Sensitivity analysis of a mixture model to determine genotype/phenotype

Ivan Matthews and Leon Aarons
School of Pharmacy and Pharmaceutical Sciences, University of Manchester, UK

Introduction

Mixture models are useful in population pharmacokinetics and pharmacodynamics to characterise underlying population distributions (two or more subpopulations) that are not adequately explained by the evaluated covariates. When applied in NONMEM, the subpopulation, to which an individual is classified, can be determined from the maximum *a posteriori* Bayesian post-hoc estimates.

Polymorphic metabolising enzymes, such as those from the cytochrome P450 (CYP) family, may give rise to a multimodal clearance distribution. Their involvement in the elimination of many drugs means that the use of a mixture model may be of value to categorise the phenotype or genotype of a patient. Several common drugs such as warfarin, metoprolol, ibuprofen, and phenytoin are eliminated via a polymorphic enzyme pathway.

A previous population pharmacokinetic analysis of warfarin with a NONMEM mixture model resulted in the model incorrectly assigning all of the poor metabolisers [1]. A sensitivity analysis, described below, was proposed to determine the potential reasons for this failure.

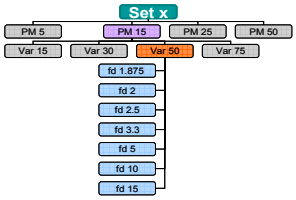
Objective

Determine the factors that most strongly affect the ability of NONMEM mixture models to correctly predict the genotype or phenotype of an individual.

Methodology

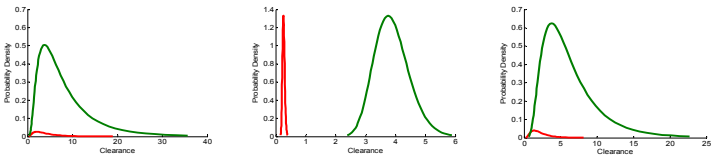
Study Design

A sensitivity analysis of a NONMEM mixture model was undertaken. A one compartment disposition model with first order input and first order elimination was used for the simulations [2]. A discrete clearance model was used to give the poor metabolisers a lower clearance. Each simulation had 100 virtual patients receiving a single dose at time zero with 15 log equispaced observations over 24 hours plus one observation at 30 hours. All patients were simulated without any additional covariates. In all there were 112 simulations per set.



The diagram shows the parameters varied in the simulation (only one arm shown in full). The percentage of poor metabolisers (5, 15, 25, 50 %), the between subject variability (15, 30, 50, 75 %) on the structural model parameters (ka, CL, V) and the typical value of clearance for the poor metabolisers (0.25, 0.375, 0.75, 1.125, 1.5, 1.875, 2 L.h^{-1}) relative to extensive metabolisers (3.75 L.h^{-1}) → a fold difference in clearance of 15, 10, 5, 3.3, 2.5, 2 and 1.875.

Fifteen unique sets were simulated and analysed using NONMEM. The results were expressed as the percentage of false negatives and the percentage of false positives. Where false negatives are poor metabolisers who are assigned as extensive metabolisers and false positives are extensive metabolisers who are assigned as poor metabolisers.



Graphs showing simulated Clearance distributions for poor metabolisers (---) and extensive metabolisers (---). Left: 75% variability, 5% poor metabolisers, 2 L.h^{-1} TVCL$_{PM}$ (fd 1.875). Middle: 15% variability, 50% poor metabolisers, 0.25 L.h^{-1} TVCL$_{PM}$ (fd 15). Right: represents warfarin on the same scale as our simulated compound, 60% variability, 6% poor metabolisers, fd 2.78 (TVCL$_{PM}$ 1.35 L.h^{-1}).

Decision Rule

Two multivariate nonlinear regressions were performed in Mathematica [3] to define the border between a mixture model failure and a success. A failure was defined as a false positive or negative incidence of greater than 2.5%. The equations for each of the regressions were used to develop 2 criterion values, Crit$_{FN}$ (1) and Crit$_{FP}$ (2). If either or both of these criterion values are negative (3) then the mixture model can be reasonably expected to fail.

