Paper 203-2012
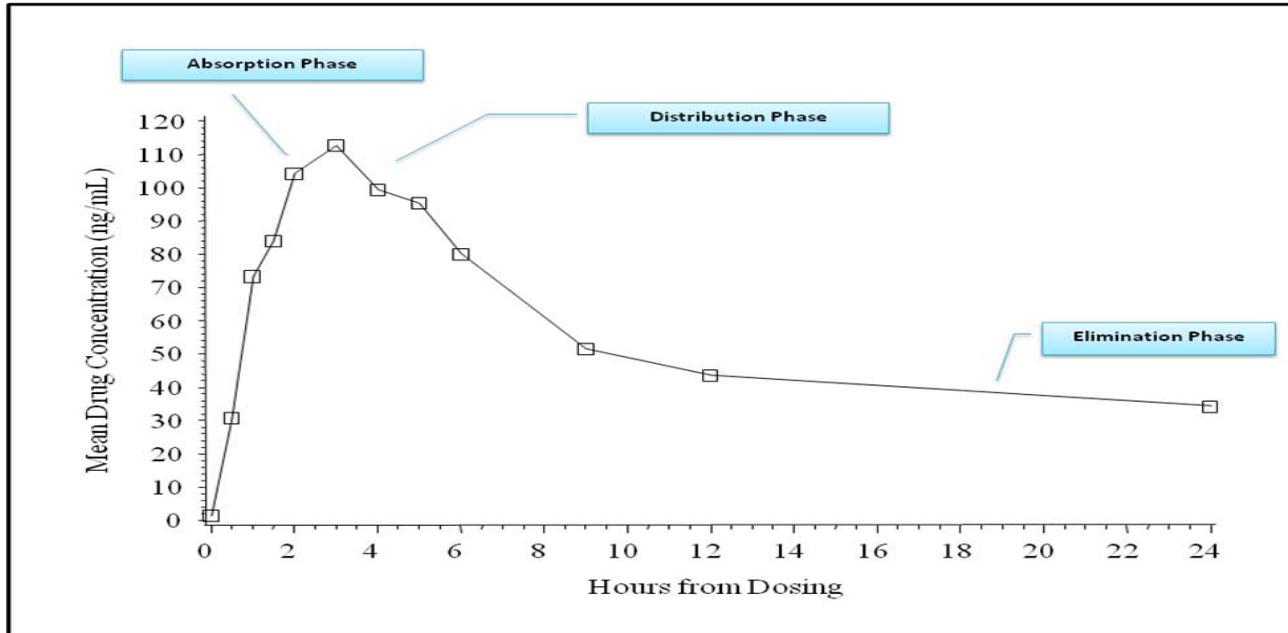
Creating Pharmacokinetic Graphs using SAS/GRAPH®
Katrina E. Canonizado, Celerion, Inc
Matthew Murphy, Celerion, Inc

ABSTRACT

Graphs are visual representation of the data. In a clinical trial setting, figures show the general trend and relationships of variables collected during the conduct of a study. In the pharmacokinetic world, graphs play an important role in showing the drug's concentration profiles. Line graph is a typical way of presenting drug's concentration. However, depending on the data available and the analysis required, different visual presentations, such as scatter plots, spaghetti plots and many more, are needed to fully communicate the information at hand. These graphs can be achieved by using SAS/GRAPH®. This paper demonstrates some available SAS/GRAPH® procedures, options and the Annotate facility for producing effective pharmacokinetic graphs.

INTRODUCTION

Pharmacokinetics (PK) may be defined as the absorption, distribution, metabolism, and excretion of an investigational drug. In a new drug development process, pharmacokinetic types of studies are conducted to examine what is happening to the drug within the body. These studies involve analyzing drug concentrations in samples collected from the blood, urine, feces and other matrices.

