# An Explainable Machine Learning Platform for Antimicrobial Resistance Prediction and Resistance Gene Identification

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# Introduction

## Antimicrobial Resistance - a Global Health Crisis

- **Antimicrobial resistance (AMR)** - bacteria or microbes evolve to resist certain antibiotics
- Worldwide, 700,000 people die annually due to AMR infections
- It is estimated that there will be 10 million deaths worldwide each year by 2050 if the trend continues
- World Health Organization (WHO) identifies AMR as one of the biggest threats to global health

# Problem

- A Challenge of treating AMR infection is long diagnosis time: two days to even a month with current culture-based diagnosis
- Current practice in clinics for AMR treatment is broad-spectrum antibiotic therapy
  - contributes to more AMR, oftentimes ineffective
- Rapid diagnosis of AMR in clinics urgently needed to save patient lives and prevent outbreaks
- Furthermore, diagnosis ought to be explainable, as this would be helpful for understanding the mechanisms behind resistance

# Proposal

**Using genomic data, Deep Convolutional Neural Networks (DCNNs) can predict AMR Accurately, and Support Vector Machine (SVM) can find the mutations that cause the resistance**

- AMR phenotype is determined by genomic data, a strain's chromosomal resistance mutations
- DCNN: advanced machine learning algorithm
  - Highly successful in image classification
  - High speed, accuracy
- Convert genomic data to image format
  - Analogous to image classification
- SVM intrinsically explainable - usable as a surrogate to DCNNs to pinpoint the genes and mutations that cause AMR

# Data Collection

## Genomic Data Retrieval

- Genomic mutations of 149 strains of *Mycobacterium tuberculosis* is collected from international hospitals
- Data on their resistance to pyrazinamide (PZA) is obtained from culture-based testing
- Genomic data is in Variant Call Format (VCF)
- Build a parser to gather all mutations from VCF. A snapshot of a strain's mutations are as below

| POSITION | MUTATION |
|---|---|
| 698 | G->A |
| 1977 | A->G |
| 4013 | T->C |

# Method

- **Convert Mutations To Genomic Images**
  - The union of all mutations from all strains form a list, with each entry having the position, the reference base, and alternate base.
  - For each strain, build an array initialized with all 0's
    - if it has a mutation that belongs to the union, the array element is set to 1
  - We pad each array with zeros so it can be reshaped an n x n array – "**Genomic Image**", similar in format to a traditional image represented with pixels
- **Represent AMR phenotypes as one-hot arrays**
  - Resistant strains are mapped to label [0, 1]; susceptible strains to label [1, 0]

# Method

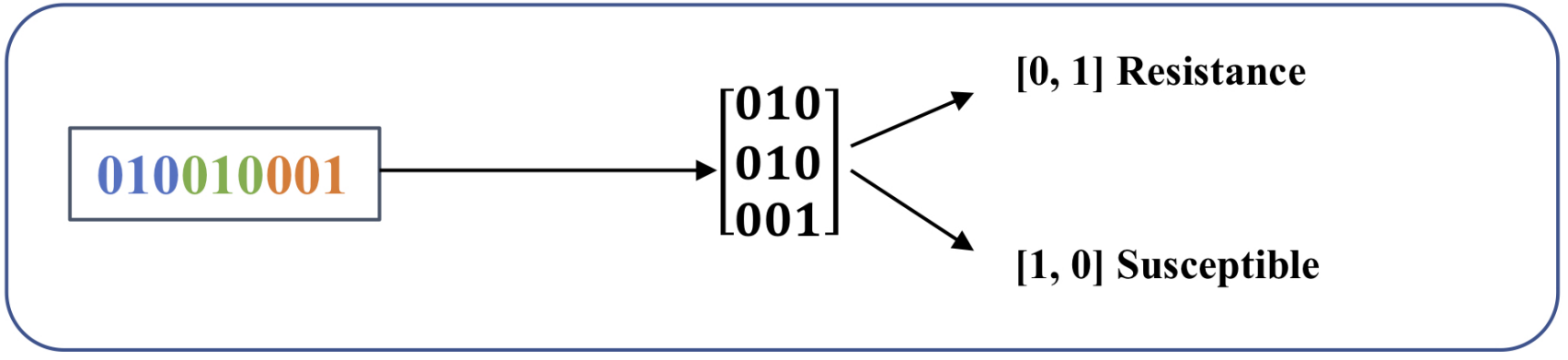
## Converting Mutations to Feature Array

