Study Title : To measure the effects of Aspirin loading dose of 300mgs in healthy controls using AA LTA and the Dynamic Platelet Function Assay (DPFA).

EudraCT number: 2015-000499-89

Results

The purpose of this study was to see if a loading dose of aspirin in normal donors could be detected in an assay termed the Dynamic Platelet Function Assay (DPFA).[1-3] This assay well described in the literature[4-8] measures the motional parameters of platelets translocating across von Willberand factor. In this study 29 donors were given a loading dose of 300mg of Aspirin and their platelet function analysed before and after taking aspirin. Of these donors 17 acted as controls with platelet function tested in the morning and afternoon in the absence of aspirin.

The aim of the analysis was to determine whether blood flow parameters obtained from the DPFA assay could distinguish pre and post-aspirin samples. Samples from control patients were used to estimate the effects of diurnal variation in blood-flow parameters and correct for these in analysis.

In addition based on some historical data and to aid with model fitting a dataset consisting of blood draws from n=164 normal subjects and n=67 stable ASA subjects was employed to identify parameters which could distinguish aspirin users from non-users.

Methods

We employed a range of linear and non-linear mixed-effects logistic regression models to attempt to identify markers in this assay on the effect of aspirin. Model fitting was attempted both using the data derived from the experiments in this study termed NASA alone, and using models fitted in the larger data set. Dimensional reduction was applied to the DPFA parameters using both PCA-based and Lasso methods. To avoid overfitting, models fitted on the larger dataset of normal and stable ASA patients were internally validated using 10-fold cross-validation before being tested on the NASA dataset. The final model chosen was that which gave the highest out-of-sample classification accuracy based on pseudo-r^2 and RMSE.

Results

- Analysis did not identify any combination of parameters which can reliably distinguish pre- and post- aspirin samples in the NASA dataset.
- Comparison of morning and afternoon samples in control subjects indicated that there is a large diurnal variation in the relevant parameters which may mask any short-term aspirin effect. Differences between morning and evening samples in all parameters were of equal magnitude in both control and ASA patients,
- Analysis of the larger stableASA/Normal dataset did identify a three-parameter model which could weakly distinguish ASA from normal patients. The three strongest predictors identified were
    - Log of the number of transiting cells (nTrans)
    - Log of the number of static cells (nStatic)
    - Surface coverage (SurfCovEnd)

  The classification accuracy of this model was weak (AUROC = 0.69) albeit highly significant ($p<0.0001$)

- However this model failed to classify pre-and post-aspirin samples in the NASA dataset

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- AUROC = 0.53, p=0.93

Conclusion

While our model is able to identify between-group due to long-term aspirin use, our results suggest that within-patients differences in DPFA parameters due to ASA are small relative to the magnitude of diurnal variation, making it difficult to accurately identify ASA effects. In addition the peak and trough times of diurnal variation are likely to differ between patients, making it impossible to correct for this effect in a single assay.

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