Paper SP08

Estimating terminal half life by non-compartmental methods with some data below the limit of quantification

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ABSTRACT

In pharmacokinetic studies it occurs more or less frequently that some measurements are reported to be below the limit of quantification. These data are usually discarded in estimating the terminal half-life by non-compartmental methods. We modify this approach to allow for incorporating data below the quantification limit into the analysis. This is achieved by using maximum likelihood methods for censored data. The two approaches yield identical estimates in case of no censoring. The performance of the standard approach and the maximum likelihood approach is compared in some one- and two-compartment models by simulation.

INTRODUCTION

Pharmacokinetic data consist of a series of concentration measurements made in some blood compartment (usually plasma) at particular intervals after (and possibly before) administration of a drug. We consider only those cases where the measured substance is not present in untreated individuals so that the observed plasma levels are entirely attributable to the administered dose of the drug. Depending on the substance measured and the assay used, it occurs more or less frequently that some values are reported to be below the limit of quantification.

The limit of quantification is the lowest concentration at which the assay has been validated. This implies that measurements below this limit, which might be perfectly reasonable estimates of the true concentration, are not reported as such because they do not meet the acceptance criteria of the validation study, for example a coefficient of variation of maximum 20%. Statistically, however, there is no need to exclude measurements from the analysis solely because the coefficient of variation is above 20% or some other limit. Beal [1] discussed further issues related to the quantification limit in pharmacokinetic analyses and points out that "the concern about just how to handle BQL observations is a little misplace; there should be greater concern about QL validation". The present paper, though, focuses only on the former, i.e. on statistical methods of dealing with data below the quantification limit.

There are two basically different approaches for estimating subject-specific pharmacokinetic parameters from individual concentration profiles. With the first approach, a parametric nonlinear regression curve is fitted to the data. Parameterisation is usually based on compartmental models. Beal [1] evaluated different ways to fit a one-compartment model to pharmacokinetic data with some values below the limit of quantification. A practical and yet adequate method seems to be to simply discard the values below the quantification limit. Another option is to impute a statistical distribution function and apply maximum likelihood methods for censored data (see e.g. [2]). According to Duval and Karlsson [4] omission of censored data from the analysis may induce a not negligible bias on the parameter estimates of a two-compartment model.

In contrast to parametric analysis, non-compartmental analysis does not resort to a parametric model for the subject-specific profiles. A crucial point in non-compartmental analysis is estimation of the terminal half-life. A frequently applied procedure for this purpose is as follows. The data are log-transformed and linearly regressed on time by the method of ordinary least squares. The regression is repeated using first the last three points, then the last four points, then the last five points, and so on. The negative regression coefficient of the fit with the largest adjusted coefficient of determination (adjusted R^2) is taken as estimate of the terminal elimination rate constant. There are a number of variants of this procedure, though (e.g. [6] and [8]). It is to be noticed that the adjusted R-squared criterion is purely empirical, i.e. there is no theoretical justification for it. Some authors do not propose any algorithm for selecting the terminal subset (e.g. [5]). If some observations are below the quantification limit it is recommended to simply discard these data (see e.g. [5]).

To account for data below the quantification limit in non-compartmental analysis a natural generalization is to fit the terminal regression lines not by ordinary least squares but rather by the method of maximum likelihood assuming a normal distribution with constant variance. In this analysis, data below the quantification limit are treated as censored observations. Without censoring, ordinary least squares and maximum likelihood yield identical estimates of the regression coefficient. For selecting the best fitting regression line the definition of adjusted R^2 must be extended to

maximum likelihood analyses with censored observations. This is accomplished by considering the relationship between R^2 and the likelihood ratio statistic in the case of no censoring.

The paper is organized as follows. First a standard non-compartmental method for estimating the terminal half-life is described. Then a definition of R^2 and adjusted R^2 in maximum likelihood analyses with censored observations is proposed. It is shown how these statistics can be easily computed with the SAS/STAT® procedure LIFEREG. Finally, the performance of the standard approach and the maximum likelihood approach is compared in some one- and two-compartment models by simulation.

METHODS

Let *C* denote drug concentration values at times *T* in some blood compartment after a single intravenous bolus administration. Let *n* be the total number of observations. The quantification limit *Q* of the measurements is greater than zero. The data *C* are either at or above the quantification limit or they are reported to be below the quantification limit. Let *Y* denote the natural logarithm of *C* or that of the quantification limit *Q*, whichever is larger:

$$Y = \log [\max (C , Q)]$$

The variable *Z* indicates censoring:

$$Z = 1 \text{ if } C < Q \quad \text{and} \quad Z = 0 \text{ if } C \geq Q$$

We compare the following two non-compartmental methods for estimating the terminal half-life.