Graphs play an important role in interpreting drug concentration-time data as they provide a quick view of the general trend of the drug concentrations over time. A simple illustration of a concentration-time graph is presented below.
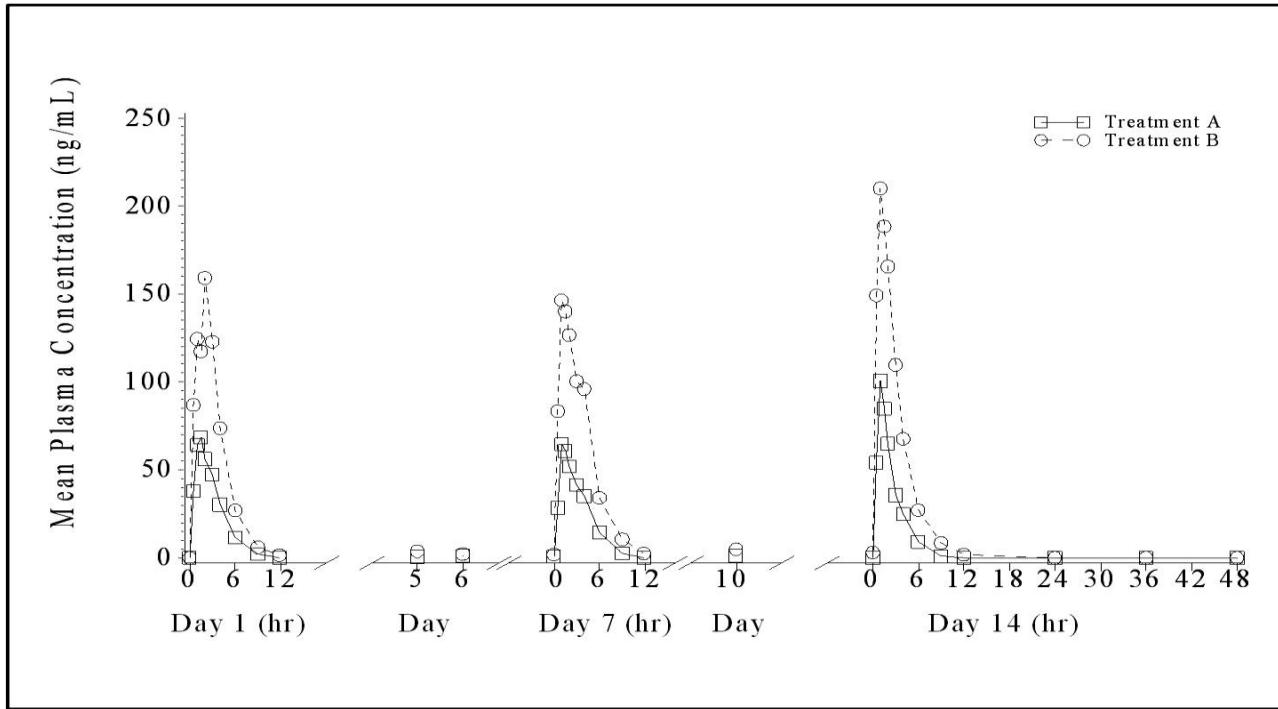

In general, this graph is very effective to show the mean concentration-time curves for a dataset with enough quantifiable concentrations over time; however not all cases of data received by programmers, biostatisticians and scientists fit into this picture. Every dataset is different, thus, requiring close attention given to the best way of presenting the information, in order that the data may be correctly interpreted. SAS/GRAPH® is the most common and available SAS tool to produce graphs. This paper highlights some components of SAS/GRAPH® which may be used to produce figures used for pharmacokinetic analysis.

ILLUSTRATION

Case 1:

