

# Installation Guide for AmiRNA and WMD3

## Release 3.1

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## 1 Introduction

This document describes the installation process for WMD3/AmiRNA. WMD3 (Web Micro RNA Designer version 3) is a web application for artificial microRNA design based on the AmiRNA Perl module library. For more information please visit the website at <http://wmd3.weigelworld.org>. We focus on a single tier deployment although multi tier installations are possible. The recommended target platform is a Ubuntu 8.10 server system, but other Linux/UN\*X flavors supporting Perl, MySQL, Apache and a C compiler will work as well.

This installation How To is split up into 7 sections: Section 1 describes the database setup. Section 2 deals with the installation of the AmiRNA Modules including the installation of its dependencies (some Perl modules, GenomeMapper, Blast, the ViennaRNA package and MySQL). Section 3 explains the AmiRNA configuration, section 4 the data preprocessing, section 5 the installation of the web application and finally section 6 the designer daemon setup.

## 2 Database server setup

We support SQLite and MySQL as database backend. If you choose SQLite as backend you have to install the appropriate DBI drivers:

```
sudo apt-get install libdbd-sqlite3 libdbd-sqlite3-perl
```

If you choose MySQL and you don't have a database server setup yet, install MySQL now:

```
$ sudo apt-get install mysql-server mysql-client
```

Login as root

```
$ mysql -u root -p
```

and create a new user and grant some permissions on the new database:

```
create database amiradb;  
create user 'amirna'@ '%' identified by 'passwd';  
GRANT ALL ON amiradb.* TO 'amirna'@ '%' identified by 'passwd';  
GRANT ALL ON amiradb.* TO 'amirna'@ 'localhost' identified by 'passwd';
```

## 3 AmiRNA Installation

### 3.1 Check and install prerequisites

#### 3.1.1 Third party Perl modules

The AmiRNA package relies on some third party Perl modules (XML::Smart , DBI, Data::Types, Error and XML::Dumper). They are all available from CPAN.

```
$ perl -MCPAN -e 'install XML::Smart'
$ perl -MCPAN -e 'install DBI'
$ perl -MCPAN -e 'install DBD::mysql'
$ perl -MCPAN -e 'install Data::Types'
$ perl -MCPAN -e 'install Error'
$ perl -MCPAN -e 'install XML::Dumper'
```

On an Debian like operating system (e.g. Ubuntu) you can use your system package management to install these modules:

```
$ sudo apt-get install libxml-smart-perl libdbi-perl libdata-types-perl
liberror-perl libxml-dumper-perl libdbd-mysql-perl libgd2-xpm-dev liblog-
log4perl-perl
```

#### 3.1.2 Build environment

Some prerequisites like GenomeMapper and ViennaRNA needs to be compiled from source. Please check if you have a C/C++ compiler and the make utility installed.

```
$ sudo apt-get install gcc g++ make
```

#### 3.1.3 GenomeMapper

Download GenomeMapper (version 0.3.1beta or higher) from the project site at <http://1001genomes.org/downloads/genomemapper.html>

```
$ cd /tmp; wget
http://www.1001genomes.org/data/software/genomemapper/genomemapper_0.3.1b
eta/genomemapper.tar.gz
```

Untar the package

```
$ tar xvfz genomemapper.tar.gz
```

and build the program

```
$ cd genomemapper && make
```

Then install the binaries on standard locations

```
$ sudo cp genomemapper gmindex /usr/local/bin
```

### 3.1.4 Optional: Vmatch

You can use Vmatch as a drop-in replacement for GenomeMapper.

More information on Vmatch could be found at <http://www.vmatch.de/>

```
$ sudo tar xvzf vmatch-x86_64-unknown-linux-gnu-64-bit.tar.gz
$ sudo mv vmatch.distribution /usr/local/bin/vmatch
$ sudo chmod ugo+x /usr/local/bin/vmatch/*
$ export PATH=$PATH:/usr/local/bin/vmatch
```

### 3.1.5 ViennaRNA

AmiRNA needs RNAup and RNAcofold from the ViennaRNA package. Download the latest release from <http://www.tbi.univie.ac.at/RNA/>.

```
$ cd /tmp; wget http://www.tbi.univie.ac.at/RNA/ViennaRNA-1.8.1.tar.gz
$ tar xvfz ViennaRNA-1.8.1.tar.gz
$ cd ViennaRNA-1.8.1
$ ./configure && make
$ sudo make install
```