(1) Discarding values below the quantification limit

The analysis dataset *D* consists of all measurements at or above the quantification limit. Let *m* be the number of these measurements. Let D_k denote the subset of *D* consisting of the chronologically last *k* points (*k*=3…*m*). For all terminal subsets D_k a linear regression model with dependent variable *Y* and independent variable *T* is fitted by the method of least squares. Let R^2_k denote the coefficient of determination and b_k the estimated regression coefficient. The adjusted coefficient of determination for a regression with *k* observations is generally defined as

$$R^2_{adj} = 1 - (1 - R^2) (k - 1) / (k - 2)$$

The estimated terminal elimination rate λ is defined as the negative regression coefficient of the fit with the largest adjusted coefficient of determination. Technically, if there is more than one fit with largest R^2_{adj} value (which occurs with zero probability) then the fit with the larger number of points is used, i.e.

$$\lambda = - b_r \quad \text{with} \quad r = \max \{ s : R^2_{s,adj} = \max \{ R^2_{k,adj} : k = 3, 4, \ldots m \text{ and } b_k < 0 \} \}$$

The estimated terminal half-life is $\log(2)/\lambda$ [5].

(2) Using censored data methods if some values are below the quantification limit

In this case, the analysis dataset *A* comprises all observations. The terminal subsets including the chronologically last *k* data points are denoted by A_k. For all terminal subsets A_k with at least three observations above the quantification limit a linear regression model with dependent variable *Y*, censoring variable *Z*, and independent variable *T* is fitted by the maximum likelihood method. The statistical distribution of *Y* is assumed to be normal with constant variance. In a simple linear regression model with *k* observations and no censoring the following relations hold between the coefficient of determination R^2, the *F*-statistic *F*, and the logarithm Λ of the likelihood-ratio statistic (see e.g. [3] for the first relation and [7] for the second):

$$R^2 = F / [F + (k - 2)] \quad \text{and} \quad 2 \Lambda = k \log [1 + F / (k - 2)]$$

It follows that

$$R^2 = 1 - \exp (- 2 \Lambda / k)$$

We thus define R^2 in case of censoring by the same equation, but with *k* replaced by *q*, the number of observations above the quantification limit:

$$R^2 = 1 - \exp (- 2 \Lambda / q)$$

A possible alternative for *q* would be to use the so-called effective sample size (see [7], Chapter 6.5.2). This, however, would require some additional programming and we have not seen an advantage for this choice in the simulation studies. As a consequence, adjusted R^2 is defined by

$$R^2_{adj} = 1 - (1 - R^2) (q - 1) / (q - 2)$$

Estimation of the terminal elimination rate λ and the terminal half-life now continues as above by selecting the fit with the largest adjusted coefficient of determination.

IMPLEMANTATION WITH SAS/STAT

Implementation of linear regression is straightforward, e.g. using the procedure REG. For the second method with censored data one needs to calculate the log-likelihood ratio statistic, which is the difference of two maximum log-likelihood values, one for the full model with intercept and independent variable *T* and the other for the null model including only the intercept. The procedure LIFEREG can be used for this purpose. It is advantageous to regress the negative value of *Y* on time because then censoring is from above and can be easily modelled with the procedure LIFEREG. With minus_Y denoting the negative of *Y* the model statement is

```
model minus_Y * Z(1) = T / d=normal;
```

for the full model and

```
model minus_Y * Z(1) = / d=normal;
```

for the null model. It must be observed that the regression coefficient with respect to $-Y$ is the negative of the regression coefficient with respect to *Y*. The following example presents the result of the adjusted R^2 calculations for an artificial dataset.

