Università degli Studi di Padova – Department of Mathematics
Master's Degree in Data Science
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Classification of Contacts in Protein Structures

1. Training Set and Data Retrieval

The first step of the project was retrieving the data from each `.tsv` file from the `features_ring` folder (1,807 PDBs in total) and storing it into a single `DataFrame`.

The data contained in each `.tsv` file consists in a tab-separated file available for each protein, in which each row represents a contact in the protein and each column represents a feature about that contact. The last column is the target label, specifying the interaction type.

Column position	Column name	Column meaning	Type of column
1	`pdb_id`		
2	`s_ch`	chain	source residue identifier
3	`s_resi`	index	
4	`s_ins`	insertion code	
5	`s_resn`	name	
6	`s_ss8`	secondary structure 8 states (DSSP)	source residue features
7	`s_rsa`	relative solvent accessibility	
8	`s_up`	half sphere exposure up	
9	`s_down`	half sphere exposure down	
10	`s_phi`	phi angle	
11	`s_psi`	psi angle	
12	`s_ss3`	secondary structure 3 states (from angles)	
13	`s_a1`	Atchley feature 1	
14	`s_a2`	Atchley feature 2	
15	`s_a3`	Atchley feature 3	
16	`s_a4`	Atchley feature 4	
17	`s_a5`	Atchley feature 5	
18	`t_ch`	chain	target residue identifier
19	`t_resi`	index	
20	`t_ins`	insertion code	

21	t_resn	name	
22	t_ss8	secondary structure 8 states (DSSP)	target residue features
23	t_rsa	relative solvent accessibility	
24	t_up	half sphere exposure up	
25	t_down	half sphere exposure down	
26	t_phi	phi angle	
27	t_psi	psi angle	
28	t_ss3	secondary structure 3 states (from angles)	
29	t_a1	Atchley feature 1	
30	t_a2	Atchley feature 2	
31	t_a3	Atchley feature 3	
32	t_a4	Atchley feature 4	
33	t_a5	Atchley feature 5	
34	Interaction	interaction type	

Table 1.1: Training Set Description

Interaction Type	Count
HBOND	333,346
VDW	155,789
PIPISTACK	10,403
IONIC	9,068
SSBOND	866
PICATION	626
Unclassified	225,412

Table 1.2: Number of Examples by Bond Type.

2. Data Preprocessing

The preprocessing pipeline starts with removing all samples where the label is unavailable. Then, the missing values of each feature are replaced using the mean of the feature itself. Only numerical features are used.

It was considered to perform *Best Subset Selection* using Logistic Regression to determine which features were most meaningful and the ones that were not influencing the decision much. However, by doing that, the overall performance dropped, so it was decided to discard this approach and keep all features for training.

Scaling is then performed to standardize all the features to values between [0,1] to be then fed to the model.

The biggest criticality in the dataset is the heavy imbalance that is evident by looking at the number of contacts by interaction type. Notably, *Hydrogen Bonds* (`HBOND`) and *Van der Waals Contacts* (`VDW`) are the most numerous, which makes them overrepresented in the training set. The remaining contact types, instead, are underrepresented. Training a model with such unbalanced datasets is sure to yield poor performance, especially when evaluating the model on new unseen data.

To mitigate this issue, oversampling the underrepresented classes is applied to encourage the model to better learn the minority interactions. It is to be noted, however, that altering the dataset, especially by oversampling, can yield overly optimistic results in the training performance, which don't necessarily transfer to as good performances at inference time on new data. Therefore, the balancing process of the training set is performed carefully and conservatively.

The use of undersampling lead to greatly diminished performance distinguishing the majority classes (*Hydrogen Bonds and VDW Interactions).* For oversampling, *SMOTE* (*Synthetic Minority Oversampling TEchnique)* is used, which uses interpolation between samples to create new artificial data points. The sampling strategy is `{IONIC: 20'000, PIPISTACK: 10'000, PICATION: 20'000, SSBOND: 10'000}`

3. Model

3.1. Deep Neural Network

The model created to classify residue-residue contacts is a *Deep Neural Network* for multiclass classification. The Deep Learning library of choice is *Keras*, a very commonly used open-source library that acts as an interface for *TensorFlow*.

The first step to be carried out is the encoding of the label of each sample into an identity vector, using a common practice called "*One-Hot Encoding*". This ensures that each class is uniquely identified and independent of the others. It helps the neural network to better understand the categorical nature of the data and prevents any ordinal relationship assumptions between the classes.

One-Hot Encoding is also performed to then set the number of neurons of the output layer to be equal to the number of classes in the dataset, in order to train each output neuron to determine the probability $\mathrm{P}(C_i \mid \text{data})$, with $\sum \mathrm{P}(C_i \mid \text{data}) = 1$. Therefore, the output layer represents the probability distribution of a contact being of a certain type.

Although the model is a relatively small Deep Neural Network with fully connected layers, overfitting is mitigated using *L2 Regularization (Weight Decay)* on each `Dense` layer, and *Early Stopping* monitoring the loss progression over the epochs. These two techniques

provide better generalization performance. The use of random dropout of neurons after each hidden layer was also tested, without any meaningful improvement on final test performance.

