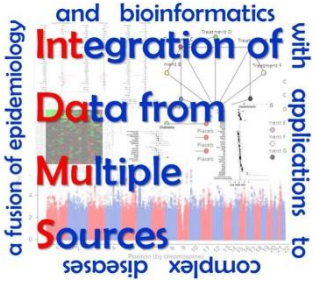

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Ευρωπαϊκό Κοινωνικό Ταμείο

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ΕΚΠΑΙΔΕΥΣΗ ΚΑΙ ΔΙΑ ΒΙΟΥ ΜΑΘΗΣΗ
επένδυση στην κοινωνία της γνώσης
ΥΠΟΥΡΓΕΙΟ ΠΑΙΔΕΙΑΣ ΚΑΙ ΘΡΗΣΚΕΥΜΑΤΩΝ
ΕΙΔΙΚΗ ΥΠΗΡΕΣΙΑ ΔΙΑΧΕΙΡΙΣΗΣ
Με τη συγχρηματοδότηση της Ελλάδας και της Ευρωπαϊκής Ένωσης

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πρόγραμμα για την ανάπτυξη
ΕΥΡΩΠΑΪΚΟ ΚΟΙΝΩΝΙΚΟ ΤΑΜΕΙΟ

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Workshop III: Bioinformatics with Perl

October 29, 2015

Instructors: Pantelis Bagos, Panagiota Kontou
University of Thessaly

What is Perl?

- Scripting language by Larry Wall
- Practical Extraction and Reporting Language
- Easy language to use
- Fast cross platform text processing.
- Good pattern matching.
- Many extensions for Life Sciences data types

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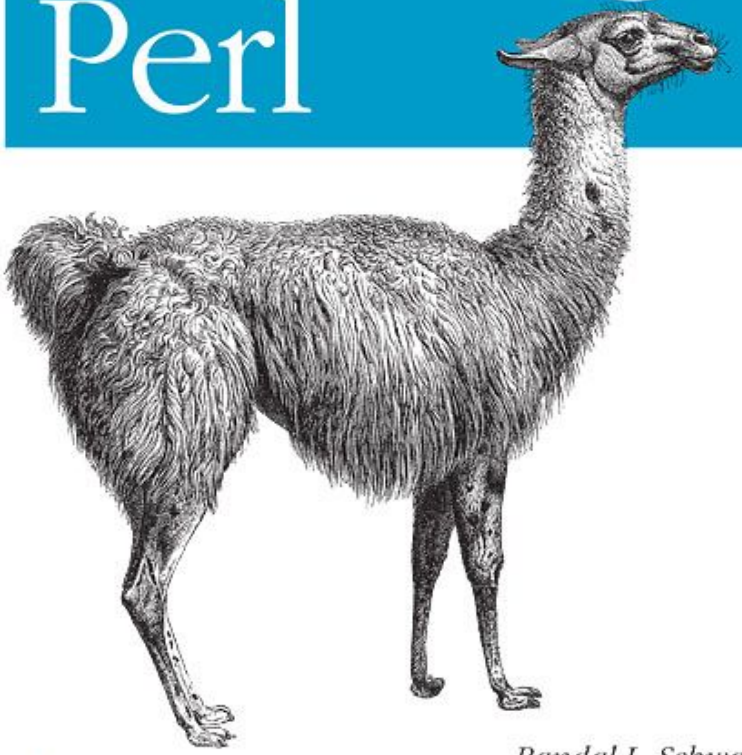