EXAMPLE

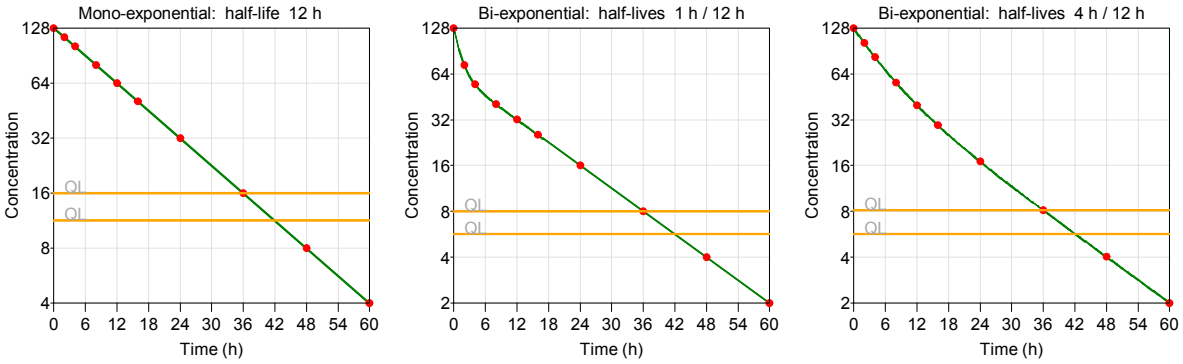
In the dataset below the quantification limit is $Q=16$. The variable names `T, Z, C, Y` refer to the variables *T*, *Z*, *C*, *Y* of the methods section above. The last two observations are censored ($Z=1$), i.e. the measured concentration *C* is below the quantification limit of 16.

```
data pk;
  input T Z C;
  Y = log(C);
  minus_Y = -Y;
cards;
 8  0  91.3
12  0  49.6
16  0  54.1
24  0  40.9
36  0  16.1
48  1  16.0
60  1  16.0
run;

proc reg data=pk(where=(not Z));
  model Y = T;
quit;

proc lifereg data=pk;
  model minus_Y * Z(1) = T / d=normal;
run;

proc lifereg data=pk;
  model minus_Y * Z(1) =   / d=normal;
run;
```

The adjusted R^2 value for the linear regression of the uncensored observations is 0.88946; the negative regression coefficient is 0.054668. For the LIFEREG procedures, the values of the log-likelihood function of the full model and the null model are 1.96953 and -8.31426, respectively, so that the log-likelihood ratio statistic Λ is 1.96953-(-8.31426), i.e. 10.28380. The number of observations above the quantification limit is $q=5$. Thus R^2 is 0.98365 and R^2_{adj} is 0.97820. The regression coefficient of the time variable T is 0.054677.

SIMULATION STUDY

Six scenarios were considered in the simulation study, i.e. three different kinetic curves and two different quantification limits (QL) for each, as depicted in the graphs below.



The terminal half-life was 12 hours in each case. The first kinetic profile is mono-exponential and the other two are bi-exponential. The initial half-life associated with the bi-exponential profiles is short (1 h) in one case and long (4 h) in the other case. Measurement times are 0, 2, 4, 8, 12, 16, 24, 36, 48, and 60 hours after administration. The quantification limit (QL) was chosen such that it was either reached at 36 h, i.e. exactly at a measurement time, or at 42 h, i.e. in the middle between two measurement times. The former is a worst case scenario for the quantification limit. The coefficient of variation of the simulated data was fixed at 0.2. The variation of the measured data comprises assay variation and variation caused by the biological system.

For each of the six scenarios a dataset with 100 000 kinetic profiles was simulated. For each scenario a newly generated seed number was used. The terminal half-life was estimated from the simulated data by the two methods described above.

RESULTS OF THE SIMULATION STUDY

The results of the simulation study are presented in the table below. The first table shows descriptive statistics of the estimated terminal half-life. For ease of reference, the method discarding data below the quantification limit will be called "discarding-method" while the method applying censored data methods will be called "censoring-method".

In the mono-exponential scenario, if the quantification limit was reached exactly at the measurement time at 36 h the log-transformed terminal half-life was overestimated by the mean of the discarding-method and underestimated by the mean of the censoring-method. The estimated bias was +0.036 for the discarding-method and -0.016 for the censoring-method, which translates into a bias of the geometric mean of +3.6% and -1.6%, respectively. The bias, however, was small compared to the variation of the estimates: the quartile ranges of the estimates were 0.26 and 0.25, respectively.

If the quantification limit was reached in the middle between two measurement times (42 h), the bias of the geometric mean value was qualitatively similar, but smaller in magnitude.

For the bi-exponential kinetic with short initial half-life the bias of the geometric mean was negative for both methods but still negligible compared to the variation of the estimates.

For the bi-exponential kinetic with long initial half-life the bias of the geometric mean was substantial. In the worst case (QL at 36 h) the estimated bias was -18% for the discarding-method and -17% for the censoring-method. In the best case (QL at 42 h) the estimated bias was -15% for the discarding-method and -13% for the censoring-method. The bias was mainly caused by the long initial phase with its half-life of 4 h. The initial phase still influences the curve at 8 h, 12 h and 18 h, but these points also contribute to estimating the terminal half-life. In other words, the terminal phase is not adequately represented by the study design. The bias is slightly smaller for the censoring-method because the first time point used for estimating the terminal elimination rate (10.7 h on average) is later and thus less influenced by the initial phase than it is for the discarding-method (8.1 h on average, see table below).