3.2. Model Details and Hyperparameters

Hyperparameters	
Batch Size	16'000
Weight Initialization	Xavier (`GlorotNormal`)
Loss Function	Categorical Cross-Entropy
Optimizer	Adam
Hidden Layers: Activation Function	ReLU
Output Layer: Activation Function	Softmax

Table 3.2.1: Model's hyperparameters (Keras)

Layer Type	Output Shape	# Param
Dense	128	2432
Dense	128	16512
Dense	128	16512
Dense	128	16512
Dense	128	16512
Dense	6	774
# Total Parameters		69,254

Table 3.2.2: Model's architecture (Keras)

4. Results

4.1 Performance

Testing data consisted of 51,608 examples, or 10% of unbalanced non-preprocessed data. Particular attention was put into not having any leak of the oversampled training set data into the test set. Well behaved loss (*figure 4.1*) and AUC (*figure 4.2*) histories are evidence of successful model improvement. As demonstrated by the accompanying metrics, the model preforms better that the provided Naive Bayes classifier, achieving *on the test set* a Total Accuracy of 71%, ROC AUC of 0.79 (*figure 4.2*), and a Matthews Correlation Coefficient of 0.38.

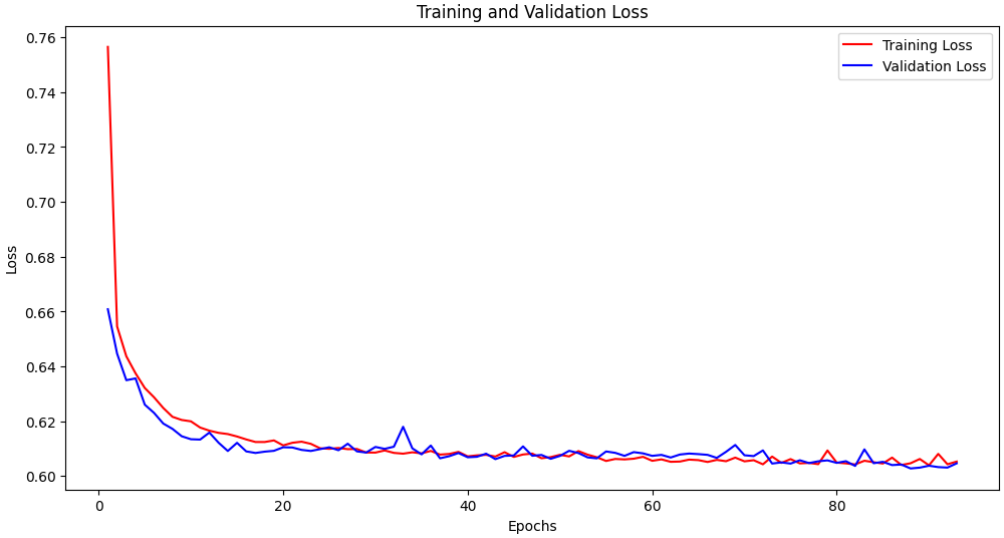

Figure 4.1: Training and Validation Loss

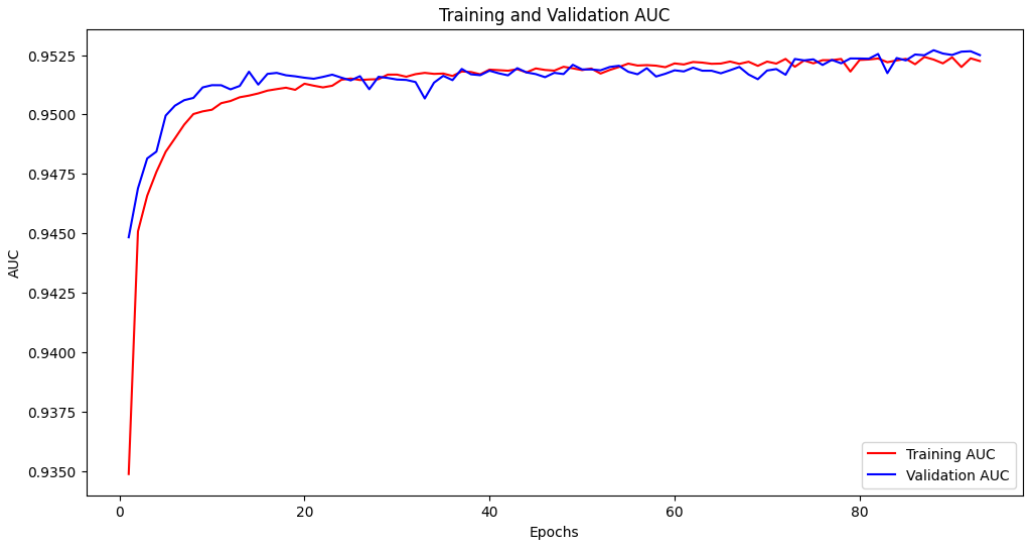

Figure 4.2: Training and Validation AUC

	precision	recall	f1-score	support
HBOND	0.77	0.85	0.81	33656
IONIC	0.46	0.26	0.33	916
PICATION	0.05	0.60	0.09	63
PIPISTACK	0.39	0.99	0.56	1050
SSBOND	0.43	1.00	0.60	88
VDW	0.61	0.41	0.49	15735
macro avg	0.45	0.68	0.48	51508
weighted avg	0.71	0.70	0.70	

