

# Documentation for Antibiotic Resistance Genes Annotation Tool

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## Contents

<b>1</b>	<b>Introduction</b>	<b>1</b>
<b>2</b>	<b>Installation</b>	<b>1</b>
<b>3</b>	<b>Program Usage</b>	<b>2</b>
3.1	Input your samples . . . . .	2
3.2	Run the annotation program . . . . .	2
3.3	Results . . . . .	2
<b>4</b>	<b>Customization (Change Parameters)</b>	<b>3</b>
4.1	Change evaluate cutoff . . . . .	3
4.2	Change percent identity . . . . .	3
<b>5</b>	<b>Citation</b>	<b>3</b>

## 1 Introduction

This program `ardbAnno.pl` is a command-line version of the antibiotic resistance genes annotation tool in Antibiotic Resistance Genes Database at <http://ardb.cbcb.umd.edu>. Basically, this program takes a set of genes or reads in FASTA format, and can identify and annotate potential antibiotic resistance genes using BLAST similarity search.

## 2 Installation

This annotation tool requires the installation of Perl, BioPerl toolkit and BLAST programs.

Uncompress the file by typing

```
tar xzvf ardbAnno.tar.gz
```

A directory named `ardbAnno1.0` should result. In this directory, there is a subdirectory named `blastdb`, which contains the reference resistance genes `resisGenes.pfasta`. To compile this data set for BLAST, within this `blastdb` subdirectory type

```
formatdb -i resisGenes.pfasta -p T -o T
```

### 3 Program Usage

#### 3.1 Input your samples

In the directory `ardbAnno1.0`, there is a file named as `genomeList.tab`, which contains a list of FASTA files that need to be annotated. Open this file using text editor, the content format is

```
>test  
test.pfasta
```

`>test` specifies the name of the sample is `test` and the second line is the location of your sample FASTA sequence file. You can include multiple samples into this file, for example

```
>test1  
test1.pfasta  
>test2  
test2.pfasta
```

#### 3.2 Run the annotation program

After you have specified your samples in the previous step, you are ready to run the annotation program. Within the directory `ardbAnno1.0`, type

```
perl ardbAnno.pl
```

#### 3.3 Results

For each sample there are two result files: (1) raw blast file (with `blastp` or `blastx` suffix); (2) annotation file (with `anno` suffix). In addition, for all the samples specified in file `genomeList.tab`, their annotations are summarized into an excel sheet `output.xls`. Format of 'anno' file

```
>NP_878016  
BestHit CAL90940
```

```
Type vana
Description VanA type vancomycin resistance operon genes ...
Resistance vancomycin teicoplanin
Require vanra vanha vansa vanya vanxa
Evaluate 0.0
QueryLength 343
HitLength 343
HSPLength 343
PercentIdentity 100.00
```

First line `>NP_878016` is the ID of the query sequence. `BestHit` means the best BLAST hit in the reference antibiotic resistance genes dataset `resisGenes.pfasta`. `Type` is the gene type name of the best BLAST hit. `Resistance` is the antibiotic resistance profile. `Require` means `vana` gene requires other cooperative genes to perform resistance function. `HSPLength` is the high scoring pair alignment length from BLAST.

## 4 Customization (Change Parameters)

For a gene to be annotated as resistance gene, two criteria are used in this program: (1) E-value; (2) percent identity. The E-value cutoff is specified in variable `$valueCutoff` in program `ardbAnno.pl`, which means all resistance genes identification using this same evaluate cutoff. The percent identity for different resistance gene types vary, and this value is specified in the last column of file `originType.tab` in directory `tabs`.

### 4.1 Change evaluate cutoff

Open program `ardbAnno.pl`, change the value of the variable `$valueCutoff` to the value you need.

### 4.2 Change percent identity

Open file `originType.tab` in directory `tabs`, for each resistance gene (row), change the value (in percentage) of the last column to your desired percent identity.

## 5 Citation

Please cite the following paper as the reference to this program.

Liu, B. and M. Pop (2009). "ARDB—Antibiotic Resistance Genes Database." *Nucleic Acids Res* 37(Database issue): D443-7.