Computation

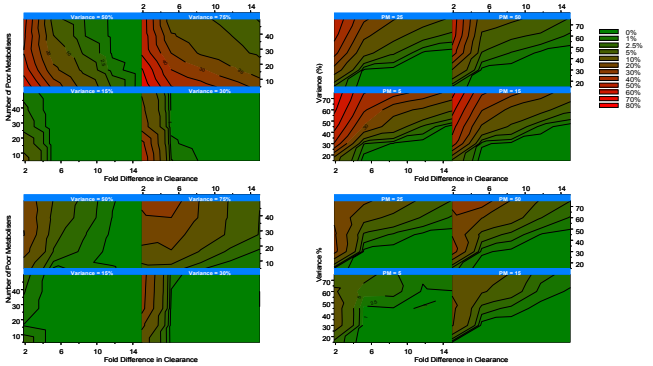
Simulations and analysis was performed using NONMEM version 6ß [4] in conjunction with Wings for NONMEM Version 611 [5] under MS-DOS on an Intel Pentium 4 3GHz PC using Microsoft Windows XP and the g77 FORTRAN compiler or on an Intel Xeon X5355 2.66GHz Server using Microsoft Windows Server 2003 and the Compaq Visual Fortran compiler Version 6.6. All analysis was performed using the first order conditional estimation method with the interaction estimation option.

ivan.matthews@postgrad.manchester.ac.uk

Results

The reliability of the mixture model to correctly classify an individual as a poor or extensive metaboliser diminishes as:

1. variability of the structural parameters increases
2. fold difference in clearance between the two populations decreases
3. proportion of poor metabolisers decreases → increased percentage of false negatives AND proportion of poor metabolisers increases → increased percentage of false positives



Contour plots showing the percentage of false negatives top and percentage of false positives below. The areas in green represent areas on the parameter surface where a mixture model could be expected to be used reliably whereas those in red represent the area where a mixture model would be inappropriate.

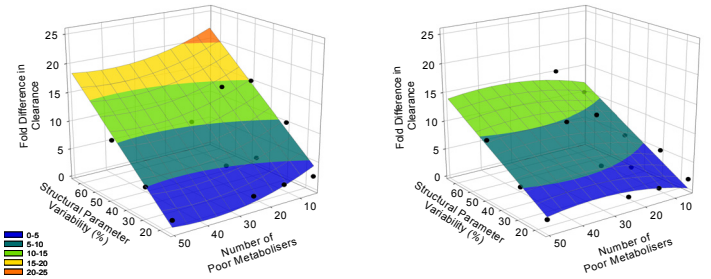
Decision Rule

An empirical equation (3) was developed to provide a decision rule for whether a mixture model may be useful for a given drug. Using warfarin as an example, it falls in a region on the parameter surfaces that would suggest that a mixture model would be inappropriate for classifying patients on warfarin therapy.

$$\text{Crit}_{FN} = -0.295\boldsymbol{Var} + 0.277\boldsymbol{PM} + 0.00355\boldsymbol{PM}^2 - 1.24 + \boldsymbol{FD} \quad (1)$$

$$\text{Crit}_{FP} = -0.191\boldsymbol{Var} - 0.210\boldsymbol{PM} + 0.00300\boldsymbol{PM}^2 + 3.45 + \boldsymbol{FD} \quad (2)$$

If Crit1<0 and/or Crit2<0 then Fail (3)



Plots showing the fit of the surfaces of the regression equations with the border values used to perform the regression. Left Panel: False negative surface. Right Panel: False positive surface. The region above the surface represents an area where the criterion value would be positive.

Discussion

The incidence of false negatives in the area with a small poor metaboliser proportion was extremely high, resulting in large numbers of the poor metabolisers being categorised incorrectly. This contrasts to a similar study by Kaila et al, 2007 [6], where they choose only to look at the percentage of patients classified correctly. Thus if there are only 5 poor metabolisers in a patient group and all are categorised incorrectly, and there are no further classification errors, then the percent classified correctly would still be rather high at 95%. The sample size is another potential variable that could be investigated.

Conclusions

Between subject variability on the structural parameters and the fold difference in clearance between the two subpopulations are the most important factors affecting correct subpopulation assignment in the NONMEM mixture model.

Acknowledgement

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References

[1] Matthews, I and Aarons, L., A population pharmacokinetic model for S-warfarin: application of a mixture model to determine genotype/phenotype. PAGE. Abstracts of the Annual Meeting of the Population Approach Group in Europe, 2006.

[2] D'Argenio DZ. Incorporating prior parameter uncertainty in the design of sampling schedules for pharmacokinetic parameter estimation experiments. Math Biosci. 1990 Apr; 99(1):105-18.

[3] Mathematica 5.2. Wolfram Research, Inc, Champaign, IL; 2005.

[4] NONMEM Users Guides, (1989-2006). Beal, S.L. and Sheiner, L.B. (Eds.) Icon Development Solutions, Ellicott City, MD.

[5] Holford N.H.G. Wings for NONMEM Version 611. http://wfn.sourceforge.net; 2006.

[6] Kaila, N., R.J. Straka, and Brundage, R.C., Mixture Models and Subpopulation Classification: A Pharmacokinetic Simulation Study and Application to Metoprolol CYP2D6 Phenotype. J Pharmacokinet Pharmacodyn, 2007 Apr; 34(2):141-56.