In silico Identification of Precursors for CYP Profiling Breath Tests

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The family of cytochrome P450 (CYPs) plays a key role in the metabolism of xenobiotics. The high degree of polymorphisms and possible transcriptional regulation make it impossible to predict the individual patient response based on genomic information. [1] Hence, personalized medicine requires high-throughput measurement of the CYP phenotype. Breath tests are established for individual CYP isoforms and proven as diagnostic tools. [2] Breath tests detect volatile organic compounds (VOCs) in the patients' exhaled air, allowing for CYP tests by detection of $^{13}CO_2$ originating from CYP-catalyzed oxidative degradation reactions of an administered precursor. These small metabolites of CYP-catalyzed degradation of xenobiotics are generally considered unimportant for further drug metabolism and thus not listed in established CYP databases.

The presented work focuses on the establishment of an *in silico* workflow aiming at the identification of novel precursor molecules, likely to result in VOCs other than CO_2 upon oxidative degradation by CYPs. The proposed workflow comprises three parts: CYP profiling to identify most-likely CYP isoforms for given substrates, reaction encoding to predict metabolites from CYP-catalyzed reactions and subsequent volatility prediction.

CYP profiling was encoded as a ligand-based decision tree based on 2D descriptors derived from established models in the literature [3] and validated against publicly available data sets extracted from the DrugBank. [4] Oxidative degradation reactions (O- and N-dealkylations) were found to be most promising in the release of VOCs. Thus, the CYP-catalyzed oxidative degradation reaction was encoded as SMIRKS to enumerate all possible reaction products. A QSPR model aiming to predict the Henry constant k_H was derived from a data set of 488 small organic compounds. [5] This QSPR equation is finally applied to identify potentially volatile compounds amongst CYP reaction products.

A list of potential novel breath test precursors was identified based on the presented three stage workflow and subsequent comparison to metabolism data from literature. These candidate precursors are currently undergoing *in vitro* testing for their release of VOCs and hence applicability as precursors for CYP profiling breath tests.

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