Some clinical trials are designed to collect data over several periods of time, such as in the case of multiple-dose studies. One example of this study design is when subjects are dosed for series of days and blood samples are drawn at the particular time points. The graph below is one example of this study design. Subjects were dosed for series of days with trough (predose) and serial plasma samples were drawn on different study days.
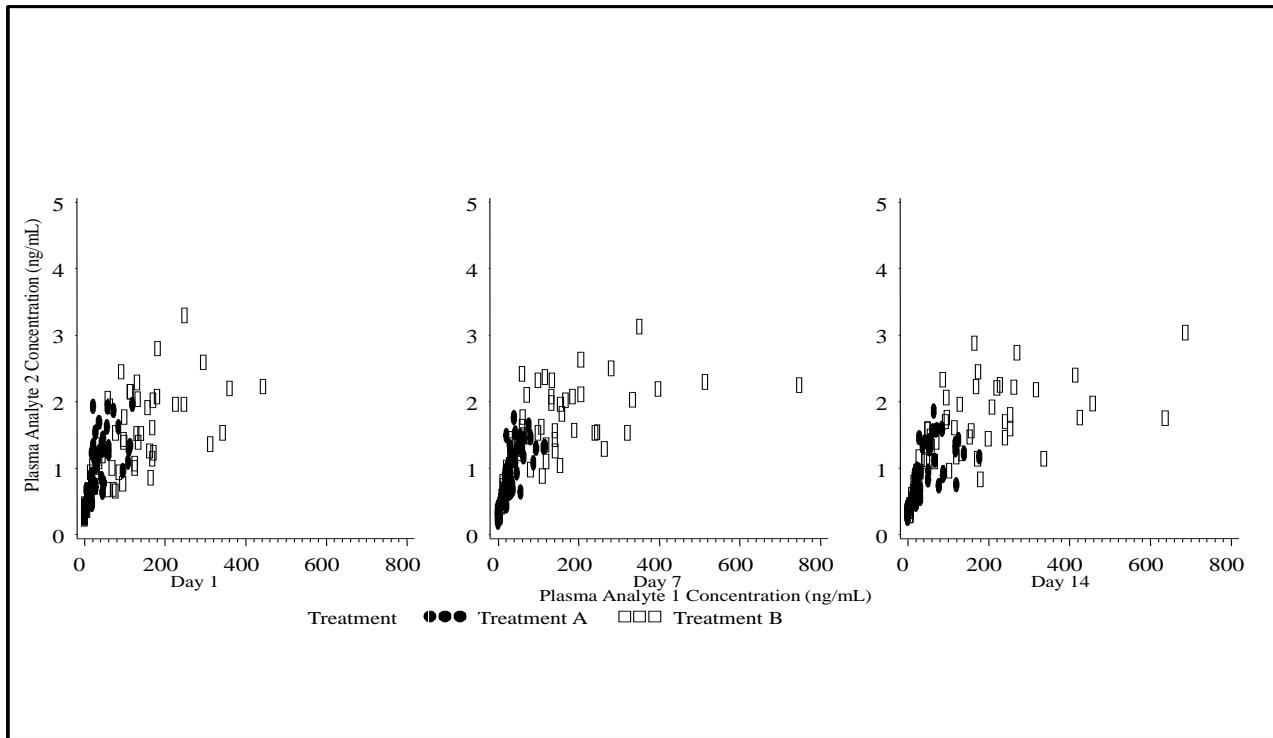

Partial SAS code:

```
PROC GPLOT DATA=ALLGRAPH GOUT=GRPH2 annotate=legend1;
PLOT c1*hour=1 / overlay HAXIS=AXIS1 VAXIS=AXIS2 nolegend noframe des=" " skipmiss;
run;
```

One useful GPLOT procedure (PROC GPLOT) option is the **skipmiss** option. The **skipmiss** option breaks a plot line when it encounters a missing value. Therefore, in order to include a break in the x-axis between days, when preparing the dataset to be plotted, an additional missing observation is added.

Case 2:

Another way of presenting multiple-dose studies is to have multiple graphs in one page. An advantage of this type of presentation is, it provides graphical comparisons of concentration profiles over different time points. The graph below compares the relationship of two plasma samples on different study days.
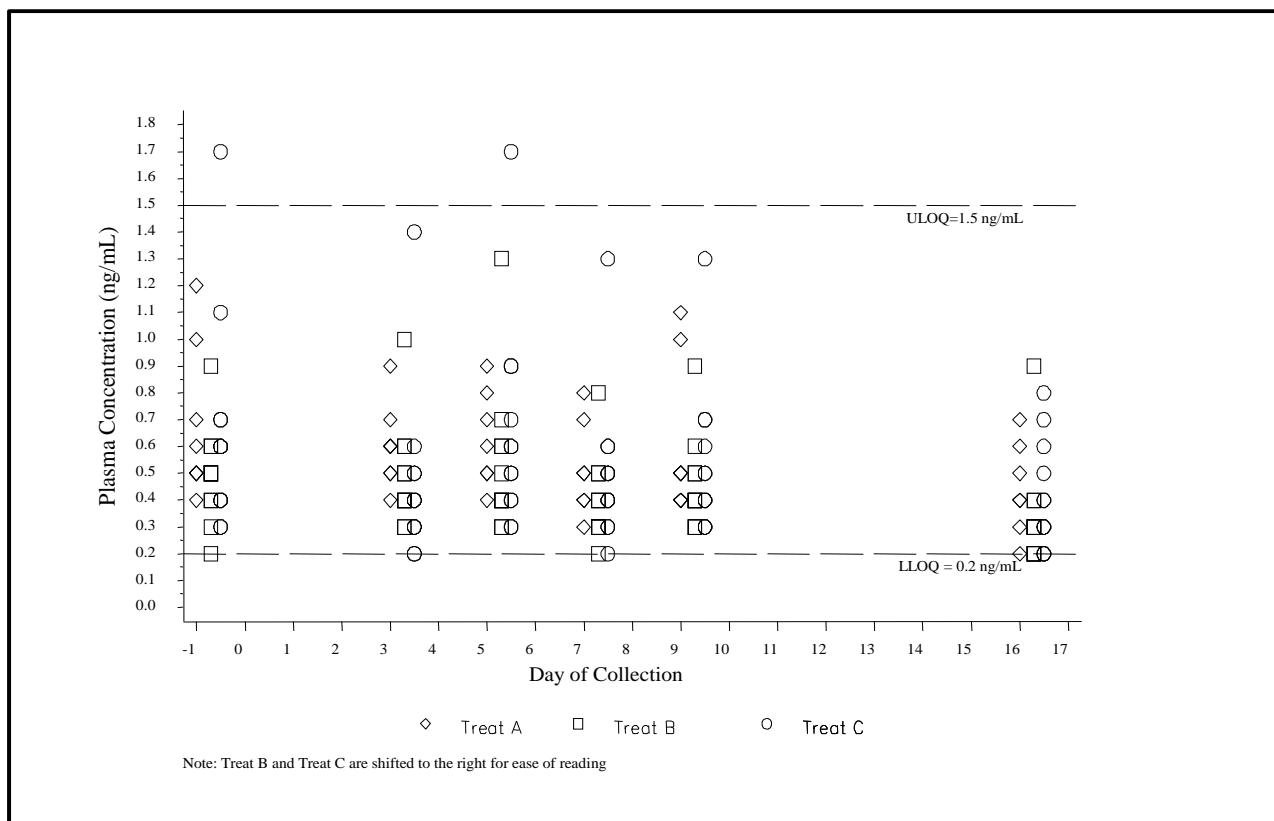

Partial SAS code:

```
PROC GREPLAY NOFS;
        TC=TEMPL1;
        TDEF TEMPL1 des=" "
            1/ llx=0    lly=0  ulx=0    uly=100 urx=33.3 ury=100 lrx=33.3 lry=0 color=white
            2/ llx=33.3 lly=0  ulx=33.3 uly=100 urx=66.6 ury=100 lrx=66.6 lry=0 color=white
            3/ llx=66.6 lly=0  ulx=66.6 uly=100 urx=100  ury=100 lrx=100  lry=0 color=white
            4/ llx=0    lly=0  ulx=0    uly=100 urx=100  ury=100 lrx=100  lry=0;
        TEMPLATE TEMPL1;
        IGOUT=GRPH1;
        list igout;
        list template;
        TREPLAY 1:1 2:2 3:3 4:4
        RUN;
 quit;
```

The GREPLAY procedure (PROC GREPLAY) can display and manage graphical outputs stored in SAS catalog and is very useful in displaying multiple graphs in one page.

Case 3:

Pharmacokinetic data can be visualized by subjects, and scatter plots are commonly used to display pharmacokinetic data by subject on a single graph. Scatter plots are particularly useful for interpreting PK data when there are a significant number of concentrations that are not quantifiable or missing, in which case mean concentration-time data may be misleading.

