Learning to predict the sites of metabolism and metabolic endpoints

by

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Abstract

When you ingest anything (e.g., food or medicine), your body will break down (metabolize) the compound's molecules; this process clearly affects the safety and the effectiveness of the compound. This breakdown is facilitated by certain proteins that catalyze this process. Thus it is important to predict whether a compound will be catalyzed by a particular protein, how it will be metabolized and what compounds will result from the process.

This thesis presents the framework and models for software systems dealing with three subtasks. The *substrate predictor* will learn to predict whether a given molecule will be catalyzed by a specific enzyme. Here we focus on the cytochrome P450 (CYP) proteins, which catalyze 90% of the drugs currently on the market. Each catalysis process involves at least one "site of metabolism" (SOM), which is the location of a single atom within the compound, where the reaction happens. We learned one *SOM predictor* for each of the 9 enzymes, that predicts which site(s) of the compound will be modified. This SOM predictor involves a novel "ranking and classification" framework, and works with simple-to-compute features. Finally, we present a simple way to generate the metabolic endpoints, given the enzyme and predicted SOMs. The empirical results on small datasets show our overall system, including *substrate predictor* and *SOM predictor*, performs quite well and is superior to state-of-art systems, in terms of computational efficiency and/or accuracy.

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Glossary

Atom environment: the atomic environment around a center molecular site within a molecule.

Atom: the smallest constituent unit of ordinary matter that has the properties of a chemical element [1].

Catalyst: the substance that facilitates the rate of a specific reaction [2].

Enzyme: the biological catalyst that catalyzes chemical reactions [3].

Inhibitor: a molecule that binds to an enzyme E and decreases E's activity [4].

Ligand: an ion or a functional group in a molecule that binds to a central metal atom [5].

MEG: Name of our metabolic endpoint generator.

Metabolism: the biochemical reactions of xenobiotics by particular enzymes [6].

Metabolites (Metabolic endpoints): the intermediates and products of metabolism [7].

Molecular site: a place within a molecule occupied by an atom.

Molecule: an electrically neutral group of two or more atoms held together by chemical bonds [8].

Notation for Carbon: any vertex in a figure that does not have an explicit atom type, is a carbon (see Figure 1.1).

Reaction type: the type of a reaction, e.g., oxidation, reduction [9].

Reaction: a process that leads to the transformation of one set of chemical substances to another [9].

Reactivity: the activation energy an atom needs during a reaction [10].

Reagent: a substance or compound that added to cause a chemical reaction [11].

Site of metabolism (SOM): the molecular site where the metabolic reaction happens.

SomPred: Name of our SOM predictor.

SubPred: Name of our substrate predictor.

Substrate-metabolite pair: a pair of substrate and its corresponding metabolite.

Substrate: the chemical species that reacts with reagent [12].

Xenobiotic: the foreign chemical substance found within an organism [13].

Chapter 1

Introduction

1.1 Motivation

When you eat food, have a drink, or ingest drugs, your body will transform these component chemicals into new compounds. The whole process is called *xenobiotic metabolism*, which involves some metabolic transformations (reactions), where the food, drink, or drugs are the xenobiotics, or substrates [6]. It also involves proteins, that work as enzymes to catalyze (facilitate) this process. The new compounds, resulting from the process, are called metabolites or metabolic endpoints. In Figure 1.1, we present a simple example to demonstrate this process, showing three possible metabolic endpoints of caffeine. Figure 1.2 provides more details about one of these three endpoints.

Metabolic transformations play an important role on the performance and safety of the drugs. It may reduce the bio-activity of a certain compound, which affects the therapeutic performance of drugs. It may also generate some by-products, even toxic metabolites. Generally speaking, metabolic transformations involve two phases: phase I transformation (addition, elimination, etc) and phase II transformation (also known as conjugation); see Figure 1.2. In this thesis, we focus on phase I metabolism.

Understanding xenobiotic metabolism is particularly important in the field of drug discovery. In the early stages of drug discovery, medicinal chemists want to know which parts of a compound are likely to be metabolized by particular enzyme. Knowing this, they can modify specific parts of the compound, thereby controlling its metabolism and improving its safety and effi-

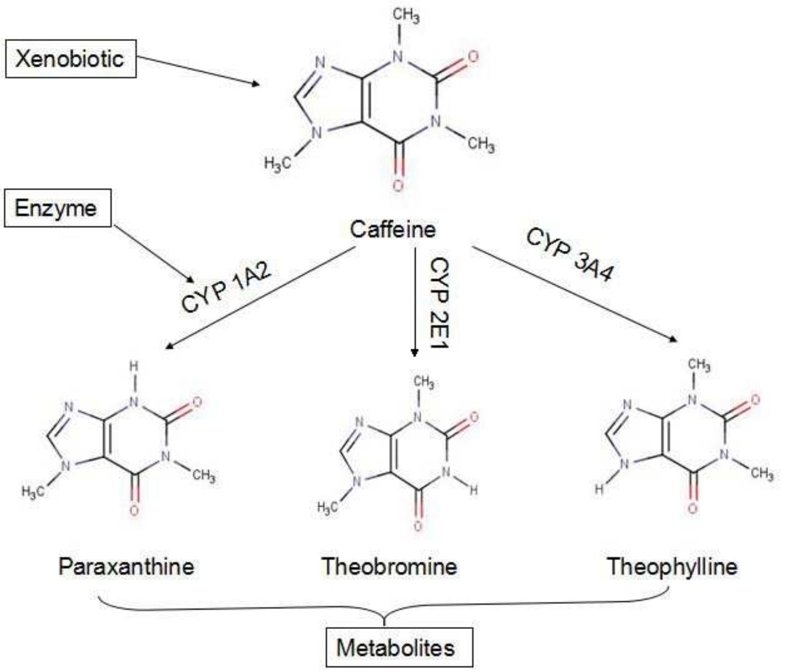


Figure 1.1: Caffeine, the input xenobiotic, breaks down into one of three metabolic endpoints during xenobiotic metabolism, each catalyzed by one of three specific enzymes. In this example, (Caffeine, Paraxanthine) is a substrate-metabolite pair. (Here and elsewhere: any vertex in a figure that does not have an explicit atom type, is a carbon atom.)

cacy [36]. But traditional experimental approaches are quite expensive and time-consuming, even for human experts with advanced scientific equipment. To avoid this, several computational predictive methods have emerged in recent years [43].

The computational approaches to predict metabolic processes typically focus on two subtasks: predicting the sites of metabolism (SOMs) of a substrate (for a given enzyme) and predicting the metabolic endpoints (again from substrate and protein) [43]. In particular, many programs attempt to predict the SOMs, especially phase I transformations mediated by cytochrome P450 enzymes, as this enzyme family are responsible for nearly 90% of current drug metabolism [36, 47]. Few projects have tried to identify the metabolic endpoints for a given substrate and enzyme [48], probably because that task is more complicated, as it requires first correctly predicting the SOMs and then identifying the metabolic transformations. This dissertation describes a sys-

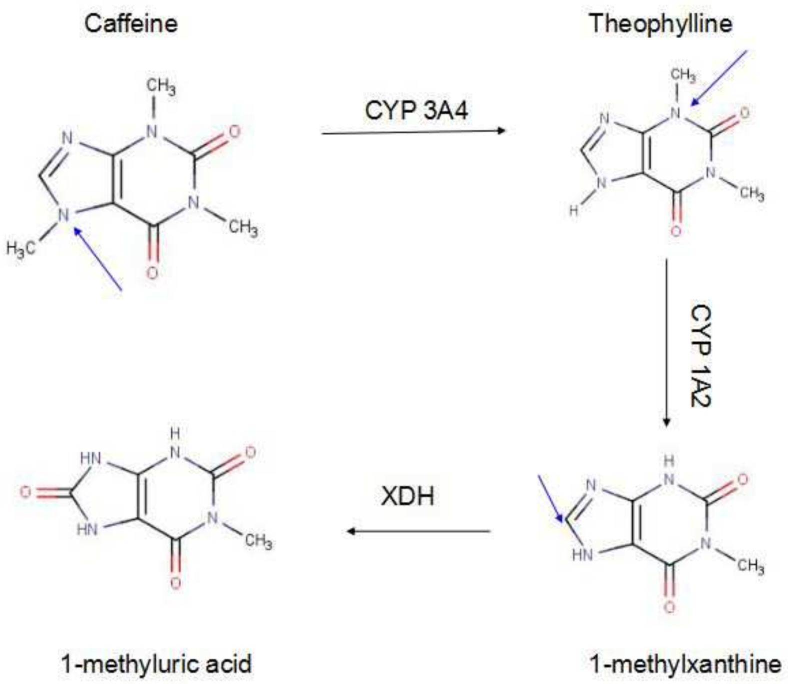


Figure 1.2: Caffeine transforms into Theophylline (using CYP 3A4) then to 1-methylxanthine (using CYP 1A2) through phase I elimination, finally into 1-methyluric acid, which is then excreted by the liver through urine. CYP 3A4, CYP 1A2 and XDH are the enzymes during this process. (The blue arrows point to the SOMs involved in each reaction.)

tem that first decides whether a molecule can be metabolized by a specific enzyme, and if so, predicts the SOMs and then the metabolic end products.

1.2 Related work

There are several computational prediction systems that try to identify the inhibitors of specific CYP450 enzymes. Zuegge et al. [61] designed a classifier based on linear models utilizing projection to latent structures (PLS) for predicting CYP450 3A4 inhibition. Each molecule was encoded with 333 descriptors, including atom type descriptors, topological descriptors, structural descriptors, etc. The model was trained on a training set containing 194 inhibitors and 117 non-inhibitors, and evaluated on a validation set consist of 29 inhibitors and 21 non-inhibitors. It achieved 95% accuracy on the training set and 90% on the validation data set. Molnr and Keser [46] built a classifier using neural network for predicting CYP450 3A4 inhibitors. They used 2D Unity fingerprints to represent each molecule in the dataset [41]. The model was trained on a training set consist of 109 inhibitors and non-inhibitors respectively and correctly predicted 91.7% of 36 inhibitors and 88.9% of 36 non-inhibitors in the test set. Later, the ensemble approach using recursive partitioning (tree) technique was used to predict CYP450 2D6 inhibitors [53]. Several hundred 2D structural descriptors were computed for the molecules, e.g. topological descriptor, electrotopological descriptor and physicochemical descriptor. The training set contained 59 inhibitors and 41 non-inhibitors, and the trained model correctly predicted all the 10 inhibitors and 76% of 41 non-inhibitors in the test set. Then Yap and Chen [57] used the support vector machine (SVM) method to predict inhibitors versus substrates for CYP450 3A4, 2D6 and 2C9. They used 1607 structural and chemical descriptors to represent the molecules. The training sets for CYP450 3A4, 2D6 and 2C9 were 312 substrates and 290 non-substrates, 169 substrates and 433 non-substrates, 130 substrates and 472 non-substrates, respectively. The trained models correctly predicted 98.2% of 56 substrates and 90.9% of 44 non-substrates, 96.6% of 29 substrates and 94.4% of 71 non-substrates, 85.7% of 14 substrates and 98.8% of 86 non-substrates, respectively.

