45	DACB	110	C	3	4.58	A
46	DACB	103	B	4	4.94	C
47	DACB	107	B	4	5.84	C
48	DACB	110	B	4	1.37	C

Cross-Over Design  
 Antibiotic Blood Levels Following Aerosol Inhalation

The GLM Procedure

Class Level Information

Class	Levels	Values
SEQGRP	4	ABDC BCAD CDBA DACB
DOSE	4	A B C D
PER	4	1 2 3 4
PAT	12	101 102 103 104 105 106 107 108 109 110 111 112
CO	5	0 A B C D

Number of Observations Read 48

Number of Observations Used 48

Cross-Over Design  
 Antibiotic Blood Levels Following Aerosol Inhalation

The GLM Procedure

Dependent Variable: AUC

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	20	225.1866862	11.2593343	2.90	0.0053
Error	27	104.8902450	3.8848239		
Corrected Total	47	330.0769312			

R-Square	Coeff Var	Root MSE	AUC Mean
0.682225	34.38658	1.970996	5.731875

Source	DF	Type I SS	Mean Square	F Value	Pr > F
SEQGRP	3	7.1111896	2.3703965	0.61	0.6142
PAT (SEQGRP)	8	48.6932667	6.0866583	1.57	0.1815
DOSE	3	134.4534729	44.8178243	11.54	<.0001
PER	3	3.9931229	1.3310410	0.34	0.7947
CO	3	30.9356342	10.3118781	2.65	0.0687

Source	DF	Type III SS	Mean Square	F Value	Pr > F
SEQGRP	3	10.3361626	3.4453875	0.89	0.4604
PAT (SEQGRP)	8	48.6932667	6.0866583	1.57	0.1815
DOSE	3	118.0792959	39.3597653	10.13	0.0001
PER	2	0.0628167	0.0314083	0.01	0.9920
CO	3	30.9356342	10.3118781	2.65	0.0687

Tests of Hypotheses Using the Type III MS for PAT(SEQGRP) as an Error Term

Source	DF	Type III SS	Mean Square	F Value	Pr > F
SEQGRP	3	10.33616258	3.44538753	0.57	0.6526

```

PROC GLM DATA = INHAL;
  CLASS SEQGRP DOSE PER PAT CO;
  MODEL AUC = SEQGRP PAT(SEQGRP) DOSE PER;
  LSMEANS DOSE / ADJ=TUKEY CL;
RUN;

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Antibiotic Blood Levels Following Aerosol Inhalation  
The GLM Procedure  
Dependent Variable: AUC

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	17	194.2510521	11.4265325	2.52	0.0129
Error	30	135.8258792	4.5275293		
Corrected Total	47	330.0769312			

Source	DF	Type III SS	Mean Square	F Value	Pr > F
SEQGRP	3	7.1111896	2.3703965	0.52	0.6694
PAT (SEQGRP)	8	48.6932667	6.0866583	1.34	0.2606
DOSE	3	134.4534729	44.8178243	9.90	0.0001
PER	3	3.9931229	1.3310410	0.29	0.8294

The GLM Procedure  
Least Squares Means  
Adjustment for Multiple Comparisons: Tukey

Least Squares Means for Effect DOSE

i	j	Difference Between Means	Simultaneous 95% Confidence Limits for LSMean(i)-LSMean(j)	
1	2	-1.010833	-3.372843	1.351176
1	3	-1.744167	-4.106176	0.617843
1	4	-4.509167	-6.871176	-2.147157
2	3	-0.733333	-3.095343	1.628676
2	4	-3.498333	-5.860343	-1.136324
3	4	-2.765000	-5.127010	-0.402990