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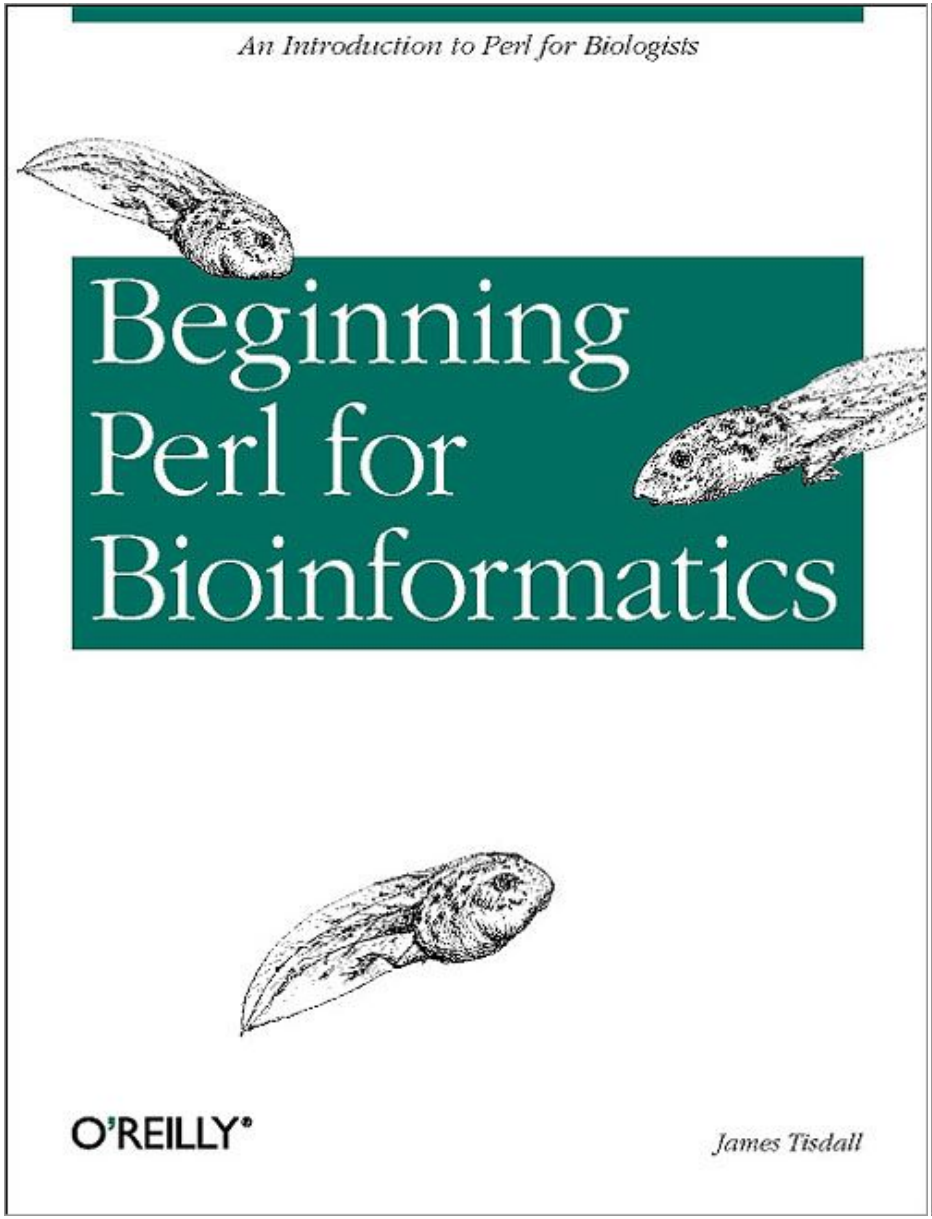
An Introduction to Perl for Biologists
Beginning Perl for Bioinformatics
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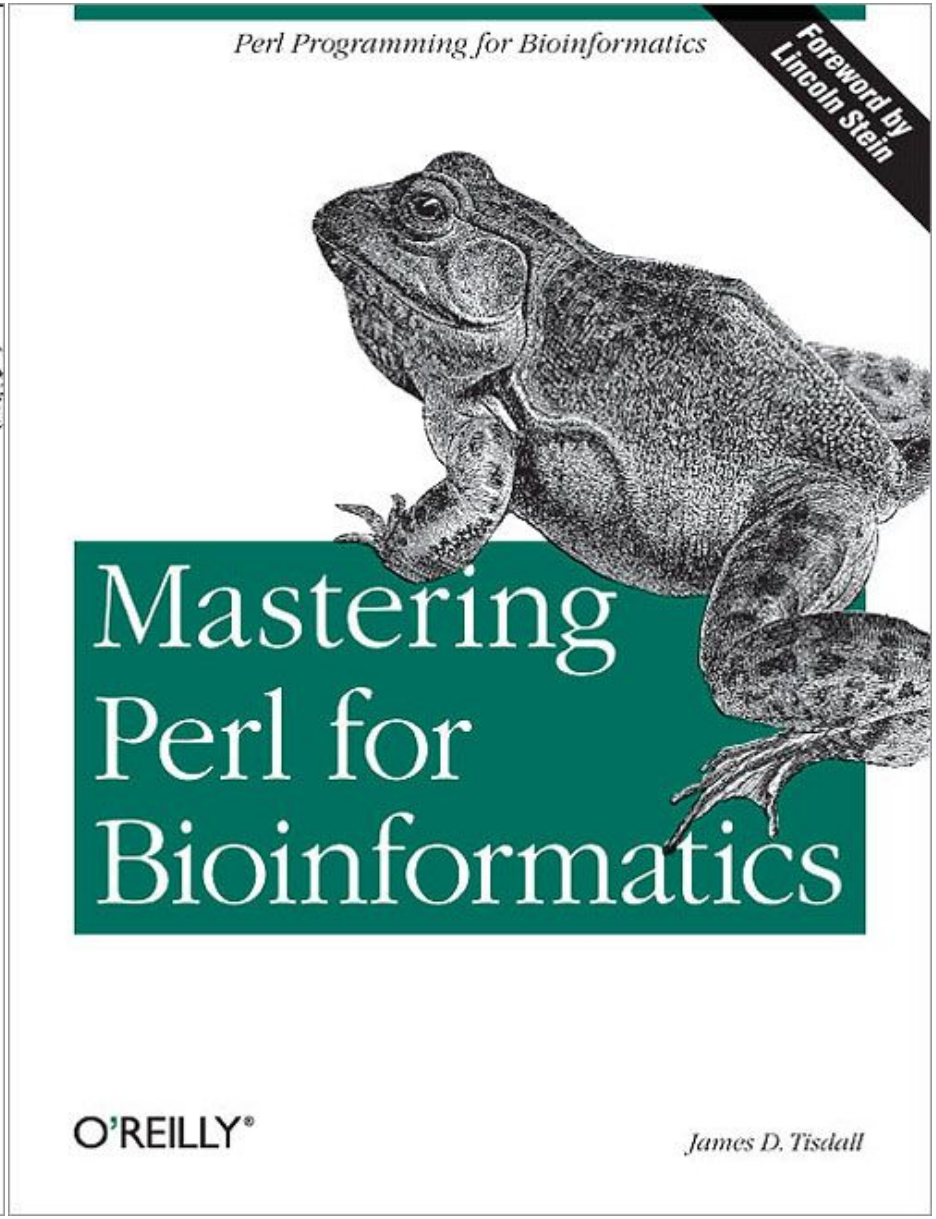
Perl Programming for Bioinformatics
Foreword by Lincoln Stein
Mastering Perl for Bioinformatics
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Getting Started

```
#!/usr/bin/perl
# First Program
print “Hello World\n”;
```