During recent years, there are several public servers predicting the CYP450 inhibition. WhichCyp is used to predict which CYP450 isoforms (among 1A2, 2C9, 2C19, 2D6 and 3A4) a given molecule is likely to inhibit, using SVM models [49]. The average test accuracy among the five enzymes are around 85% on a dataset of 3000 molecules. Another tool is CypRules, which is a rule-based CYP inhibition prediction server for CYP450 1A2, 2C19, 2C9, 2D6 and 3A4 [51]. It achieves an average accuracy of around 79% on a dataset of over 16000 compounds.

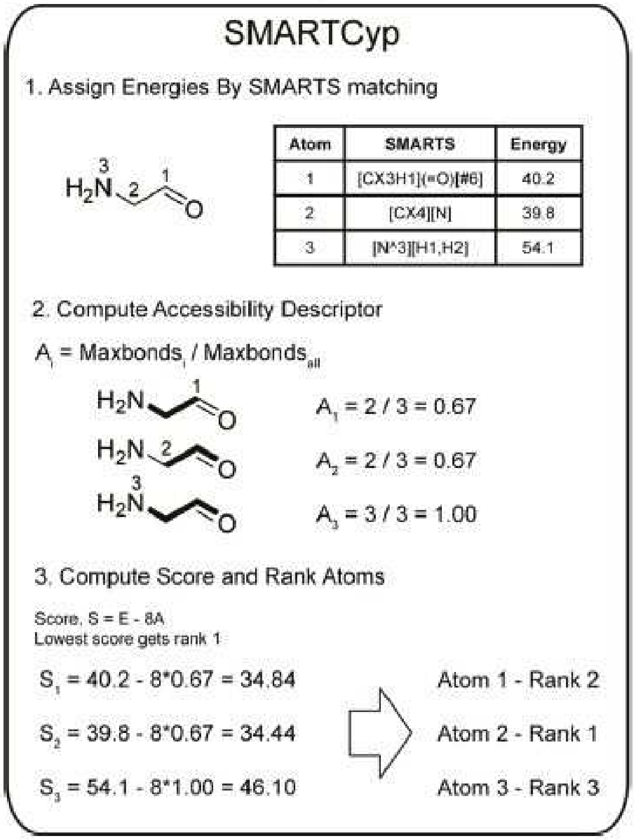
There are now several tools that try to predict SOMs for any query molecule. SMARTCyp is a reactivity-based SOM predictor, which predicts phase I metabolism for CYP450 enzymes [50]. It uses a reactivity descriptor to estimate activation energy that the specific CYP enzyme needs to react at a certain molecular site[1],

[1] They use the term "atom" instead.

which is calculated by certain quantum chemical methods, for various ligand fragments in its database. To make a prediction on a new query molecule, it calculates the reactivity for each site by SMART pattern matching, where it tries to match the ligand fragments (functional groups) contained in the query molecule with the ones in their library [14]. If no pattern matches for any of the sites in a ligand fragment, then no reaction will take place for this ligand. Otherwise, the sites whose activation energy are less than the corresponding one in the database, are predicted as SOMs. Finally, SMARTCyp uses an accessibility descriptor to produce a ranking over the predicted SOMs. On a dataset of 394 CYP 3A4 substrates, SMARTCyp's top two ranked positions include at least one true SOM, 76% of the time. Figure 1.3 gives an example for this process.

Metaprint2D is a tool for predicting SOMs based on circular fingerprints [22, 24, 25]. The model is built by mining similar atom environments of molecules in a large biotransformation database, Accelrys Metabolite Database [40], which contains more than 100,000 metabolic transformations. At training time, for each substrate-metabolite pair in the database, all the sites that are SOMs are encoded with a circular fingerprint [55, 56]. Then all the fingerprints are saved in the database. For example, consider the SOM for the (Caffeine, Theophylline) transformation in Figure 1.2 – i.e., the "N" where the blue arrow points. The associated four-level circular fingerprint (see Figure 1.4), that encoding the atom environment, as well as with the specific reaction (dealkylation), will be saved in the database. After training, to deal with a new query molecule, Metaprint2D calculates the atom environments for each molecular site in the molecule and searches the database for similar environments. In order to derive the likelihood for a specific site undergoing a metabolic reaction (i.e., for being a SOM), it calculates an occurrence ratio (OR), measuring the number of reactions (occurring at this site) that involve this or similar atom environment, divided by the total number of reactions containing this or similar atom environment in the database.[2] The calculated OR for each site in

[2] Assuming there are a total of 100 reactions in the database containing a particular atom environment that matches the one in the query molecule, 30 of which actually undergo a



SMARTCyp

1. Assign Energies By SMARTS matching

Atom	SMARTS	Energy
1	[CX3H1](=O)[#6]	40.2
2	[CX4][N]	39.8
3	[N^3][H1,H2]	54.1

2. Compute Accessibility Descriptor

A_i = Maxbonds_i / Maxbonds_{all}

$A_1 = 2 / 3 = 0.67$

$A_2 = 2 / 3 = 0.67$

$A_3 = 3 / 3 = 1.00$

3. Compute Score and Rank Atoms

Score, S = E - 8A
Lowest score gets rank 1

$S_1 = 40.2 - 8*0.67 = 34.84$

$S_2 = 39.8 - 8*0.67 = 34.44$

$S_3 = 54.1 - 8*1.00 = 46.10$

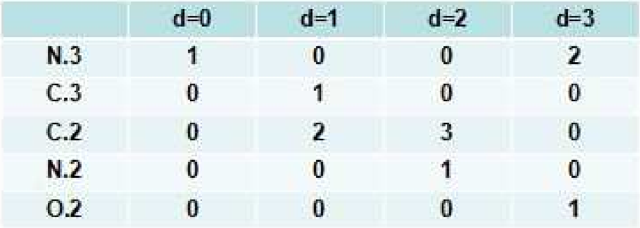
Atom 1 - Rank 2
Atom 2 - Rank 1
Atom 3 - Rank 3

Figure 1.3: In step 1, the patterns in the functional group are identified by SMARTS matching, then SMARTCyp calculates the activation energy for each atom in the functional group. In step 2, the accessibility descriptor is calculated for each atom. In step 3, the scores are calculated and a rank is derived according to the score. (Taken from [50].)

the query molecule is then normalized, so that the OR of the highest scoring site always has a score of one; these are called NORs. All the sites are then ranked according to their NORs, and the list of the ranked sites are returned. One of the shortcomings of MetaPrint2D is that it can not make predictions for novel atom environments – i.e., environments that are not in its database.

Moreover, Metaprint2D does not predict metabolic endpoints. Metaprint2D-React is an extension of MetaPrint2D that can make such predictions. It applies different types of reactions at each predicted SOM (see Figure 3.5) to generate corresponding metabolites. Unfortunately, we do not know how well

particular reaction occurring at that site, then the OR for that particular site being a SOM is $30/100 = 0.3$.



	d=0	d=1	d=2	d=3
N.3	1	0	0	2
C.3	0	1	0	0
C.2	0	2	3	0
N.2	0	0	1	0
O.2	0	0	0	1

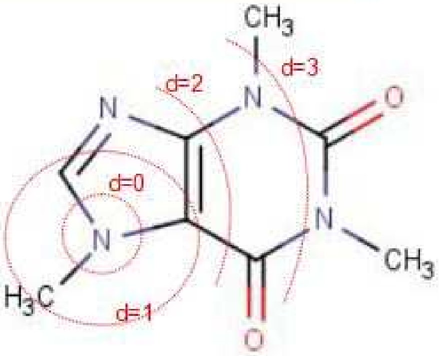


Figure 1.4: To construct the circular fingerprint, Metaprint2D counts the occurrences of SYBYL atoms around a certain central site up to six levels [15]. The circular fingerprint represents the atom environment. In this figure, d refers to the number of levels, and we just show four levels.

it works in practice, as there are no empirical results reporting its effectiveness, and its code is not available.

There are also some SOM predictors based on machine learning techniques. One example is the RegioSelectivity (RS)-predictor [58, 60], which uses a Support Vector Machine (SVM) model to predict SOMs, encoding each site with 148 topological descriptors, 392 quantum chemical descriptors and SMARTCyp reactivity descriptor. Later, Xenosite, which also adopts these descriptors with additional molecular and fingerprint descriptors, learns a neural network model to predict SOMs [59]. We compare its performance to our system in Chapter 4.

However, it is computationally expensive to calculate these quantum chemical features for the predictors (up to several hundred CPU hours for small molecules [44]). This motivated FAst MEtabolizer (FAME) [42], a metabolism prediction tool that applies random forest models, trained on the Accelrys Metabolite Database. FAME calculates six atomic descriptors and one molecular descriptor using the Chemistry Development Kit (CDK) [52], and makes SOM predictions in around three seconds per molecule, using these simple features. FAME also has some other benefits that others do not: it is not just a predictor for a specific enzyme family, but covers the broader enzyme reactions recorded in its database; it is not limited to human metabolism, but also has various models for rat and dog metabolism; it supports both phase I and phase II metabolism. On a dataset of 680 CYP450 substrates, FAME's top two ranked positions include at least one true SOM, 73% of the time[3].

Identifying SOMs for compounds is important for the drug discovery and designing processes. Medicinal chemists can optimize the properties of drugs based on the metabolic predictions. There are a number of limitations to many of today's SOM predictors:

1. Some predictors involve quantum chemical calculations, which is computationally expensive, leading to long processing time (up to several hundred CPU hours for small molecules) when making prediction for new molecules, which means it is not convenient for interactive use.

2. Most of these projects focus on detailed description for the calculation of the features, but they just briefly mention the process for learning their model, which makes it hard to understand/extend their learning algorithms.

3. Some predictors are not easily accessible due to license issues – i.e., the predicted results are not downloadable.

1.3 Contributions

This dissertation describes the new BioTransformer System for predicting xenobiotic metabolism, involving a pipeline (see Figure 3.1) to predict whether a compound will be catalyzed, and if so, how it will be transformed and what compounds will be produced. This requires addressing three different metabolism tasks:

[3] The definition of SOM in FAME differs a bit from our definition, for more details, see [42].

1. Given a particular enzyme and a query molecule, predict whether the molecule interacts with the enzyme.
2. Assuming the molecule interacts with the enzyme, predict which parts of the molecule will be metabolized by the enzyme (predicted SOMs).
3. Given the predicted SOMs and the enzyme, generate possible metabolic endpoints.