### 3.1.6 NCBI Blast

The Blast tool is needed for the AmiRNA data preprocessing procedure and the WMD3 web application.

```
$ sudo apt-get install blast2
```

## 3.2 Install AmiRNA

### 3.2.1 Download AmiRNA/WMD3

```
$ cd /tmp; wget http://wmd3.weigelworld.org/downloads/wmd3-3.1.tar.gz
```

### 3.2.2 Install AmiRNA Modules

```
$ tar xvfz wmd3-3.1.tar.gz
```

```
$ cd wmd3-3.1/AmiRNA/  
$ perl Makefile.PL  
$ make  
$ sudo make install
```

### 3.2.3 Install AmiRNA Tools

```
$ sudo cp tools/Designerd/designerd.pl /usr/sbin  
$ sudo cp tools/Designerd/designerd /etc/init.d/  
$ sudo mkdir /etc/AmiRNA/  
$ sudo cp tools/Designerd/designerd.xml /etc/AmiRNA/  
$ sudo cp tools/Preprocess/* /usr/local/bin  
$ sudo cp tools/amirna_initdb.pl /usr/local/bin
```

## 4 AmiRNA Configuration

### 4.1.1 Copy AmiRNA configuration to /etc/AmiRNA

```
$ sudo cp etc/AmiRNA.xml /etc/AmiRNA/
```

### 4.1.2 Adjust AmiRNA.xml

The configuration file is `/etc/AmiRNA/AmiRNA.xml`. Open it with your favorite text editor and adjust it for your needs.

```
$ sudo vim /etc/AmiRNA/AmiRNA.xml
```

Set the path where your transcript libraries will be stored.

```
<index_path>/var/biodata/amirna/genomemapper</index_path>
```

Please create this directory if it does not exist.

```
$ sudo mkdir -p /var/biodata/amirna/genomemapper
```

Set the directory where temporary files will be created. The default should work in the most cases.

```
<tmp_path>/tmp</tmp_path>
```

Configure the database. Use the same database name and user as in the database setup above. For MySQL please specify the database name, host and port in the `<dsn>` directive:

```
<database>
  <engine>MySQL</engine>
  <dsn>DBI:mysql:database=amirnadb;host=localhost:port=3306</dsn>
  <user>amirna</user>
  <password>passwd</password>
</database>
```

For SQLite please specify the location of the database file (dbname) in the <dsn> section:

```
<database>
  <engine>SQLite</engine>
  <dsn>dbi:SQLite:dbname=/var/biodata/amirna/amirna.db</dsn>
  <user>amirna</user>
  <password>passwd</password>
</database>
```

Choose the mapping tool. Currently supported are genomemapper and vmatch. Please specify the name of the binary and the path in the mapping section. For the folding section specify the path of RNAup and RNAcifold. If you are unsure about the paths, try 'which genomemapper' and 'which RNAcifold'.

```
<external_programs>
  <mapping>
    <path>/usr/local/bin</path>
    <binary>genomemapper</binary>
  </mapping>

  <folding>
    <path>/usr/local/bin</path>
  </folding>
</external_programs>
```

#### 4.1.2.1 Setup the database tables

Initially create the queue and results relations. amirna\_initdb.pl uses the database defined in /etc/AmiRNA/AmiRNA.xml.

```
$ amirna_initdb.pl
```

This initialization script should create three database tables. You can verify this with:

```
$ mysql -u amirna -D amirnadb -p -e "show tables;"
```

The output should look like

```
+-----+
| Tables_in_amirnadb |
+-----+
| queue               |
| results             |
| sequences           |
```

## 5 Data preprocessing

### 5.1 Generate mapping index

Get some Fasta files and run generate\_mapping\_index.pl:

```
$ cd /tmp; wget
http://wmd3.weigelworld.org/downloads/TAIR8_cdna_20080412.gz
$ gunzip TAIR8_cdna_20080412.gz
$ sudo generate_mapping_index.pl -d "A. thaliana CDNA" -f
TAIR8_cdna_20080412
```