Getting Started

- perl program.pl
- ./program.pl
- perl –e ‘…perl program…’

<STDIN>

```
#!/usr/bin/perl
# Second Program

print “What is your name\?\n”;
$name =<STDIN>;
chomp $name;
print “Hello $name\n”;
```

Scalars και Variables

There are two basic data types in Perl: numbers and strings.

Numbers

- 5
- -245
- 1.25
- 7.25e45
- -12e44
- -1.2e-25

Strings

- Double quotes or single quotes may be used around literal strings:

```
print "Hello, world";
print 'Hello, world';
```

- However, only double quotes "interpolate" variables and special characters such as newlines (\n):

```
print "Hello, $name\n";      # works fine
print 'Hello, $name\n';      # prints $name\n literally
```

- Escape (\t, \n, \U, \L)

printf, sprintf

```
$x=3.7009;
print $x;
printf (“%.3f\n”, $x);
$y=sprintf (“%.3f”, $x);
print “$y\n”;
```

Operators

- Numbers

  +, -, *, /, **, %, <, >, ==, !=

- Strings

  ., x, eq, ne, lt, gt, le, ge

Variables

$variable_name=String or number or expression;

- Special variables ($_, $/, $1,etc)
- $number=5;
- $name=“George”;
- $exp=3*$number+($number+1);
- $a+=5;
- $b*=3;
- $c .=“ ”;
- ++$a; or $a++;
- $a--;

substr, index

- $name=“Takis”;
- $x=substr($name,0,1);
- $y=substr($name,0,1,“L”);
- print “$name\t$x\t$y\n”;
- $position=index($name, “k”);
- $position=rindex($name, “k”);

chomp, chop

- chomp ($x)
- chop ($x)

Interpolation

```
$test=‘bla bla bla’;
print “this is a $test\n”;
print ‘this is a $test\n’;
print ‘this is a \$test\n’;
print “this is a ${test}bla bla\n”;
```

Upper Case, Lower Case

```
$bigname="\Uname";
$name="name";
$bigname="\U$name";
$capname="\u$name"
```

Lists-Arrays

List:
(1, 2, 3)
(“perl”, 3, 15)
($x, 3, $x+2, “$y$x”)
(1..10)

Array
@name=(1..10);

Arrays

- @name=(1..10);
- @table=1;
- @table=@name;
- @name=(“John”, “George”, ”Mike”);
- @name=qw(John George Mike);
- @table=(1, 2, @name, 7);
- ($a, $b, $c)=(1,2,3);
- ($a,$b)=($b,$a);
- ($a, @table)=($c, $d, $e);
- $table=(1,2,3);
  $x=@table;
  ($y)=@table;