Partial SAS code:

```
AXIS2 length=%eval(&vl.+0) cm
      label=(h=0.45 cm font="&font." angle=90 "&name. (&un.)")
      value=(height=&h_v. cm)
      reflabel=(position=bottom j = r h=&h. cm font="&font."  "LLOQ = 0.2 &un." "ULOQ=1.5 &un.")
      order=(0 to 1.8 by 0.1)
      offset=(0.25 cm);

proc gplot data=all;
   plot conc*day1 = ptno / haxis=axis1 vaxis=axis2  vref=0.2 1.5 lvref=4 annotate=legend1
nolegend noframe;
```
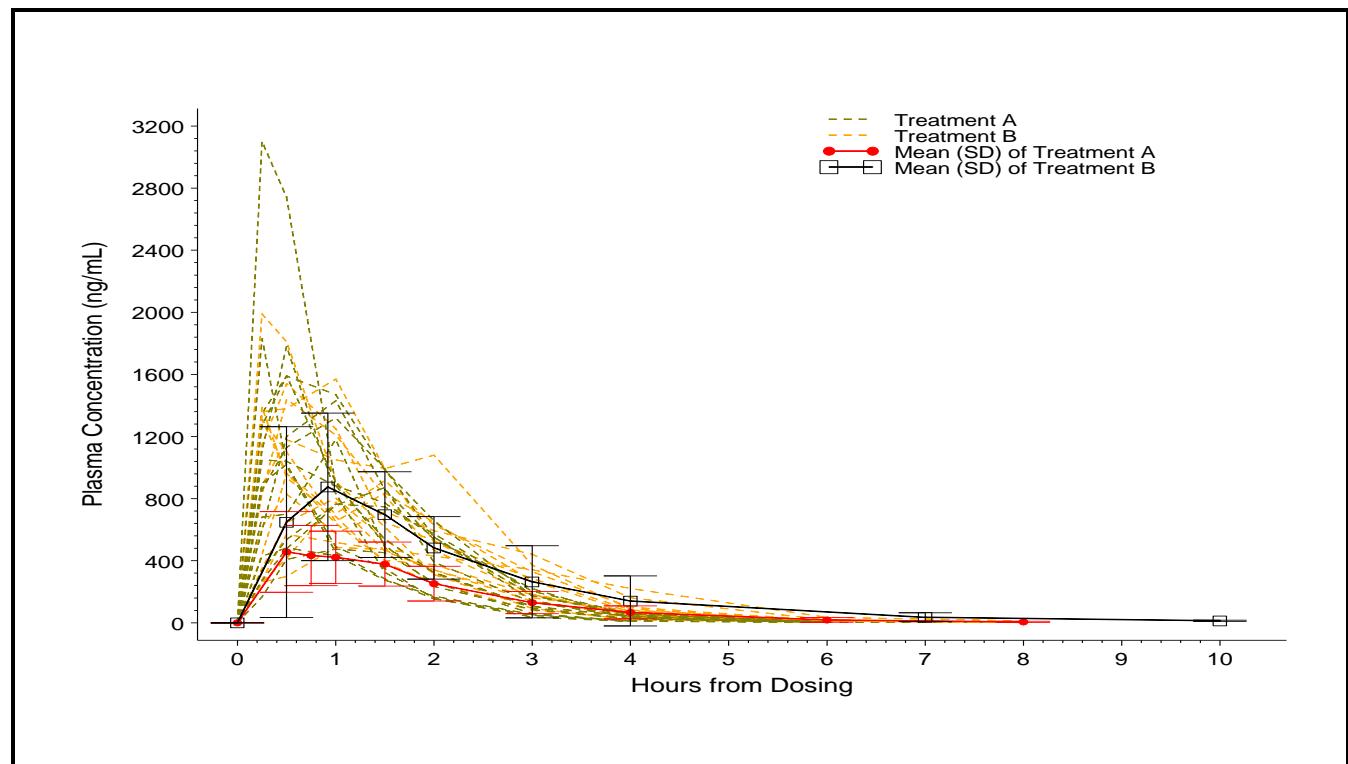
Another useful PROC GPLOT options are **vref** and **href**. These options are commonly used when there is a need to include reference lines. In general, **vref** and **href** draw horizontal and vertical lines, respectively in the graph. The **lvref** specifies the line type to be used to draw the reference lines. The **reflabel** adds a label or text that goes with the reference line as shown in the code above**.**

Case 4:

Another option for presenting individual concentration-time data is a spaghetti plot. Spaghetti plots show variability between individual concentration data at a given time. Moreover, there are cases where there is a need to overlay the mean (+/- standard deviation [SD]) concentration profile to have an overall clear understanding of the concentration-time data.