**Union of mutations for all strains**

| | Position | Mutation | Gene |
|---|---|---|---|
| **1** | 7361 | C/T | Rv0006 |
| **2** | 7362 | G/C | Rv0006 |
| **3** | 7539 | A/G | Rv0006 |
| **4** | 2289215 | T/G | Rv2043c |
| **5** | 2289221 | T/G | Rv2043c |
| **6** | 2289227 | T/G | Rv2043c |
| **7** | 4247351 | G/A | Rv3795 |
| **8** | 4247577 | A/C | Rv3795 |
| **9** | 4247607 | A/G | Rv3795 |

**Mutations of strain BTB13-128**

| | Position | Mutation | Feature array |
|---|---|---|---|
| **1** | N/A | N/A | 0 |
| **2** | 7362 | G/C | 1 |
| **3** | N/A | N/A | 0 |
| **4** | N/A | N/A | 0 |
| **5** | 2289221 | T/G | 1 |
| **6** | N/A | N/A | 0 |
| **7** | N/A | N/A | 0 |
| **8** | N/A | N/A | 0 |
| **9** | 4247607 | A/G | 1 |



# Method

## Build a DCNN Model for AMR Prediction

- Four convolutional layers–filter size 4x4, and five filters per layer, as shown in Figure 2.
- At the last convolutional layer (Conv4 Layer), a large stride is used to reduce the dimension before a fully connected(FC) layer.
- No pooling layer is used, though they are widely used in image classification
  - Pooling would cause info loss in Genomic Image resulting in lower prediction accuracy
  - Use strides in convolution layer for dimension reduction instead of using pooling layers

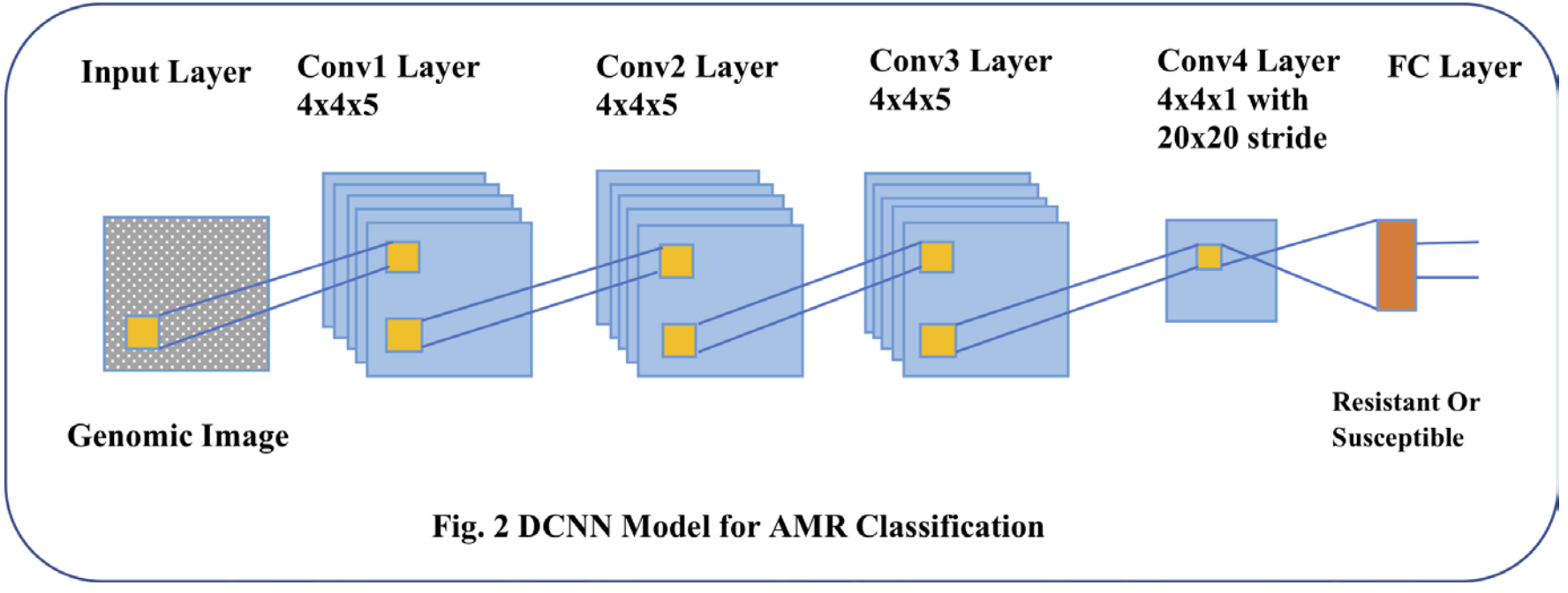



Fig. 2 DCNN Model for AMR Classification

# Method

## Build a Support Vector Machine Model to Identify Genes/Mutations that Harbor Resistance

- DCNN achieves high accuracy, but does not identify genes and mutations that harbor resistance
  - In medical applications, important to explain diagnosis
  - Explainable diagnosis help understand root cause and provide better therapies
- A SVM model is built as a surrogate to DCNN model
  - SVM is inherently explainable
  - The weights of the trained SVM indicate the importance of a feature
- A SVM model finds a hyperplane that divides the inputs to two classes with the maximum margin between the two
  - The resistant strain's input array is mapped to -1
  - The susceptible strain's input array is mapped to 1