Index number of the elements

- $table=(1,2,3);
- $x=$table[0]
- $table[1]++;
- ($table[0], $table[1])= ($table[1], $table[0]);
- @table[0,1]=@table[1,0];
- @table[0,1,2]=@table[1,1,1];
- @table=(1,2,3);$x=$table[3];
- $table[5]=12;
- print $#table;
- print $table[$#table];

push, pop

- push @table,$scalar;
- @table=(@table, $scalar);
- $last=pop(@table);

shift, unshift

- unshift(@table, $scalar);
- @table=($scalar, @table);
- $x=shift(@table);
- ($x, @table)=@table;

reverse, sort, splice

- @table=(1,2,3);
- @new_table=reverse(@table);
- @name=(“John”, “George”, ”Mike”);
- @sort_names=sort(@name);
- splice(@table,1,1);

Hashes

- A hash is a collection of zero or more pairs of scalar values, called keys and values
- An array variable begins with the % sign followed by a legal variable name

```
%genes=('gene1'=>'AACCCGGTTAACCG',
        'gene2'=>'CCAAATTCCCCTTG');
```

Hashes

- %password=(“pbagos”, 123, “pkontou”, 321);
- %table=@table;
- @table=%table;
- %names=reverse %password;

- %password=(“pbagos”, 123, “pkontou”,321);
- @list=keys(%password);
- @list2=values(%password);

Hashes

%password=(“pbagos”=> “123”, “pkontou”=>”321”);

@password{“pbagos”, “pkontou”}=(123, 321);

Hashes

$hash{“key”}=“value”

- $password{“pbagos”}=“123”;
- $password{“pkontou”}=“321”;
- print $password{“pbagos”};
- $name=“pbagos”;
- print $password{$name};

Conditions and Loops

- if/unless
- while/until
- do{}while/until
- for
- foreach
- last, next, redo

if

```
if (some condition)
{
}
elsif
{
}
else
{
}
```

while/until

```
while (condition)
{
…
}
until (condition)
{
…
}
```

do {}while/until

```
do
{
…
} while (condition)
```

```
do
{
…
} until (condition)
```

for

```
for (initialization; condition; update)
{
…
}

for ($i=1; $i<=10;$i++)
{
print “$i\n”;
}
```

foreach

```
foreach $i(@list)
{
…
}

@a=(1,2,3,4,5);
foreach $i(@a)
{
print “$i\n”;
}
```

last

```
while (condition 1)
{
…
if(condition 2)
{
…
last;
}
}
```

Can be used only in: for, foreach, while, until

next

```
while (condition 1)
{
…
if(condition 2)
{
…
next;
}
```

redo

```
while (condition 1)
{
#
…
if(condition 2)
{
…
redo;
}
}
```

<>

```
while(<>)
{
print $_;
}
```

Run:
Perl program.pl file

Input-Output

- open FILEHANDLE, “filename”;
- open IN, “/etc/passwd”;
- $x=<IN>;
- print $x;
- close IN;
- open OUT, “>(>)tempfile”;
- print OUT “bla bla bla\n”;

@ARGV

- perl program.pl file1 file2 …
- file1: $ARGV[0]
- file2: $ARGV[1]

…

Regular Expressions

- A *regular expression* is a simple way of matching a series of symbols to a pattern you have in mind

```
$pattern=~/abc/;

if(/abc/){
print “I found it”;
}
```

Regular Expressions Syntax

char	meaning
^	beginning of string
$	end of string
.	any character except newline
*	match 0 or more times
+	match 1 or more times
?	match 0 or 1 times; or: shortest match
|	alternative
( )	grouping; “storing”
[ ]	set of characters
{ }	repetition modifier
\	quote or special

Regular Expressions Syntax

Matching

\w matches any single character classified as a “word” character
\W matches any non-“word” character
\s matches any space character
\S matches any non-space character
\d matches any digit character, equiv. to [0-9]
\D matches any non-digit character

Examples

Expression	Matches...
abc	abc (that exact character sequence, but anywhere in the string)
^abc	abc at the *beginning* of the string
abc$	abc at the *end* of the string
a|b	either of a and b
^abc|abc$	the string abc at the beginning or at the end of the string
ab{2,4}c	an a followed by two, three or four b's followed by a c
ab{2,}c	an a followed by at least two b's followed by a c
ab*c	an a followed by any number (zero or more) of b's followed by a c
ab+c	an a followed by one or more b's followed by a c
ab?c	an a followed by an optional b followed by a c; that is, either abc or ac
a.c	an a followed by any single character (not newline) followed by a c
a\.c	a.c exactly
[abc]	any one of a, b and c
[Aa]bc	either of Abc and abc
[abc]+	any (nonempty) string of a's, b's and c's (such as a, abba, acbabcacaa)
[^abc]+	any (nonempty) string which does *not* contain any of a, b and c (such as defg)
\d\d	any two decimal digits, such as 42; same as \d{2}
\w+	a "word": a nonempty sequence of alphanumeric characters, such as foo and 12bar8 and foo_1

Doing Substitutions

Replace every “Hello” with a “Hi"