Table 4.1: Test Performance by Interaction Type

4.2 Issues

In attempt to address poor precision on minority classes, many alternative approaches were tested, including:

- Ensemble Methods: One-vs-One, One-vs-All, and Model Stacking
- Different architectures
- Regularization techniques: Dropout, L2, Batch Normalization, Class Weighting
- Balancing

Nonetheless, it was found that ordinary Deep Neural Networks had the best compromise between minority class and majority class precision. Models that showed improvements in one of these areas consistently suffered in the other.
Implementing dataset balancing greatly improved model recall on minority classes. This would indicate that the model is effectively learning information about the classes. However, these classes still suffer from poor precision with resampling likely attributed to the relatively extremely low unique examples not allowing for a good representation of the class to be learned. The model additionally showed great sensitivity to undersampling, quickly leading to diminished ability to distinguish *Hydrogen Bonds* and *VDW Interactions*, the largest influence on total accuracy. The final model relied primarily on having the minority classes oversampled. Though, oversampling additionally quickly lead to poor generalization on minority classes due to the extremely low number of unique examples making learning a good representation of the classes difficult.

5. Usage

To predict the contacts of a new `PDB` file using the pretrained model, run the following command in your terminal, using arguments:

```
> python3 contact_net.py --predict --pdb your_pdb_id
> python3 contact_net.py --predict --pdb your_pdb_id --unprocessedfeats
> python3 contact_net.py --predict --pdb your_pdb_id --full
```

To perform retraining or the model, instead, run:

```
> python3 contact_net.py --train
```

The full documentation of the software is available in the GitHub repository at the following link

5.1 Example and Comparison with RING

An illustrative demonstration of usage can be seen with comparing the outputs of the model with the output of RING for *Deoxy Human Hemoglobin* (1A3N):

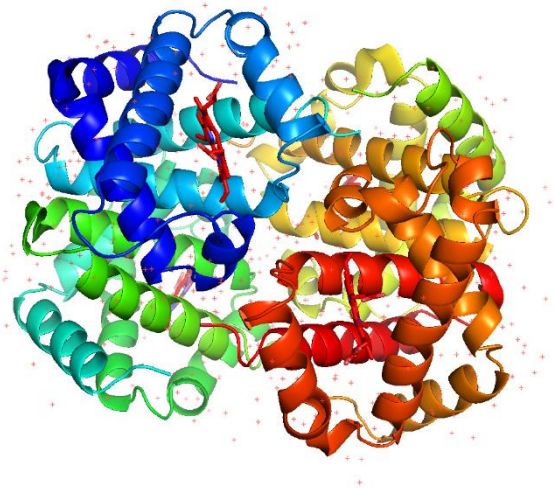

Figure 5.1.1: Deoxy Human Hemoglobin (PDB: 1a3n)

```
> python3 contact_net.py --predict --pdb 1a3n --full
```

↳ generates `/data/output/1a3n_pred.csv`

Submitting a corresponding `1a3n.pdb` to RING allows us to compare the outputs of the two models (*table 5.1.1*). Note that because the output of `calculate_features.py` and RING will provide multiple classifications for the same contact, the mode will be taken for each so accuracy can be calculated. Because the example of *Deoxy Human Hemoglobin* is a well-ordered globular protein, we can see that the model here provides favourable results, delivering an overall accuracy of 80 % on the one protein.

index	resA	resB	RING	pred	success
0	3	6	HBOND	HBOND	true
1	3	7	HBOND	HBOND	true
2	4	7	HBOND	HBOND	true
3	4	8	HBOND	HBOND	true
4	5	8	HBOND	HBOND	true
5	5	9	HBOND	HBOND	true
6	6	9	HBOND	HBOND	true
7	6	10	HBOND	HBOND	true
8	6	127	IONIC	HBOND	false
9	7	10	HBOND	HBOND	true
10	7	11	HBOND	HBOND	true
11	7	74	IONIC	VDW	false
12	7	132	IONIC	IONIC	true
13	8	11	HBOND	HBOND	true
14	8	12	HBOND	HBOND	true
15	8	78	VDW	HBOND	false
16	8	79	IONIC	HBOND	false
17	9	12	HBOND	HBOND	true
18	9	13	HBOND	HBOND	true
19	9	124	VDW	HBOND	false
20	10	13	HBOND	HBOND	true
21	10	14	HBOND	HBOND	true
22	11	14	HBOND	HBOND	true
23	11	15	HBOND	HBOND	true
24	11	130	VDW	VDW	true

Table 5.1.1: Comparison between ContactNet Predictions and RING.