### 5.2 Generate Blast index

```
$ sudo mkdir /var/biodata/amirna/blast
$ sudo cp /var/biodata/amirna/genomemapper/TAIR8_cdna_20080412
/var/biodata/amirna/blast/
$ cd /var/biodata/amirna/blast/
$ sudo formatdb -p F -i TAIR8_cdna_20080412
```

## 6 WMD3 Setup

Install the apache web server and imagemagick:

```
$ sudo apt-get install apache2 imagemagick
```

Install the blast tools if not yet happened:

```
$ sudo apt-get install blast2
```

The web application WMD3 needs some third party Perl modules:

```
$ sudo apt-get install libcgi-application-perl libcgi-application-
plugins-perl libhtml-template-perl libspreadsheet-writeexcel-perl
```

and

```
$ sudo perl -MCPAN -e "install
CGI::Application::Plugin::View::HTML::Template"
$ sudo perl -MCPAN -e "install CGI::Application::Plugin::Stash"
```

Create an application directory and copy the CGI programs:

```
$ sudo mkdir /usr/lib/cgi-bin/wmd3
```

```
$ cd /tmp; sudo cp -r wmd3-3.1/WMD3/WMD3 wmd3-3.1/WMD3/webapp.cgi wmd3-3.1/WMD3/wmd3.xml /usr/lib/cgi-bin/wmd3
```

Create a static webroot directory and copy the layout files:

```
$ sudo mkdir -p /var/www/wmd3  
$ sudo cp -r wmd3-3.1/WMD3/layout_files /var/www/wmd3/
```

Configure the web application:

```
$ sudo vim /usr/lib/cgi-bin/wmd3/wmd3.xml
```

and adjust the URL prefix of the static files (htdocs), the URL of the web application (cgi), the path of the static web root (webroot) and the administrator email address.

Do not change the 'standard\_project' though we have currently only one software project.

In the section 'amirna' please specify the location of your AmiRNA.xml configuration file. If you followed the instructions above the default values will work for you.

```
<web_app>  
  <htdocs>http://localhost/wmd3</htdocs>  
  <cgi>http://localhost/cgi-bin/wmd3/webapp.cgi</cgi>  
  <webroot>/var/www/wmd3</webroot>  
  <admin_mail>wmdhelp@localhost</admin_mail>  
  <standart_project>stdwmd</standart_project>  
</web_app>  
  
<amirna>  
  <configfile>/etc/AmiRNA/AmiRNA.xml</configfile>  
  <default_genome>TAIR8_cdna_20080412</default_genome>  
</amirna>  
  
<blast>  
  <db_location>/var/biodata/amirna/blast</db_location>  
</blast>
```

## 7 designerd

### 7.1 Prerequisites

You need a configured AmiRNA installation (see section 3 and 4), a Database (section 1) and preprocessed data (section 5).

If you intend to setup a computation node hosting only designerd, you can configure designerd to use a remote database. Index files must be accessible for this host over the filesystem either as a local copy or shared via a network filesystem. For more information see Appendix A.

## 7.2 Configure designerd

Open the configuration file for designerd:

```
$ sudo vim /etc/AmiRNA/designerd.xml
```

Please specify the location of your AmiRNA configuration file (amirna\_config), your mailhost, email address and the base URL of your WMD3 installation:

```
<config>
  <amirna_config>/etc/AmiRNA/AmiRNA.xml</amirna_config>
  <max_childs>2</max_childs>
  <no_jobs_sleep_time>120</no_jobs_sleep_time>
  <pid_file>/var/run/designerd.pid</pid_file>
  <log_file>/var/log/designer_log</log_file>
  <mailhost>mailhost.tuebingen.mpg.de</mailhost>
  <from_email>wmd@tuebingen.mpg.de</from_email>
  <wmd3_webapp_url>http://localhost/cgi-
bin/wmd3/webapp.cgi</wmd3_webapp_url>
</config>
```

Please note that <mailhost> must reference a valid SMTP server.

## 7.3 Start designerd

Designerd is implemented as a UN\*X daemon running in the background. Use init.d scripts to start and stop the daemon:

```
$ sudo /etc/init.d/designerd start
```

You can monitor designerd's activity watching it's log file:

```
$ tail -f /var/log/designer_log
```

If you would like designerd to be run at bootup you can add it to the default runlevels using update-rc.d.

```
$ sudo update-rc.d designerd defaults
```

Now you are ready to fire up your web browser and browse to <http://localhost/cgi-bin/wmd3/webapp.cgi>. WMD3 should launch.