Partial SAS code:

```
%let num=1;
%let stat=std;
%macro build;
if group=2 then do;
   color='red';
   line=1;
end;
else if group=3 then do;
   color='black';
   line=1;
end;
%mend build;
data anno2 (drop=conc mean std);
  length color function style $8.;
  retain xsys ysys '2' when 'a';
  set b;
  if std ne .;
/* Lower tick */
function='move'; xsys='2'; x=hour; y=mean-(&num.*&stat.); output;
function='draw'; xsys='9'; x=-2; y=mean-(&num.*&stat.); %build; output;
function='draw'; xsys='9'; x=+4; y=mean-(&num.*&stat.); %build; output;
/* Upper tick */
```

```
function='move'; xsys='2'; x=hour; y=mean+(&num.*&stat.); output;
function='draw'; xsys='9'; x=-2; y=mean+(&num.*&stat.); %build; output;
function='draw'; xsys='9'; x=+4; y=mean+(&num.*&stat.); %build; output;
/* Join upper and lower*/
function='move'; xsys='2'; x=hour; y=mean-(&num.*&stat.); output;
function='draw'; xsys='2'; x=hour; y=mean+(&num.*&stat.); %build; output; run;
```

Using Annotate facility allows for better control of graph settings. The user can specify exactly the x and y coordinates, where to draw a line, and many more features that can produce more customized graph. Some common but very powerful functions used to create the graph above include:
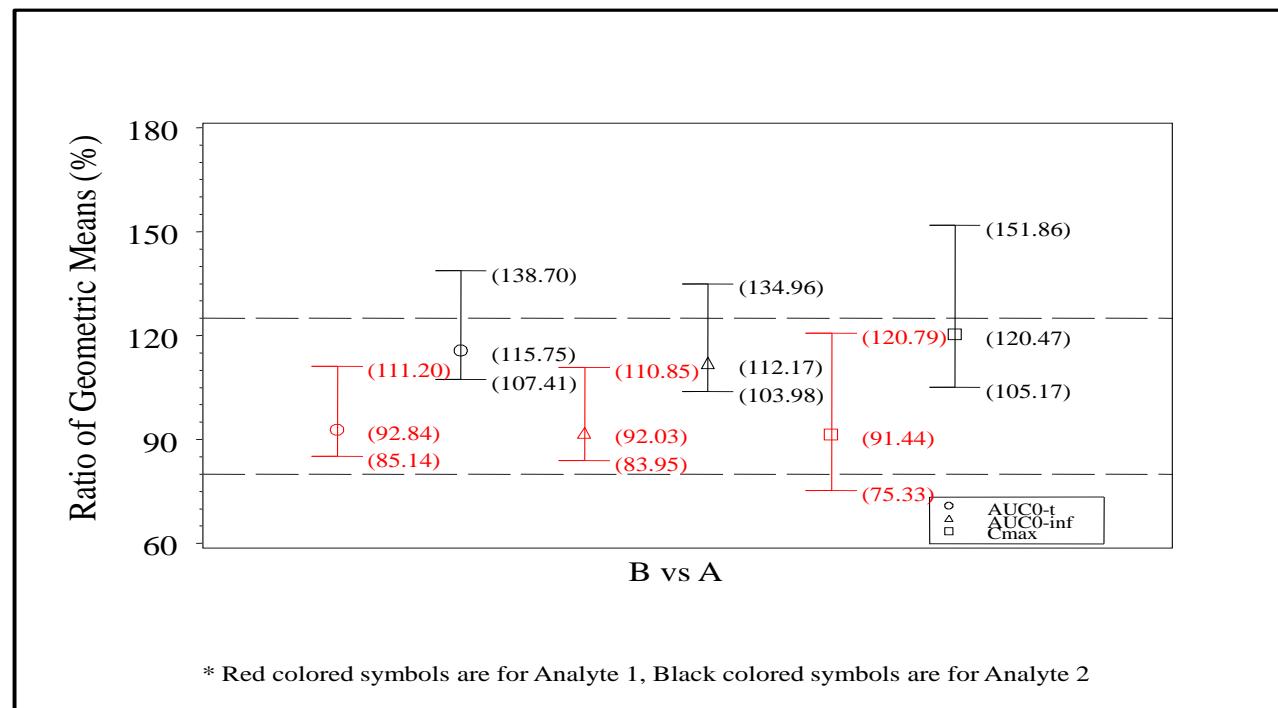
- **Move** allows the user to move the pointer to a specific location.
- **Draw** allows the user to draw a line.
- **Xsys** and **Ysys** specify a value that represents a coordinate system.

