# Getting started with CRISPRDirection v.1.0

CRISPRDirection is developed in Fedora 14 with perl 5.10, and tested in Red hat linux and MacOS (Version-10.6.8). If you already have 'RNAfold', 'seqret', and 'water' pre-installed and accessible, then you don't need anything else. To test CRISPRDirection_v1.0.pl program, open a terminal and issue the following commands:

First download the **CRISPRDirection_v1.0.zip** file:

```
$ wget http://bioanalysis.otago.ac.nz/CRISPRDirection/CRISPRDirection_v1.0.zip
```

Then unzip **CRISPRDirection_v1.0.zip** file (say in your desktop),

```
$ unzip CRISPRDirection_v1.0.zip
```

Move to CRISPRDirection_v1.0 folder,

```
$ cd CRISPRDirection_v1.0
```

and issue the following command :

```
$ perl CRISPRDirection_v1.0.pl -h
```

[Note: $ is the GNOME Terminal 2.32.0 prompt]

A detailed report will be printed, showing if the required programs are installed and accessible. Alternately, you may need Emboss tools (version 6.3.1 or higher) and Vienna RNA package (version 2.0.7 or higher) to be installed. The 'other_executables' folder contains the executables of **RNAfold**, **water** and **seqret** but they may not work with your version of Linux. Check each of them by the following commands from the current directory:

```
./other_executables/RNAfold -h

./other_executables/water -h

./other_executables/seqret -h
```

If you see all the programs are running successfully, then copying them to a location like '/usr/local/bin' [require root access] will solve the issue. Alternately, exporting the 'other_executables' folder to your path will work. To know how to export a directory refer to http://www.troubleshooters.com/linux/prepostpath.htm

***Prerequisites***

1. You must have a working perl installation preferably >5.8 (comes with pre-installed in most operating system except Windows).

2. CRISPRDirection uses **'water'** and **'seqret'** from EMBOSS tools. Please follow the instructions from http://emboss.sourceforge.net/download/ to get and install EMBOSS tools.

We have noticed, that the '**water**' executable works in most unix based operating systems. If 'water' works, then simply copying the program to /usr/local/bin or any other folder which is in your system path will make it accessible.

'**seqret**' is required to extract genomic sequences from genbank annotation files (.gbk). If you want to use CRISPRDirection only for fasta sequence files, and if '**water**' is working, then you may skip installation of EMBOSS tools.

3. **RNAfold**: RNAfold comes with Vienna RNA package and is required (http://www.tbi.univie.ac.at/RNA/) for the method "Analysis of RNA secondary structure". If you don't have a pre-installed working version of RNAfold, you can install RNAfold program from

http://www.tbi.univie.ac.at/~ronny/RNA/index.html#download

For more details on installation of Vienna RNA package please refer to

http://www.tbi.univie.ac.at/RNA/INSTALL.html

4. CRISPR arrays are predicted by both **PILER-CR** and **CRT**. Both the executable are provided with this distribution. However, if either of them fails to run, you should obtain a working version of
PILER-CR (http://www.drive5.com/pilercr/) & CRT (http://www.room220.com/crt/)
and replace the existing versions of the program kept in this directory.

[**Note**, if the directory name of the version differes (for PILER-CR), then modify the CRISPRDirection_v1.pl script accordingly. To do so, open the CRISPRDirection_v1.pl with a standard text editor (e.g. gedit), and search for "./pilercr1.06/pilercr" (currently in line number 529). Replace the "./pilercr1.06" with the toplevel directory name of the PILER-CR program (e.g. ./pilercr1.02). If **'pilercr'** is in your system path (e.g. /usr/local/bin), then simply remove the './pilercr1.06' from the line. Similarly, if you have a different version of CRT (e.g. CRT1.1-CLI.jar), then find "CRT1.2-CLI.jar" (currently in line 534) and replace the existing "java -cp CRT1.2-CLI.jar" with the name of your CRT jar file.]