Our contributions include:

- Task 1: We learn to produce a Substrate Predictor, SubPred, that learns a model that can predict whether a given molecule will be catalyzed by a particular enzyme. The empirical results demonstrate excellent performance, when trained on a relatively small dataset.
- Task 2: We formulate SOM prediction a ranking and classification problem. We present a novel perspective where we learn the preferences between a pair of sites, each described using very simple-to-compute features. We also build the regression model that provides the probability that each site is a SOM. Both empirically and theoretically, we show that our preference learning approach, embodied in our SOM prediction system, SomPred, is superior in terms of computational efficiency, and at least competitive in terms of accuracy.
- Task 3: We built a Metabolic Endpoints Generator, MEG, that uses the results of the previous two predictors to predict metabolic endpoints.

1.4 Outline

Chapter 2 describes the chemical foundations. Section 2.1 introduces how we will represent molecules. Section 2.2 introduces how we will represent reactions. Section 2.3 describes how we will generate features for building SubPred and SomPred.

Chapter 3 describes the BioTransformer System for metabolism prediction. Section 3.1 describes the model for building SubPred. Section 3.2 presents a detailed introduction of preference learning and illustrates how it is applied to the SOM prediction problem. Section 3.3 introduces the procedures for generating metabolic endpoints.

Chapter 4 presents the empirical results on public datasets. Section 4.1 gives the detailed description for the datasets. Section 4.2 describes the evaluation criterion. Section 4.3 presents the experimental results of different models. Section 4.4 gives a short discussion on the experimental results.

Chapter 5 describes the future work and contributions.

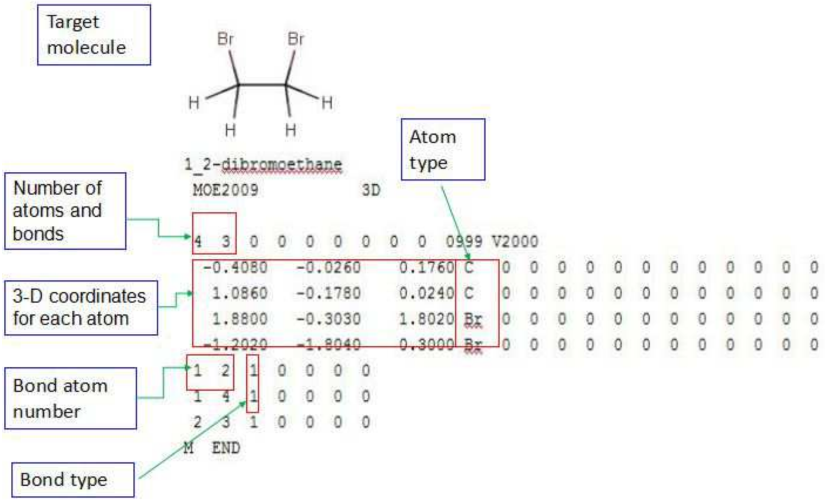
In Appendix A, we introduce an extension of the pairwise learning framework.

Chapter 2

Chemical foundations

2.1 Representations for molecules

We adopt the Daylight specifications for representing molecules and reactions [16], which represents a molecule as a graph, also known as a connection table, where the nodes are atoms and the edges are bonds. Each atom has several properties, like its atomic number, weight, etc. The properties of a bond are even simpler: single, double, triple, or aromatic. Figure 2.1 shows a description of a simple molecule.



```
1_2-dibromoethane
 MOE2009            3D

  4  3  0  0  0  0  0  0  0  0999 V2000
   -0.4080   -0.0260    0.1760 C   0  0  0  0  0  0  0  0  0  0  0  0
    1.0860   -0.1780    0.0240 C   0  0  0  0  0  0  0  0  0  0  0  0
    1.8800   -0.3030    1.8020 Br  0  0  0  0  0  0  0  0  0  0  0  0
   -1.2020   -1.8040    0.3000 Br  0  0  0  0  0  0  0  0  0  0  0  0
  1  2  1  0  0  0  0
  1  4  1  0  0  0  0
  2  3  1  0  0  0  0
M  END
```

Figure 2.1: Daylight description of a target molecule

Simplified Molecular Input Line Entry System (SMILES) is another way to represent molecules [54]. Like some ordinary languages (English, Chinese, etc), it has a very simple vocabulary (atom and bond symbols) and a few grammar rules to store the chemical information of molecules. One of the advantages of SMILES is that there is a unique representation for each molecule, which means that people can use this format to determine if two molecules are isomorphic. Figure 2.2 gives a few examples of how molecules are encoded as SMILES.

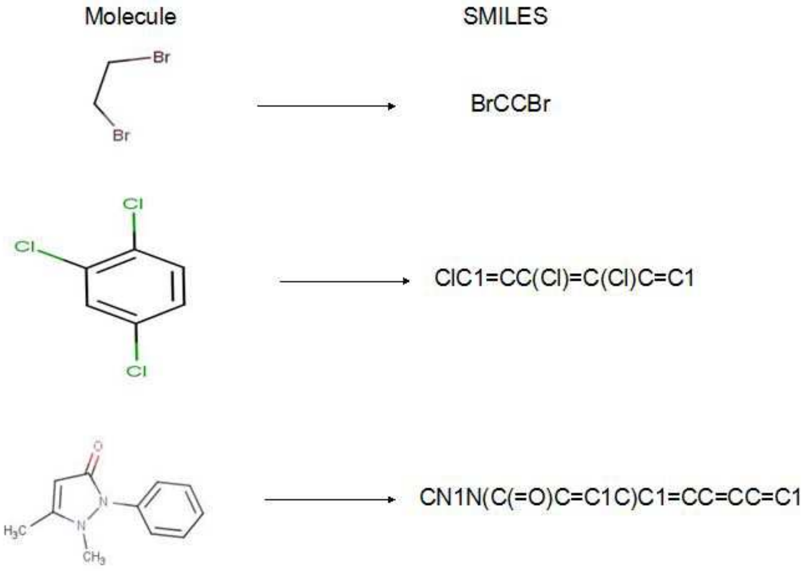
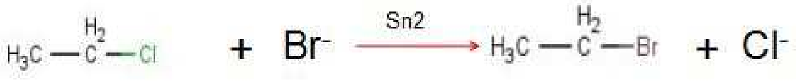
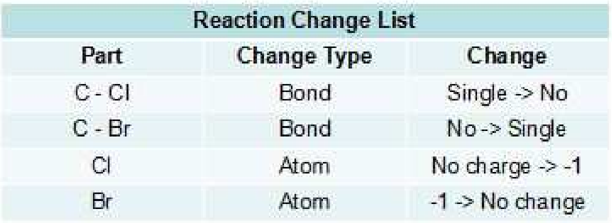


Figure 2.2: Encoding molecules with SMILES

2.2 Representations for reactions

A reaction usually involves a substrate and an enzyme to produce possible metabolites (e.g., in Figure 1.1, Caffeine goes through the dealkylation and transforms to one of its metabolites catalyzed by an enzyme.). To represent a reaction, first we should identify the patterns contained in a substrate, which is called substructure searching. SMiles ARbitrary Target Specification (SMARTS) is a language for specifying substructure patterns in molecules, using extended rules of SMILES [14]. Using SMARTS, flexible and efficient substructure searching queries can be easily made. For example, people can use the SMARTS string **[OH]c1ccccc1** to search for phenol-containing structures in a large database.





Reaction Change List		
Part	Change Type	Change
C - Cl	Bond	Single -> No
C - Br	Bond	No -> Single
Cl	Atom	No charge -> -1
Br	Atom	-1 -> No change

Figure 2.3: During a Sn2 reaction of chloroethane with bromide ion, the atom Cl changes from charge 0 to charge -1, and its associated bond changes from single to no bond. The Br ion goes through the reverse changes. In this formalism, any reaction that undergoes the same set of atom and bond changes is regarded as the same generic reaction [17].

In this thesis, we will represent the reaction as the list of atom/bond changes (see Figure 2.3), using SMIles Reaction Keying System (SMIRKS) [17], which is a reaction transform language used to represent generic reactions. This representation is common and easy, as any reaction that undergoes the same atom and bond changes, can be regarded an example of the given generic reaction, regardless of the substrate.

2.3 Feature generation

One of the main disadvantages for previous SOM predictors is that they require calculating complex features for potential SOMs of the molecule. Instead, we calculate some simple features with CDK for each site in the molecule.

Our SubPred first computes, then uses a number of molecular descriptors, that are shown in Table 2.1.

For building the SOM Predictor, in addition to the molecular descriptors, the following atomic descriptors are calculated with CDK for each site in the molecule:

- Atomic features (10 features): describing the chemical properties of each atom at the site, including SYBYL Atom Type [15], Degree, Hybridiza-

Name of Descriptors	**Definition**	**Type**
ALOGP	Atom additive logP and molar refractivity values	Real value
APol	The sum of the atomic polarizabilities	Real value
HBondAcceptorCount	The number of hydrogen bond acceptors	Integer
HBondDonorCount	The number of hydrogen bond donors	Integer
MomentOfInertia	The principal moments of inertia and its ratios	Real value
RotatableBondsCount	The number of non-rotatable bonds	Real value
TPSA	Topological polar surface area	Real value
Weight	Molecular weight	Real value
XLogP	Prediction of logP	Real value
ASA	Accessible surface area	Real value

Table 2.1: Molecular descriptors calculated with CDK

tion, Valence and so on (see Table 2.2).

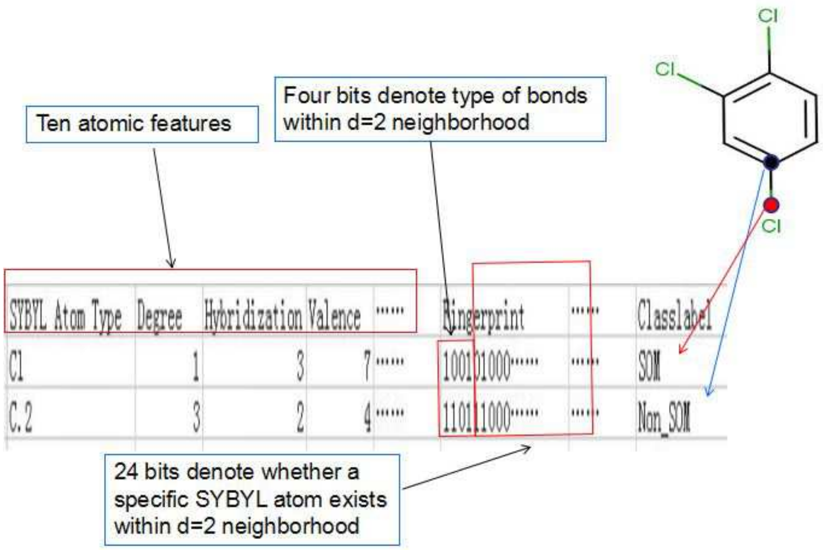
- Environmental features (56 features): describing each atom that is within depth d=2 of the target site (see Figure 1.4), using a bit to denote whether this set of atoms includes each specific SYBYL atom (like C.1, C.2, N.1). Four extra bits denote whether this d=2 neighborhood includes a single, double, triple or aromatic bond.