__Case 5__:

In addition to graph drug concentrations-time data, graphs that show relationships of pharmacokinetic parameters are often done as part of the analysis. Common pharmacokinetic endpoints are presented below:

Parameter	Definition
Cmax	Maximum plasma concentration
AUC0-t	Area under the plasma concentration-time curve from zero to time t, where t is the time of the last measurable concentration
AUC0-∞	Area under the plasma concentration-time curve from zero to extrapolated infinity

As per FDA guidelines, bioequivalence criteria are met if the 90% confidence intervals of the ratios of the mean fell within the range of 80% to 125% for logarithmically transformed Cmax, AUC0-t and AUC0-∞. The graph below shows an effective way of visualizing whether bioequivalence criteria are met, per this guidance.
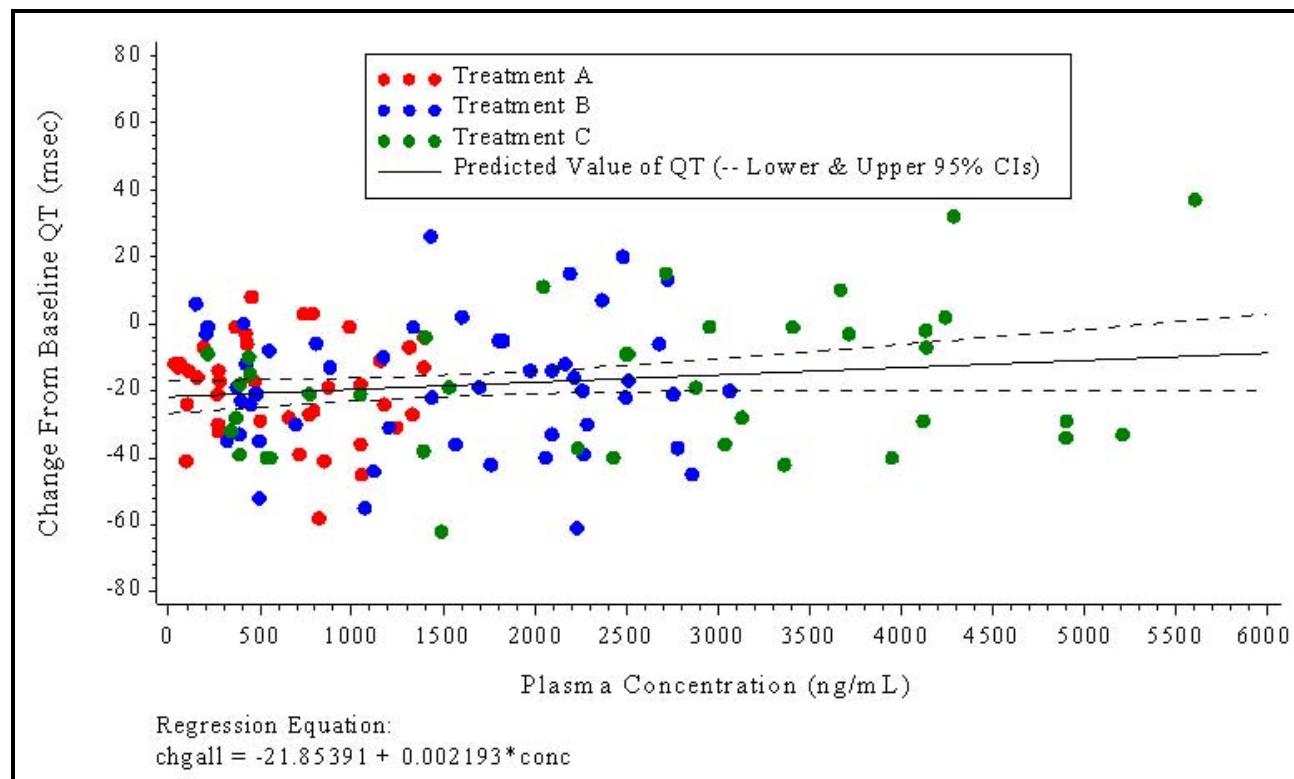
Partial SAS code:

```
data anno2;
  length color function style $8.;
  retain xsys ysys '2' when 'a';
  set ratio;
function='label';
     x=point + 0.3;  y=ratio; position='6';
     text='   ' || trim(left(cratio));  size=1.3;
     output;
function='label';
     x=point +0.3 ;  y=LL; position='6';
     text='   ' || trim(left(cratiol));  size=1.3;
     output;
function='label';
     x=point + 0.3;  y=UL; position='6';
     text='   ' || trim(left(cratiou));  size=1.3;
     output;
run;
```