# Method

## A Recursive Feature Addition Algorithm uses SVM to find resistance gene list, PZA_genes

1. From all the resistance genes, pick one gene a time to find the gene, gene_0, that produce the most accurate prediction using the SVM model. Add gene_0 to PZA_genes, so PZA_genes = {gene_0}.
2. Take another gene from the remaining genes one by one, gene_i, train the SVM model with gene_i and the genes in PZA_genes list. Pick the gene_i that produces the most accurate prediction, and is better than the result without gene_i by at least a margin, delta. If gene_i is found, add it to PZA_genes, so PZA_genes = {gene_0, …, gene_i}. If gene_i can not be found, stop.
3. Repeat step 2

# Procedure

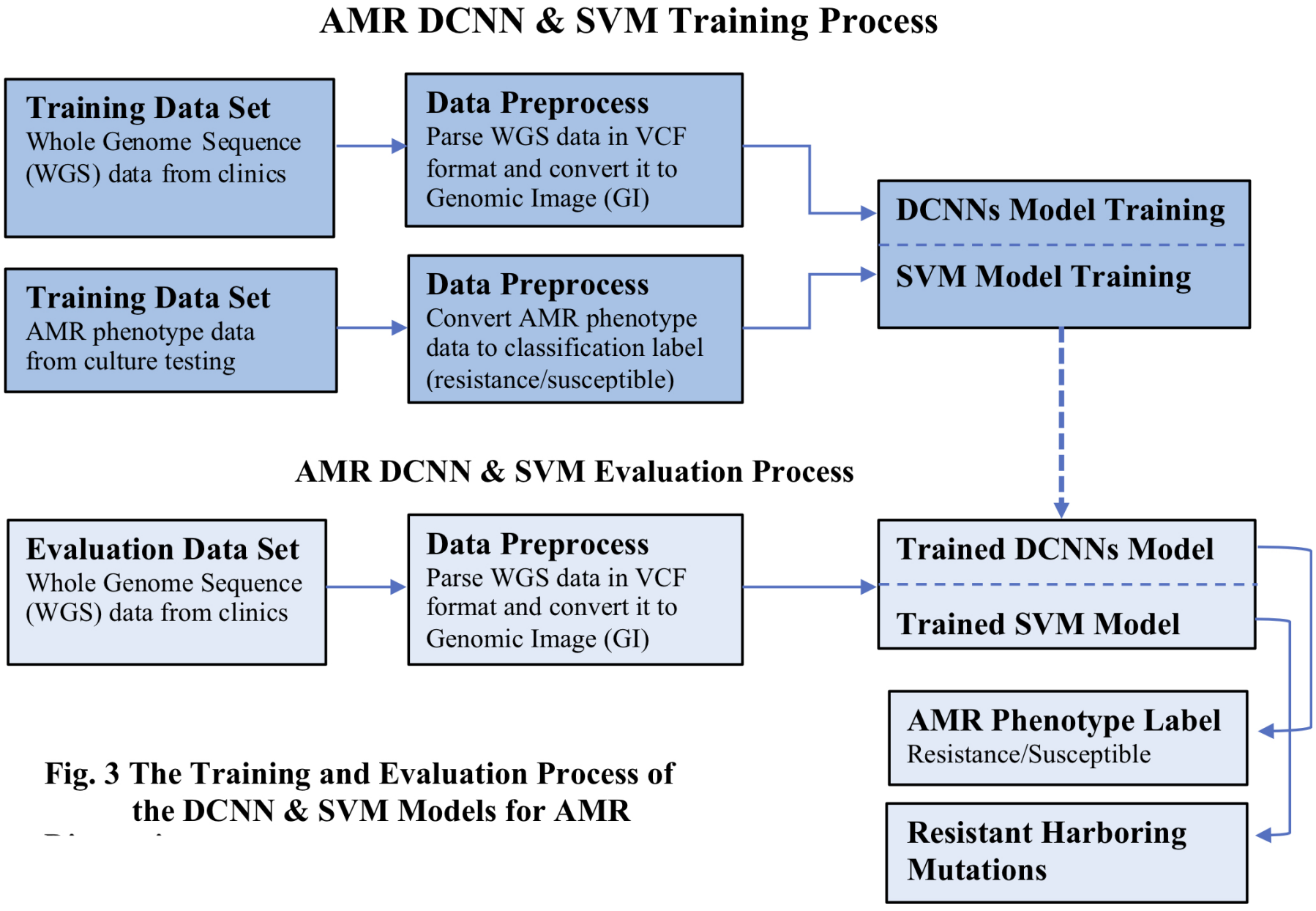



**Fig. 3 The Training and Evaluation Process of the DCNN & SVM Models for AMR**

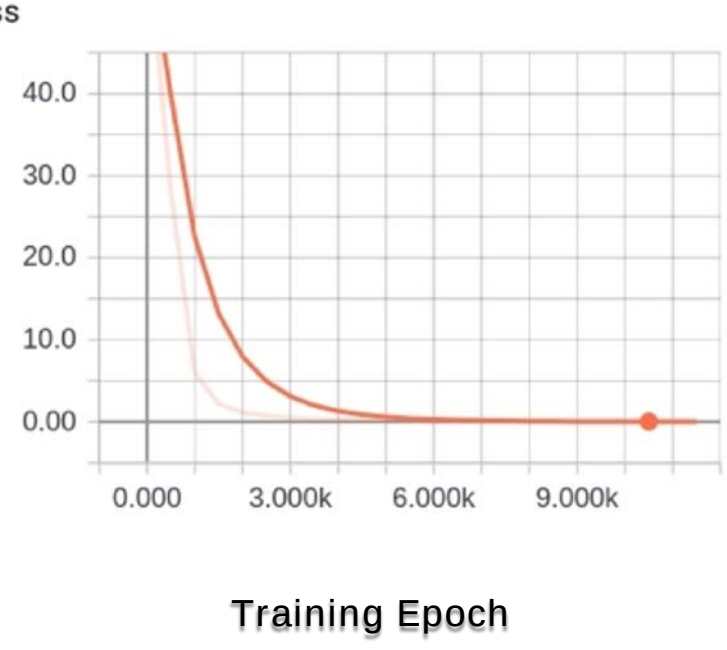
# Result

## Test Case 1: Prediction of M. Tuberculosis Resistant to Pyrazinamide (PZA)

120 strains for training and 29 strains for evaluation

### Training Result:

- Loss function is reduced to 0 in 9000 epochs
- Loss function is defined as the quadratic of the difference between model output and the actual label
- Loss reaching 0 means model output and label obtained from bacterial database fully match




### Evaluation Result:

- Only 2 out of 29 strains has wrong prediction – 93.1% accuracy.
- Prediction time is less than 1 second

# Result

## Test Case 2: Find Resistance Genes Using the SVM Model and the Recursive Feature Addition Algorithm

| Genes | Accuracy with training data | Accuracy with evaluation data | Total Mutations | Unique Mutations |
|---|---|---|---|---|
| pncA | 87% | 64% | 77 | 42 |
| pncA+embB | 85% | 78% | 199 | 79 |
| pncA+embB+gyrA | 89% | 80% | 704 | 104 |
| embB+gyrA | 79% | 72% | 627 | 62 |

- The algorithm starts with the gene *pncA*, a well-known PZA resistance gene