- Atom fingerprint (132 features): describing functional group features, using a bit to denote whether the substructure that includes the target site, belongs to each of the 132 specific groups (e.g., hydroxyl group, or carboxyl group, etc).

Figure 2.4 shows an example of a sample molecule in one of the datasets.

Descriptor name	**Type**
AtomDegreeDescriptor	Integer
AtomHybridizationDescriptor	Integer
AtomValenceDescriptor	Integer
EffectiveAtomPolarizabilityDescriptor	Real value
PartialSigmaChargeDescriptor	Real value
PartialTChargeMMFF94Descriptor	Real value
PiElectronegativityDescriptor	Real value
SigmaElectronegativityDescriptor	Real value
StabilizationPlusChargeDescriptor	Real value
SYBYLAtomTypeDescriptor	Vector of 24 bits

Table 2.2: Ten atomic descriptors calculated with CDK


Ten atomic features
Four bits denote type of bonds within d=2 neighborhood
Cl
Cl
Cl
24 bits denote whether a specific SYBYL atom exists within d=2 neighborhood

SYBYL Atom Type	Degree	Hybridization	Valence	······	Fingerprint	······	Classlabel
Cl	1	3	7	······	100101000······	······	SOM
C.2	3	2	4	······	11011000······	······	Non_SOM

Figure 2.4: Encoding for one molecule.

Chapter 3

BioTransformer model

In this thesis, we describe hwo some of the tools were constructed in the BioTransformer System for metabolism prediction, including the Substrate Predictor, SubPred; the SOM Predictor, SomPred; and the Metabolic Endpoint Generator, MEG. The whole BioTransformer pipeline is shown in Figure 3.1.

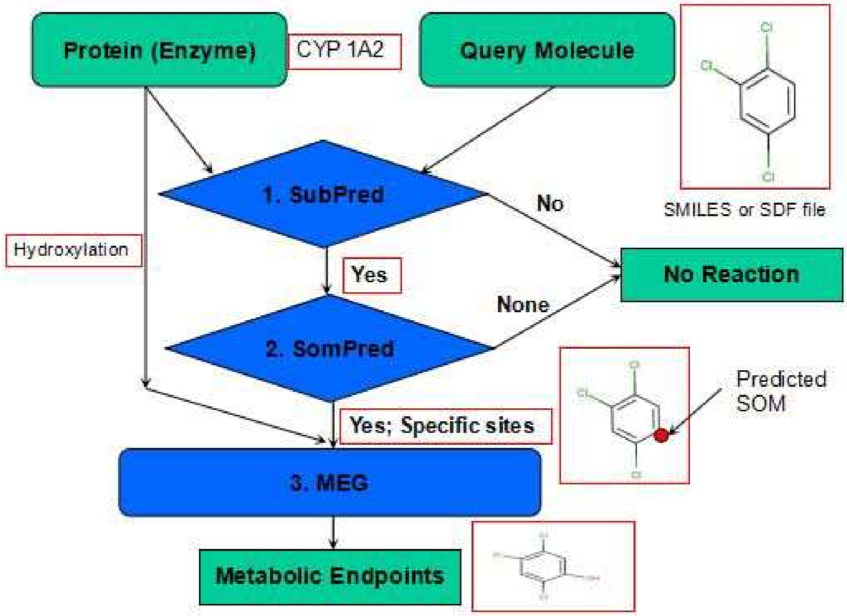


Figure 3.1: Pipeline for the BioTransformer System

3.1 SubPred

Our SubPred program takes a molecule M and an enzyme E as input, and determines whether M is a substrate of the enzyme E (i.e., whether M can be metabolized by E). We formulate substrate prediction, for a fixed enzyme E, as a standard learning problem, where a set of molecules is used as the training data, along with a label determining whether that molecule reacts with E or not. We used a support vector machine (SVM) method for supervised data classification [23, 32]. The training and testing processes are done with LIBSVM [27]. The details of the dataset and the performance are reported in Chapter 4.

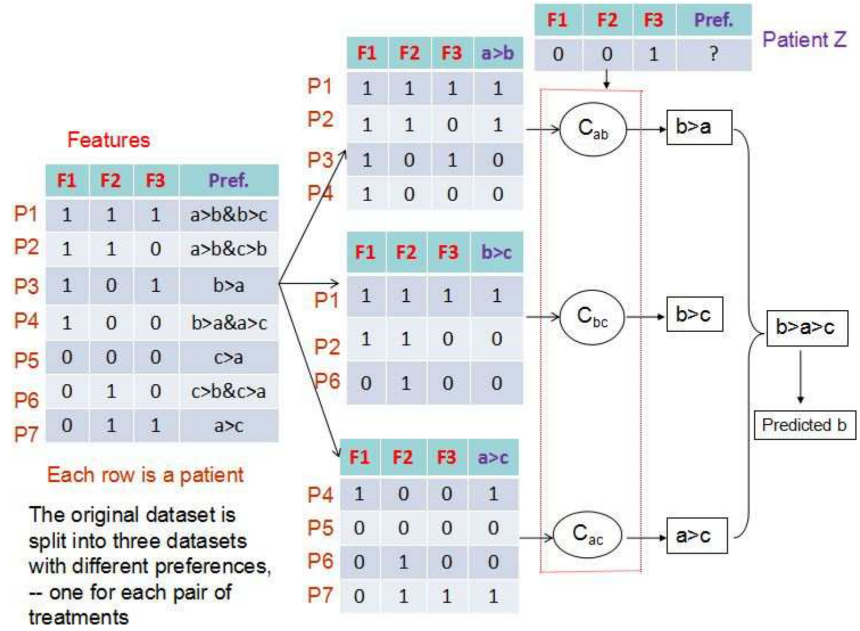
Given a set of training instance-label pairs $D = \{(x_i, y_i) | x_i \in R^m, y_i \in \{-1, 1\}\}_{i=1}^{n}$, SVM tries to find the solution for the following optimization problem:

$$\begin{aligned} \min_{\mathbf{w},b,\boldsymbol{\xi}} \quad & \frac{1}{2}\mathbf{w}^T\mathbf{w} + C\sum_{i=1}^{n}\xi_i \\ \text{subject to} \quad & y_i(\mathbf{w}^T\boldsymbol{x_i}) + b \geq 1 - \xi_i, for\ all\ 1 \leq i \leq n \\ & \xi_i \geq 0. \end{aligned} \tag{3.1}$$

where $\mathbf{w}$ is the weight vector we want to learn. SVM tries to find a linear separating hyperplane with the maximal margin. C is a regularization parameter for the error. We use the linear kernel and select the regularization parameter C from $\{10^{-5}, 10^{-4}, \ldots, 10^{2}\}$ by five fold cross validation – i.e., at each C, four folds are used as the training set while the remaining one as the validation set. We then identify the setting with the minimum average error. Once the optimal parameter C is selected, we build the best learner with optimal parameters on training set and run on test set, reporting its performance.

3.2 SomPred: Preference learning

Many previous works [22, 24, 25, 42, 50, 58, 60] regard SOM prediction as a standard classification problem, where a set of single sites is used as the training data, and the goal is building a model that can predict whether a new site (in a new molecule) is a SOM or not. For reasons discussed below, we instead formulate our prediction task as a "pairwise ranking and classification" problem, where during the training time, we learn a model that tries to optimize the ranking, so that all true SOMs are ranked in the top positions. Once the model is learned, for a new query molecule, we will consider all of its sites, and predict a site is a SOM if it has sufficiently high ranking.



	F1	F2	F3	Pref.
P1	1	1	1	a>b&b>c
P2	1	1	0	a>b&c>b
P3	1	0	1	b>a
P4	1	0	0	b>a&a>c
P5	0	0	0	c>a
P6	0	1	0	c>b&c>a
P7	0	1	1	a>c

The original dataset is split into three datasets with different preferences, -- one for each pair of treatments

	F1	F2	F3	a>b
P1	1	1	1	1
P2	1	1	0	1
P3	1	0	1	0
P4	1	0	0	0

	F1	F2	F3	b>c
P1	1	1	1	1
P2	1	1	0	0
P6	0	1	0	0

	F1	F2	F3	a>c
P4	1	0	0	1
P5	0	0	0	0
P6	0	1	0	0
P7	0	1	1	1

F1	F2	F3	Pref.
0	0	1	?

Figure 3.2: An example for learning by pairwise comparison. (Taken from [38])

The notion of preference, which is mainly used in e-commerce tasks before, has gained a great deal of attention in artificial intelligence recently [33]. It is now widely used in the fields of machine learning, information retrieval and recommender systems [35, 38, 45]. Typically, there are two types of preference learning (PL) problems [38]: one is learning from object preferences [30], the other is learning from label preferences [38]. Motivated by the latter, we will extend its learning framework in our scenario. We first introduce a general label preference learning scenario below, then we describe how it is extended and applied on our SOM prediction problem.

3.2.1 Label preference learning

Label preference learning (LPL), also called label ranking, attempts to learn a mapping from instances to rankings over a finite number of labels. It first induces a set of preference functions from the training data. Then it derives a ranking from the set of preference relation.

For example, in Figure 3.2, we are trying to learn a model to predict the preferences between all the candidate labels (treatments) for all the training instances (patients), based on our training set. The training dataset has seven instances, each of which is a patient who has three features ($F1, F2, F3$). There are also three different treatments (labels: a, b, c) to choose for each patient. For each patient, we know some of its preferences between treatments. For example, patient P1 prefers a to b and prefers b to c; we will write $a >_{P1} b$ to mean P1 prefers treatment a to treatment b. (Or just $a > b$ if the context is obvious. Note that transitivity does not hold in general.) For each pair of treatments – e.g., {a, b} – we collect all of the explicitly given relationships – either $a > b$ or $a < b$ – into a data subset; see the top middle dataset in Fig 3.2. We then use this labeled dataset to learn the base classifier $C_{ab}(\cdot)$; we can similarly assemble two other data subsets, to learn two other base classifiers: $C_{bc}(\cdot)$ and $C_{ac}(\cdot)$. To deal with a new patient Z, we first compute $C_{i,j}(Z)$ using each base classifier – i.e., over all (i,j) (where $i < j$)pairs. Then we try to derive a ranking (priority order) using preference aggregation for all the labels (treatments), given the set of $\{C_{i,j}(Z)\}_{i,j}$ values. Finally, the ranked 1st label (best treatment) is returned as the appropriate treatment for this target instance Z.

The detailed learning scenario is described below.