Another Annotate feature is the **label** function. The **label** function allows to add text in the specified x and y coordinates.

Case 6:

It can be useful to plot clinical data such as vital signs and electrocardiogram (ECG) data against pharmacokinetic concentration data in order to spot existing relationships between the two parameters. Below is an example of a graph that shows ECG data (Change from Baseline QT measurement) plotted against the time-matched plasma concentration for each subject by treatment. Regression lines can then be added to display predicted values and confidence limits.

Partial SAS code:

```
symbol1  i=none  v=dot  h=0.4 cm  c=red;
symbol2  i=none  v=dot  h=0.4 cm  c=red;
symbol3  i=none  v=dot  h=0.4 cm  c=red;
symbol4  i=rlclm95 c=black;      /*mean predicted values*/
/*symbol4  i=rlcli95 c=black;*/  /*individual predicted values*/

proc gplot data=final;
plot chga*conc=1 chgb*conc=2 chgc*conc=3 chgall*conc=4 / vaxis=axis1 haxis=axis2 legend=legend2
noframe overlay regeqn;
```

In the symbol statements below, the **Interpol** option can be used to produce linear regression plots.

- **i=rlclm95** requests a linear regression with 95% confidence limits based on the mean predicted values.
- **i=rlcli95** requests a linear regression with 95% confidence limits based on the individual predicted values.
- In the plot statement, the **regeqn** option allows the derived regression equation to be displayed below the graph.

CONCLUSION

Datasets that are not presented well can lead to misinterpretation of the data which may lead to an erroneous decision. This paper demonstrated some helpful ways to visualize pharmacokinetic data effectively. As seen in this paper, creating graphs using SAS® procedures such as PROC GPLOT and PROC GREPLAY, together with the available plotting options, keeps the programming from becoming more complicated than it needs to be. The ultimate goal is to visually communicate the data at hand that scientists can use to interpret and draw conclusions.

REFERENCES AND RECOMMENDED READING

SAS 9.1.3 Online Documentation.

Kirsila,Tiina. *PK Plots With SGPLOTS.*PhUSE, 2011.

ACKNOWLEDGMENTS

We would like to thank our colleagues in the Biostatistics and Pharmacokinetics groups in Clinical Pharmacological Sciences at Celerion who provided comments on earlier drafts. We would like to thank especially Dr. Toni Grant for her insights and guidance.

CONTACT INFORMATION

Katrina E. Canonizado
Celerion (Remote)
West Covina, CA 91792
katrina.canonizado@celerion.com

Matthew Murphy
Celerion
22-24 Lisburn Road
Belfast BT9 6AD Northern Ireland, UK
matthew.murphy@celerion.com