$string =~ s/Hello/Hi/;

Doing Translations

Translations are like substitutions, except they happen on a letter by letter basis instead of substituting a single phrase for another single phrase

```
$string =~ tr/[a,e,i,o,u,y]/[A,E,I,O,U,Y]/;
$string =~ tr/[A,E,I,O,U,Y]/[1,2,3,4,5]/;
$string =~ tr/[a-z]/[A-Z]/;
```

split

```
#!/usr/bin/perl
$data = 'Becky Alcorn,25,female,Melbourne';
@values = split(',', $data);
  foreach $val (@values) {
    print "$val\n";
  }
```

This program produces the following output:

```
Becky Alcorn
25
female
Melbourne
```

join

```
#!/usr/bin/perl

$string = join( "-", "one", "two", "three" );
Print "Joined String is $string\n";
```

This program produces the following output:

```
Joined String is one-two-three
```

Application in Bioinformatics

Genetic code

You have to write a Perl program that:

1) Takes as input a DNA sequence

2) Finds the complementary strand and the mRNA that is produced

3) Translates the DNA using all 6 possible reading frames and outputs the possible ORFs and the putative protein sequences.

```
%genetic_code = (
'GCA'=>'A', #Alanine
'GCC'=>'A', #Alanine
'GCG'=>'A', #Alanine
'GCT'=>'A', #Alanine
'AGA'=>'R', #Arginine
'AGG'=>'R', #Arginine
'CGA'=>'R', #Arginine
'CGC'=>'R', #Arginine
'CGG'=>'R', #Arginine
'CGT'=>'R', #Arginine
'AAC'=>'N', #Asparagine
'AAT'=>'N', #Asparagine
'GAC'=>'D', #Aspartic acid
'GAT'=>'D', #Aspartic acid
'TGC'=>'C', #Cysteine
'TGT'=>'C', #Cysteine
'GAA'=>'E', #GlTtamic acid
'GAG'=>'E', #GlTtamic acid
'CAA'=>'Q', #GlTtamine
'CAG'=>'Q', #GlTtamine
'GGA'=>'G', #Glycine
'GGC'=>'G', #Glycine
'GGG'=>'G', #Glycine
'GGT'=>'G', #Glycine
'CAC'=>'H', #Histidine
'CAT'=>'H', #Histidine
'ATA'=>'I', #IsoleTcine
'ATC'=>'I', #IsoleTcine
'ATT'=>'I', #IsoleTcine
'TTA'=>'L', #LeTcine
'TTG'=>'L', #LeTcine
'CTA'=>'L', #LeTcine
'CTC'=>'L', #LeTcine
'CTG'=>'L', #LeTcine
'CTT'=>'L', #LeTcine
'AAA'=>'K', #Lysine
'AAG'=>'K', #Lysine
'ATG'=>'M', #Methionine
'TTC'=>'F', #Phenylalanine
'TTT'=>'F', #Phenylalanine
'CCA'=>'P', #Proline
'CCC'=>'P', #Proline
'CCG'=>'P', #Proline
'CCT'=>'P', #Proline
'AGC'=>'S', #Serine
'AGT'=>'S', #Serine
'TCA'=>'S', #Serine
'TCC'=>'S', #Serine
'TCG'=>'S', #Serine
'TCT'=>'S', #Serine
'ACA'=>'T', #Threonine
'ACC'=>'T', #Threonine
'ACG'=>'T', #Threonine
'ACT'=>'T', #Threonine
'TGG'=>'W', #Tryptophan
'TAC'=>'Y', #Tyrosine
'TAT'=>'Y', #Tyrosine
'GTA'=>'V', #Valine
'GTC'=>'V', #Valine
'GTG'=>'V', #Valine
'GTT'=>'V', #Valine
'TAA'=>'-', #STOP
'TAG'=>'-', #STOP
'TGA'=>'-', #STOP
);
```

```
$seq="AAAAAAATTAATAGATGAACATATATATAGATTTTCTATATAGACCCTCTACCCGATAAGGCTAC";
$seq2=$seq;
$seq2=~tr/ATCG/TAGC/;
$seq2=reverse($seq2);

for($i=0;$i<=length($seq)-3;$i++)
{
    $x=substr($seq,$i,3);

    if ($x  eq 'ATG')
    {
            for($j=$i;$j<=length($seq)-3;$j=$j+3)
            {
                $y=substr($seq,$j,3);
                $k=$genetic_code{$y};
                if($k eq '-')
                {

                print"\n";
                last;
                    }

                print "$k";

            }
    }
}
```

```
for($l=0;$l<=length($seq2)-3;$l++)
{
     $m=substr($seq2,$l,3);

     if ($m  eq 'ATG')
     {
              for($n=$l;$n<=length($seq2)-3;$n=$n+3)
              {
                   $o=substr($seq2,$n,3);
                   $p=$genetic_code{$o};
                   if($p eq '-')
                   {

                   print"\n";
                   last;
                       }

                   print "$p";

              }
     }
}
```