Given:

- a set of training instances $X = \{x_k |\ k = 1 \ldots n\}$
  - e.g., the patients {P1, . . . , P7} in Figure 3.2
- a set of labels $L = \{\lambda_i |\ i = 1 \ldots m\}$
  - e.g., the treatments {a, b, c} in Figure 3.2
- for each training instance x_k: a set of pairwise preferences in the form of $\lambda_i \succ_{x_k} \lambda_j$, where every $\lambda_i \succ_{x_k} \lambda_j$ means that, for instance x_k, label λ_i is preferred than λ_j
  - e.g., the 4th column in Figure 3.2

Learn:

- a set of base classifiers $C_{i,j}(\cdot)$ for each pair of labels $\lambda_i, \lambda_j \in L$ and $i < j$
  - e.g., the classifiers $C_{ab}(\cdot)$, $C_{ac}(\cdot)$, $C_{bc}(\cdot)$

After this learning process, to deal with a novel instance (e.g., patient Z above),

Predict:

- a $C_{i,j}(x)$ for each new instance x, over L, where $\lambda_i, \lambda_j \in L$ and $i < j$
  - e.g., for patient Z, $C_{a,b}(Z) = 0$, which means $b > a$. Similarly for $C_{a,c}(Z) = 1$ and $C_{b,c}(Z) = 1$

Derive by preference aggregation:

- a ranking $\tau_x : L \to \{1 \ldots m\}$, from the set of $\{C_{i,j}(x)\}$ for each new instance x.
  - e.g., for patient Z, $\tau_Z(b) = 1$, $\tau_Z(a) = 2$, $\tau_Z(c) = 3$
- a preferred label (top 1 label) from the ranking τ_x for instance x
  - e.g., as $\tau_Z(b) = 1$, we say that b is the preferred treatment for patient Z

For solving this label preference learning problem, the key part is how to represent the pairwise preferences $\lambda_i \succ_{x_k} \lambda_j$. Below, we motivate an approach, called ranking by pairwise comparison (RPC) [38].

The key idea of RPC is to represent the preferences as pairwise relations $C_{i,j}(x)$, which turns a multi-class classification (e.g., predict the best treatment for a patient among multiple alternative treatments) into a number of binary classifications (e.g., predict the preferred treatment for a patient between each pair of treatments). For each pair of labels (λ_i, λ_j), we learn a binary base classifier $C_{i,j}(\cdot)$, where for any x, $C_{i,j}(x) = 1$ means $\lambda_i \succ_x \lambda_j$ for instance x. To deal with a new instance, we first compute the predictions for all pairwise label preferences $\{C_{i,j}(x)\}$, and then derive a ranking by some ranking procedures.

In the example of Figure 3.2, we separate all the patients into three different datasets, where each dataset only contains the patients whose preferences between two treatments are known. We use a binary preference relation for each pair, and train three classifiers, which here leads to the following simple rules for each classifier.

- For $C_{ab}(\cdot)$, if $F2 = 1$, then $a > b$.
- For $C_{bc}(\cdot)$, if $F3 = 1$, then $b > c$.
- For $C_{ac}(\cdot)$, if $F1 = 1$ or $F3 = 1$, then $a > c$.

When a new patient Z ($F1 = 0$, $F2 = 0$, $F3 = 1$) comes, we use these classifiers to make the three predictions independently, and then derive a ranking combining all the predictions. Finally, as we find $a > b$, $b > c$, $a > c$, we predict treatment b for this patient Z.

In the above example, we use a binary mapping, and the model is trained with all the examples x, where either $\lambda_i \succ_x \lambda_j$ or $\lambda_j \succ_x \lambda_i$ is known. If there is no preference between λ_i and λ_j for some training instance x, then this is not included.

In addition, if we use a valued mapping, and $C_{ij}(x)$ is in the unit interval [0, 1], it can be interpreted as the probability of the preference $\lambda_i \succ_x \lambda_j$.

During the process of building the model on training set, we assume that the training data only provide partial order information about the ranking for training instances. For example, for patient P3 in our dataset, we only know he/she prefers b to a, there is no information for his/her preferences on treatment c. Also there may be some conflicts between the pairwise preferences due to some errors in training data. In general, transitivity is not held for the set of $C_{i,j}(x)$. That is to say, we might have $\lambda_i \succ_x \lambda_j$, $\lambda_j \succ_x \lambda_k$, and $\lambda_k \succ_x \lambda_i$. For more details about the non-transitivity, see [38].

3.2.2 Preference aggregation

Preference aggregation combines the set of $\{C_{i,j}(x)\}_{i,j}$ into a consensus ranking τ_x for instance x, we consider two methods for deriving the ranking [39].

- Voting: A scoring function $S : L \rightarrow \Re$ is used to evaluate every label λ_i, defined as:

  $$S(\lambda_i) = \sum_{\lambda_j \neq \lambda_i} C_{i,j}(x) \tag{3.2}$$

  Then sort the scores from maximum to minimum, and derive the ranking, – e.g., $\tau_x(\arg\max_i S(\lambda_i)) = 1$, and so forth.

- Choice: A ranking is derived with an iterative process: the top one label is chosen from the whole candidate set with the choice function; then the top second label is chosen from the remaining sets, and this process continues repeatedly, until reaching a total ranking.

  The choice function is defined by maximizing the probability of a particular label is selected among all the candidates. We define the following expression – all for a fixed instance x:

  1. $Pr(i)$: the probability that label λ_i is most preferred among the candidates.
  2. $Pr(i, j)$: the probability that either label λ_i or label λ_j is most preferred among the candidates.
  3. $Pr(i|(i, j))$: the probability that label λ_i is most preferred, given either label λ_i or label λ_j is most preferred among the candidates. Notice this corresponds to $C_{i,j}(x) = Pr(i|(i, j))$.

  Note $Pr(i) = Pr(i, (i, j)) = Pr(i, j)Pr(i|(i, j))$.

  Given the label set $L = \{\lambda_1 \ldots \lambda_m\}$, we have :

  $$(m-1)Pr(i) = \sum_{j=1, j\neq i}^{m} Pr(i) = \sum_{j=1, j\neq i}^{m} Pr(i, j)Pr(i|(i, j)) \tag{3.3}$$

  As $Pr(i, j) = Pr(i)+ Pr(j)$, and letting $C_{i,j}(x) = Pr(i|(i, j))$, we have:

  $$(m-1)Pr(i) = \sum_{j=1, j\neq i}^{m} C_{i,j}(x)(Pr(i) + Pr(j)) \tag{3.4}$$

  Note that Equation 3.4 corresponds to m linear equalities, over the m variables $Pr(i)$. Note we also have the m+1'st constraint:$\sum_{i=1}^{m} Pr(i) =$

1. Once we solve for $\{Pr(i)\}_i$, we can get a solution, then pick the label $i^* = \arg\max_i Pr(i)$ as the index with the maximum $Pr(\cdot)$ value. On the r-th iteration of the algorithm, this index becomes the r-th element of the ordering τ_x: $\tau_x(i^*) = r$. We then remove this i^*, and repeat this process to find the $r+1$'st most likely label, and so forth, until we derive a ranking over all of labels.

We implement the classification model with PL using voting ($C_{i.j}(x) \in \{0, 1\}$) and regression model with PL using choice ($C_{i.j}(x) \in [0, 1]$). For more details, see Chapter 4.

3.2.3 Applying label preference learning to SOM prediction

There are two main differences between label ranking and SOM prediction problems, which makes the latter more complicated than the former:

1. The set of labels for each instance in label ranking problem is fixed, while the size of the set of sites for each molecule in the SOM prediction problem is varied.

2. The former predicts the top 1 label as the true label while the latter may predict several sites as SOMs.

The extended learning framework is shown in Figure 3.3.

The detailed scenario of applying preference learning to SOM prediction is described below.

Given:

- a set of molecules $M = \{m_k |\ k = 1 \ldots n\}$

- a set of sites $A_k = \{a_k^i |\ i = 1 \ldots l_k\}$, where l_k is the number of sites contained in each molecule m_k

- for each molecule m_k: a set of pairwise preferences in the form of $a_k^i \succ_{m_k} a_k^j$, where every $a_k^i \succ_{m_k} a_k^j$ indicates that, for molecule m_k, site a_k^i is more preferred than a_k^j, which means a_k^i is a SOM and a_k^j is a Non-SOM

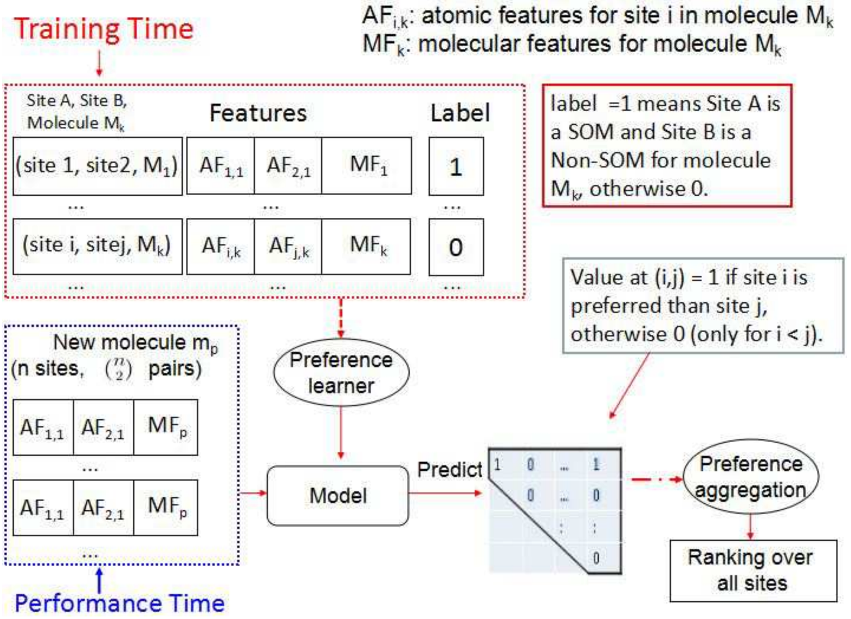

Training Time
$AF_{i,k}$: atomic features for site i in molecule M_k
MF_k: molecular features for molecule M_k
Site A, Site B, Molecule M_k
Features
Label
(site 1, site2, M_1)
$AF_{1,1}$
$AF_{2,1}$
MF_1
1
(site i, sitej, M_k)
$AF_{i,k}$
$AF_{j,k}$
MF_k
0
label =1 means Site A is a SOM and Site B is a Non-SOM for molecule M_k, otherwise 0.
Value at (i,j) = 1 if site i is preferred than site j, otherwise 0 (only for i < j).
New molecule m_p
(n sites, $\binom{n}{2}$ pairs)
$AF_{1,1}$
$AF_{2,1}$
MF_p
Preference learner
Model
Predict
Preference aggregation
Ranking over all sites
Performance Time

Figure 3.3: At training time, all the pairwise sites in the molecules of the training set, are used to build a classifier to predict preference. At performance time, the preferences between every pair of sites in a new molecule are predicted by the classifier. Then the preferences are combined to produce a ranking over all the sites by preference aggregation.

