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In vitro **ADME & PK**

Cytochrome P450 Induction:

Relative Induction Score (RIS) Analysis

Background Information

'It is recommended to first evaluate the induction potential using the basic model. If the basic method indicates induction via PXR, the evaluation can continue using the mechanistic static model and/or the RIS correlation model provided it is possible to apply sufficiently high concentration of the investigational drug for E_{max} and EC_{50} to be determined.'

[3]EMA (2012) Guideline on the investigation of drug interactions

- Correlation methods are additional tools to aid the prediction of drug-drug interactions due to induction using *in vitro* induction data.
- Using the RIS method, batches of hepatocytes are qualified for subsequent induction studies.
- The qualification process assesses a set of known inducers, covering *in vivo* induction potency from non-inducers to strong CYP3A4 inducers.
- E_{max} and EC_{50} values are determined for all inducers and applied alongside predicted clinical unbound C_{max} to calculate RIS. RIS values for each inducer are subsequently correlated vs. observed *in vivo* change in the AUC of CYP3A4 victim.
- The relationship between *in vitro* inducing potency (RIS) and observed *in vivo* effect on CYP3A4 victim drug for the qualification data (RIS calibration curve), is then used to predict magnitude of *in vivo* induction of an investigational drug.
- If this is greater than a predefined cut-off of 20% decrease in AUC of CYP3A4 victim (AUCR ≤ 0.8), the investigational drug is considered positive for induction *in vivo* and follow-up is recommended either using mechanistic modelling or conducting a clinical DDI study.

Protocol

Pre-requisite for RIS Correlation Analysis
Induction *in vitro* data in matching conditions to RIS validation set (same donors, 72 hr dosing period)
CYP3A4 induction >2-fold and concentration-dependent in at least one donor[4]
Clinical parameters provided in order to determine [I] including MW, C_{max}, fu

CYP Isoform
CYP3A4

Negative Control
Flumazenil (non-inducer)

Positive Control
Rifampicin (clinical strong CYP3A4 inducer)
Phenobarbital (clinical moderate CYP3A4 inducer)

Data Delivery
Excel sheet containing RIS data analysis, predicted magnitude of clinical induction effect, plus charts plotting test compound on RIS calibration curves.

R_3 (R value for basic model of induction) will also be determined[2].

Written standalone summary report available on request.