Uniprot to FASTA

In this practical you will have to write a simple Perl script to convert the Uniprot Format to Fasta Format

```
$/="\/\/\n";
while (<>)
{
    if ($_=~/^AC\s{3}(.*?)\;/m)
    {
        print ">$1\n";
    }
    while ($_=~/^      (.*)/mg)
    {
        $line=$1;
        $line=~s/\s//g;
        print $line;
    }
    print "\n";
}
$/="\n";
```

Shuffle sequences

In this practical you will have to write a Perl script to shuffle sequences within a protein dataset

```
while(<>)
{
 $c=0;
 if ($_=~/^>/)
 {
     push @id, $_;
     $seq=<>;
     push @seq, $seq;
     push @c, $c;
     $c++;
 }
}
for ($x=$#c; $x>=0; $x--){
 $rnd=int(rand($x));
 #print $rnd;
 print $id[$rnd].$seq[$rnd];
 splice(@id,$rnd,1);
 splice(@seq,$rnd,1);
 splice(@c,$rnd,1);
}
```

Random Sequences

In this practical you will have to write a Perl script to generate random protein sequences in Fasta format

```
@BASES = ( 'A', 'T', 'C', 'G','D', 'E', 'F','H', 'I','K', 'L', 'M',
'N', 'P', 'Q', 'R','V' ,'W', 'Y', 'S' );

for ( $i=0; $i<500; $i++ ) {
  print '>Random', "$i\n";
  for( $j=0; $j<200; $j++ ) {
    $r = $BASES[ int (rand 20)];
    print $r;
    print "\n" if ($j+1)%60 == 0 and $j;
  }
  print "\n";
}
```

Amino acid Composition

In this practical you will have to write a Perl script to calculate the amino acid composition of protein sequences.

```
$arxeio_sequence = $ARGV[0];
open IN, $arxeio_sequence;
while (<IN>)
{
    if ($_=~/^>/)
    {
        $id=$_;
        chomp $id;
        print $id."\t";
        $seq=<IN>;
        chomp $seq;
    }
        $counter=0;
        @split_seq = split(//, $seq);
        foreach $a(@split_seq)
        { $counter++; }

    print $counter." AA\n";
    @amino_acids = (A,C,D,E,F,G,H,I,K,L,M,N,P,Q,R,S,T,V,W,Y);

    foreach $z(@amino_acids)
        {
        $count = $seq =~s/$z//g;
        $diairesi = $count/$counter;
        $pososto=sprintf( "%.3f", $diairesi );
        }
    print "\n";
}

close IN;
```

Lipoprotein signals

- The purpose of this practical is to find the presence of lipoprotein signal peptide in bacterial proteins using regular expressions
- You will have to download the 63 sequences from Gram-negative Bacteria
- In the first place you will have to use the scripts that you wrote in the previous practicals and convert the sequences to Fasta format.
- For making the subsequently calculations easier you could choose a single-line fasta format.

Lipoprotein signals

- Afterwards, you should write a simple program to test the existence of the lipo-box.
- The regular expression patterns that could be used are:

- LA[GA]C
- [LVI][ASTG][GA]C
- [^DERK]{6}[LIVMFWSTAG]{2}[LIVMFYSTAGCQ][AGS]C
- [MV].{0,13}[RK][^DERK]{6,20}[LIVMFESTAG][LVIAM][IVMSTAFG][AG]C

References
•Juncker AS, Willenbrock H, Heijne GV, et al. **Prediction of lipoprotein signal peptides in Gram-negative bacteria**. 2003;12:1652-1662. [PDF]
•Sutcliffe IC, Harrington DJ. **Pattern searches for the identification of putative lipoprotein genes in Gram-positive bacterial genomes**. *Microbiology (Reading, England)*. 2002;148(Pt 7):2065-77. [PDF]
•Bagos PG, Tsirigos KD, Liakopoulos TD, Hamodrakas SJ. **Prediction of lipoprotein signal peptides in Gram-positive bacteria with Hidden a Markov Model**, 2008, *J Proteome Research*, 7(12):5082-93 [PDF] [Pubmed] [Google Scholar]

Lipoprotein signals

- Check which of the proposed patterns performs better
- Afterwards, you should remove the mature part of the protein and keep only the sequence of the signal peptide (including the Cysteine) and write a program for aligning the sequences to the right.
- For instance if there were two sequences with lengths of signal peptide equal to 25 and 30 respectively, the former should have five gaps (-) preceeding the initial Methionine.
- This special form of a multiple alignment should be used for performing analyses of the aminoacid frequencies in the lipobox and an easy way to perform such an analysis is using the WebLogo sever (http://weblogo.berkeley.edu/)