Learn:

- a classifier $C_{a_k^i,a_k^j}(\cdot)$ that predict $C_{a_k^i,a_k^j}(m_k)$ for each pair of sites $a_k^i, a_k^j \in m_k$ and $i < j$

After this learning process, to deal with a novel molecule m_p

Predict:

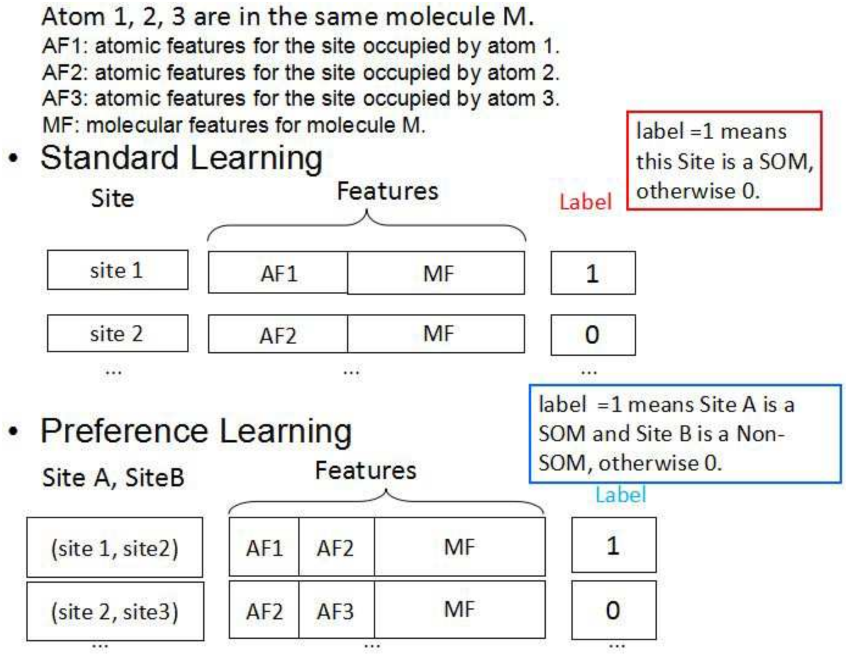
- a $C_{a_p^i,a_p^j}(m_p)$ for molecule m_p, over the set of sites A_p, where $a_p^i, a_p^j \in A_p$ and $i < j$

Derive:

- a ranking $\tau_p : A_p \rightarrow \{1 \ldots l_p\}$, from the set of $C_{a_p^i,a_p^j}(m_p)$ for each new molecule m_p
- the top few sites from the ranking τ_p for molecule m_p as SOMs

3.2.4 Comparison with standard learning

In this part, we compare the difference between standard learning (SL) and preference learning (PL) on SOM prediction problem, and the schema comparison is showed in Figure 3.4.



Atom 1, 2, 3 are in the same molecule M.

AF1: atomic features for the site occupied by atom 1.

AF2: atomic features for the site occupied by atom 2.

AF3: atomic features for the site occupied by atom 3.

MF: molecular features for molecule M.

label =1 means this Site is a SOM, otherwise 0.

label =1 means Site A is a SOM and Site B is a Non-SOM, otherwise 0.

Figure 3.4: Comparison between standard learning and preference learning

In standard learning problem, a site a_k^i of $m_k \in M$ that is a SOM, is considered to be positive, and each remaining non-SOM site is considered to be negative. Then a model is trained to fit this data. The problem with this framework is that there are certain preferences between the SOMs and non-SOMs, and the top k evaluation criterion (see Chapter 4) motivates us to optimize the ranking. Thus it is reasonable to have a rank over all the sites, rather than just predicting a value 1 or 0.

In preference learning, we are using pairwise comparison and optimizing for correctly ranking the sites. From the original dataset S, we are trying to construct a ranking τ_k of $\{1 \dots l_k\}$ for each molecule $m_k \in M$. For example, if a site $a_k^i \in m_k$ is a SOM, then it is preferable over all the non-SOMs in that molecule. For the sites in the molecule that are from the same set, either both are from SOMs or non-SOMs, we cannot infer any preference.

Standard learning:

- Each training instance is the description of one site, which is labeled 1 if it is a SOM.
- The goal is to learner a classifier, to predict which sites in a new molecule, are SOMs.

Preference learning:

- Each training instance is the description of two sites, which is labeled 1 if the first site (called site 1 in Figure 3.4) is a SOM and the second site (called site 2 in Figure 3.4) is a Non-SOM.
- The goal is to derive a ranking, over all sites in a new molecule, using the classifiers, learned from the pairs of instances.

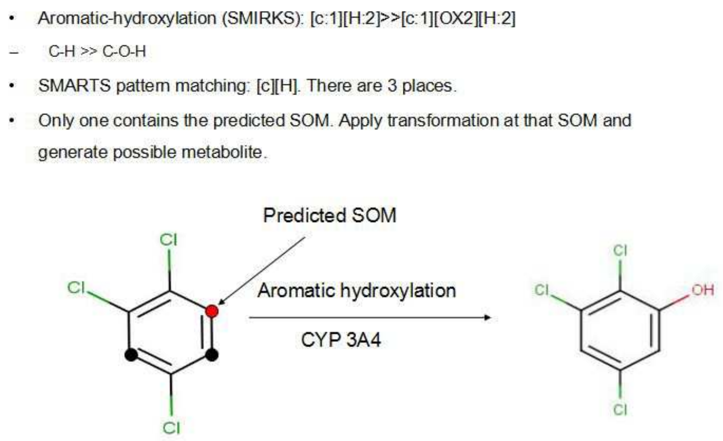
3.3 MEG

Given the predicted SOMs for a molecule and a specific enzyme, our metabolic endpoint generator, MEG, is able to generate all possible metabolites. We identify the different types of reactions catalyzed by each of the 9 CYP enzymes, i.e., we go through the literature to see what types of reactions each CYP enzyme catalyzes.

An example for illustrating the process of metabolic endpoint generation is shown in Figure 3.5.

Given a predicted set of SOMs S for a molecule M, after identifying a set of reactions that the specific enzyme can catalyze, the procedure for generating all metabolic endpoints is shown as following:

1. A reaction R is expressed in SMIRKS: X >> Y;
2. A SMARTS pattern P is identified in molecule M (P may be found in several sites in M);
3. Identify the sites containing the predicted SOMs in X, apply reaction R at that site and generate possible metabolite.



- Aromatic-hydroxylation (SMIRKS): [c:1][H:2]>>[c:1][OX2][H:2]
  - C-H >> C-O-H
- SMARTS pattern matching: [c][H]. There are 3 places.
- Only one contains the predicted SOM. Apply transformation at that SOM and generate possible metabolite.

Figure 3.5: In the figure, we have the input substrate and predicted SOM (red circle), and want to generate the metabolic endpoint obtained with enzyme CYP 1A2. We know that CYP 1A2 can catalyze aromatic hydroxylation, which involves aromatic carbons. The reaction is represented in SMIRKS. By SMARTS pattern matching, we find there are 3 sites (aromatic carbon) matched. But the other two (black circles) are not predicted by our SOM predictor. Thus we apply reaction at the SOM and generate the metabolic endpoint shown.

Chapter 4

Experiments

4.1 Datasets

To build the SOM predictor, we used the same datasets (DS 1) that Xenosite used, which is the largest publicly available repository of CYP450 substrates, containing 680 molecules for the nine enzymes[1]. All the molecules in the dataset have at least one experimentally verified SOM. For the substrate prediction task, we need to have a set of molecules, that include substrates, inhibitors and non-reactive molecules. For now, we just consider the first two groups: substrates and inhibitors. We collected the datasets of enzyme specific inhibitors from DrugBank (Datasets 2) [18]. We built our substrate predictor SubPred on Dataset 3 (DS3), which is the union of Datasets 1 (DS1) and 2 (DS2). The details for DS1 are shown in Table 4.1 and for DS3 are shown in Table 4.2.

4.2 Evaluation criterion

The standard evaluation metrics for the substrate predictor, SubPred, are as follows: sensitivity (recall), specificity, accuracy and precision [19].

Given a labeled dataset, $D \subset \{(x, y)|\ x \in S,\ y \in \{-1, 1\}\}$, where S is a set of molecules and y is the label, where -1 means negative (not metabolized) and 1 means positive (metabolized). Here, a classifier $C : S \rightarrow \{-1, 1\}$ takes an molecule $x \in S$, and returns either -1 or 1. We define the following terms:

[1] We modify the datasets(DS 1) a bit by deleting the molecule that contains the element boron B in DS 1, as some atomic features of B is not supported in CDK.

Name of Enzyme	Number of molecules	Number of SOMs	Number of Non-SOMs	Average sites/molecule	Percentage of SOMs
1A2	270	621	4681	19.6	11.7%
2A6	105	201	1429	15.5	12.33%
2B6	151	294	2564	18.9	10.29%
2C8	142	304	2780	21.7	9.9%
2C9	225	461	4269	21.0	9.8%
2C19	217	402	4146	21.0	8.9%
2D6	269	503	5101	20.8	9.0%
2E1	145	305	1936	15.5	13.6%
3A4	473	1067	10801	25.1	9.0%

Table 4.1: Details for the DS1

Name of Enzyme	Number of enzyme catalyzed molecules (DS1)	Number of inhibitors (DS2)
1A2	270	108
2A6	105	34
2B6	151	43
2C8	142	89
2C9	225	150
2C19	217	106
2D6	269	193
2E1	145	57
3A4	473	214

Table 4.2: Details for the DS3

- True positive: a molecule is metabolized by the enzyme and also predicted to be metabolized. The set is:

$$TP(C, D) = \{(x, y) \in D \mid C(x) = 1, y = 1\} \tag{4.1}$$

- True negative: a molecule is not metabolized by the enzyme and also predicted to be not metabolized. The set is:

$$TN(C, D) = \{(x, y) \in D \mid C(x) = -1, y = -1\} \tag{4.2}$$

- False positive: a molecule is not metabolized by the enzyme but predicted to be metabolized.

$$FP(C, D) = \{(x, y) \in D \mid C(x) = 1, y = -1\} \tag{4.3}$$

- False negative: a molecule is metabolized by the enzyme but predicted to be not metabolized.

$$FN(C,D) = \{(x,y) \in D \mid C(x) = -1, y = 1\} \tag{4.4}$$

Then the accuracy, precision and recall are define as:

$$\text{Accuracy}(C,D) = \frac{|TP(C,D)| + |TN(C,D)|}{|TP(C,D)| + |TN(C,D)| + |FP(C,D)| + |FN(C,D)|} \times 100\% \tag{4.5}$$

$$\text{Precision}(C,D) = \frac{|TP(C,D)|}{|TP(C,D)| + |FP(C,D)|} \times 100\% \tag{4.6}$$

$$\text{Recall}(C,D) = \frac{|TP(C,D)|}{|TP(C,D)| + |FN(C,D)|} \times 100\% \tag{4.7}$$

The F_1 measure is another common choice for performance evaluation, defined as the harmonic mean of precision and recall:

$$\text{F}_1(C,D) = \frac{2 \times \text{Precision}(C,D) \times \text{Recall}(C,D)}{\text{Precision}(C,D) + \text{Recall}(C,D)} \times 100\% \tag{4.8}$$

For the SOM prediction, a common evaluation metric is top-k accuracy, where a molecule is considered to be correctly predicted if one of its experimentally confirmed SOMs is ranked among the k top-ranked positions. Many competitions [42, 59, 60] use $k = 2$ [2].