Have fun!



# Appendix

## A. Setting up a designerd computation node

It's possible to run only designerd on a host without a local database or web application installed on this host. One or more designerds share the queue, results and sequences database. This allows a distributed setup for parallel designer job processing.

Install GenomeMapper, ViennaRNA and other prerequisites.

```
# Prerequisites
$ sudo apt-get install libxml-smart-perl libdbi-perl libdata-types-perl
liberror-perl libxml-dumper-perl libdbd-mysql-perl libgd2-xpm-dev liblog-
log4perl-perl gcc g++ make

# GenomeMapper
$ cd /tmp; wget
http://www.1001genomes.org/data/software/genomemapper/genomemapper_0.3.1b
eta/genomemapper.tar.gz
$ tar xvfz genomemapper.tar.gz
$ cd genomemapper && make
$ sudo cp genomemapper gminindex /usr/local/bin

# ViennaRNA
$ cd /tmp; wget http://www.tbi.univie.ac.at/RNA/ViennaRNA-1.8.1.tar.gz
$ tar xvfz ViennaRNA-1.8.1.tar.gz
$ cd ViennaRNA-1.8.1
$ ./configure && make
$ sudo make install
```

Install AmiRNA.

```
$ cd /tmp; wget http://wmd3.weigelworld.org/downloads/wmd3-3.1.tar.gz
$ tar xvfz wmd3-3.1.tar.gz
$ cd wmd3-3.1/AmiRNA/
$ perl Makefile.PL
$ make
$ sudo make install

$ sudo cp tools/Designerd/designerd.pl /usr/sbin
$ sudo cp tools/Designerd/designerd /etc/init.d/
$ sudo mkdir /etc/AmiRNA/
$ sudo cp tools/Designerd/designerd.xml /etc/AmiRNA/
$ sudo cp etc/AmiRNA.xml /etc/AmiRNA/
```

Now configure AmiRNA. You need to specify a local path to the GenomeMapper / Vmatch index files and a remote database.

Next, run the data preprocessing script and generate the index files (See section “Generate mapping index”, you do not need to generate blast indices). As final step you have to configure and start designerd (see the appropriate section above).

## B. Upgrading

### Clean old Installation

```
# Stop services
$ sudo /etc/init.d/designerd stop
$ sudo /etc/init.d/apache2 stop

# or redirect vhost and restart apache
$ sudo vim /etc/apache2/sites-available/wmd3
# Disable CGI
AliasMatch ^/cgi-bin(.*) /var/www/wmd3/down.html
$ /etc/init.d/apache2 restart

# WMD3 web application
$ sudo rm -rf /usr/lib/cgi-bin/wmd3/ /var/www/wmd3/

# designerd
$ sudo rm -rf /usr/sbin/designerd.pl /etc/init.d/designerd

# AmiRNA Perl modules
$ sudo rm -rf /usr/local/lib/perl/5.8.8/auto/AmiRNA /usr/local/share/man/
man3/AmiRNA* /usr/local/share/perl/5.8.8/AmiRNA

# Reset queue
$ mysql -u wmd_user -p wmd3 -e "UPDATE queue SET started=0 WHERE
started=1; SELECT id, started FROM queue;"
```

Now you can proceed with the installation of AmiRNA and WMD3.

## C. Deinstalling

```
# Stop services
$ /etc/init.d/designerd stop
$ /etc/init.d/apache2 stop # or redirect vhost

# WMD3 web application
$ rm -rf /usr/lib/cgi-bin/wmd3/ /var/www/wmd3/

# designerd
$ rm -rf /usr/sbin/designerd.pl /etc/init.d/designerd

# AmiRNA Perl modules
$ rm -rf /usr/local/lib/perl/5.8.8/auto/AmiRNA /usr/local/share/man/man3/
AmiRNA* /usr/local/share/perl/5.8.8/AmiRNA

# Configuration files
$ rm -rf /etc/AmiRNA/

# Empty the database and delete index files
$ mysql -u amirna -D amirnadb -p -e "delete from queue; delete from
results; delete from sequences;"
$ rm -rf /var/biodata/amirna/genomemapper /var/biodata/amirna/vmatch
```