LA[GA]C

```
while (<>){
   if    ($_=~/^>(.*)/)
{
        $name=$1;
        $seq=<>;

        if($seq=~/(.*LA[GA]C)/)
        {
            $x=length($1);

            print "$name\t LIPOPROTEIN \t $x\t $1\n";

            $a=$a+1;     }
     else
     {
         print "$name\t NO LIPOPROTEIN\n";      }
}
}
print "$a LIPOPROTEINS FOUND";
```

[LVI][ASTG][GA]C

```
while (<>){
   if    ($_=~/^>(.*)/)
{
        $name=$1;
        $seq=<>;

        if($seq=~/(.*[LVI][ASTG][GA]C)/)
        {
            $x=length($1);

            print "$1\n";
            $a=$a+1;
        }
     else
     {
          print "$name\t NO LIPOPROTEIN\n";      }
}
}
print "$a LIPOPROTEINS FOUND";
```

[^DERK]{6}[LIVMFWSTAG]{2}[LIVMFYSTAGCQ][AGS]C

```
while (<>){
   if    ($_=~/^>(.*)/)
{
        $name=$1;
        $seq=<>;

        if($seq=~/(.*[^DERK]{6}[LIVMFWSTAG]{2}[LIVMFYSTAGCQ][AGS]C)/)
{
            $x=length($1);

         print "$name\t LIPOPROTEIN \t $x\t $1\n";
         $a=$a+1; #YPOLOGIZEI POSES LIPOPROTEINES VRISKEI
        }
     else
     {
          print "$name\t NO LIPOPROTEIN\n";
     }
}
}
print "$a LIPOPROTEINS FOUND";
```