4.3 Experimental results

Each dataset is divided into two parts: 80% of the molecules used for training and 20% for testing. We ran 5-fold cross validation (CV) on training set to select the best base learner and optimal parameters. Finally we run the best learner with optimal parameters on the test set and report its performance.

The motivation of using SVM for substrate prediction is illustrated in [57], where Yap and Chen demonstrated the advantages of SVM over some other classifiers for this task, e.g. logistic regression, decision tree, k-nearest neighbor. So we adopt SVM as base learner for our substrate predictor, SubPred. The performance of substrate predictor SubPred using SVM is shown in Table 4.3.

[2] The accuracy for SOM predictor refers to the "top 2" accuracy.

Name of Enzyme	Baseline	Test accuracy	Test precision	Test recall	Test F1-measure
1A2	71.43%	93.65%	97.67%	93.33%	0.9545
2A6	75.54%	100%	100%	100%	1.0
2B6	77.84%	100%	100%	100%	1.0
2C8	61.47%	97.37%	100%	95.65%	0.9778
2C9	60.00%	95.16%	92.5%	100%	0.9610
2C19	67.18%	100%	100%	100%	1.0
2D6	58.23%	98.70%	97.78%	100%	0.9888
2E1	71.78%	93.94%	95.83%	95.83%	0.9583
3A4	68.85%	100%	100%	100%	1.0

Table 4.3: Results of SubPred using SVM

For building the SOM predictor, SomPred, we first run five-fold CV validation experiments with different base learners on the datasets, to decide which learner yields better accuracy. We show one sample result on one of the nine datasets in Figure 4.1. The results are similar for other datasets.

In Figure 4.1, random forest yields better CV accuracy than others, so we decide to use random forest as the base learner [26]. The RF classification model (RFC) is implemented with calibrated PL using voting, and the RF regression model (RFR) is implemented with PL using choice (see Section 3.2.2). We use internal CV to tune the hyper-parameters: the number of trees n and the number of randomly selected features split m. A bootstrap sampling data is used to generate a full tree without pruning. The best parameters (m, n) for both models are chosen from $\{1, \sqrt{M}, \frac{M}{3}, \frac{M}{2}, M\} \times \{10, 20, \ldots, 100\}$ based on the CV accuracy, where M is the number of features. Our final model for SomPred is the RFC model with PL using voting.

The performance for using random forest model is shown in Table 4.4. We also present its results against other methods and predictors, which are shown in Figure 4.3.

Then we compare its performance with Xenosite [3]. In Figure 4.2, we show the five-fold CV results of SomPred and Xenosite. We also did a 2-sided paired

[3] Xenosite is reporting leave one out cross validation accuracy (LOOCV). For more details about the performance comparison of Xenosite with other predictors, see Figure 4.3 and [59].

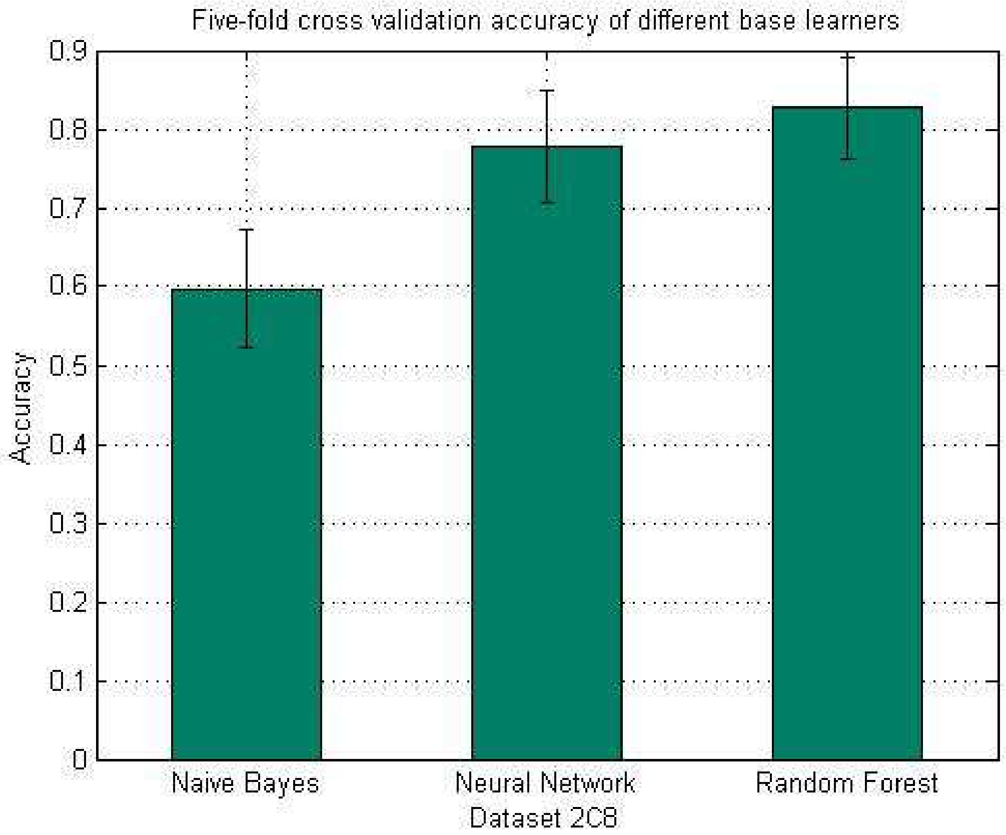


Figure 4.1: Five fold CV accuracy of different base learners (Naive Bayes, Neural Nets, Random Forest) on one dataset (CYP 2C8) for SOM prediction.

t-test for comparing our SomPred with Xenosite, over all 9 CYP enzymes [20]. The calculated value $\hat{t}$ is around 1.0, and the number of degrees of freedom $9 + 9 - 2 = 16$, leading to the tabulated value for "p=0.05" is $t^* = 2.12$ and P value $p^* = 0.33$. As $\hat{t} < t^*$ and $p^* > p$, we can not conclude that there is significant statistical difference between the performance of our SomPred and Xenosite. We repeat this similar procedure and compare our model's performance with other tools. The calculated values $\hat{t}$ and p^* are around 1.61 and 0.13 for SMARTCyp and 0.62 and 0.54 for RS-Predictor. As both $\hat{t} < t^*$ and $p^* > p$, so our model's performance is comparable with other predictors.

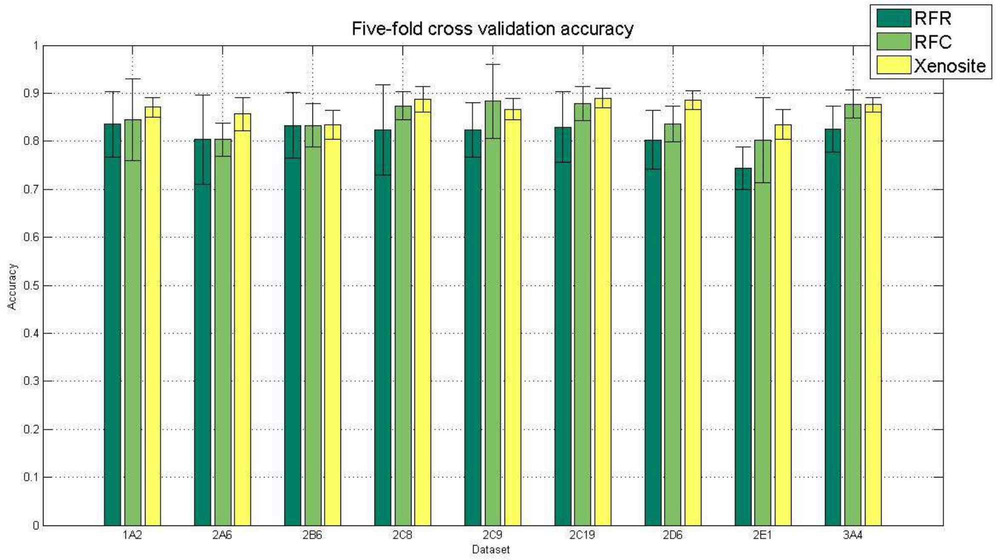


Figure 4.2: Five fold CV results of SomPred. Within each cluster of bars, the first bar represents five fold CV accuracy for RFR; the second bar represents five fold CV accuracy for RFC; the third bar represents leave one out CV accuracy for Xenosite.

4.4 Discussion

From the results in Table 4.3, we can see our substrate predictor, SubPred, is quite accurate, which indicates the molecular features selected are very expressive. This is what we want, as the errors in this predictor will propagate to the later parts, which could significantly affect the predictions for SOMs and metabolic endpoints. When training the substrate predictor, we are dealing with binary classification, i.e. the metabolized substrates are considered as positive examples and the inhibitors are considered as as negative examples. In general, it is more reasonable to add the group of non-reactive compounds and train a three-class classifier distinguishing substrate, non-reactive molecule and inhibitor. We will leave this as an extension of our substrate predictor.

For the SOM prediction model, from the results in Table 4.4, we can see the preference classification model is better than the regression model, but the latter can provide a probability for each site in a molecule being a SOM. While the prediction accuracy reflects the confidence of a model in its prediction for a molecule, the probability represents the statistical likelihood of that partic-

Name of Enzyme	Baseline	RFR accuracy	RFC accuracy	Xenosite
1A2	26.0%	84.44%	86.67%	**87.1%**
2A6	31.9%	76.47%	76.47%	**85.7%**
2B6	24.8%	76.92%	**86.00%**	83.4%
2C8	22.6%	78.26%	86.96%	**88.7%**
2C9	22.2%	81.89%	**88.19%**	86.7%
2C19	20.2%	86.11%	88.89%	**89.0%**
2D6	21.1%	84.09%	84.09%	**88.5%**
2E1	36.5%	79.17%	79.17%	**83.5%**
3A4	21.0%	86.46%	**89.03%**	87.6%

Table 4.4: Results of SomPred. For each CYP enzyme, the optimal model is shown in **bold**.

ular site is actually metabolized by a particular enzyme. Our generation for the features are efficient as we avoid quantum chemical calculation. The accuracy of our model is also competitive with others, which shows the preference learning framework fits well for this problem.