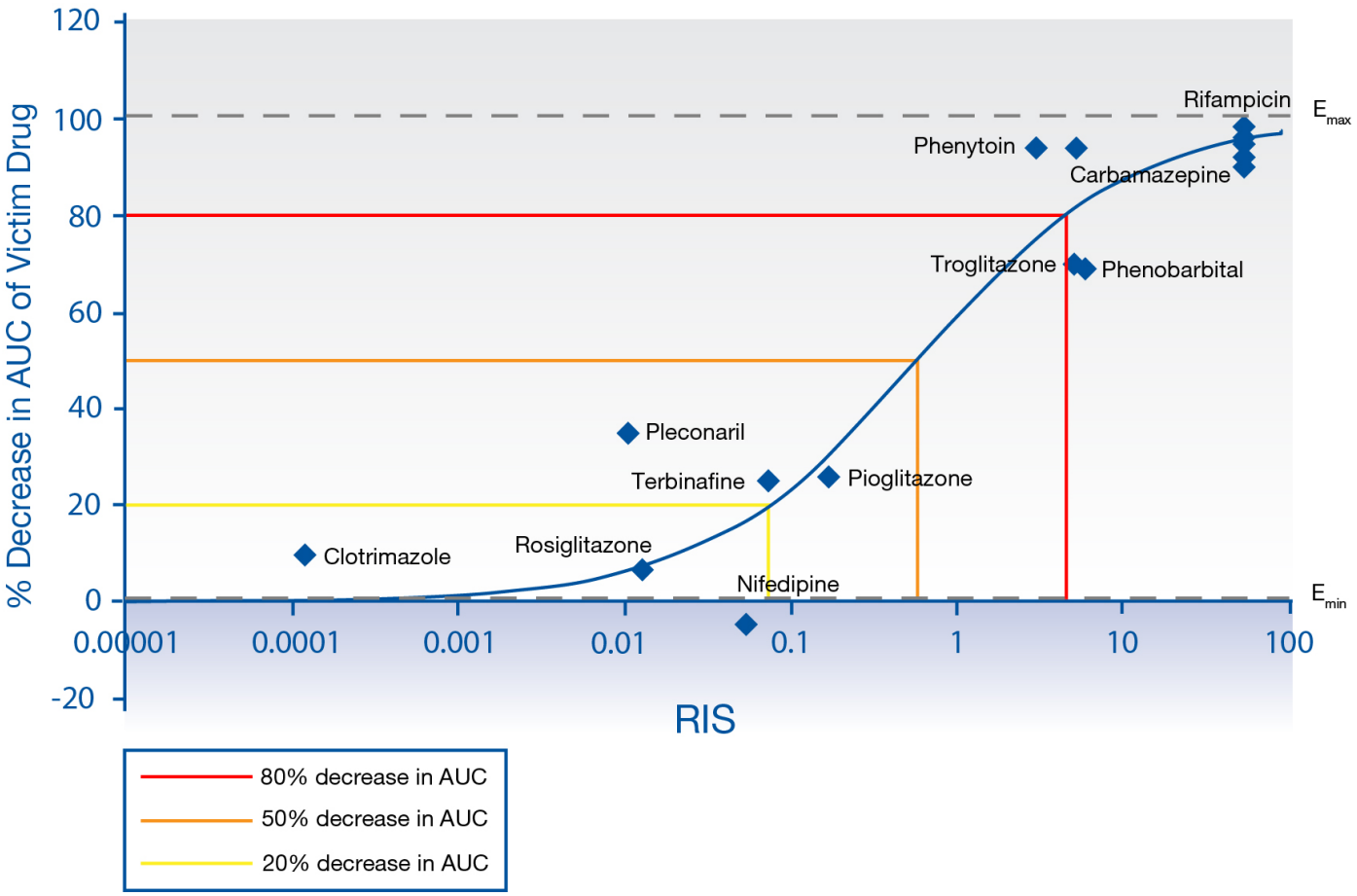
Evaluating the potential for DDIs due to CYP induction is an important part of the drug development process. The US, European and Japanese regulatory agencies have published guidance documents on how to conduct these studies and have recommended approaches by which to access the potential for DDIs utilising *in vitro* induction data and predicted human exposure.

Basic methods of assessing *in vitro* induction simply apply a cut-off to fold induction observed i.e. concentration-dependent increase of mRNA expression with fold change ≥2-fold relative to vehicle control or increase >20% of positive control, at expected hepatic concentrations of drug. This provides a conservative potential risk of clinical DDI due to induction.

The relative induction score (RIS) correlation method however, characterises a batch of hepatocytes by assessing a panel of clinical inducers, and on defining the *in vitro* induction parameters E_{max} and EC_{50}, correlating this with clinical inductive effect and *in vivo* exposure data to determine RIS score[1]. Utilising this calibration, the magnitude of clinical inductive effect can be predicted for test compounds enabling investigators to model and better understand the DDI risk for improved decision-making before the need to progress to clinical trials.

Figure 1

RIS calibration curve for HUM182351 mRNA, based on $I_{max,u}$



References

[1] Fahmi OA *et al.*, (2008) Prediction of drug-drug interactions from *in vitro* induction data: application of the relative induction score approach using cryopreserved human hepatocytes. *Drug Metab. Dispos* **36(9)**: 1971-1974

[2] US Food and Drug Administration (2020) Final guidance for industry: *in vitro* drug interaction studies – cytochrome P450 enzyme- and transporter-mediated drug interactions guidance for industry

[3] European Medicines Agency Guideline on the investigation of drug interactions EMA/CHMP/EWP/125211/2010

[4] Kenny JR *et al.*, (2018) Considerations from the innovation and quality induction working group in response to drug-drug interaction guidances from regulatory agencies: focus on CYP3A4 mRNA *in vitro* response thresholds, variability and clinical relevance. *Drug Metab. Dispos.*, **46**:1285-1303