Results of simulation study: Statistics of estimated terminal half-life

Kinetic profile	Time QL reached	Statistic	Method for data below QL	Mean	Median	Standard deviation	Quartile range
Mono-exponential	36 h	Bias of log-transformed half-life estimates	Discarded	0.0362	0.0269	0.262	0.263
			Censored	– 0.0157	– 0.0228	0.235	0.254
		Bias of half-life estimates	Discarded	0.933	0.327	4.77	3.26
			Censored	0.152	– 0.271	3.03	3.00
		First time point used in regression	Discarded	6.60	4	6.13	12
			Censored	8.87	12	5.64	8
	42 h	Bias of log-transformed half-life estimates	Discarded	0.0092	– 0.0092	0.209	0.232
			Censored	– 0.0115	– 0.0143	0.192	0.253
		Bias of half-life estimates	Discarded	0.405	– 0.110	3.24	2.78
			Censored	0.089	– 0.171	2.45	3.00
		First time point used in final regression	Discarded	8.33	8	6.85	16
			Censored	10.67	12	6.13	12
Bi-exponential (short initial phase)	36 h	Bias of log-transformed half-life estimates	Discarded	– 0.0069	– 0.0222	0.276	0.314
			Censored	– 0.0232	– 0.0368	0.242	0.273
		Bias of half-life estimates	Discarded	0.440	– 0.263	4.74	3.70
			Censored	0.084	– 0.433	3.13	3.19
		First time point used in regression	Discarded	7.11	4	5.80	10
			Censored	9.26	12	5.23	8
	42 h	Bias of log-transformed half-life estimates	Discarded	– 0.0066	– 0.0361	0.218	0.250
			Censored	– 0.0153	– 0.0223	0.194	0.262
		Bias of half-life estimates	Discarded	0.245	– 0.425	4.41	2.94
			Censored	0.048	– 0.264	2.47	3.10
		First time point used in regression	Discarded	8.95	8	6.32	14
			Censored	11.02	12	5.63	12
Bi-exponential (long initial phase)	36 h	Bias of log-transformed half-life estimates	Discarded	– 0.1808	– 0.2095	0.256	0.302
			Censored	– 0.1690	– 0.1875	0.245	0.290
		Bias of half-life estimates	Discarded	– 1.617	– 2.268	3.32	2.99
			Censored	– 1.547	– 2.052	2.75	2.93
		First time point used in regression	Discarded	6.40	4	6.20	12
			Censored	8.90	12	5.66	8
	42 h	Bias of log-transformed half-life estimates	Discarded	– 0.1489	– 0.1904	0.205	0.234
			Censored	– 0.1282	– 0.1497	0.195	0.265
		Bias of half-life estimates	Discarded	– 1.418	– 2.081	2.59	2.37
			Censored	– 1.234	– 1.668	2.26	2.78
		First time point used in regression	Discarded	8.09	8	6.86	16
			Censored	10.70	12	6.07	12

QL: quantification limit; each statistic is based on 100 000 simulated datasets.

We compared the two methods with respect to the absolute deviation of the estimated half-life from the true half-life. The absolute deviation was rounded to one decimal place and the two methods were compared by the signed-rank statistic. The censoring-method compared favourably to the discarding-method for all six scenarios. The same held true if the squared deviation was used. The same held also true if the absolute or squared deviation between the estimated log-half-life and the true log-half-life was employed as criterion (rounded to two decimal places).

SUMMARY

For a non-compartmental analysis of pharmacokinetic data a method was introduced that incorporates values below the quantification limit for estimating the terminal half-life. The method uses a linear regression model of the log-transformed concentration values on time. The estimates are derived by the maximum likelihood method. This "censoring-method" was compared to a "discarding-method" where the values below the quantification limit are excluded from the analysis. The two methods yield identical estimates if all data are above the quantification limit.

A simulation study was carried out to compare the performance of the two methods. Only a very limited set of scenarios was considered and different study characteristics may lead to different conclusions.

In the scenarios considered, the censoring-method was superior to the discarding-method. The margin, however, was very small compared to the variation of the estimates so that not much seems to be lost if data below the quantification limit are ignored in the analysis.

For both methods it is equally essential that the terminal phase of the kinetic profile is adequately covered by the data and that the quantification limit is small enough to obtain sufficient measurements above the quantification limit during the terminal phase.

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