- It recursively adds one gene a time to get better accuracy
- The algorithm finds two other genes, *embB* and *gyrA*, which harbor PZA resistance
- The *embB* gene was also found to be correlated with PZA resistance in literature with different dataset, and *gyrA* is a novel gene

# Result

**Test Case 3: Find Mutations that Cause the Resistance**

| Position/Gene | Mutation | Resistant Strains | Susceptible Strains |
|---|---|---|---|
| 4247607/pncA | A->G | 15 | 7 |
| 4247609/pncA | G->A | 12 | 5 |
| 2289076/embB | G->C | 5 | 1 |
| 2288969/embB | A->C | 4 | 0 |
| 7581/gyrA | G->T | 2 | 0 |

- Using the weights of the trained linear SVM model, I have found 9 mutations that contribute the most to resistance (5 of them in the table).
- As PZA resistance is mapped to “-1”, and susceptible is mapped to label “1”, the mutation of a strain corresponding to the most negative weight, is the mutation that contributes the most to the resistance.

# Conclusion

- AMR diagnosis is tackled as a Genomic Image Classification problem using DCNN
- Diagnosis accuracy of 93.1% in less than a second
- A surrogate SVM model is built to identify resistance genes and mutations
- Create a Recursive Feature Addition algorithm that identifies three genes that harbor PZA resistance
- SVM pinpoints 9 mutations that caused the PZA resistance
- The rapid diagnosis could guide doctors to use the right antibiotics to save patients' life and prevent the outbreak of the deadly AMR infections.
- Identifying the genes and mutations helps understanding the biological mechanism of the resistance and combating it

# Future work

- Understand the biological mechanisms by which mutations cause the PZA resistance
- Run the ML platform side-by-side with culture-based diagnosis in a hospital lab

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# Questions & Answers