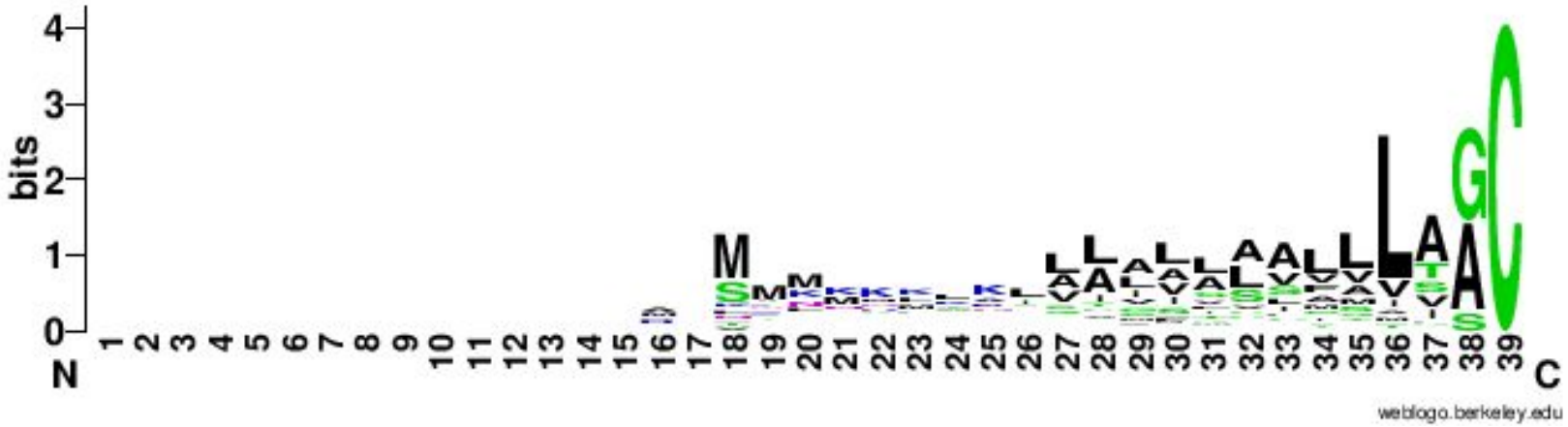
[MV].{0,13}[RK][^DERK]{6,20}[LIVMFESTAG][LVIAM][IVMSTAFG][AG]C

```
while (<>){
if   ($_=~/^>(.*)/)
{
        $name=$1;
        $seq=<>;
            if($seq=~/^([MV].{0,13}[RK][^DERK]{6,20}[LIVMFESTAG][LVIAM]
[IVMSTAFG][AG]C)/)
{
            $x=length($1);

            print "$name\t LIPOPROTEIN \t $x\t $1\n";           $a=$a+1;
}
else
{
print "$name\t NO LIPOPROTEIN\n";
}

}
}
print "$a LIPOPROTEINS FOUND";
```

```
open out,(">>Out.txt");
while (<>){
  if   ($_=~/^>(.*)/)
  {
     $name=$1;
        $seq=<>;

        if($seq=~/^([MV].{0,13}[RK][^DERK]{6,20}[LIVMFESTAG][LVIAM][IVMSTAFG][AG]C)/)
        {
           $x=length($1);

        }

       if ($max<$x)
       {

              $max=$x;
       }

  }
}
continue {
    if (eof) {    #an eisai sth teleutaia  grammi tou arxeiou
        seek ARGV, 0, 0; #gurizei to filehandler sthn 1h grammh tou arxeiou mas
        last;
    }
}
while (<>){
  if   ($_=~/^>(.*)/)
  {
     $name=$1;
        $seq=<>;
        if($seq=~/^([MV].{0,13}[RK][^DERK]{6,20}[LIVMFESTAG][LVIAM][IVMSTAFG][AG]C)/)
        {
           $x=length($1);

              #print out ">$name\n ";
              for($i=0; $i<=$max-$x; $i++)
              {
                     print out "-";
              }
              print out "$1\n";
           $a=$a+1;
       }
}
}
```

```
------------------MRRCMPLVAASVAALMLAGC
-----------------MKLKQLFAITAIASALVLTGC
------------------MKLLSKIMIIALAASMLQAC
--------------MNKNRGFTPLAVVLMLSGSLALTGC
-------------------MKRQALAAMIASLFALAAC
--------------------MRLLPLVAAATAAFLVVAC
-----------------------MRIVIFILGILLTSC
---------------------MFKRFIFITLSLLVFAC
---------------------MLKKVYYFLIFLFIVAC
---------------------MKKILLTVSLGLALSAC
-----------------MVKKAIVTAMAVISLFTLMGC
-----------------MKQLIVNSVATVALASLVAGC
-------------------MKLKTLALSLLAAGVLAGC
--------------------MKAYLALISAAVIGLAAC
--------------------MKLKATLTLAAATLVLAAC
------------------MQKTPKKLTALCHQQSTASC
----------------MPLPDFRLIRLLPLAALVLTAC
----------------MKNQVKKILGMSVVAAMVIVGC
---------------------MKKFLPLSISITVLAAC
-----------------MKRLFLSFVALALLAGSIAAC
---------------------MCGKILLILFFIMTLSAC
---------------------MSKRLLSLASLALLFGC
-------------------MFKRRYVTLLPLFVLLAAC
-------------------MKKIIKLSLLSLSIAGLASC
-----------------MGRSKIVLGAVVLASALLAGC
-----------------MKAKIVLGAVILASGLLAGC
-----------------MNNVLKFSALALAAVLATGC
--------------MKLTTHHLRTGAALLLAGILLAGC
-------------MAYSVQKSRLAKVAGVSLVLLLAAC
------------MSAGSPKFTVRRIAALSLVSLWLAGC
```

```
-MDKGEGLRLAATLRQWTRLYGGCHLLLGAVVCSLLAAC
-------------------MKPFLRWCFVATALTLAGC
-----------------MNIATKLMASLVASVVLTAC
----------------MQNAKLMLTCLAFAGLAALAGC
----------------------MKKYLLGIGLILALIAC
----------------------MRLLIGFALALALIGC
--------------MFVTSKKMTAAVLAITLAMSLSAC
-----------------MNKNMAGILSAAAVLTMLAGC
-----------------MHVSSLKVVLFGVCCLSLAAC
----------------MYKNGFFKNYLSLFLIFLVIAC
-------------------MNKFVKSLLVAGSVAALAAC
-------------------MKKTNMALALLVAFSVTGC
----------------MSLTHYSGLAAAVSMSLILTAC
-------------------MLRYTRNALVLGSLVLLSGC
---------------------MRNFILFPMMAVVLLSGC
---------------------MRKQWLGICIAAGMLAAC
-------------------MRYLATLLLSLAVLITAGC
-------------------MNMTKGALILSLSFLLAAC
----------------MNKKIFTLFLVVAASAIFAVSC
---------------------MVKRGRFALCLAVLLGAC
-----------------MKVKYALLSAGALQLLVVGC
-----------------MNNPLVNQAAMVLPVFLLSAC
----------------MNAHTLVYSGVALACAAMLGSC
----------------MKLKSLVFSLSALFLVLGFTGC
-----------------MREKWVRAFAGVFCAMLLIGC
---------------MKHNVKLMAMTAVLSSVLVLSGC
---------------------MKLRLSALALGTTLLVGC
-----------MRKRISAIINKLNISIIIMTVVLMIGC
-------------------MRKRISAIIMTLFMVLVSC
-----------MRKRISAIINKLNISIMMMIVVLMIGC
```


bits
4
3
2
1
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C
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39
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