A general prediction system should be able to take an arbitrary molecule, and predict SOMs for specific enzyme. Notice other SOM predictors assume the input molecule is a substrate, so will always return some SOMs. But they do not consider if the molecule is not a substrate. We anticipate that our prediction system, that includes SubPred as a filter, will give a better prediction than others in general, as this combined system would first predict whether the given compound will be metabolized by a specific enzyme, and if not, will not predict any sites are SOMs, while other tools will always predict some number (typically 2) of SOMs for these molecules. To evaluate this combination prediction system (SubPred + SomPred) and compare with other tools, we will use Jaccard score as the evaluation criterion [21]. For computing Jaccard score, each algorithm need to decide how many SOMs should be returned for each molecule. In our learning scenario, we will extend to calibrated preference learning, which is described in Appendix A. We will leave this extension of our SOM predictor as future work.

Our metabolic endpoint generator is fast and efficient. Based on a set of 50 compounds, it generates 225 phase I metabolites in around 14 seconds on

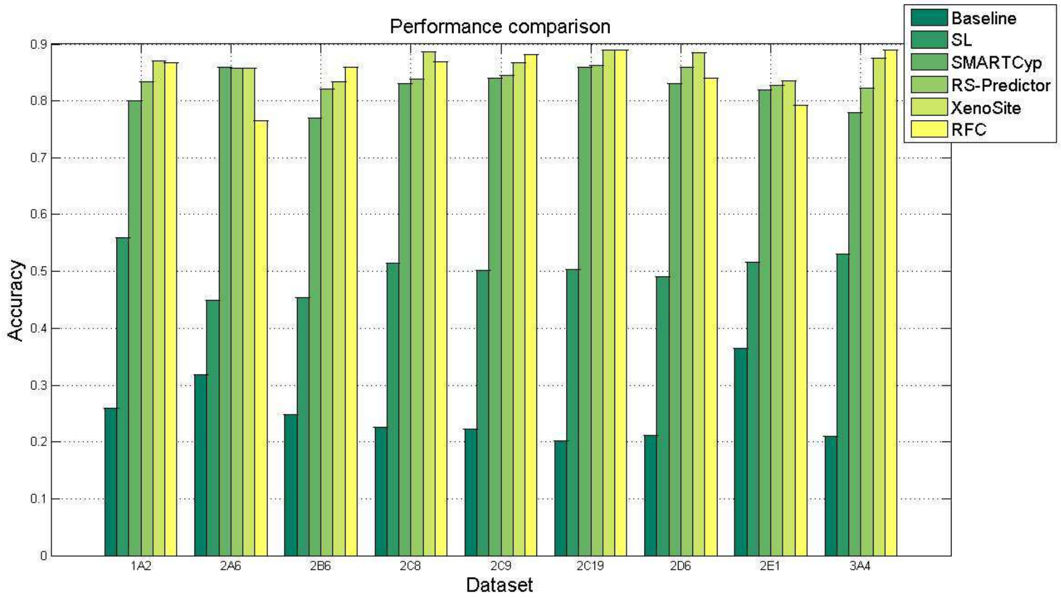


Figure 4.3: Comparison of different methods for SOM prediction. Within each cluster of bars, the first bar represents baseline result; the second bar represents test accuracy of Standard learning (SL); the third bar represents test accuracy of SMARTCyp [50]; the fourth bar represents test accuracy of RS-Predictor [58]; the fifth bar represents the accuracy of Xenosite [59]; the sixth bar represents the accuracy of RFC.

our computer (Windows 64-bit Operating System with an Intel i5 dual-core CPU, using 4 GB RAM).

Regarding the same set of 50 compounds, the computational time for generating all the features is around 26 seconds. The total time that SomPred requires to predict SOMs for these 50 compounds is around 63 seconds.

However, when we submit the file that contains 50 compounds to Xenosite server, it takes around 7 minutes to make SOM prediction, and another 12 minutes to produce the results, which is very slow and not convenient for interactive use [4].

Our results are based on relatively small datasets, of only 680 molecules. We anticipate getting better results if we have larger datasets.

[4] The Xenosite server only returns 19 compounds' result out of 50, it seems that the server can only handle 19 compounds at one time.

Chapter 5

Conclusion

5.1 Future work

5.1.1 Quantifying the preferences

One direction for extending the preference model is to further quantify the preferences between a pair of sites. In our training set (DS 1), we have three types of SOMs: primary, secondary and tertiary, which presumably refers to the likelihood that a reaction may happen at that site, e.g., primary SOM means a reaction takes place most of time at that site for a molecule. In this thesis, we treat all these as known SOMs, and the remaining as non-SOMs. However, there should be some preferences between them. For example, primary SOM's should be preferred than secondary SOM's. So if we could find a way to quantify these preferences, I expect the learning model could give a better performance.

5.1.2 Feature selection

Another direction that is worth exploring is to consider feature selection on atom fingerprints. The atom fingerprint features we generate now includes a variety of functional groups that a wide range of enzymes could target, not just for CYP enzymes. If we could generate/select enzyme specific atom fingerprint, the model will be less complicated and may give better performance.

5.1.3 Handling imbalance

For the SOM prediction, both the standard learning and preference learning encounter a common problem: data imbalance. This is due to the situation that, in our dataset, most of the sites are non-SOMs (the overall class ratio of SOM versus nonSOM is 1 : 8.7 on average, see Table 4.1 for more details). This problem makes model training more complicated:

1. With very few examples of one class, it is very difficult to learn the patterns for the minority class.

2. It is common to train the model for optimizing for 0-1 loss, which is not proper in this case, as one can achieved high accuracy by predicting everything as the majority class.

In this thesis, we are using cost sensitive classifiers, implemented by WEKA [37], to handle imbalance, where we give a much higher penalty for misclassifying the minority class, versus the majority. Typically, there are two common ways solving this problem. One is down-sampling the majority class [29], and the other is oversampling the minority class [28]. The disadvantage for the former is that it discards certain information in the dataset, and both of them changes the distribution of the classes in the dataset and may make the model biased.

A good alternative for handling data imbalance is to directly optimize the area under curve (AUC). However, one of the difficulties for using AUC as the objective function is that it is non-differentiable and its complexity is $O(n^2)$ in the number n of training instances. But if a classifier's objective function is close to the AUC statistic, then it usually produces a model with good AUC [31]. So if we could find a good way to do this, I anticipate the performance could be further improved.

5.2 Contributions

In this thesis, we propose a pipeline to predict xenobiotic metabolism, which includes SubPred that predicts whether a compound will interact with a particular enzyme, SomPred that predicts which part of the compound will be changed, and a Metabolic Endpoint Generator, MEG that produces the metabolites.

Comparing with previous methods, our approach has the following advantages:

- It includes a whole system from substrate prediction to metabolite generation, while others only work on certain parts (e.g., SOM prediction), which makes our system more applicable and scalable.

- It predicts the interactivity of the molecule with a specific enzyme using our SubPred, which is very accurate and effective to filter the non-reactive molecules and identify enzyme specific inhibitors.

- It provides a novel view of the SOM prediction problem, which allows people to compute simpler features and apply pairwise learning methods (e.g., preference learning) to this problem.

- It proposes an easy way to generate the metabolic endpoints, which is very fast and efficient.

The empirical results show that our framework is superior in terms of computational efficiency, and also competitive in terms of accuracy. Given that this success is based on a relatively small datasets, we anticipate a better performance on larger datasets with this approach.

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Appendix A

Calibrated label ranking

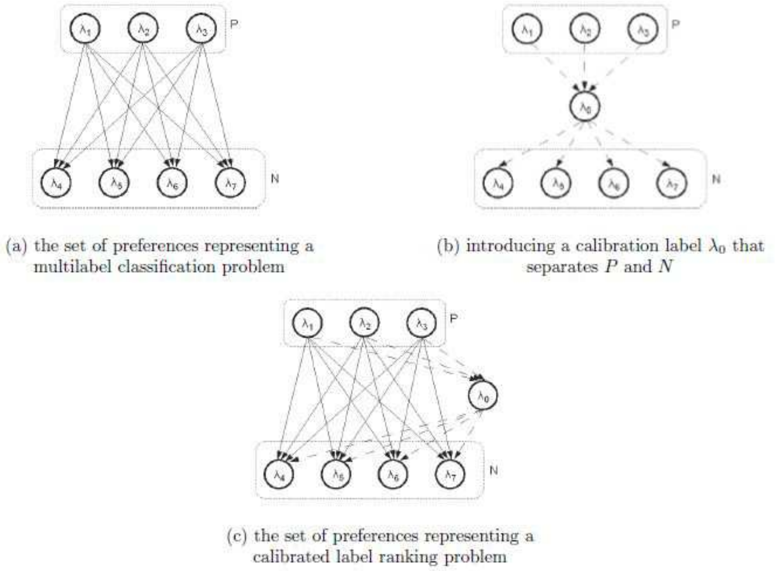
In conventional label ranking, the algorithm produces a ranking for all the candidates. We need a particular cutoff to make the predictions; this is then considered as the calibrated label ranking (CLR) [34].

The key idea of CLR is to add an additional calibration label λ_0 into the candidate set, producing the set of labels $L = \{\lambda_i |\ i = 0 \ldots m\}$. The idea is to use the label λ_0 as a split between positive (SOMs) and negative (non-SOMs) labels. For a given instance x, let P=$\{i|\ \lambda_i$ is a positive label$\}$, N=$\{j|\ \lambda_j$ is a negative label$\}$, then let $C(x) = \{C_{i,j}(x)|\ i \in P, j \in N\}$ be the conventional preference set, with calibration, the preference becomes:

$$\hat{C}(x) = C(x) \cup \{C_{i,0}(x)|i \in P\} \cup \{C_{0,j}(x)|j \in N)\} \tag{A.1}$$

With this new framework, when deriving a ranking $\hat{\tau}$ of $\{1 \ldots l + 1\}$ from the preference set $\hat{C}(x)$, all and only the labels who are ranked higher than λ_0 are predicted as positive. Figure A.1 gives a comparison for the new framework with previous one.

When applying the conventional pairwise preference learning for SOM prediction, we can derive a ranking for all the candidate sites, where the likely SOMs are ranked in the top positions. However, we lack a natural "zero" point to separate the set of SOMs from the set of non-SOMs. We can solve this problem by extending the idea to calibrated preference learning, by adding an artificial "zero" label to model the preferences between the candidates from the same set. Then we have:



(a) the set of preferences representing a multilabel classification problem

(b) introducing a calibration label λ_0 that separates P and N

(c) the set of preferences representing a calibrated label ranking problem

Figure A.1: Comparison between conventional label ranking and calibrated one. (Taken from [34].)

$$C_{i,j}(x) \leftarrow \begin{cases} 1 & if \quad \lambda_i \succ_x \lambda_j \\ -1 & if \quad \lambda_j \succ_x \lambda_i \\ 0 & \quad otherwise \end{cases} \tag{A.2}$$

With calibrated preference learning, when we induce a ranking τ, the sites whose score $S(a_i)$ (see Equation 3.2) is above "zero" (here, λ_0 is equivalent to "0"), are predicted as SOMs, i.e., $S(a_i) > 0$.